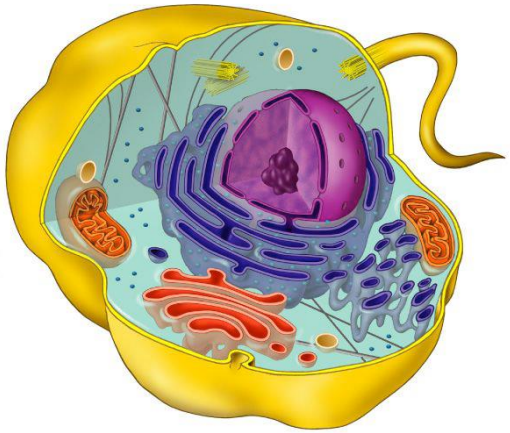
Cytology 2

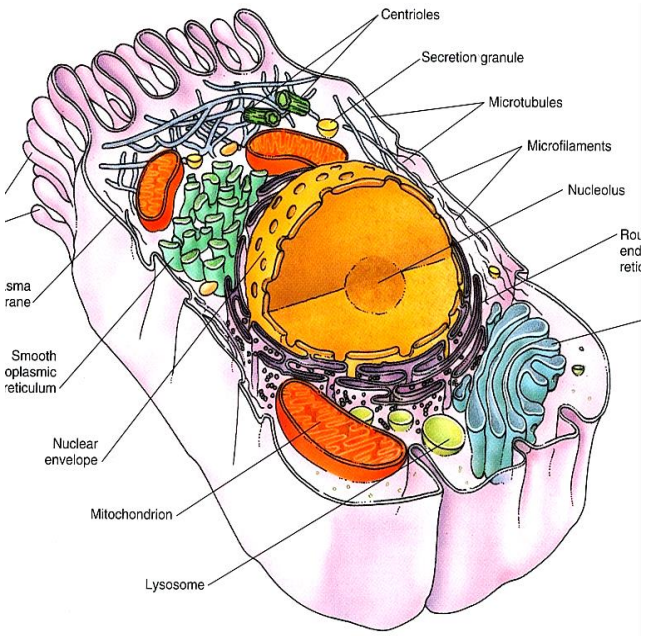
Aleš Hampl

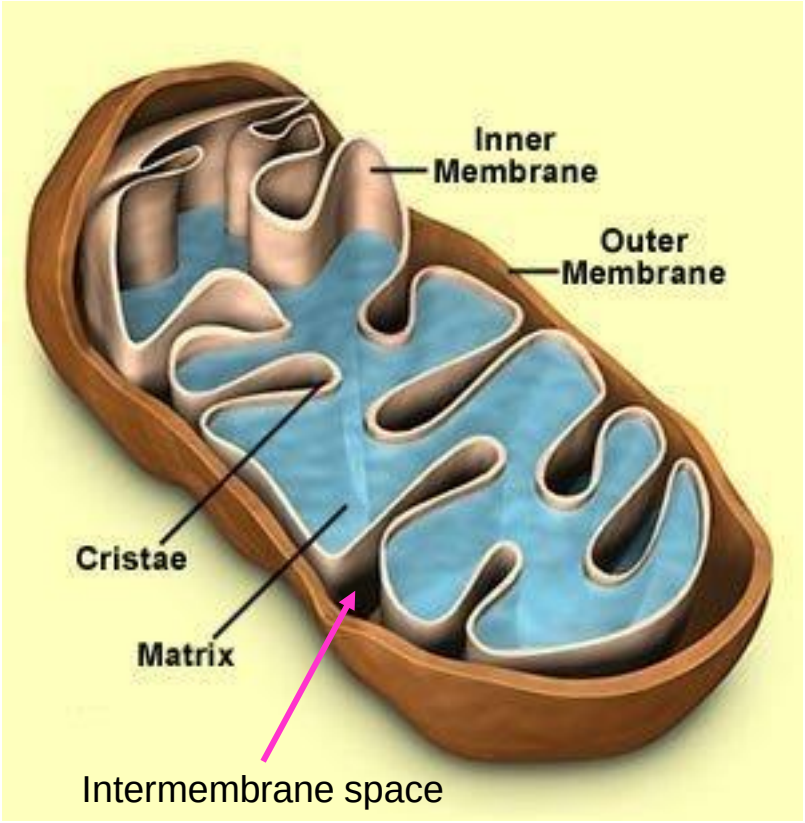
2022



- Mitochondria
- Lyzosomes + Peroxisomes
- Cytoplasmic inclusions
- Cytoskeleton
- Cell surface specialisations
- Cell cycle, cell division, cell differentiaion

Mitochondria 1

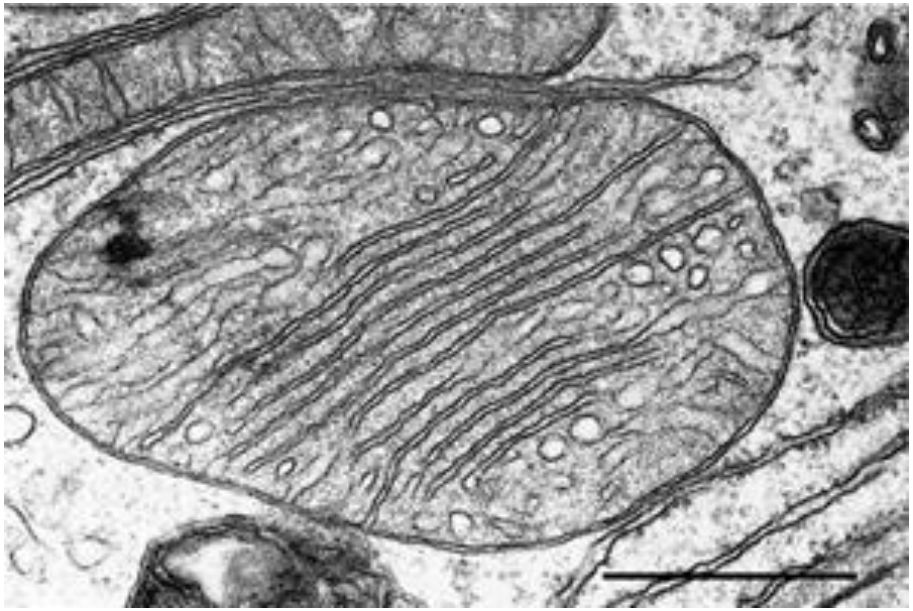




- all cells except erythrocytes
- double membrane
- diameter cca 0,5 μm
- length up to 50 (100) μm
- oxidative metabolism (glucose – ATP + CO_2 + H_20)
- cytochrome c – activation of apoptotic pathway
- origin in oocyte
- mtDNA (circular)
- brown fat thermogenesis

- both membranes with low fluidity
- both membranes equipped with many protein molecules
- growth and divission of mitochondria

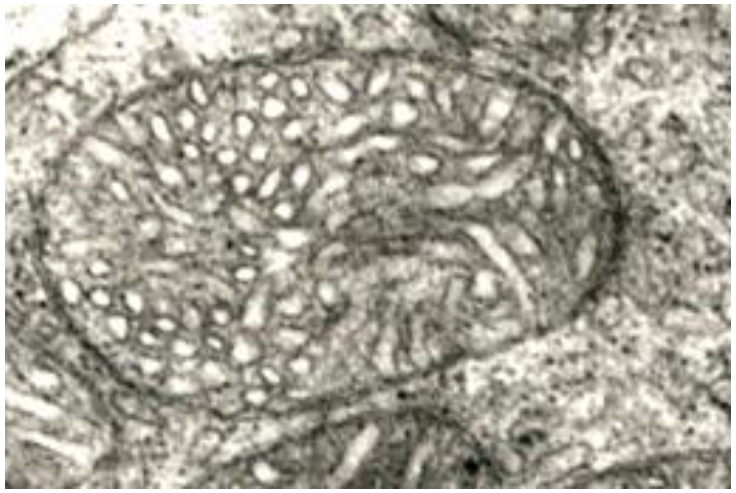
Mitochondria 2



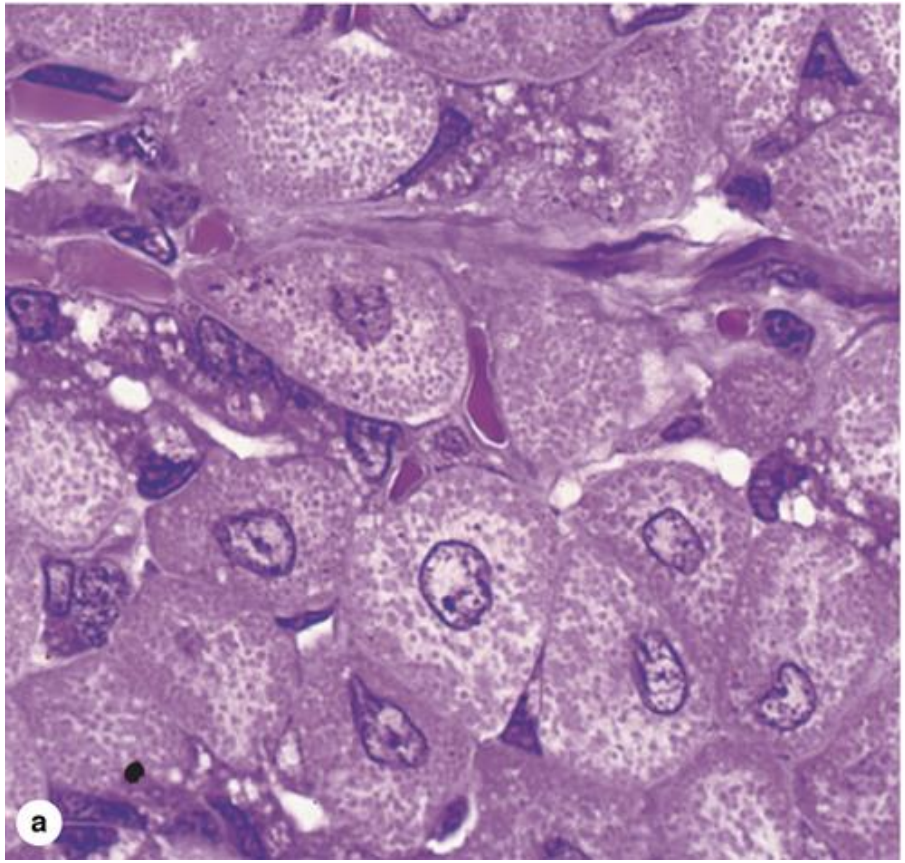
Mitochondria 3

with crists

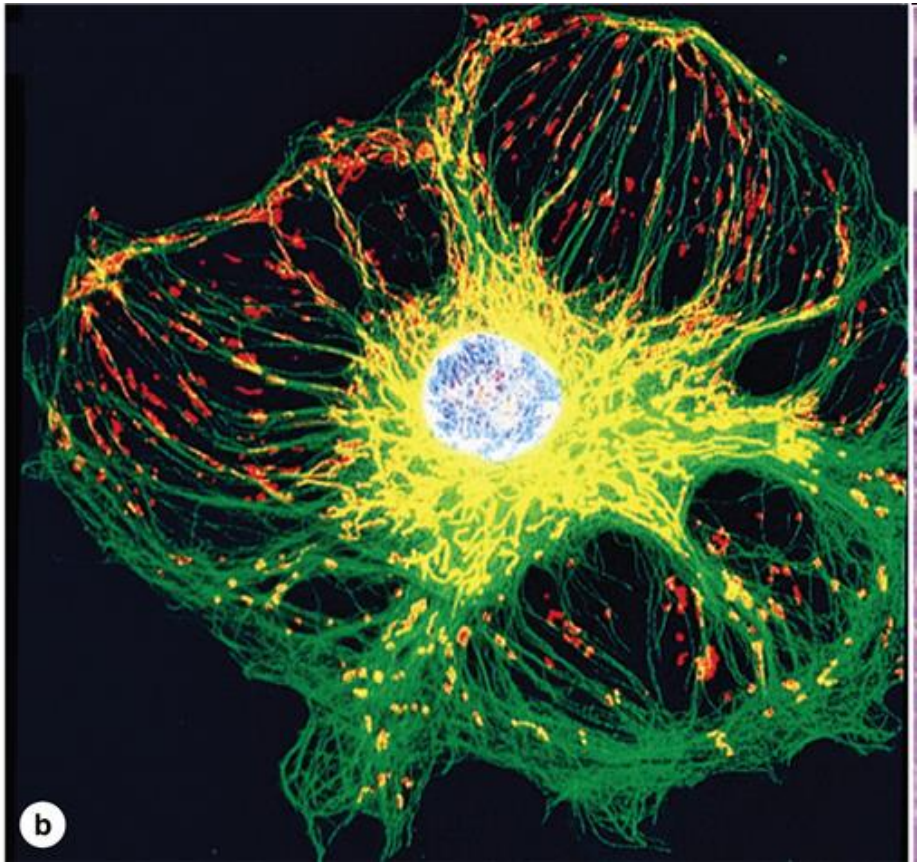
with tubuli (in steroid producing cells)



Mitochondria 4



mitochondrial eosinophilia



mitochondria
microtubuli

Lysosomes 1

endosome-lysosome system

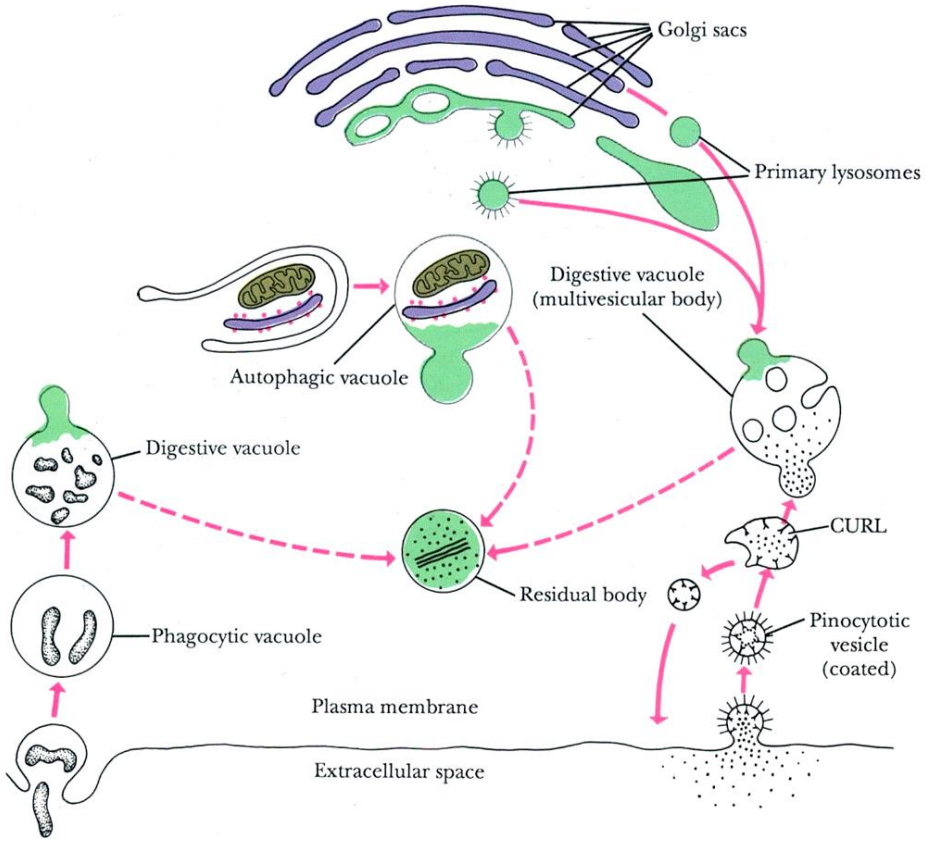


Figure 2.17. Origins of primarily lysosomes from the Golgi and trans-Golgi network. Primary lysosomes fuse with and discharge hydrolytic enzymes into autophagic, pinocytotic (or endosome), and phagocytic vacuoles to form secondary lysosomes (digestive vacuoles). Residual bodies contain undigested residue. Endosomes fuse to form a compartment where uncoupling of the ligands and surface receptors occurs (CURL, see text for explanation). The compartment containing the free ligands subsequently fuses with the lysosome; the receptors remain bound to the membrane of vesicles which is partitioned off from the CURL and recycle to the plasma membrane. (Modified from Novikoff AB, Holtzman E: *Cells and Organelles,* 2nd ed. New York, Holt, Rinehart and Winston, 1976.)

- in all cells except for erythrocytes
- vesicles about 0,05 – 0,5 μm
- membrane-bound
- highly acidic internal space (cca pH 5)
- hydrolytic enzymes inside (min. 50 types)
- tagging by mannose-6-fosphate

Lysosomes 2

primary x secondary

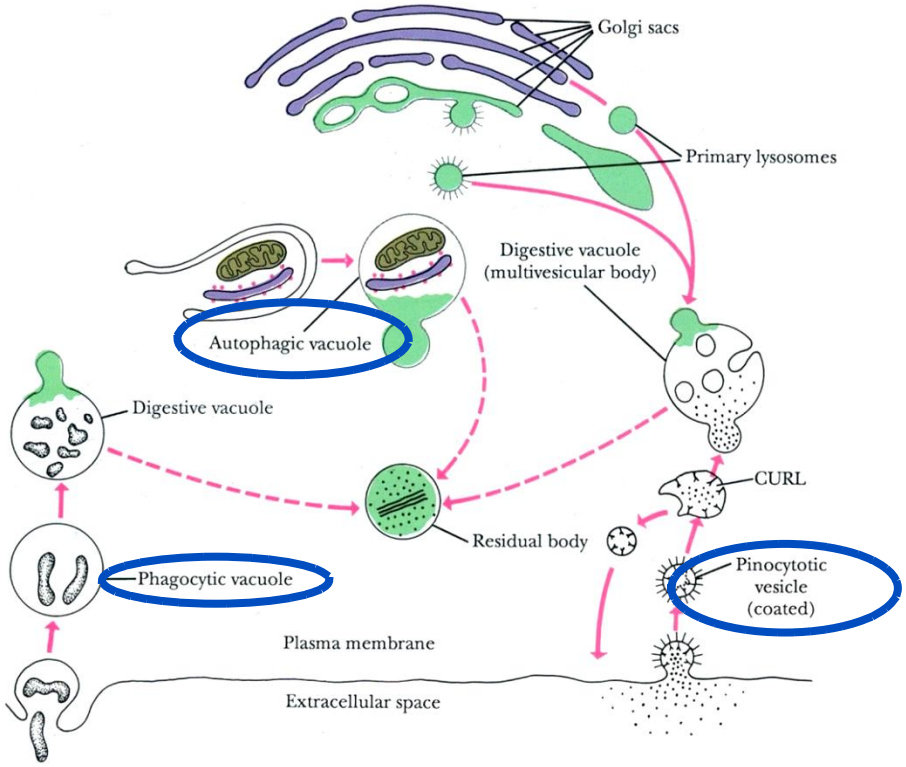
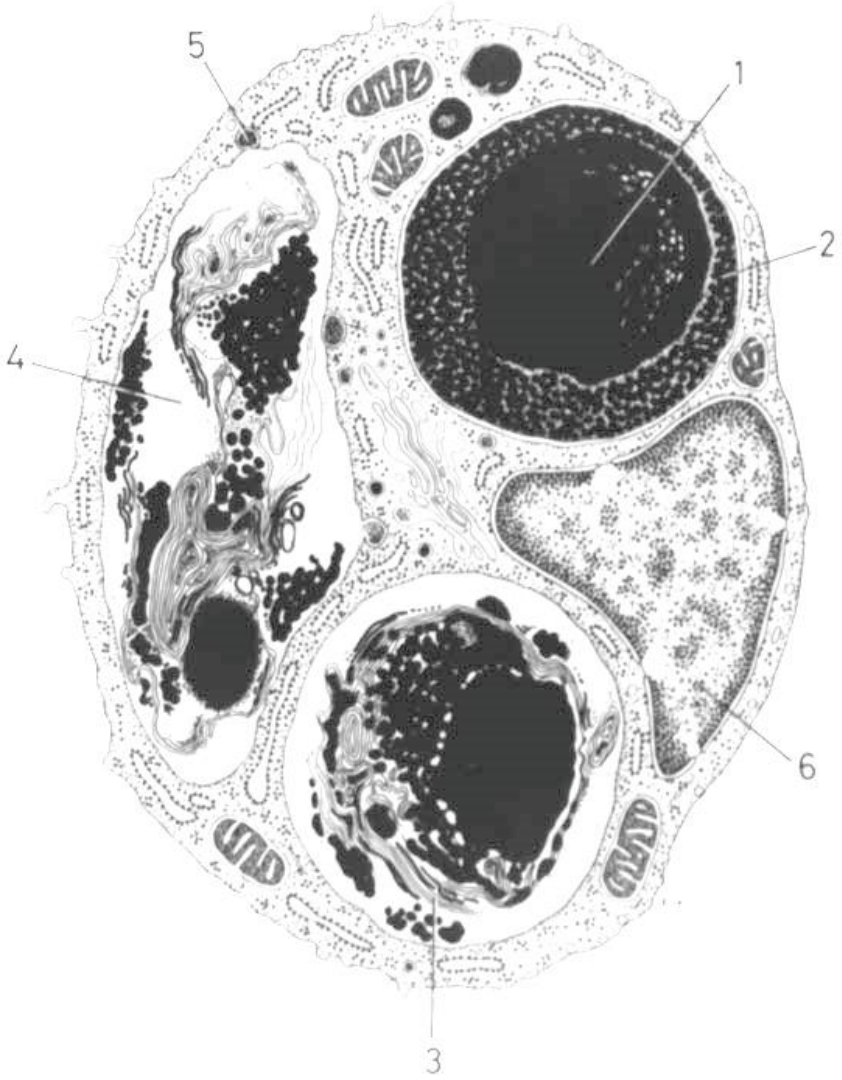


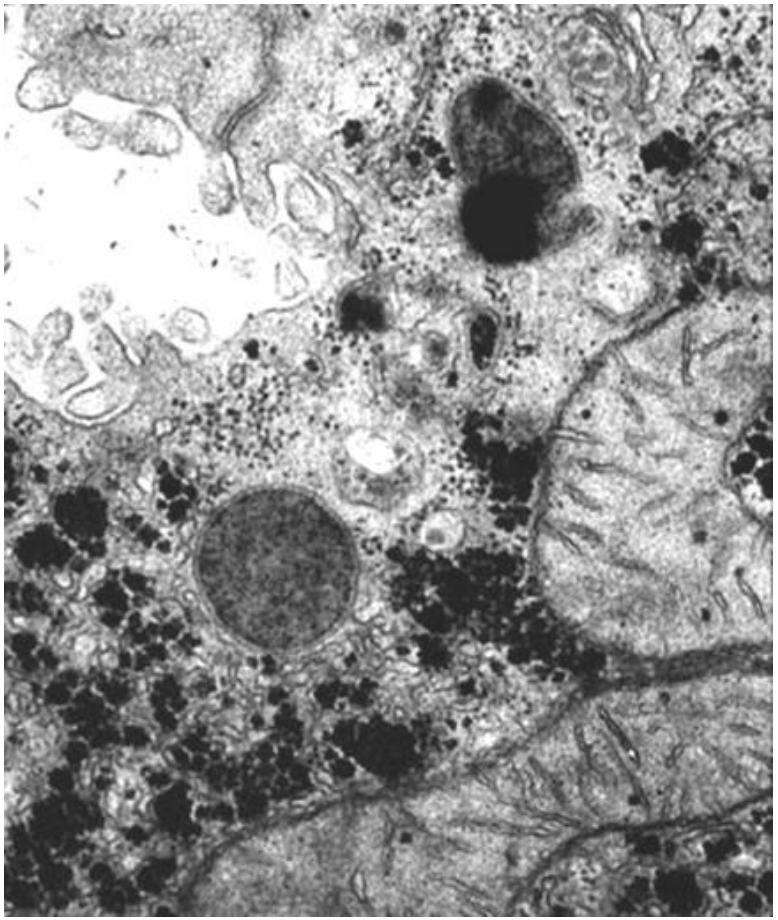
Figure 2.17. Origins of primarily lysosomes from the Golgi and trans-Golgi network. Primary lysosomes fuse with and discharge hydrolytic enzymes into autophagic, pinocytotic (or endosome), and phagocytic vacuoles to form secondary lysosomes (digestive vacuoles). Residual bodies contain undigested residue. Endosomes fuse to form a compartment where uncoupling of the ligands and surface receptors occurs (CURL, see text for explanation). The compartment containing the free ligands subsequently fuses with the lysosome; the receptors remain bound to the membrane of vesicles which is partitioned off from the CURL and recycle to the plasma membrane. (Modified from Novikoff AB, Holtzman E: *Cells and Organelles,* 2nd ed. New York, Holt, Rinehart and Winston, 1976.)

- primary lysosomes
- secondary lysosomes (fagolysosomes)
- residual bodies (lipofuscin)

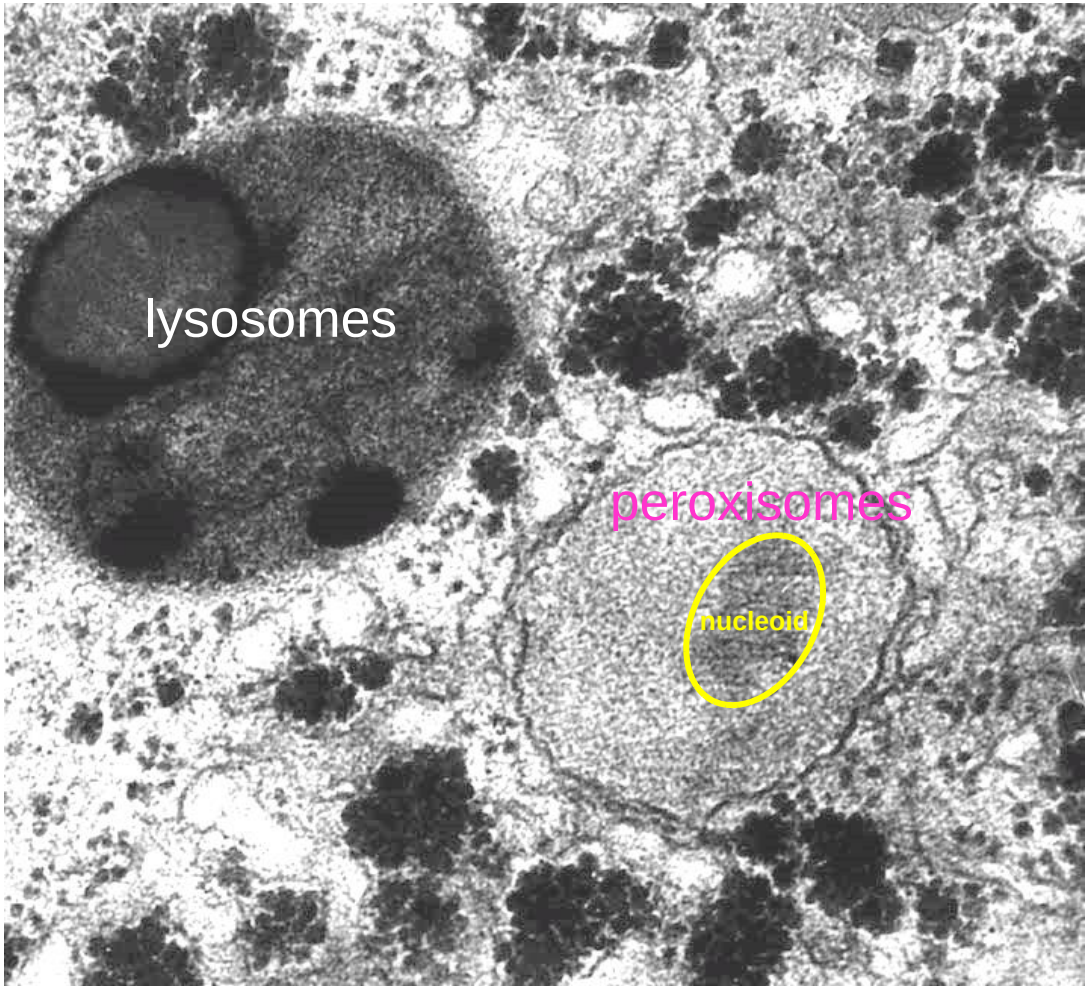
Lysosomes 3

secondary lysosomes





Peroxisomes



- structuraly similar to lysosoms
- functionally similar to mitochondria
- „nucleus “ = nucleoid
- degradation of fatty acids (H_20_2, H_2O, 0_2)
- detoxification (complement SER)
- origin: growth from ER or division

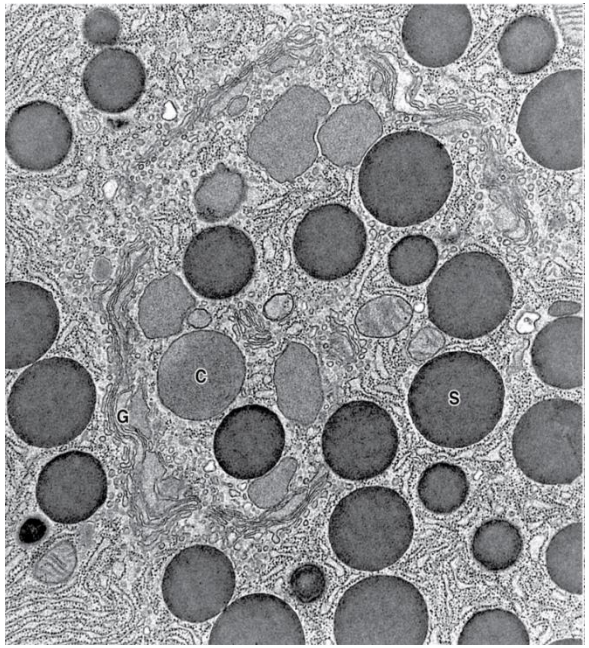
Cytoplasmic inclusions 1

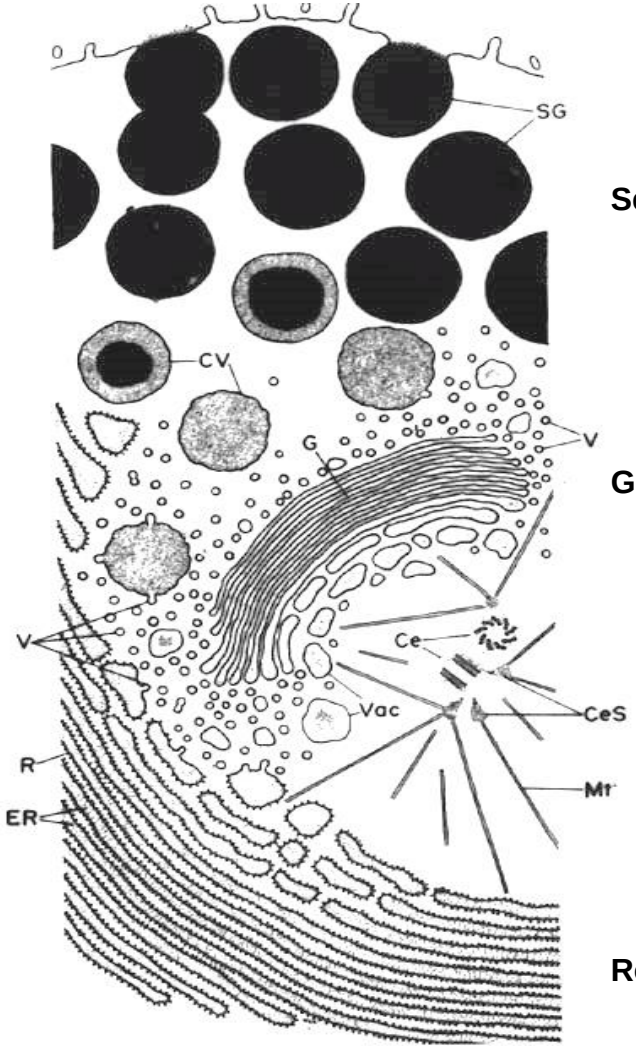
(no or only little metabolic activity on themselves)

- **secretory granules**
- **storage compounds**: sugars (glycogen), lipids
- **crystals** (proteins)
- **pigments**: endogenous (autogenic and hematogenic) + exogenous

Cytoplasmic inclusions 2

Secretory granules





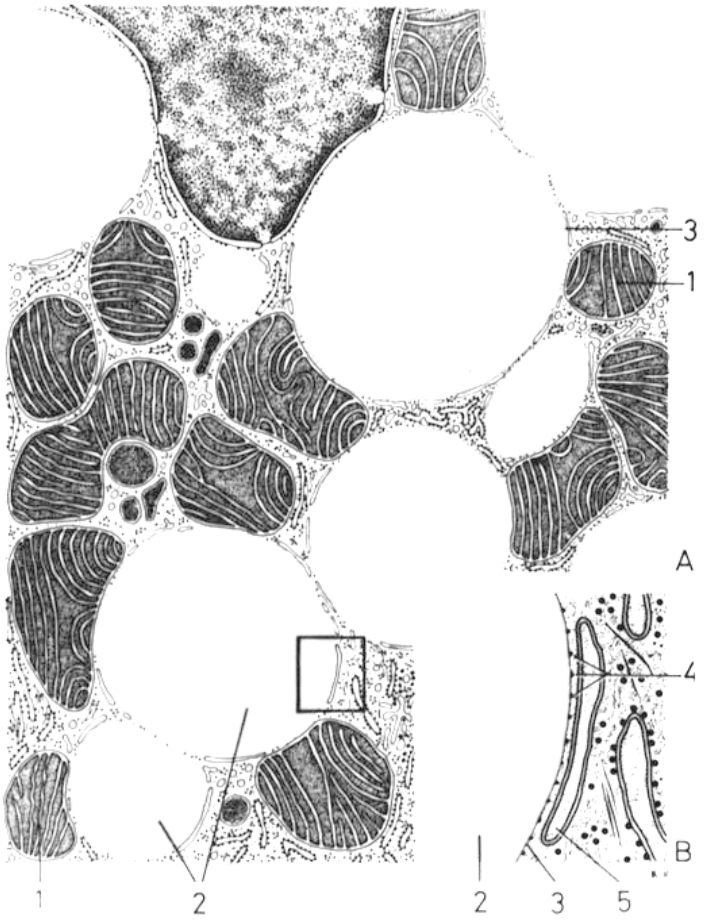
Secretory granules

Golgi apparatus

Rough ER

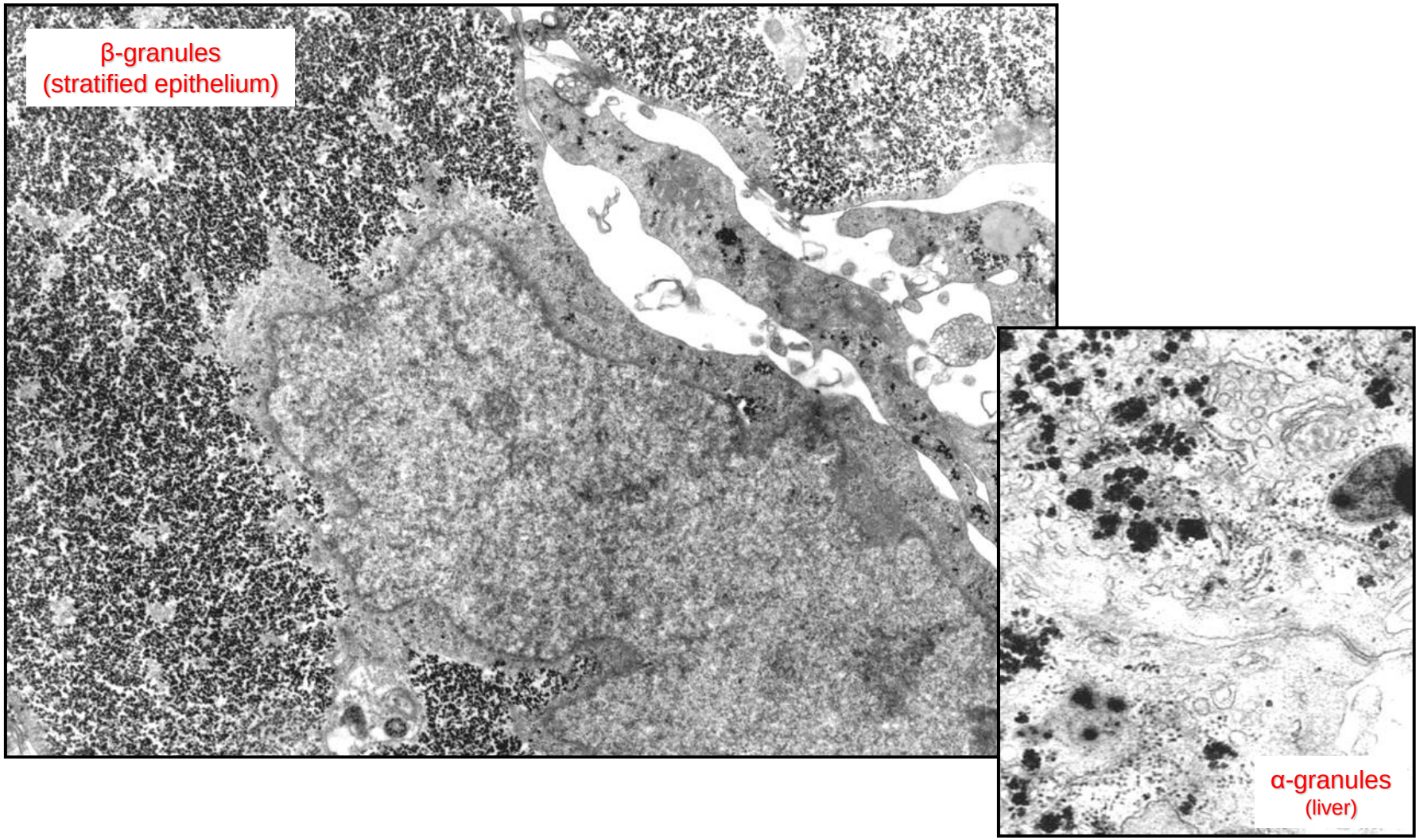
Cytoplasmic inclusions 3

Lipid inclusions



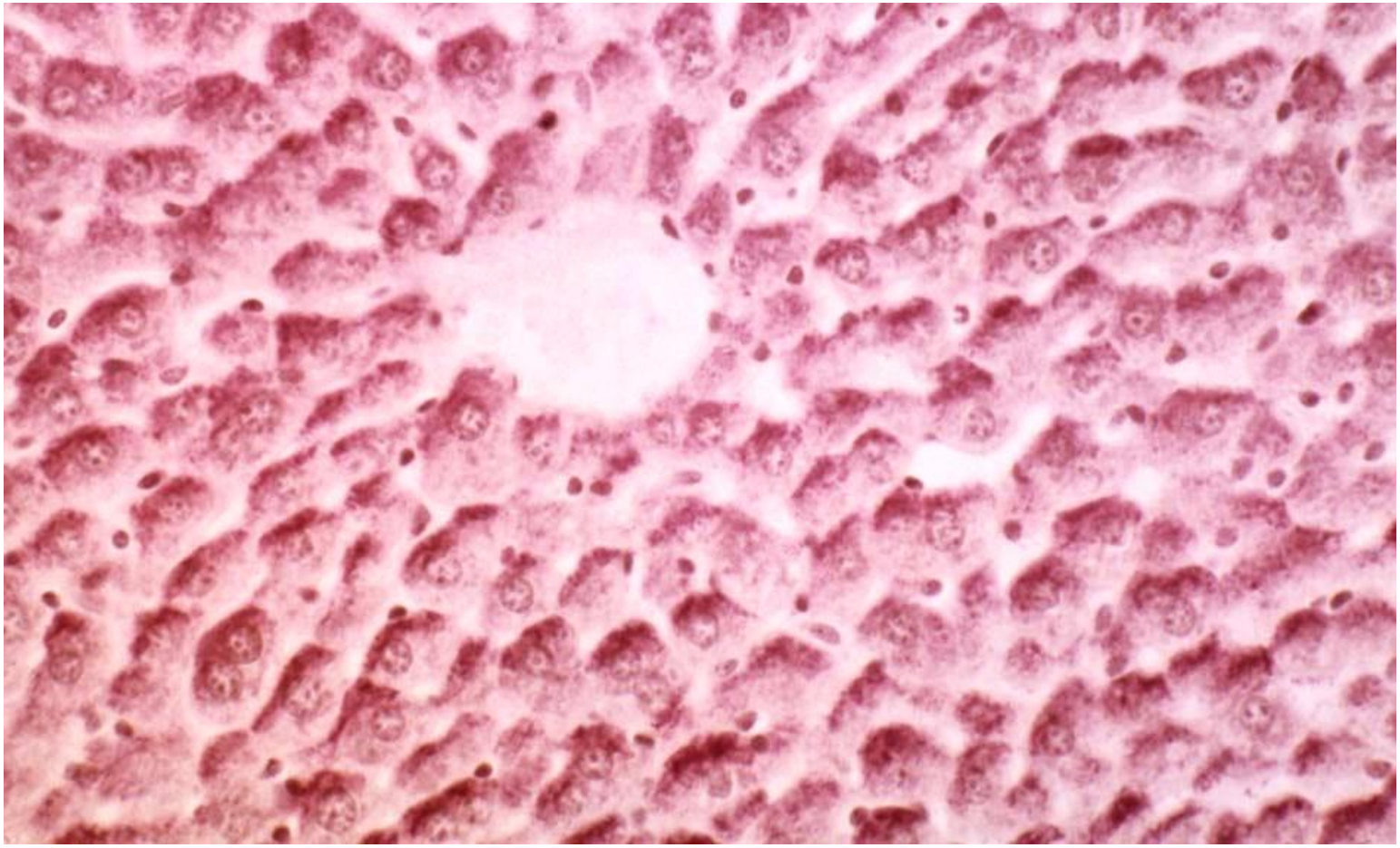
Cytoplasmic inclusions 4

Glycogen



Cytoplasmic inclusions 5

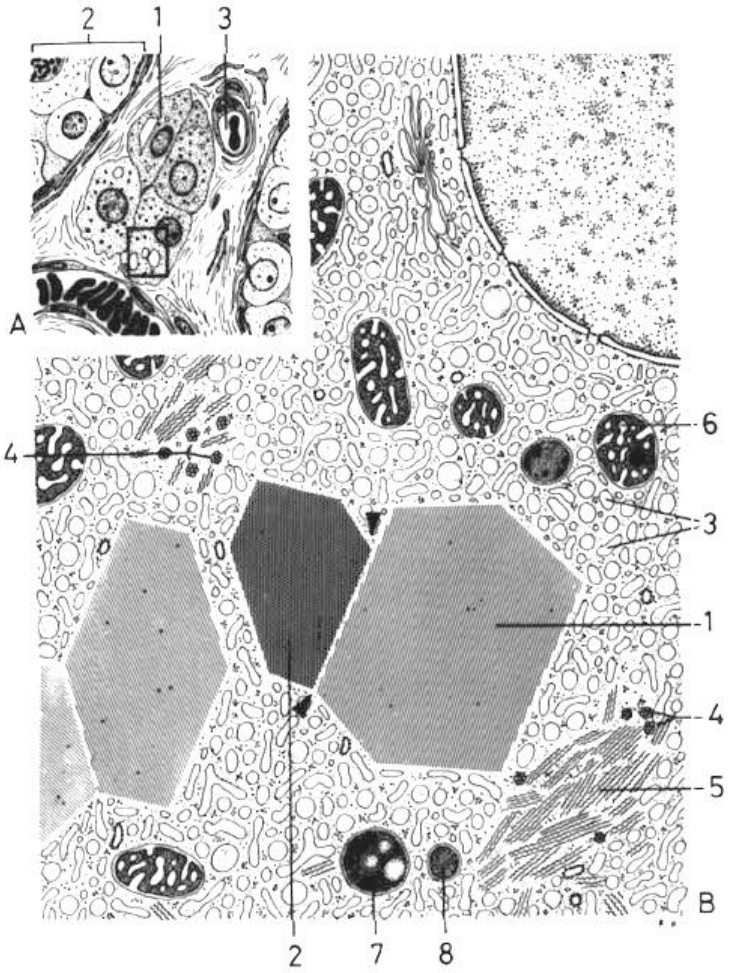
Glycogen

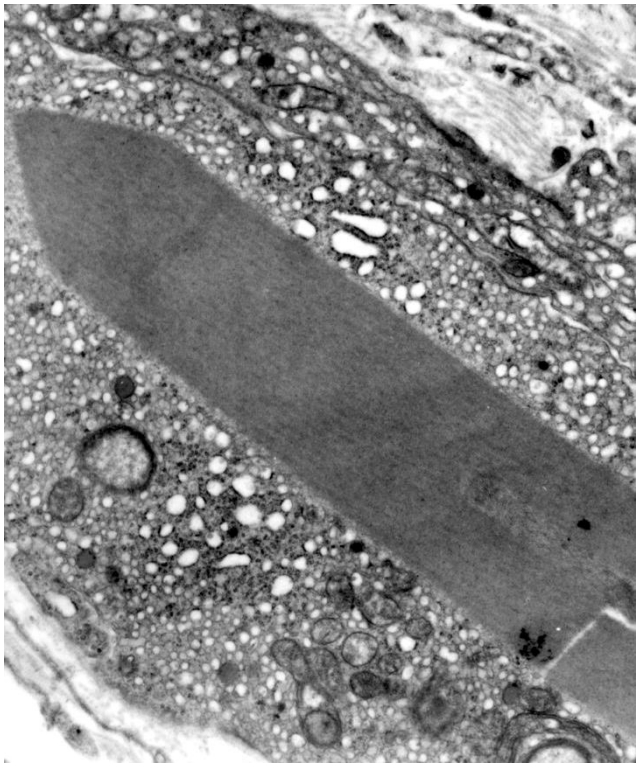


Glycogen in liver cells (light microscope; PAS reaction)

Cytoplasmic inclusions 6

Crystals



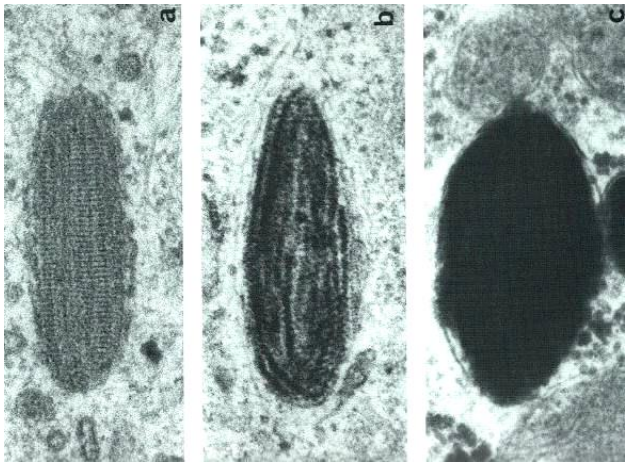


Protein inclusions in Leydig cells

Cytoplasmic inclusions 7

Pigments (colour inclusions): Exogenous x Endogenous

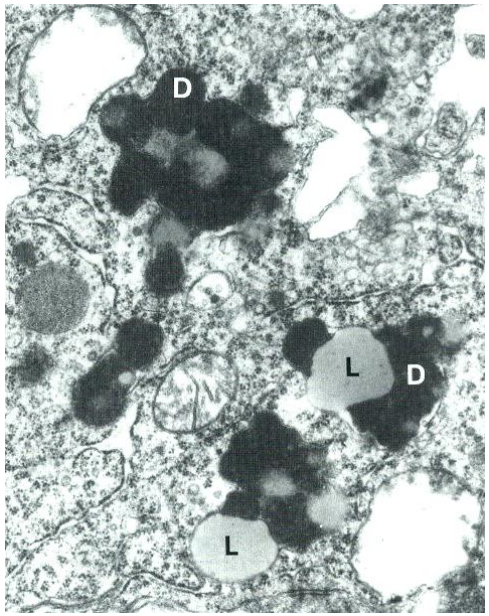
- **Autogenous**
  Specific functions – **melanin**



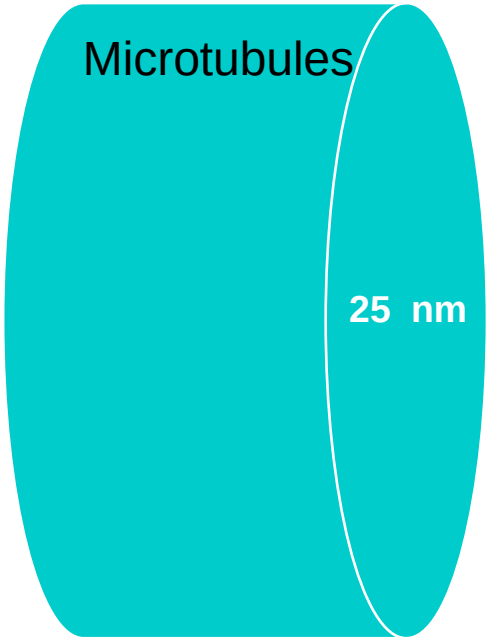
- **Hematogenous**
  Hemoglobin decomposition – **hemosiderin**, **biliverdin**, **bilirubin**

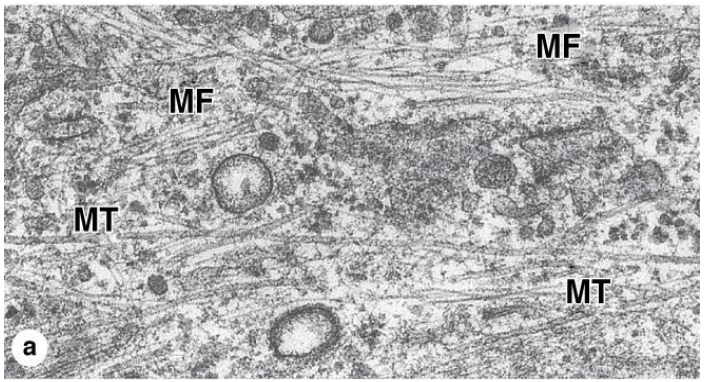
Pigment in aged cells

lipofuscin – accumulation of residual bodies in long-lived cells (neurones, kardiomyocytes)

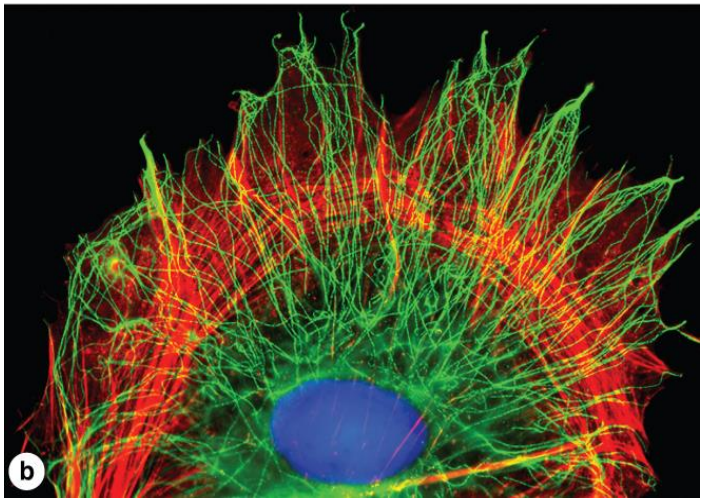


Cytoskeleton 1



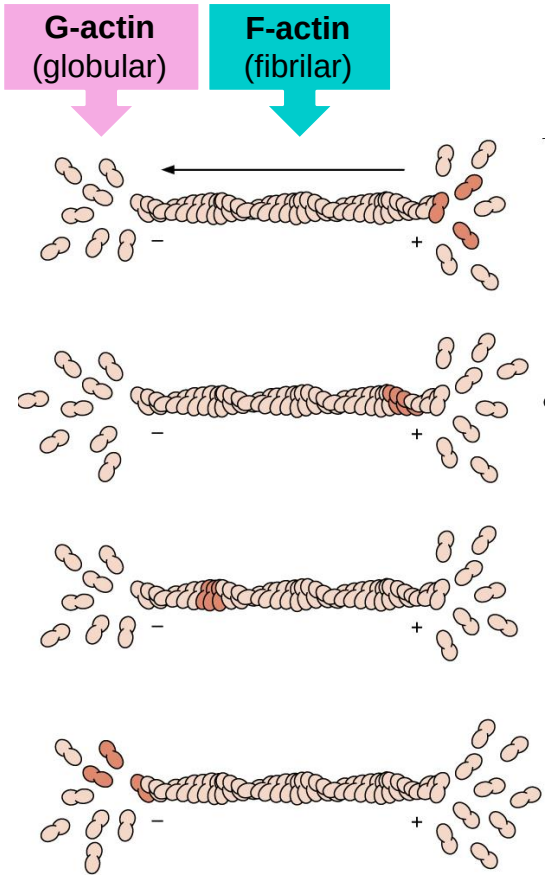


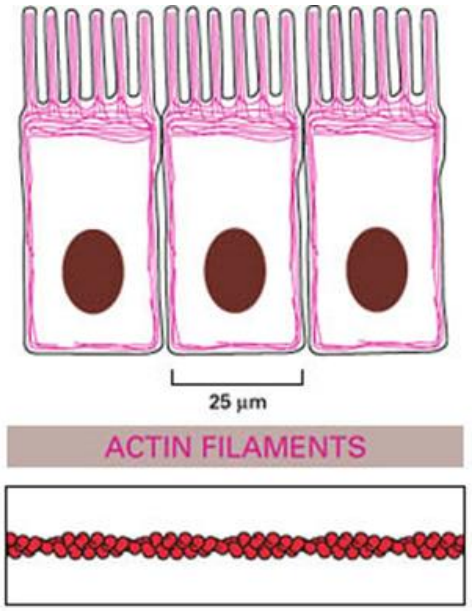
microtubules
microfilaments - actin



Cytoskeleton 2

Microfilaments (actin)

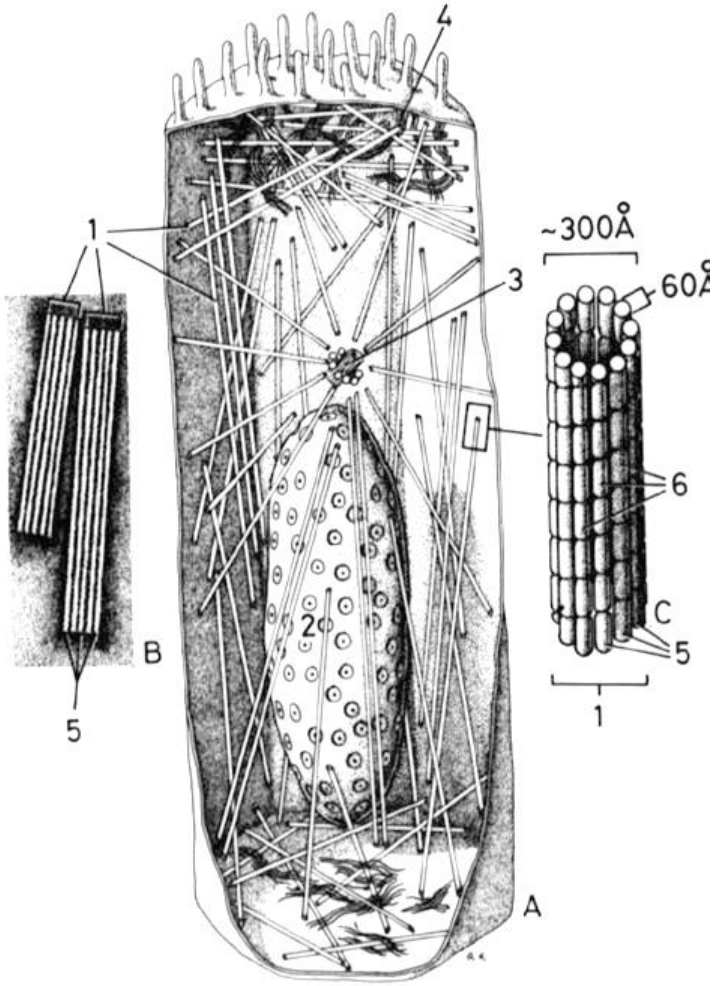


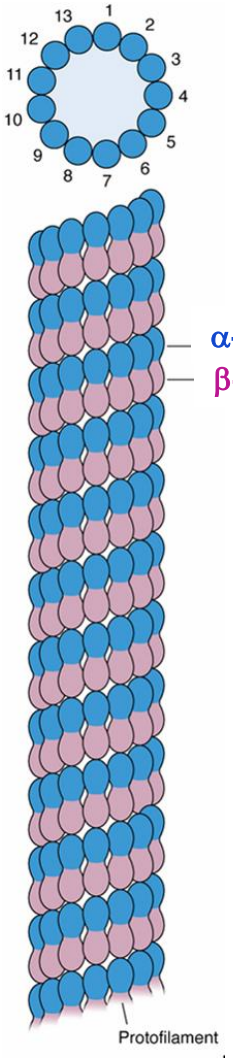


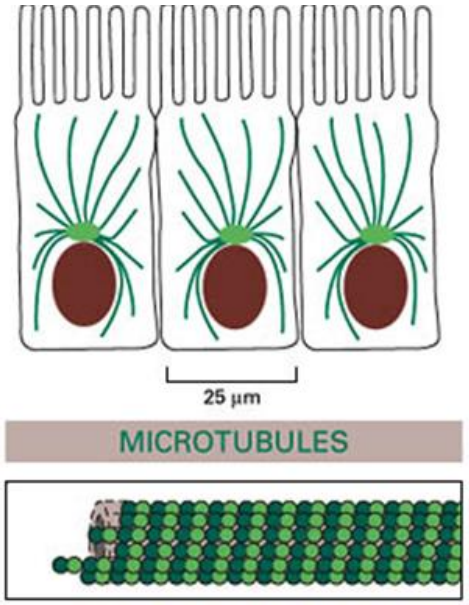
- actin isoformes (α, β, γ)
- fast polymerisation and depolymerisation
- polarisation (+ a – ends)
- stabilisation by associated proteins (tropomyosin – myofibrils)
- crosslinking by associated proteins (fimbrin, filamin, …)
- anchoring to cell membrane (vinculin, tallin, …)
- cortical actin – membrane skeleton
- myosin motors *(analogous to dynein + kinesin on microtubuli)*

Cytoskeleton 3

Microtubules



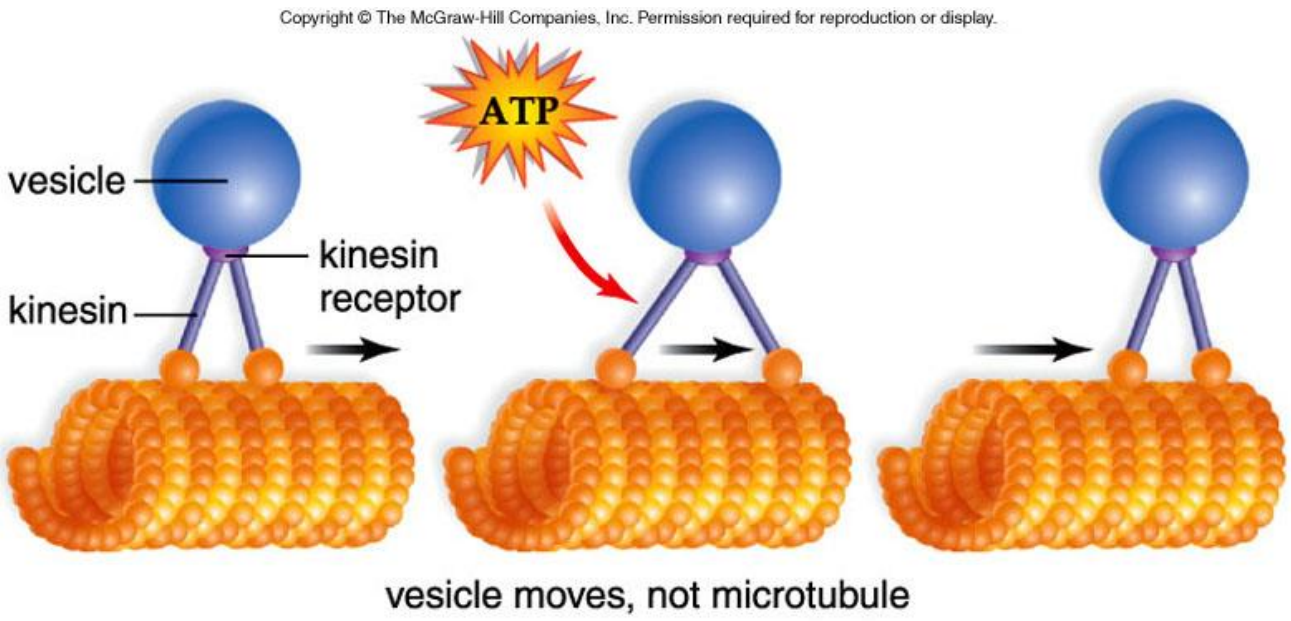




- hollow tubes
- α-tubulin + β-tubulin – dimers
- fast polymerisation and depolymerisation
- polarisation (+ a – ends)
- MAP (proteins associated with microtubuli)
- MTOC – microtubules organizing centre (centrosome; γ-tubulin)
- mechanical support
- intracellular transport
- mitotic spindle
- cilia and flagella
- mitotic poisons (colchicin, taxol, …)

Cytoskeleton 4

Microtubules - motors

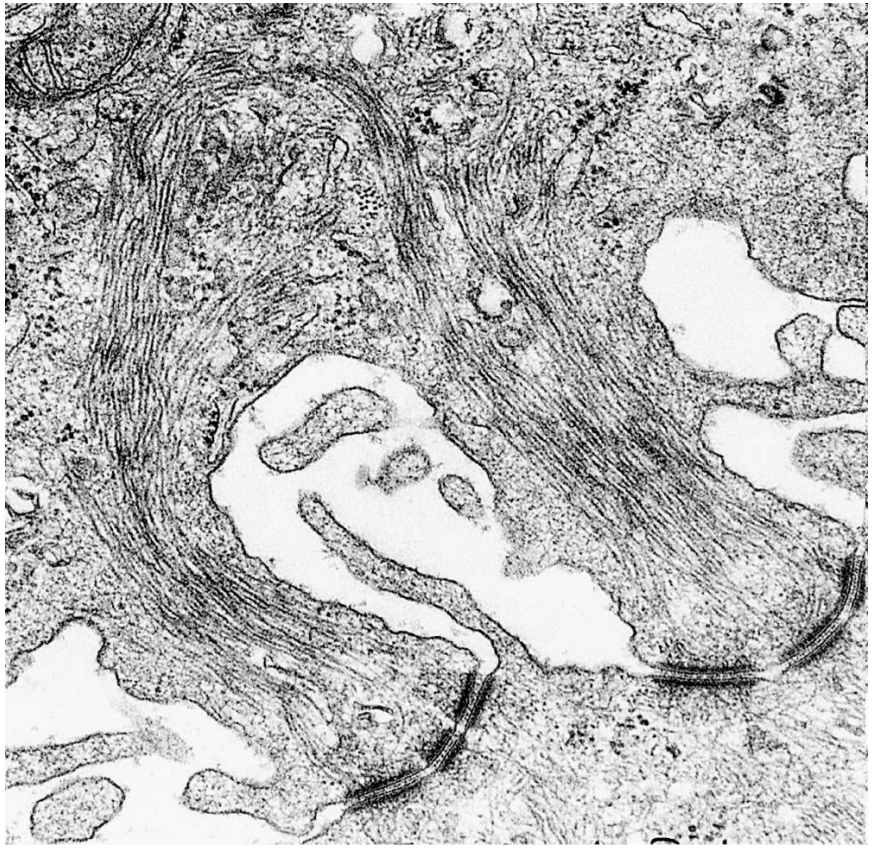


Kinesins
- move towards „plus“ end of microtubuli
- transport from centrosome

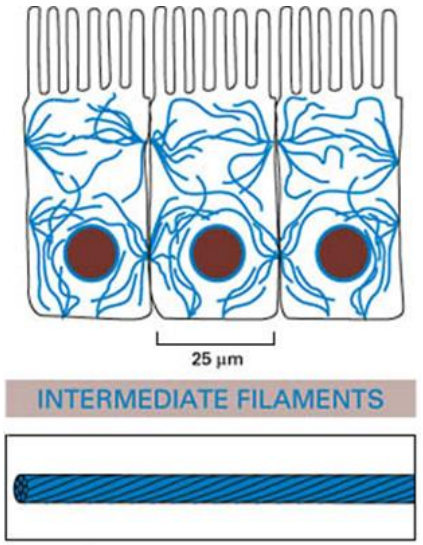
Dyneins
- move towards „minus“ end microtubuli
- transport towards centrosome
- axonal transport – long distance

Cytoskeleton 5

Intermediate filaments



Cytokeratin intermediate filaments in stratum basale of epidermis



- „chemically“ highly heterogenou group
- common composition (tetramers) “thread like“
- more stable than actin and tubulin structures
- cell type specific:

Cytokeratins (epithelia)

Vimentin (cells of mesenchymal origin)

Desmin (muscle cells)

Neurofilaments (neurons)

Glial fibrial acidic protein (neuroglia)

Lamins (nuclear envelope)

Cell surfaces 1

Free

- **microvilli** (*irregular*, *regular* – striated border, brush border)
- **cilia**

Lateral

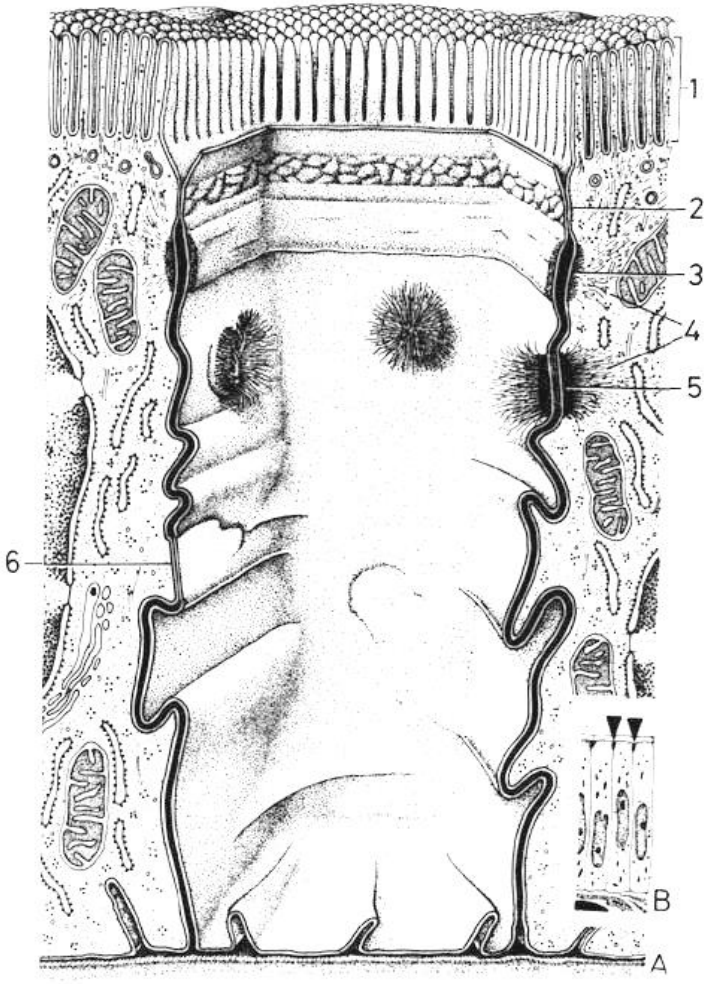
Cell-to-cell junction:

- *sealing:* tight junction=zonula occludens
- *adhesion:* zonula adherens, desmosom
- *communication:* nexus (Gap junction)

Basal

- focal adhesions
- hemidesmosomes
- basal labyrinth

free surface

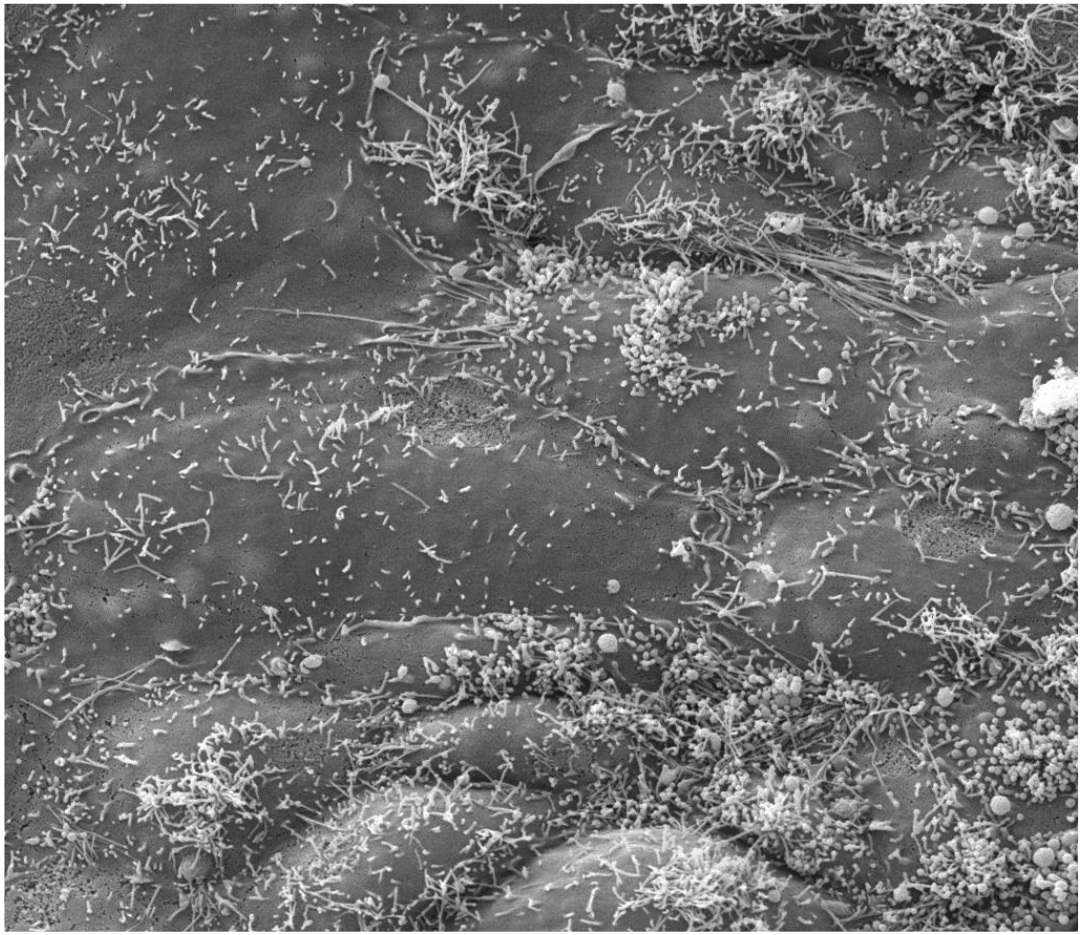


lateral surface

basal surface

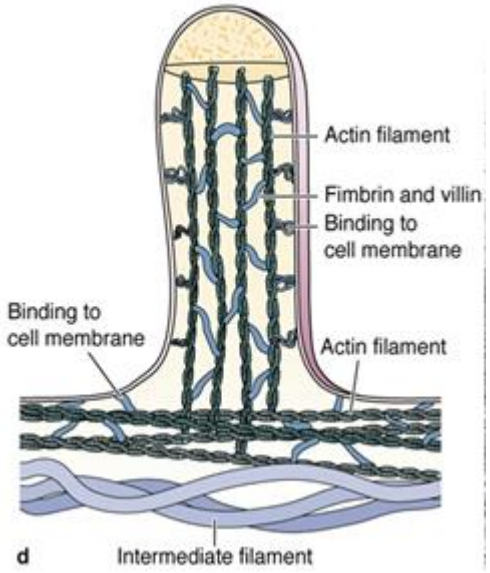
Cell surfaces 2

Microvilli



Free surface of cultured human embryonic stem cells

Cell surfaces 3



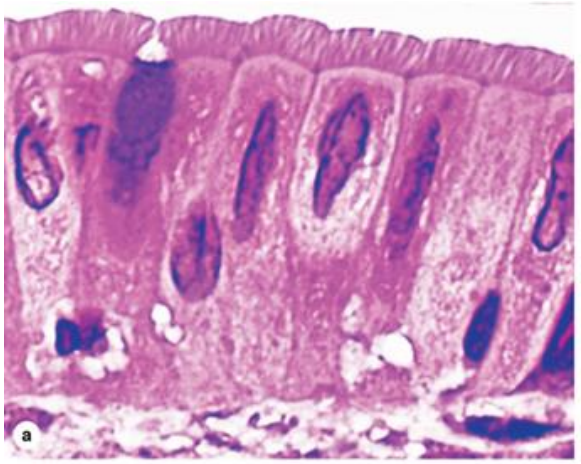
Microvilli

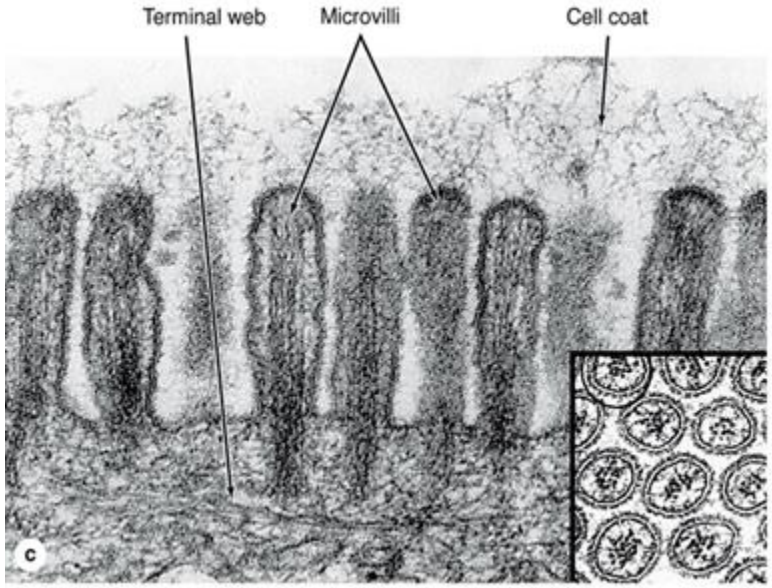
Thickness about 0,1 μm
Length about 1-6 μm

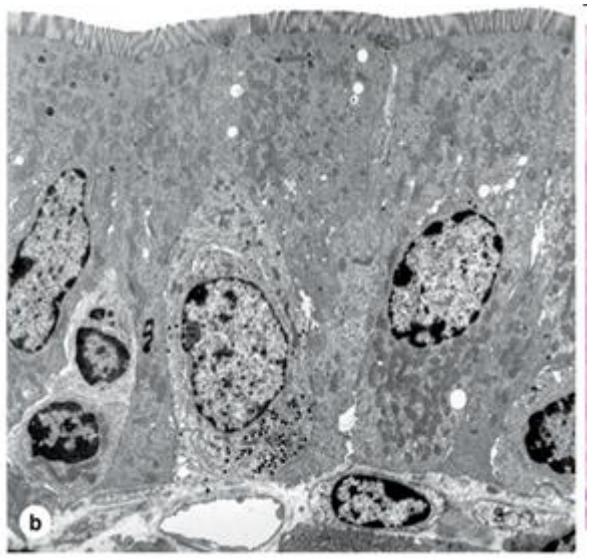
Actin filaments in microvilli

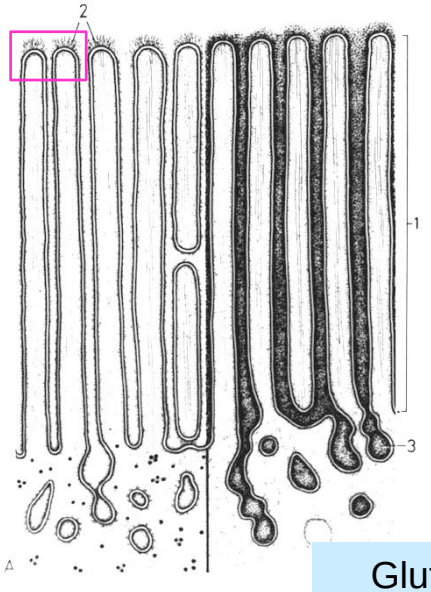
- 20 in microvilli of epithelial cells
- several hundreds in stereocilia of hair cells

Regularly organised microvilli
= striated border + brush border







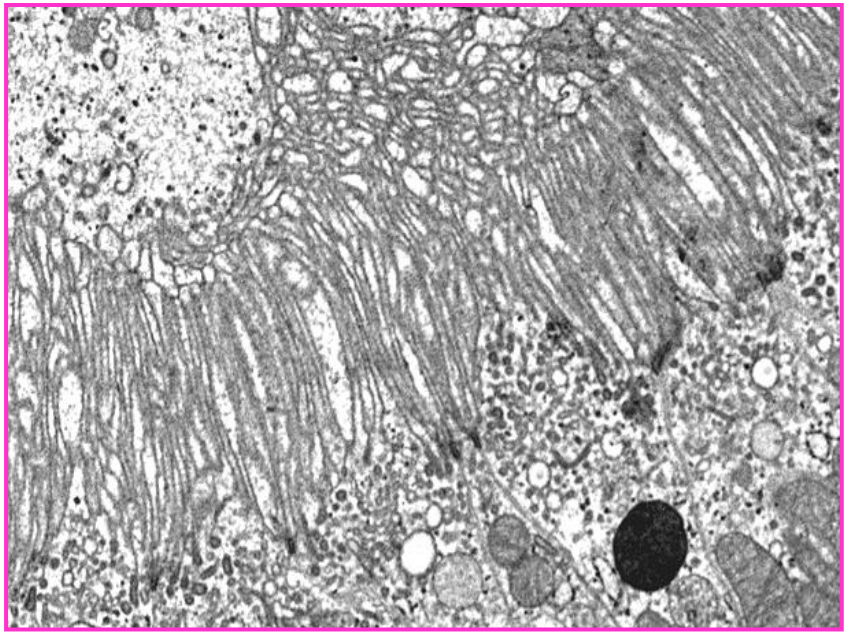


Cell surfaces 4

Microvilli

Gluten – Celiac disease

striated border
(tops of enterocytes)



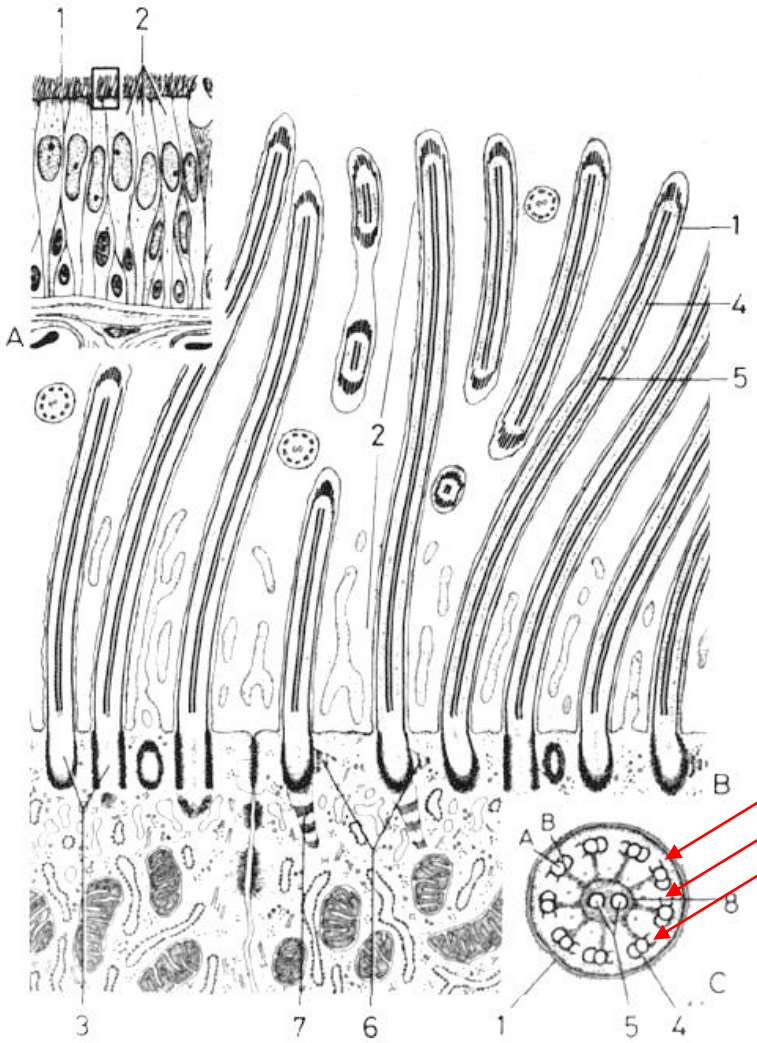
brush border
(proximal tibuli of kidney)

Cell surfaces 5

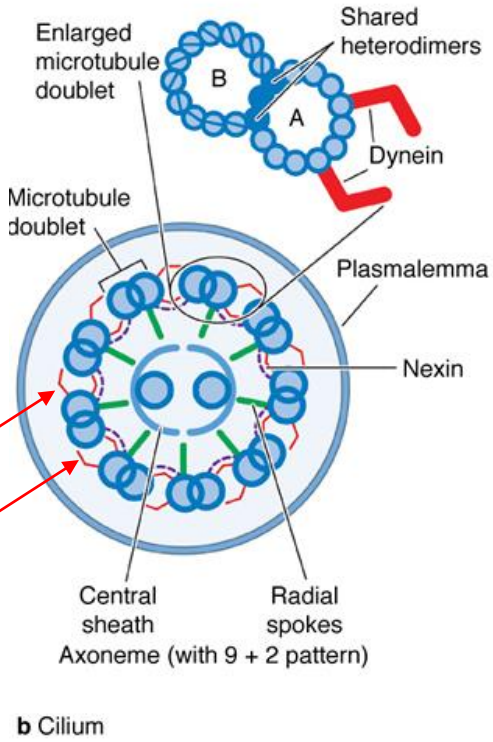
Cilia + Flagella

Thickness about 0,25 μm
Length about 7-10 μm

Axonema
20 microtubuli (9x2 + 2)



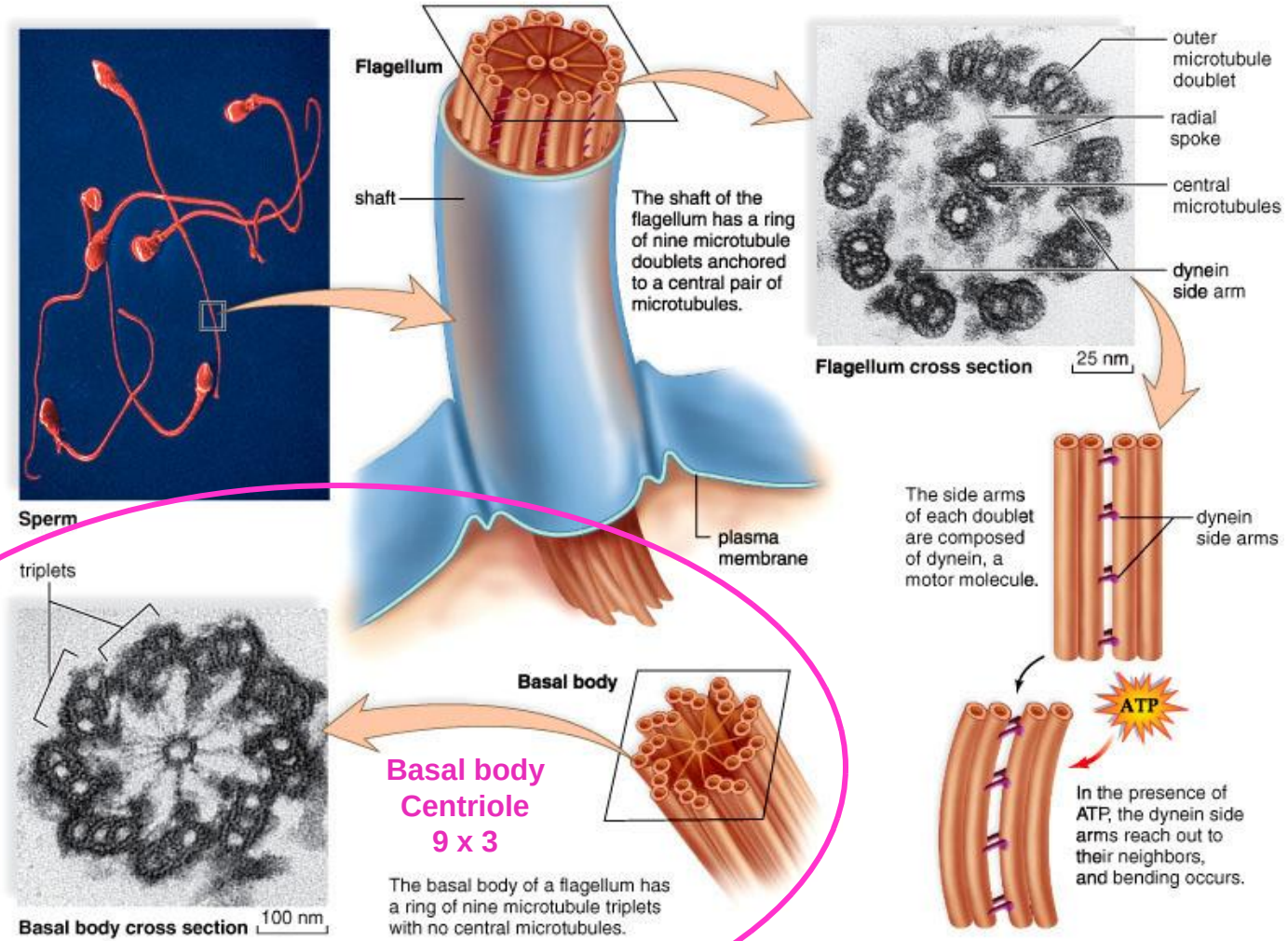
Dynein arms
(movement)



b Cilium

Cell surfaces 6

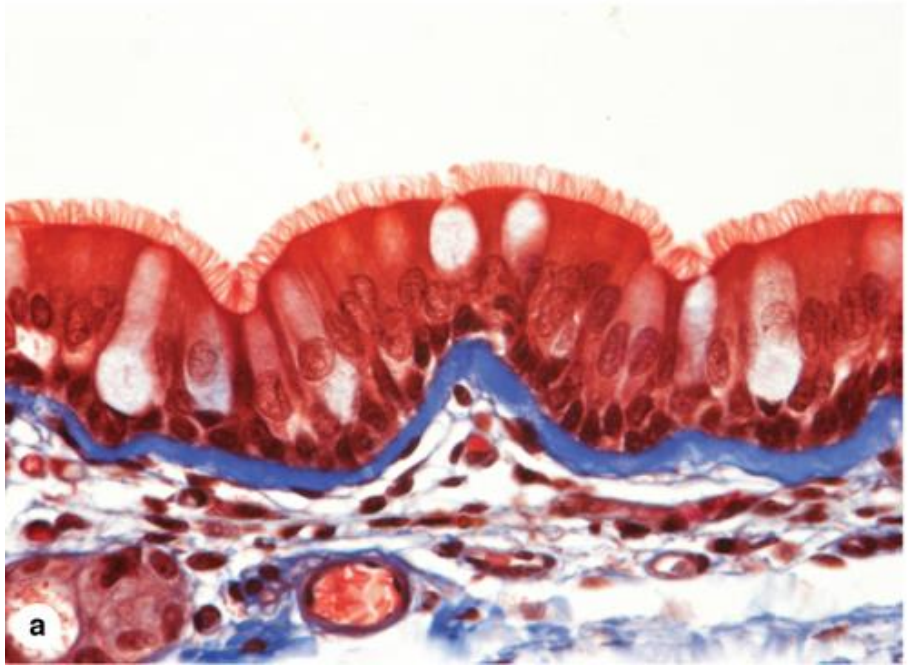
Cilia + Flagella



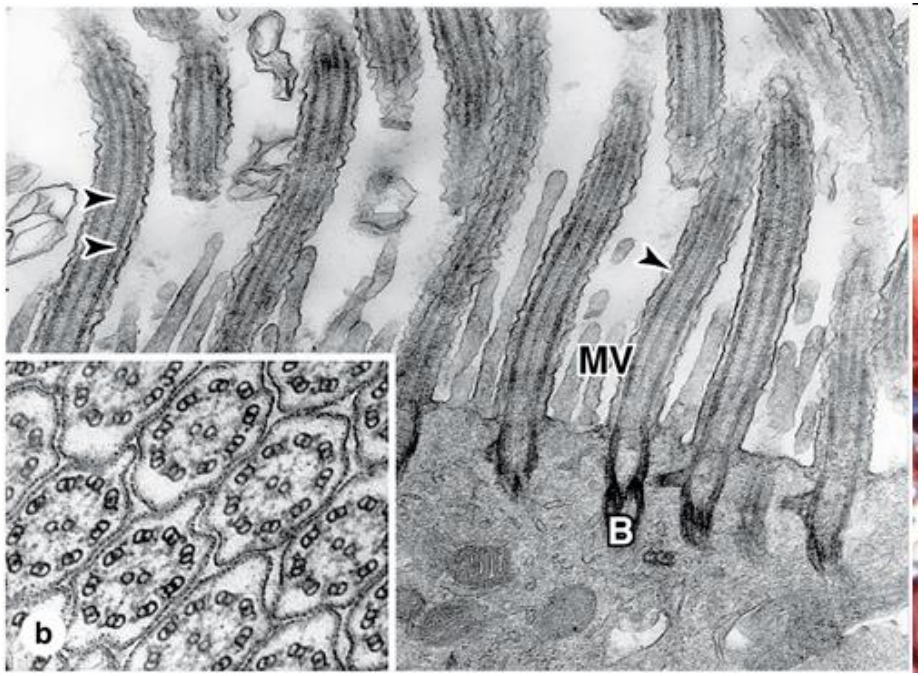
Cell surfaces 7

Cilia + Flagella

in light microscope

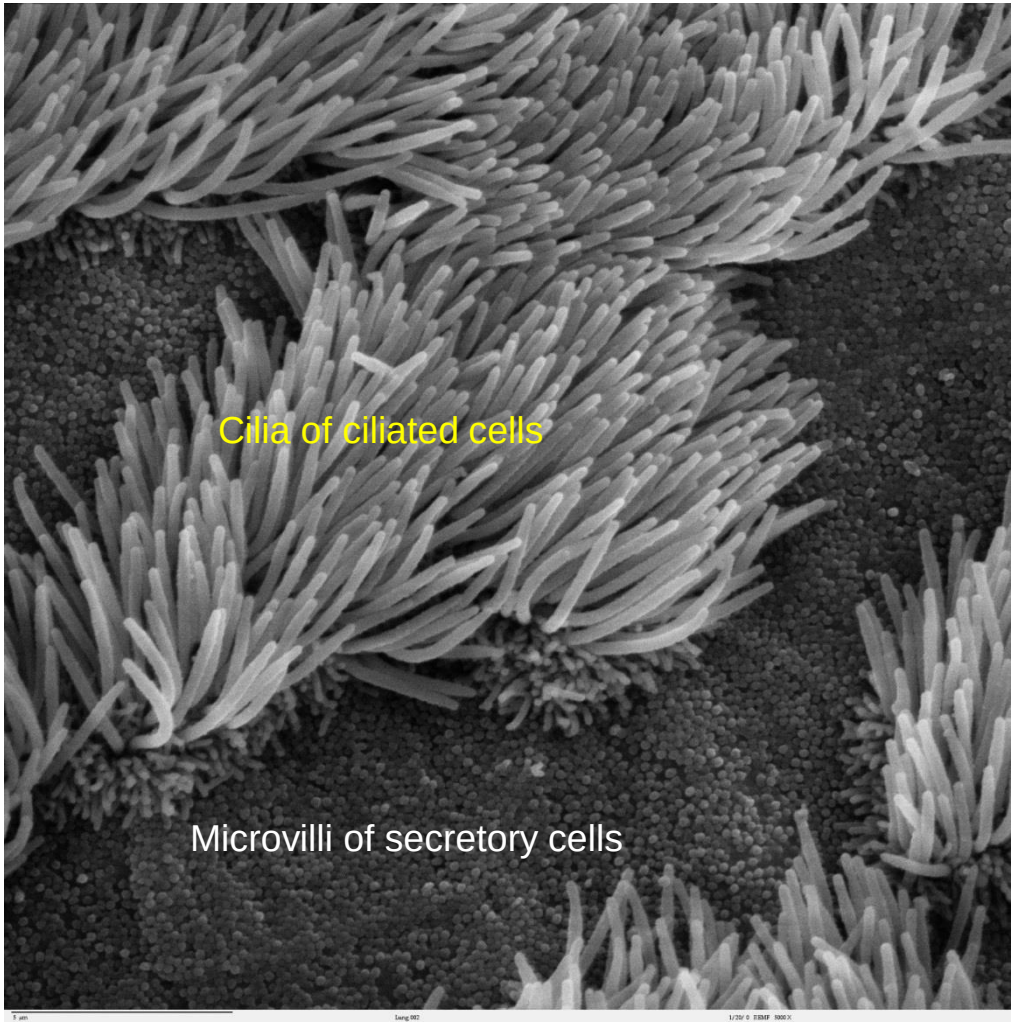


in electron microscope

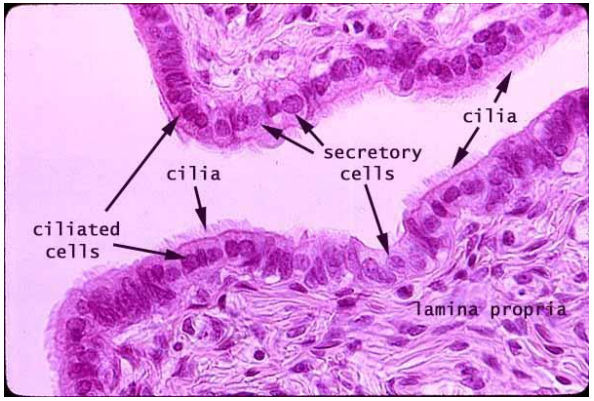


Cell surfaces 8

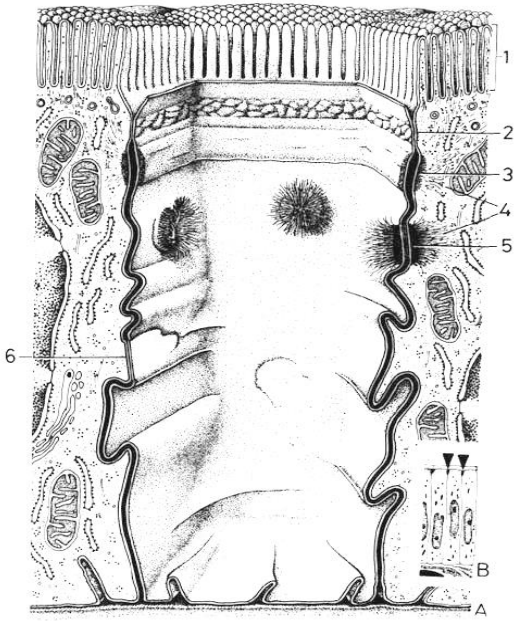
Cilia + Flagella



oviduct

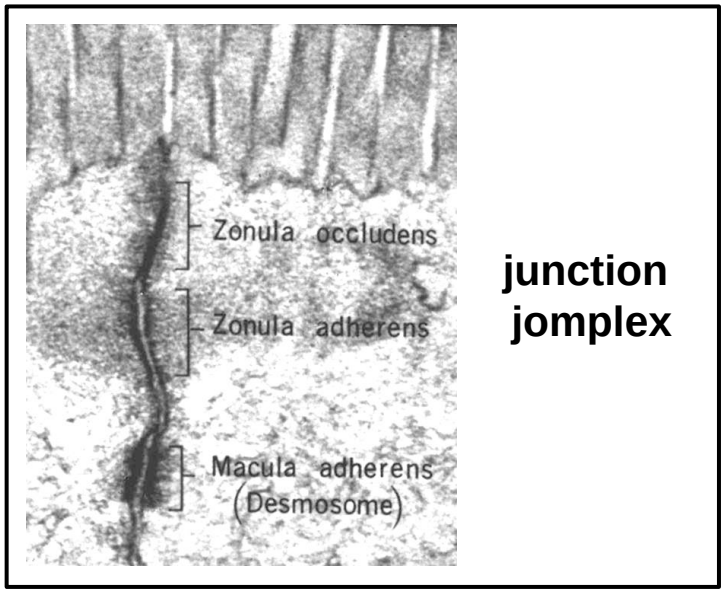


Adhesions and Junctions 1



lateral surface

Basal surface



junction jomplex

Adhesion

- **Macula adherens** (desmosome)
- **Zonula adherens**
- **Hemidesmosome**
- **Focal adhesion**

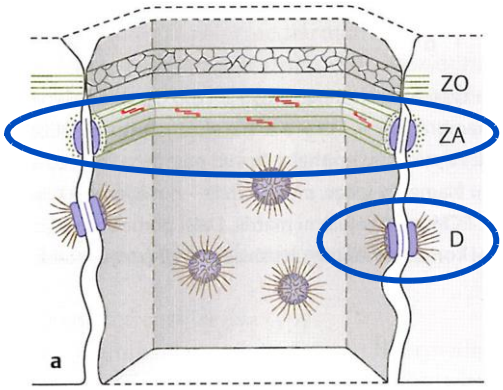
Sealing

- **Zonula occludens** (tight junction)

Communication

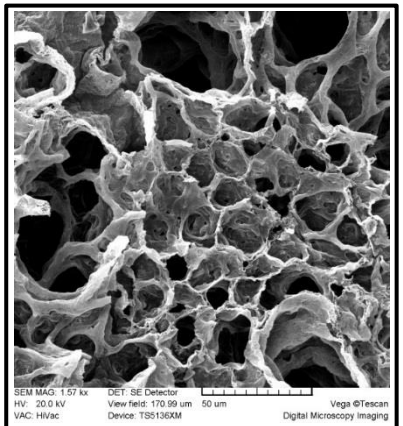
- **Gap junction** (nexus)

Adhesions and Junctions 2

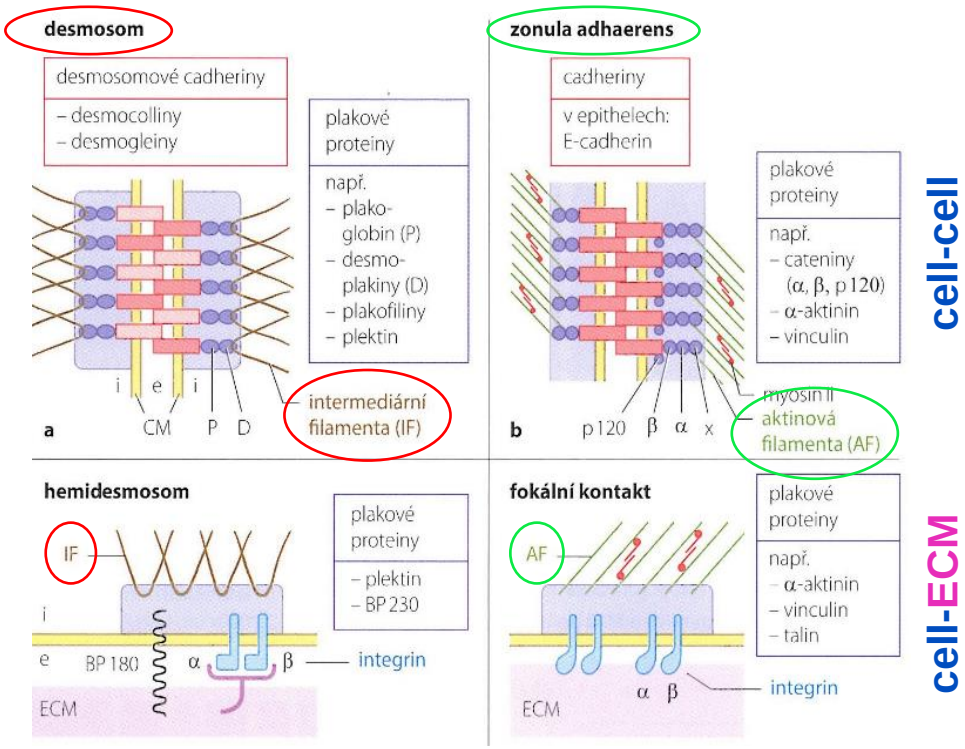


Adhesion

- **Macula adherens** (desmosom)
- **Zonula adherens**
- **Hemidesmosome**
- **Focal adhesion**

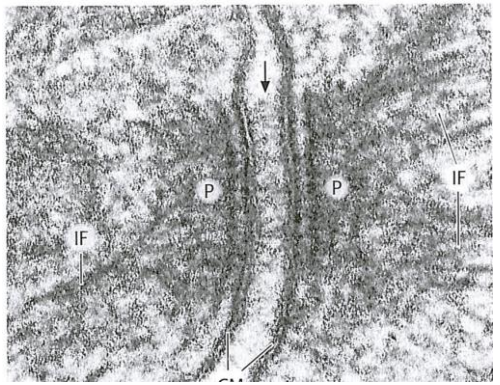


Lung ECM - alveolar region

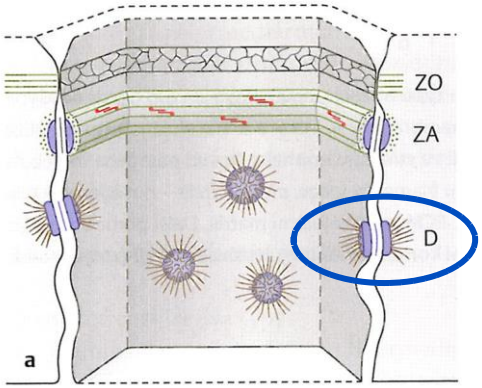


Unified composition

- **Transmembrane proteins** (cadherins+ integrins)
- **Adaptor (plak) proteins**
- **Cytoskelelal fibers**



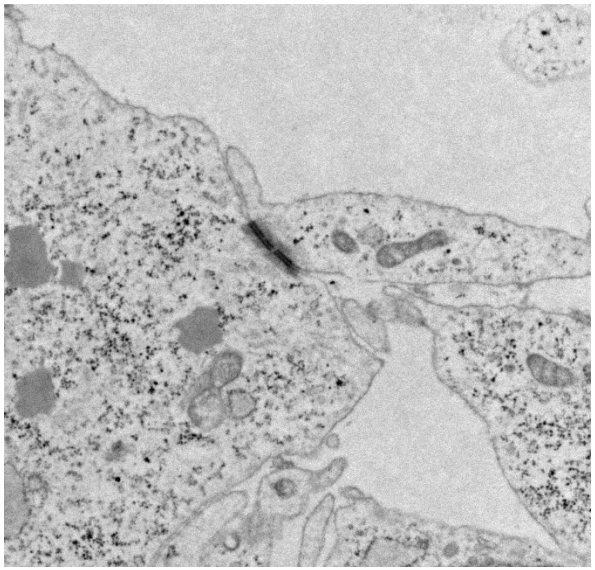
Adhesions and Junctions 3

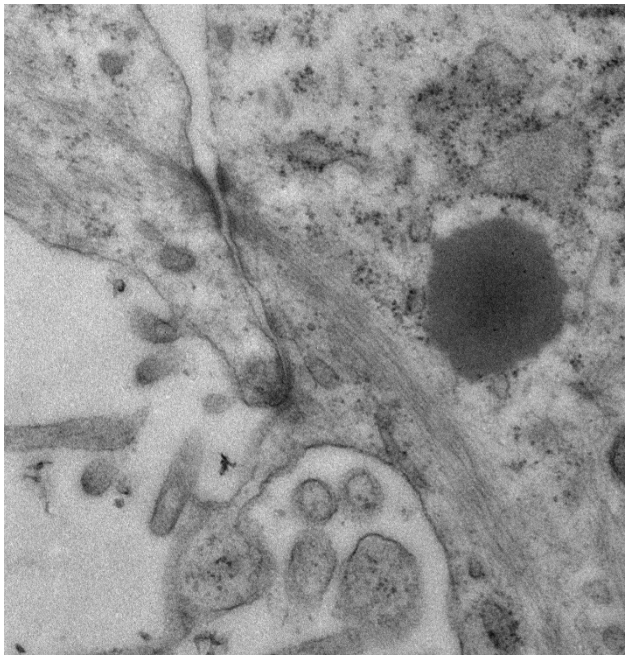


Adhesion

- **Macula adherens** (desmosome)

Diameter about 0,3 μm
Distance between membranes about 20-40 nm

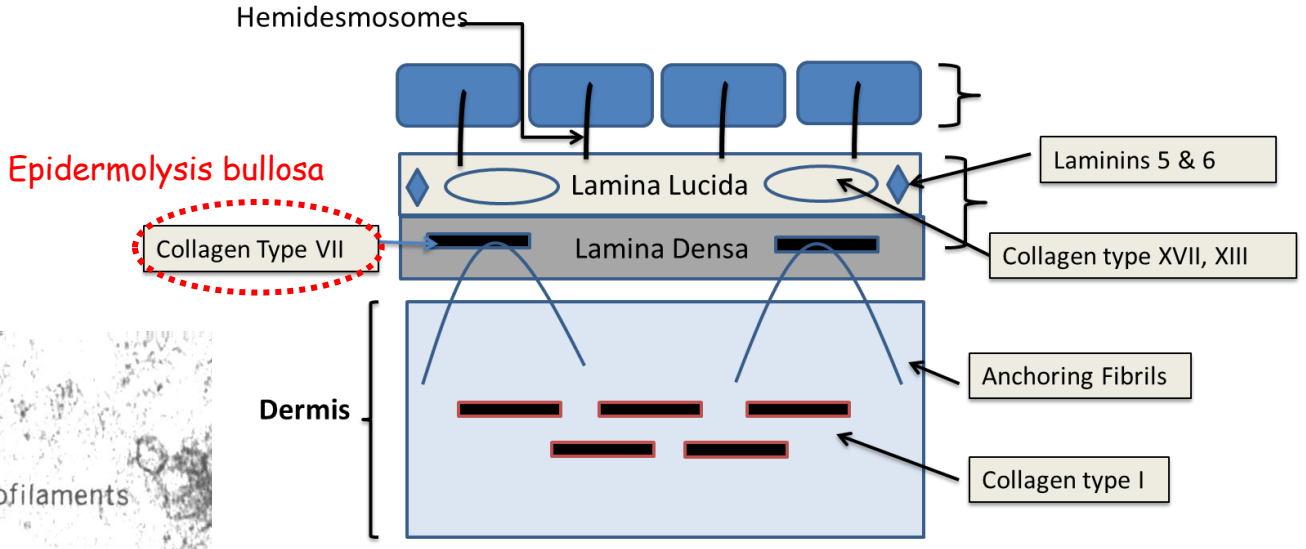


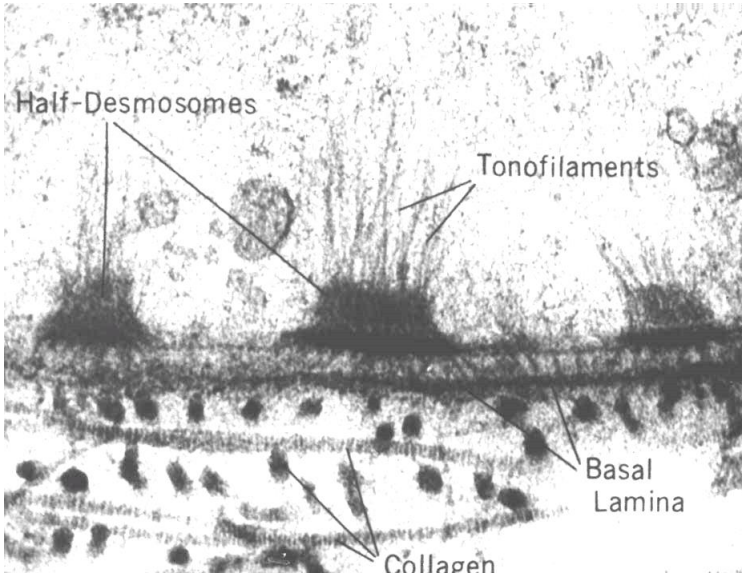


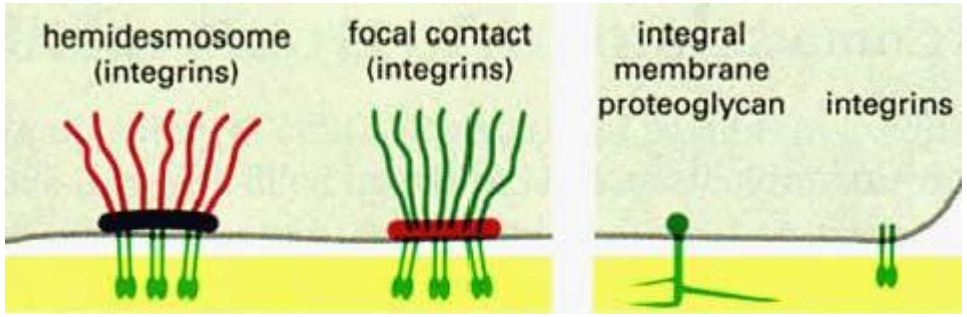
Adhesions and Junctions 4

Adhesion

- **Hemidesmosome**
- **Focal adhesion**

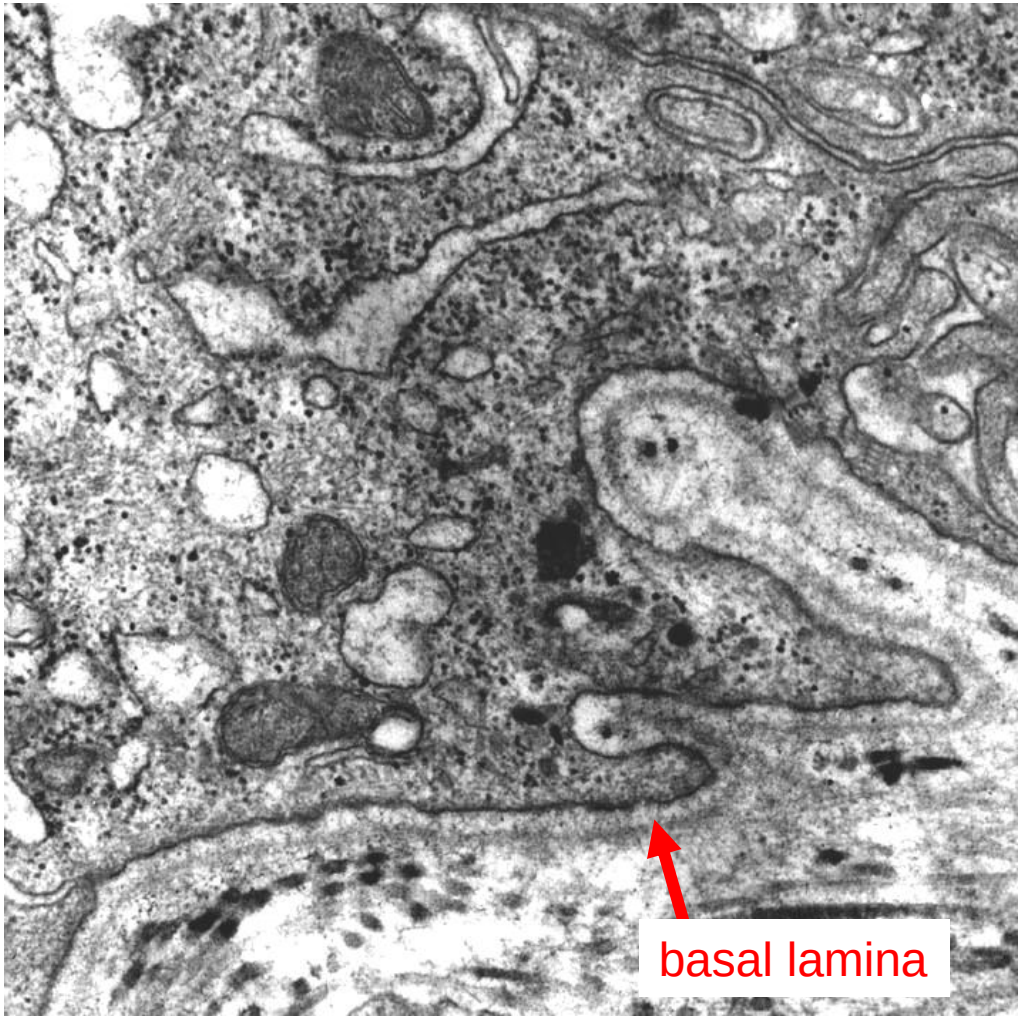






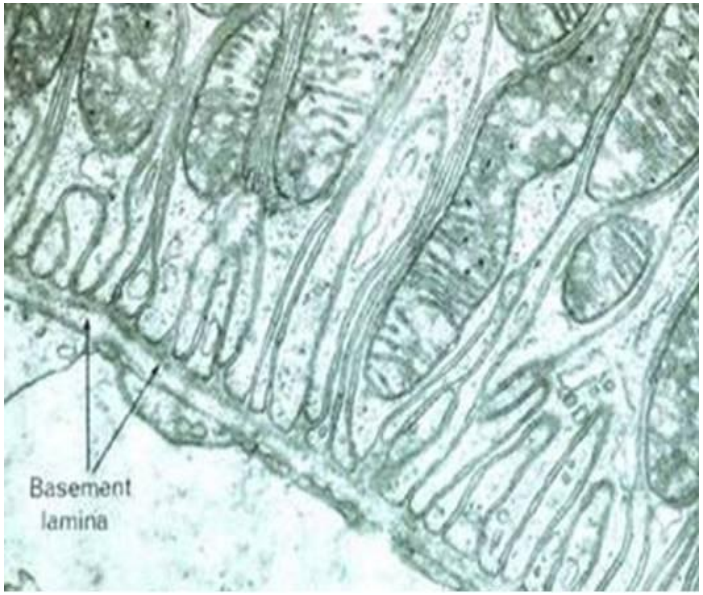
Adhesions and Junctions 5

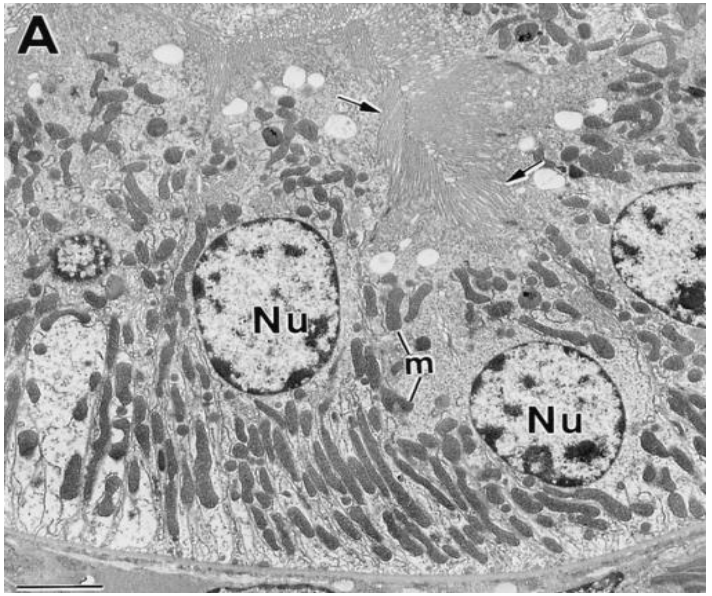
- **Focal adhesion**



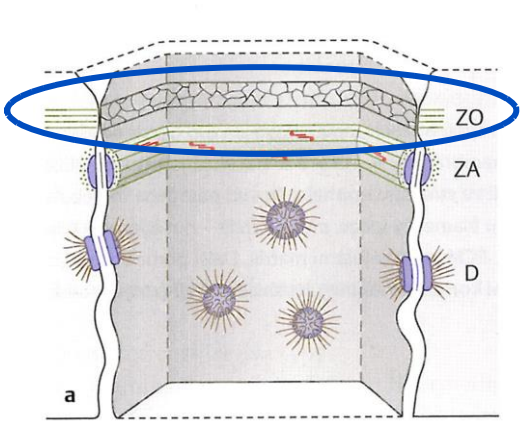
Adhesions and Junctions 6

Basal labyrinth



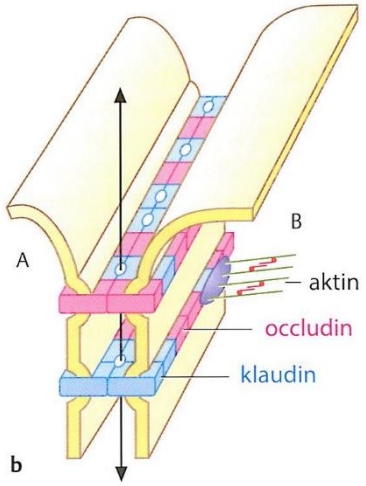


Adhesions and Junctions 7

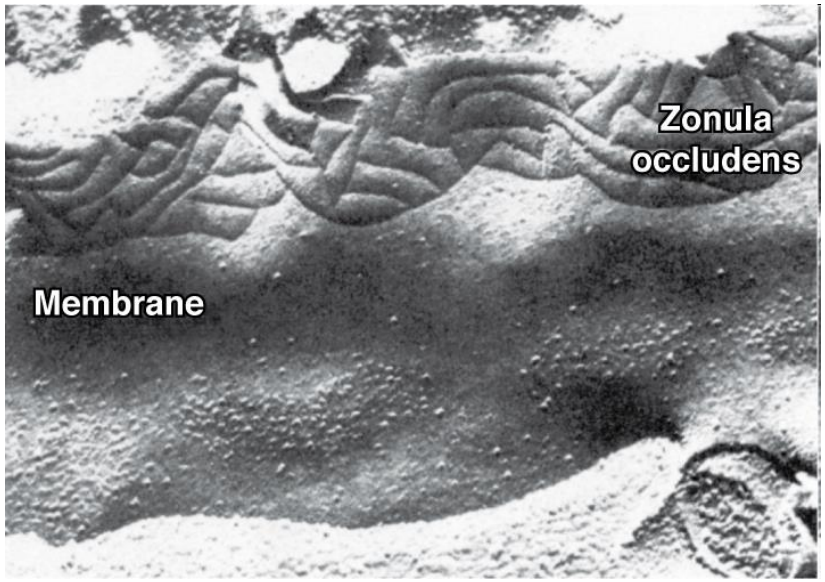


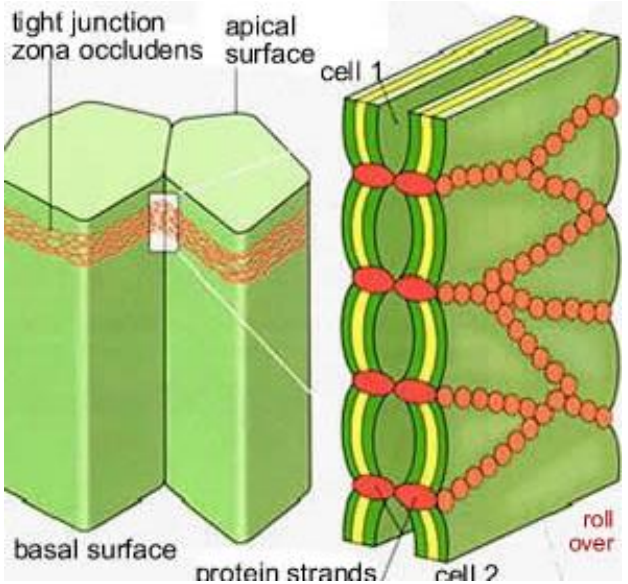
Sealing

- **Zonula occludens** (tight junction)

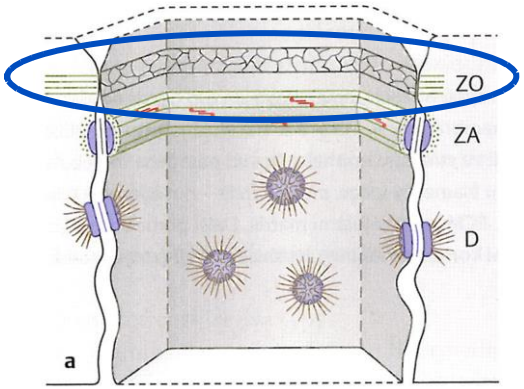


Damage by:
Clostridium perfringens
Helicobacter pylori (ZO-1)



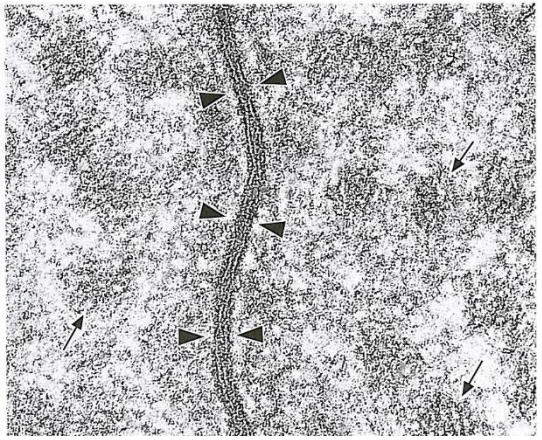


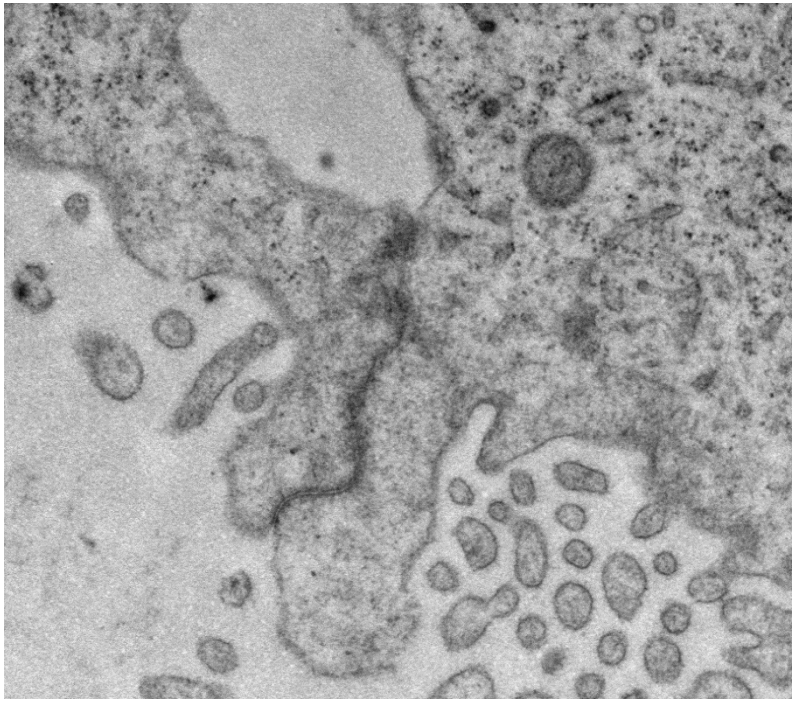
Adhesions and Junctions 8



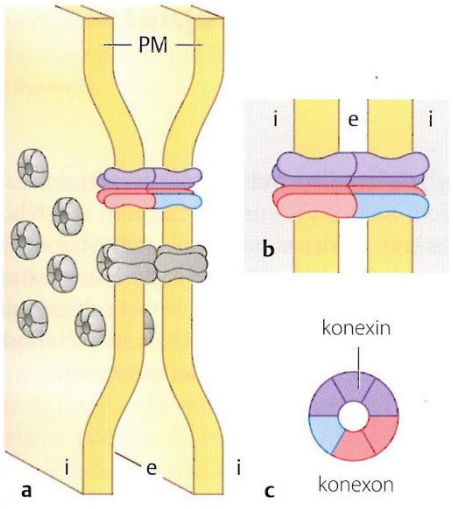
Sealing

- **Zonula occludens** (tight junction)





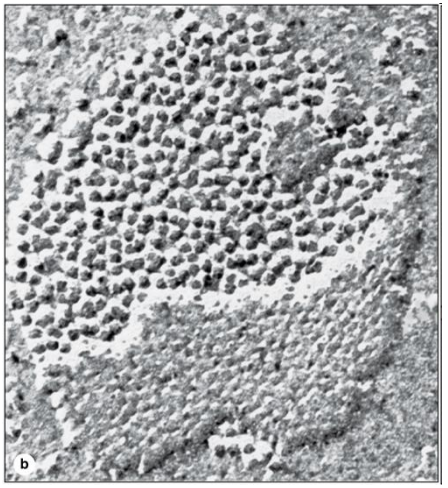
Adhesions and Junctions 9

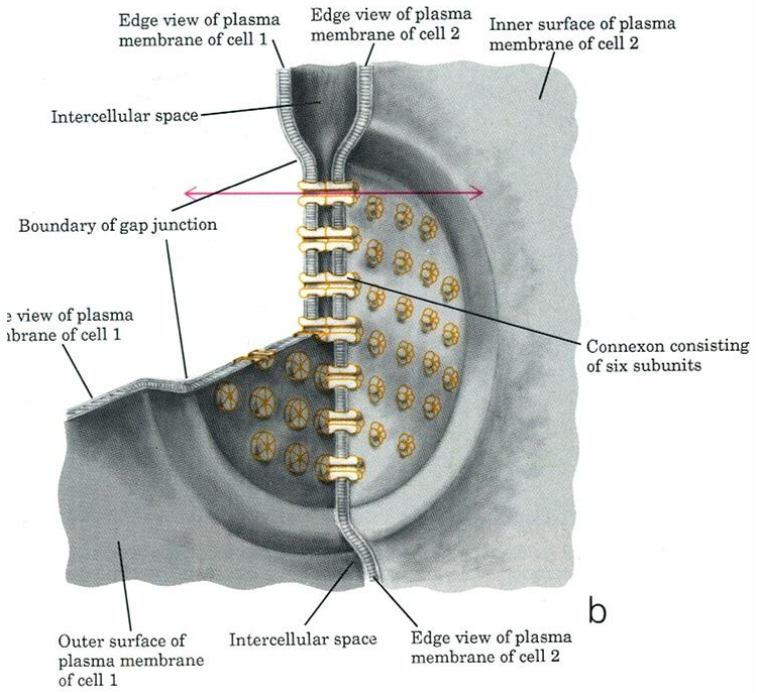


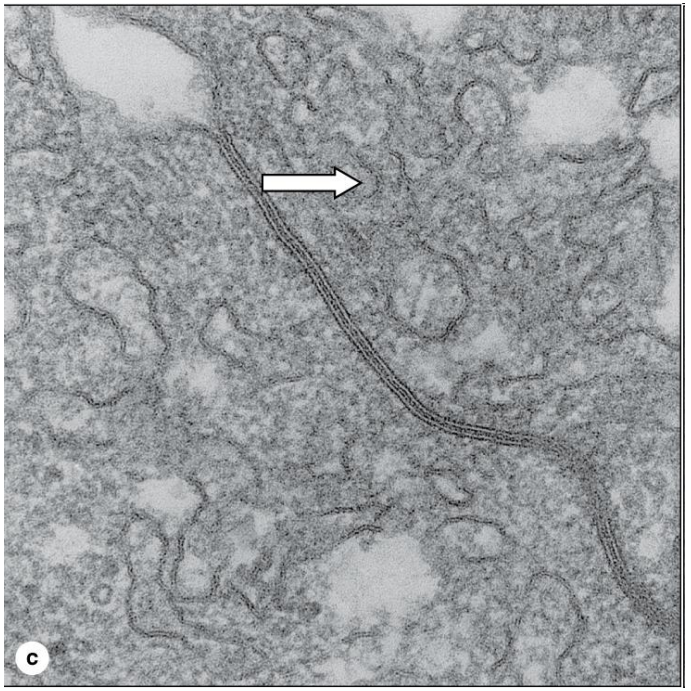
Communication

- **Gap junction** (nexus)

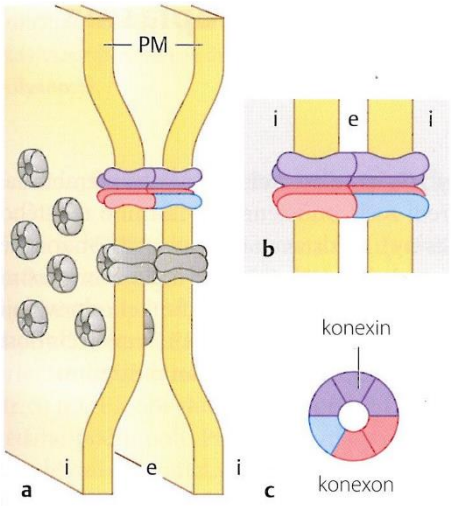
Diameter about 0,3 μm
Distance between cell membranes about 3 nm
Internal diameter of the channel about 2 nm





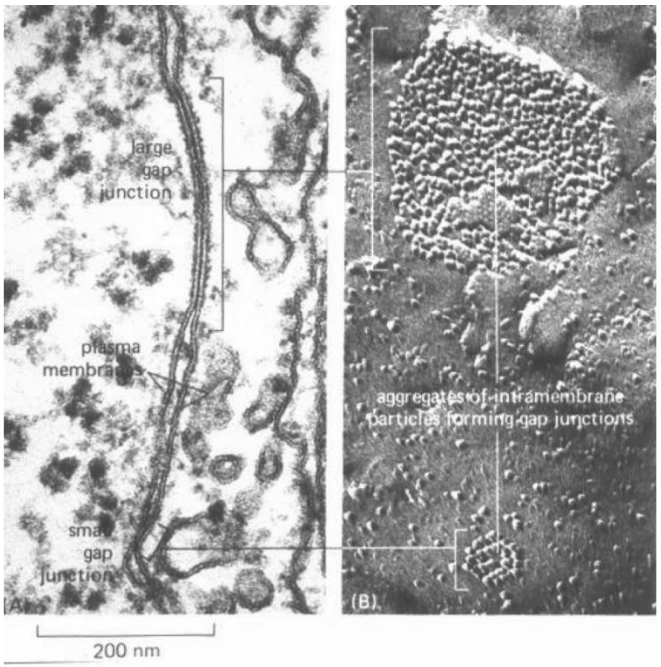


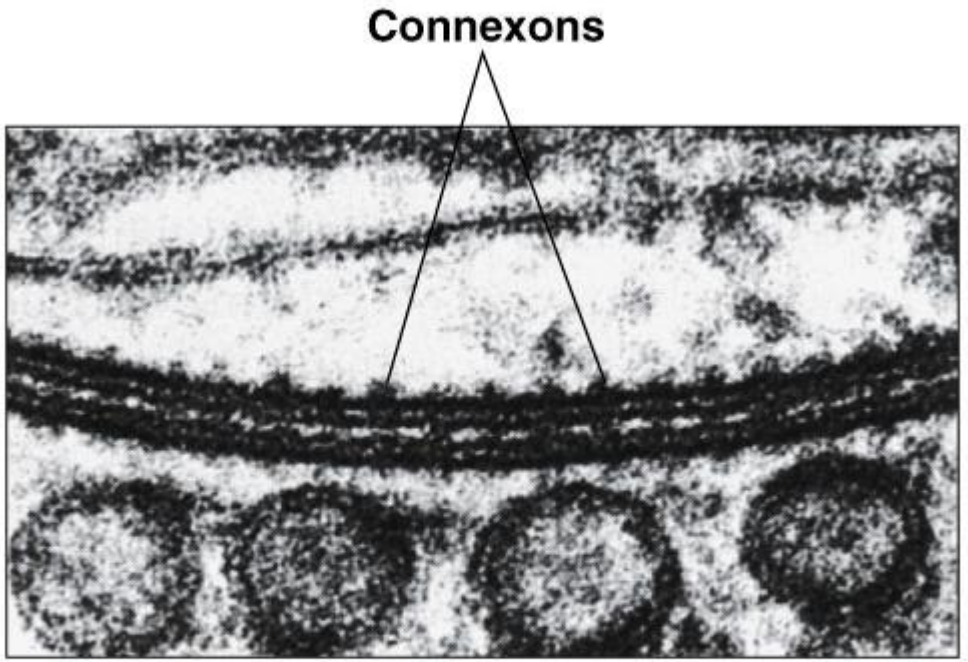
Adhesions and Junctions 10



Communication

- **Gap junction** (nexus)





(b) Electron micrograph of a gap junction

Activities of cells

- **Movement** – intracellular, amoeboid, cilia, flagella
- **Metabolism** – income, processing, outcome
- **Responsivenes**
- **Growth**
- **Differentiation**
- **Division (amplification)**

Division x Differentiation of cells 1

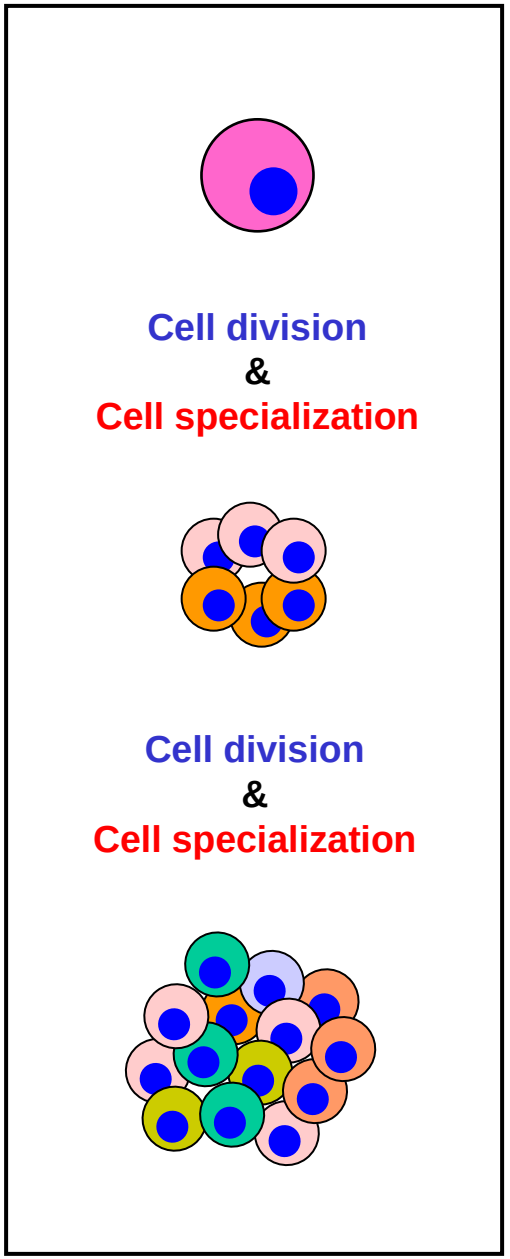
Zygote

Multicellular embryo

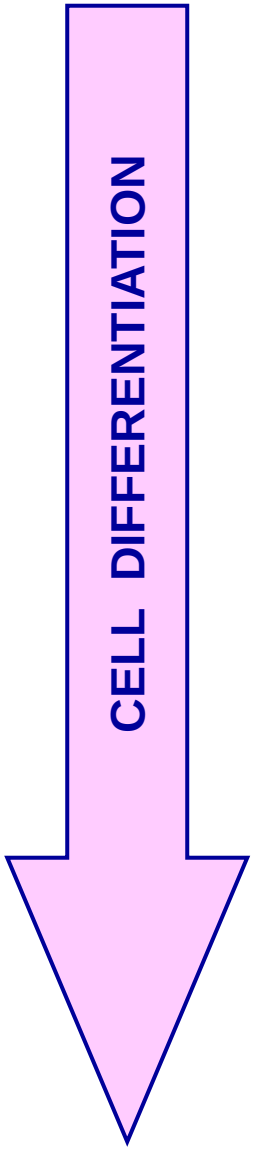
Dynamic multicellular organism

CELL DIVISION

CELL DIFFERENTIATION



Division x Differentiation of cells 2



STABLE GENOME

↕

Genomic equivalence

(= equal amount of DNA and the same nucletide sequence in all cells of a organism – cloning)

X

VARIABLE TRANSCRiPTOME

↕

Transcription Regulators

+ other regulations:

- translation
- posttranslational modification

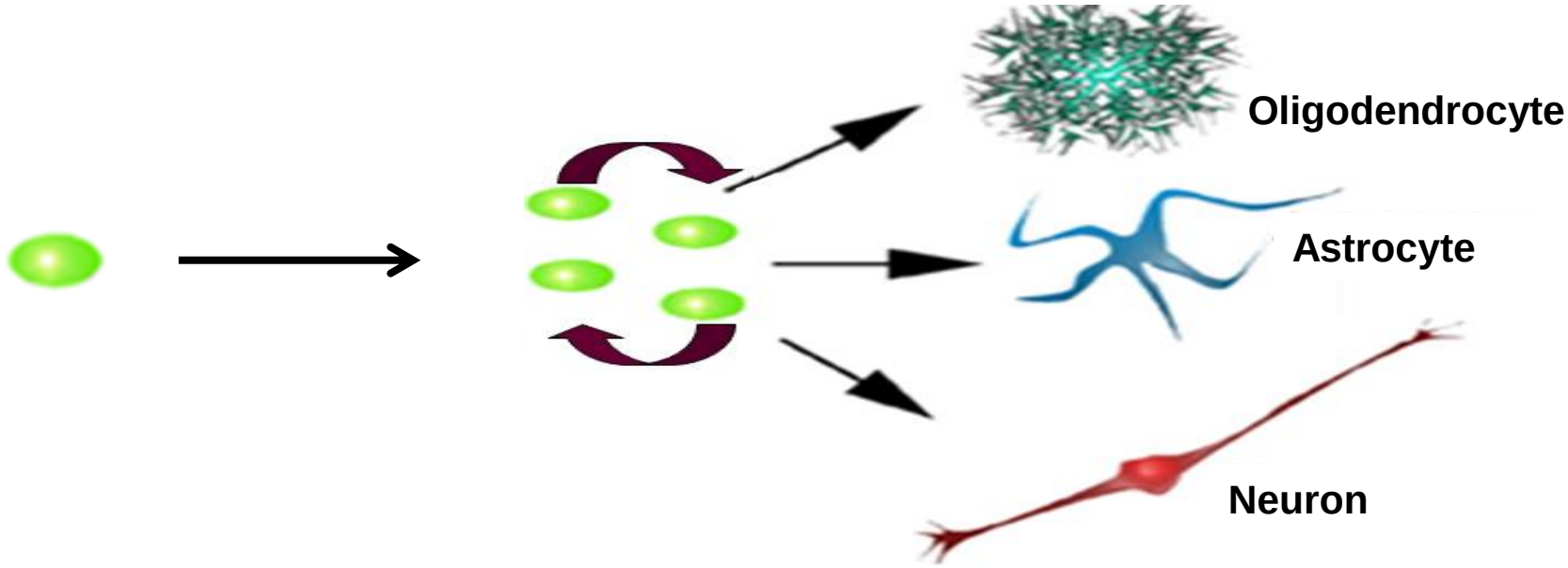
Division x Differentiation of cells 3

Tissue renewal and regeneration

Stem cells
- slowly dividing (usually)
- multipotent

Progenitor cells
- „transit amplifying cells“
- fast proliferation
- multipotent

Terminally diferentiated cells
- nondividing



Mother nature and scientists supply us with many

Stem cells generate and regenerate our body

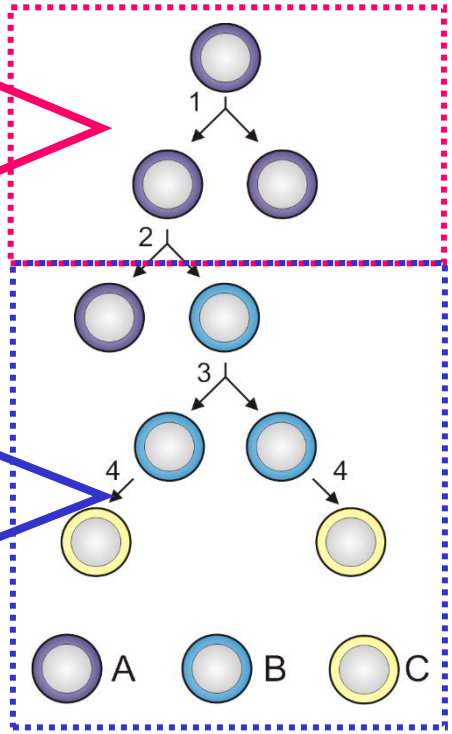
Capability to produce identical copies of itself

Self-renewal

Capability to differentite into specialized cell types

Pluripotency

1. Undifferentiated growth

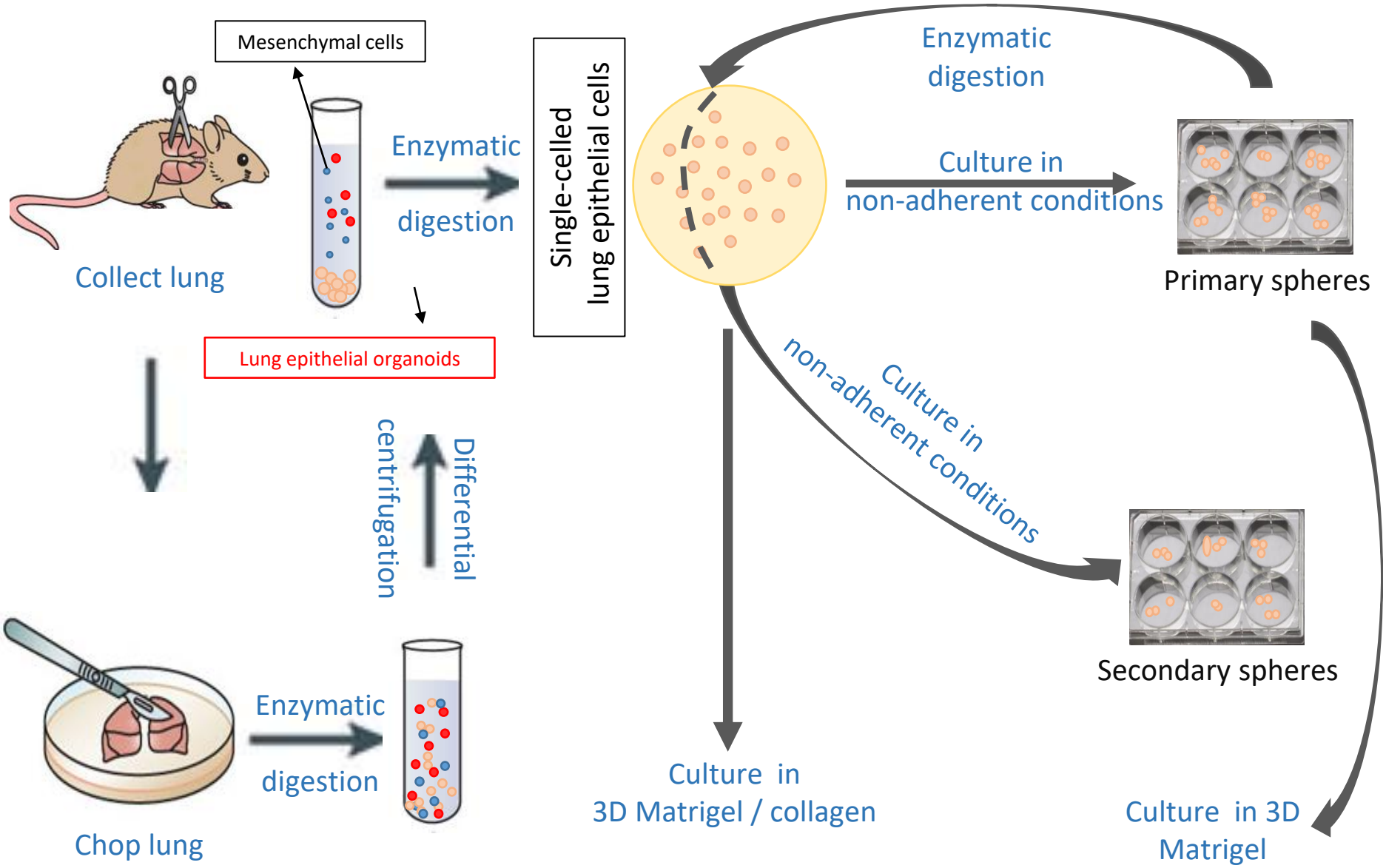


2. Differentiation

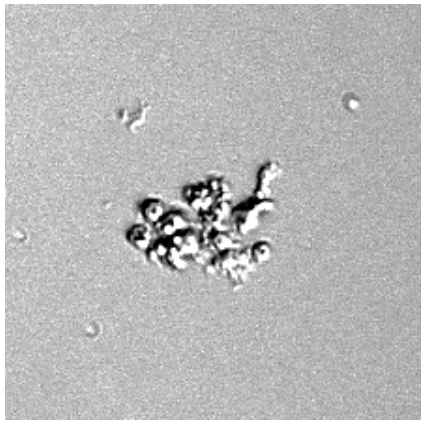
- Embryonic stem cells
- Adult stem cells
  - Fetal
  - Organ
  - Tissue
- Induced pluripotent stem cells
- Cancer stem cells

Different properties

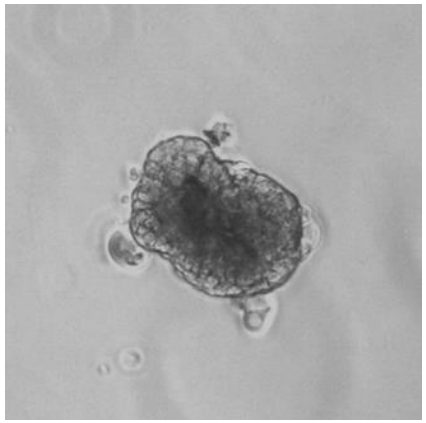
Stem cell can be isolated from tissues and studied in vitro 1



Stem cell can be isolated from tissues and studied in vitro 2

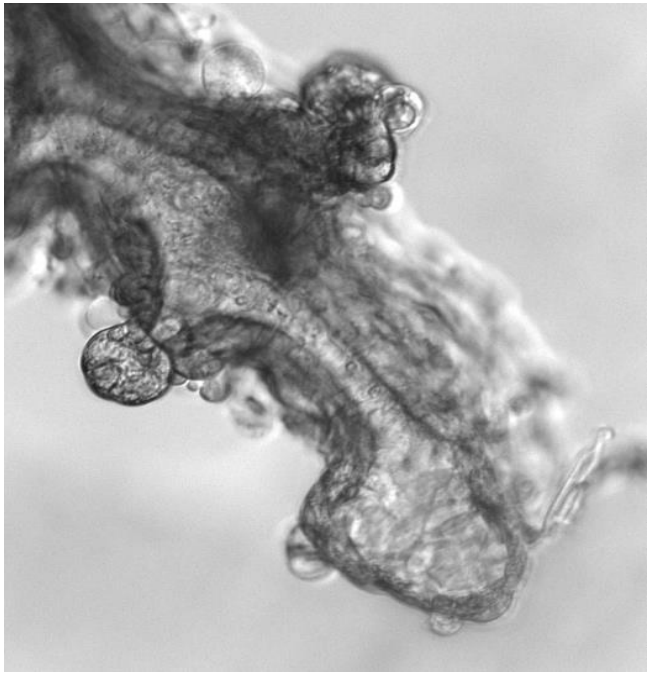


SCs after isolation



Spheroid growing from SC
„lungosphere“

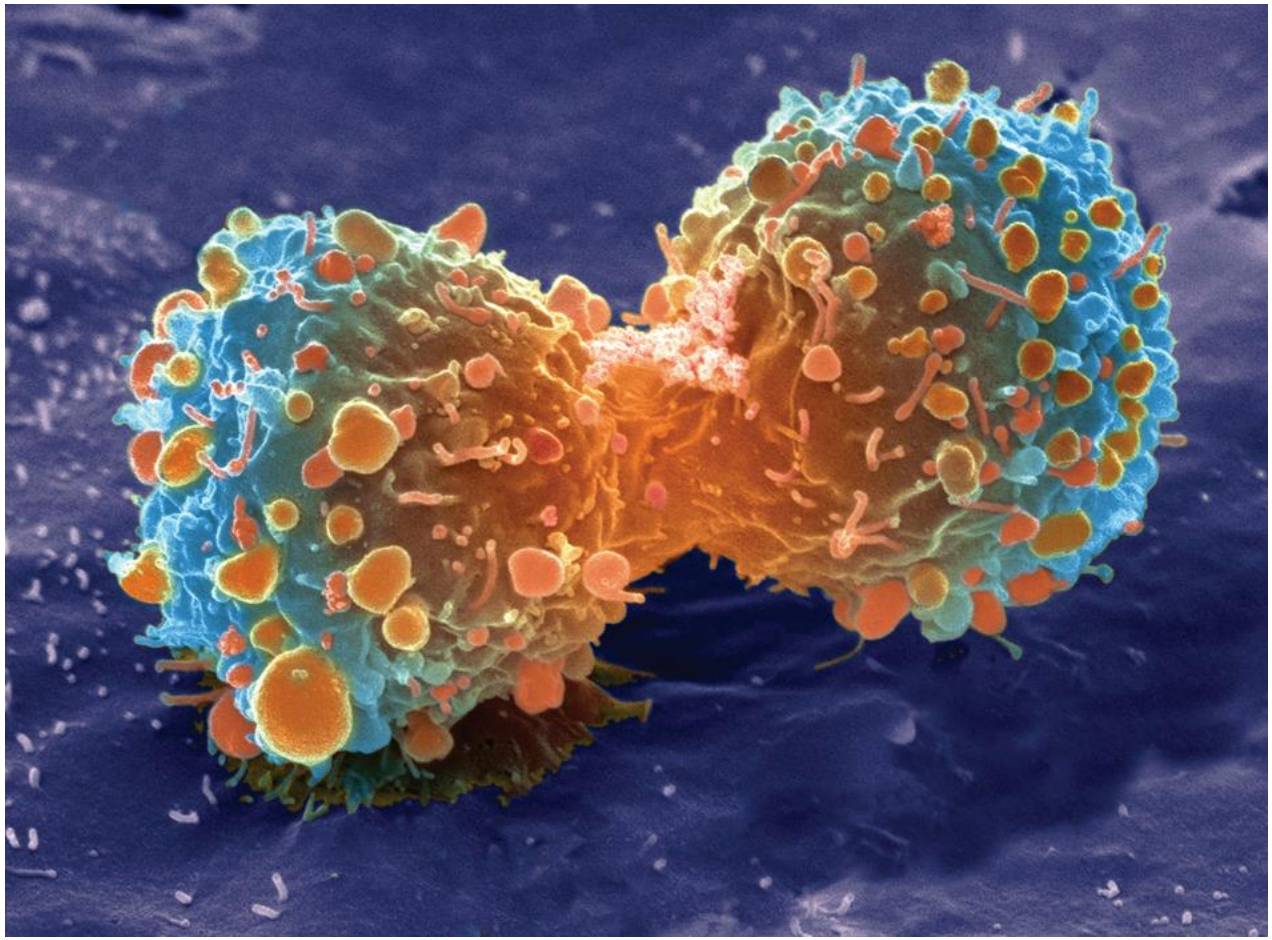
Organoid
Morphogeneis in 3D environment



Cell division 1

Basic concept 1

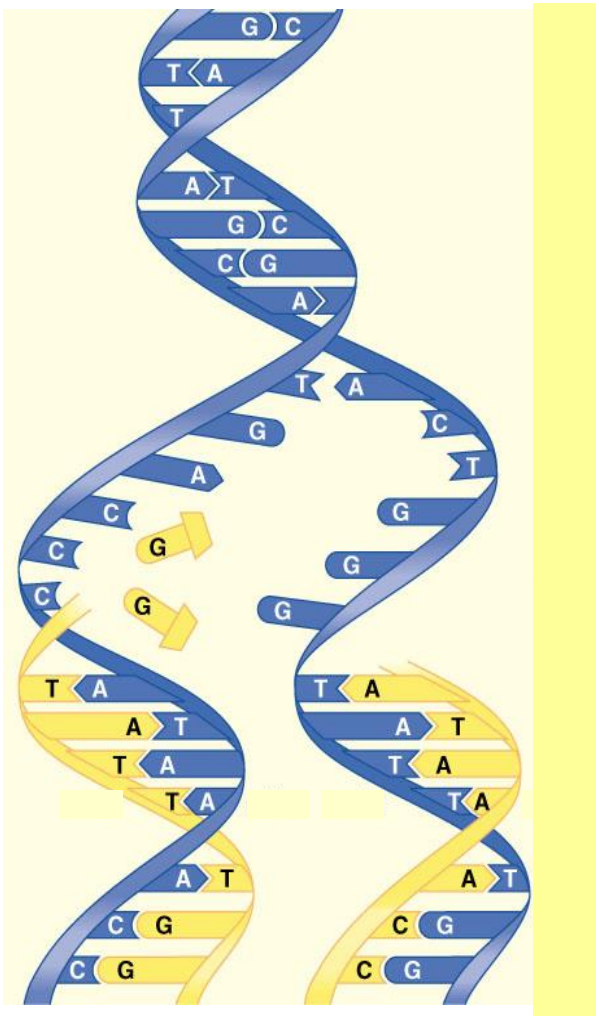
MITOSIS and CYTOKINESIS produce genetically identical cells

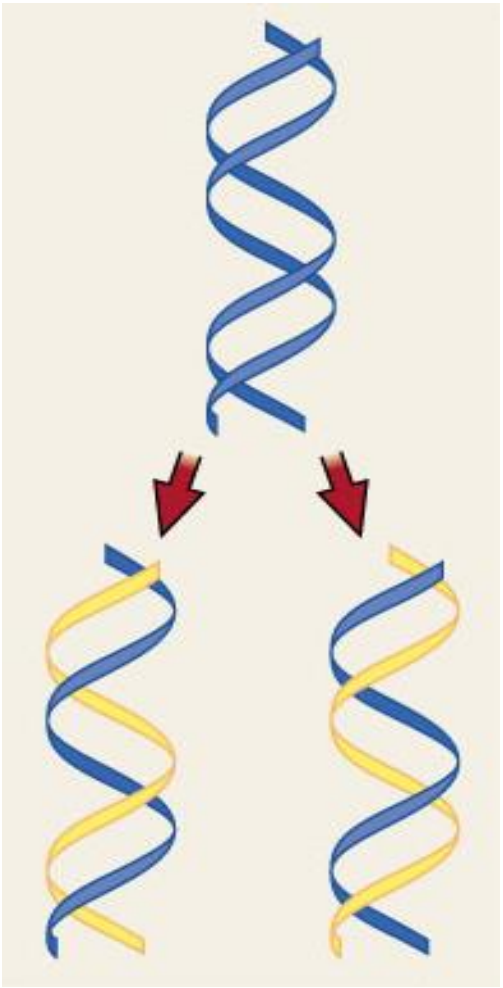


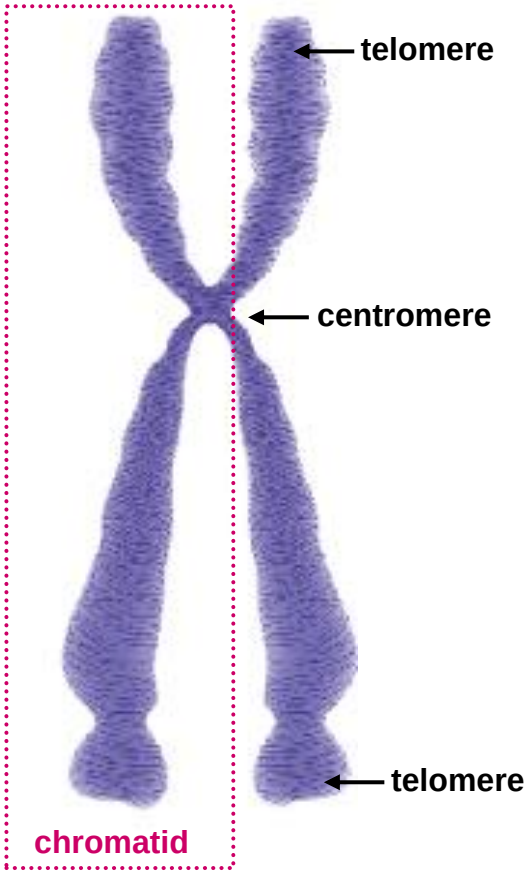
Cell division 2

STABLE (non-changing) GENOME

Due to semiconservative duplication of DNA



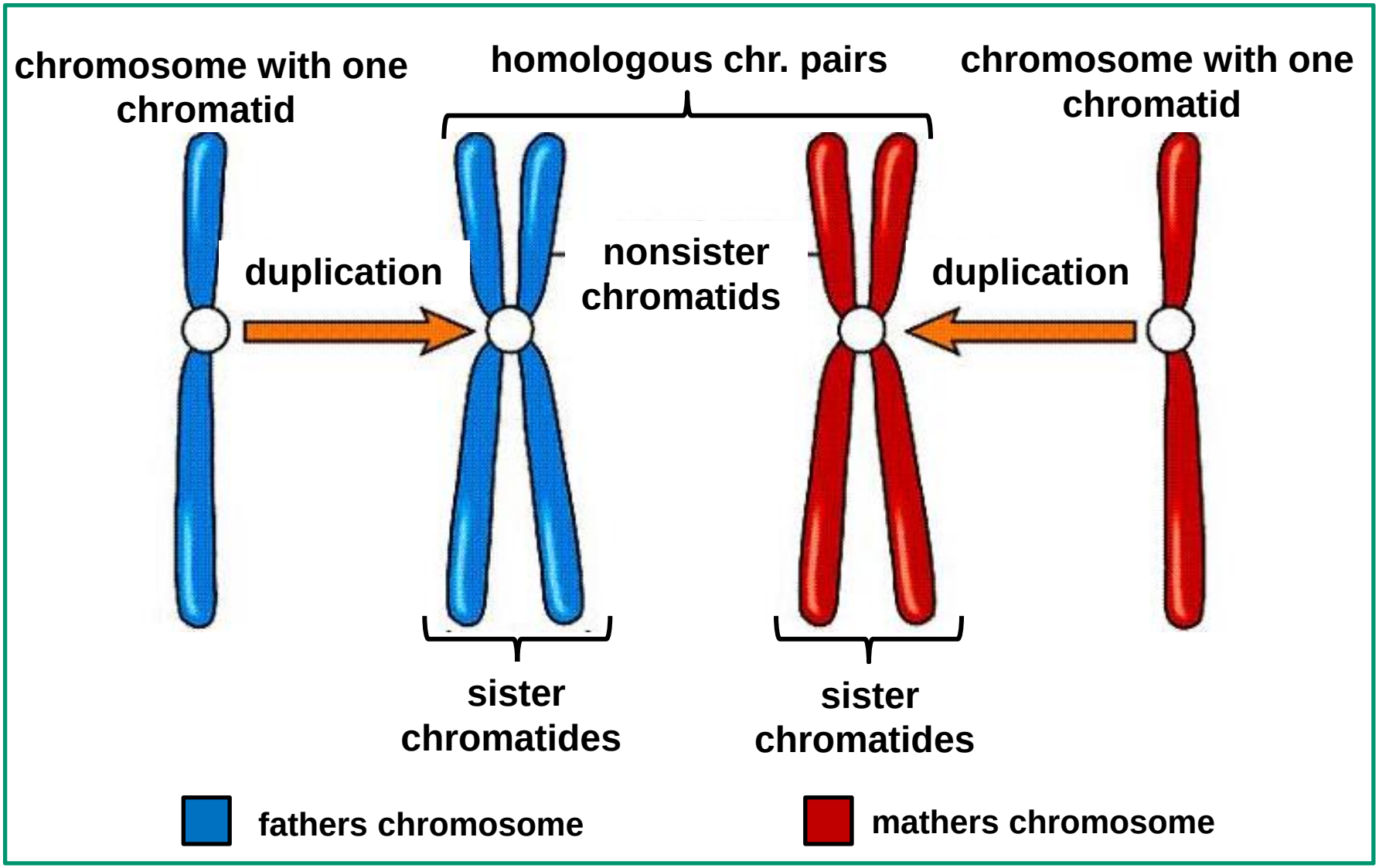




Condensed duplicated chromosome

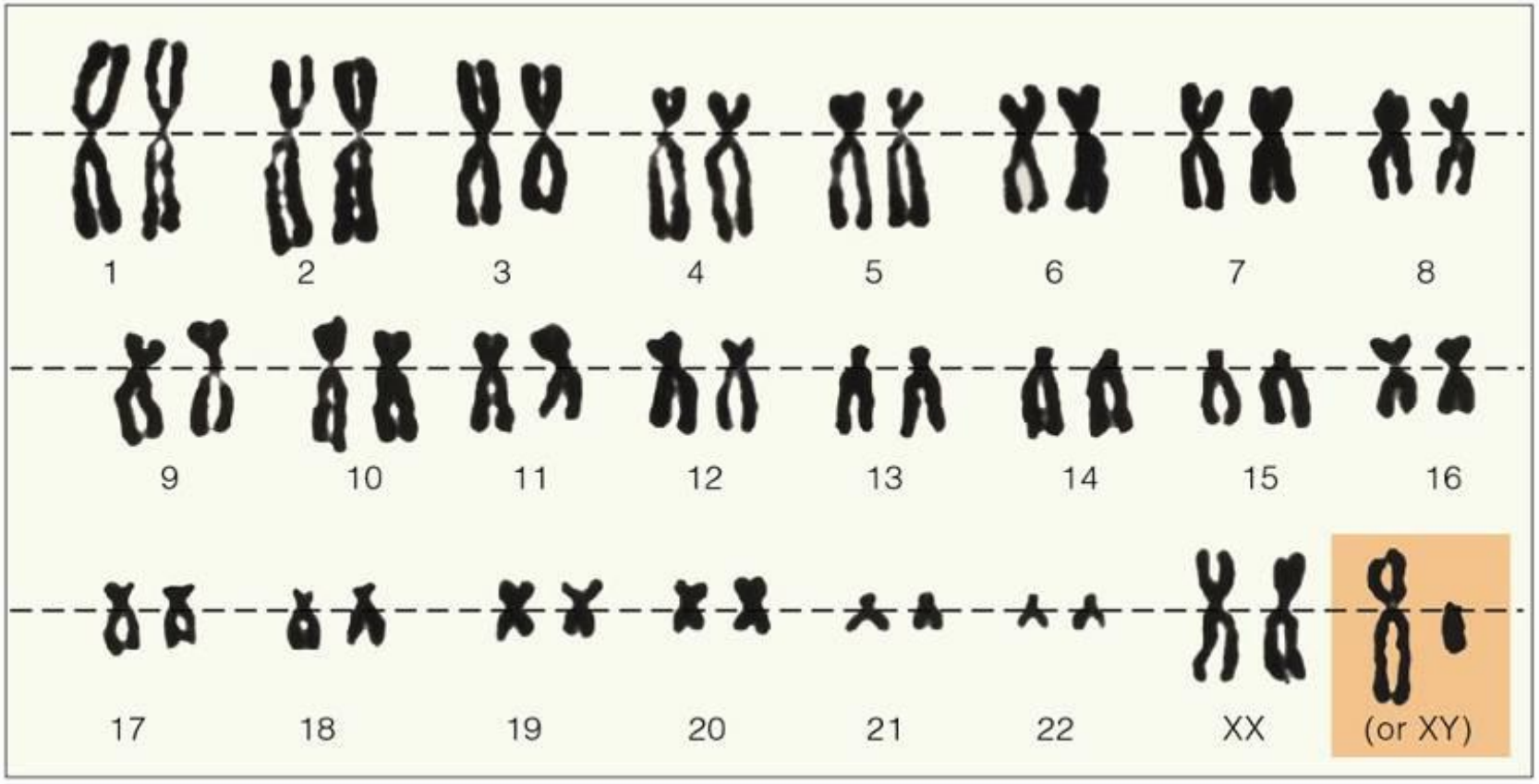
Cell division 3

Metabolism of chromosomes – Homologous chromosomes



Cell division 4

Pairs of homologous chromosomes (2N) organized into so called „KARYOTYPE“



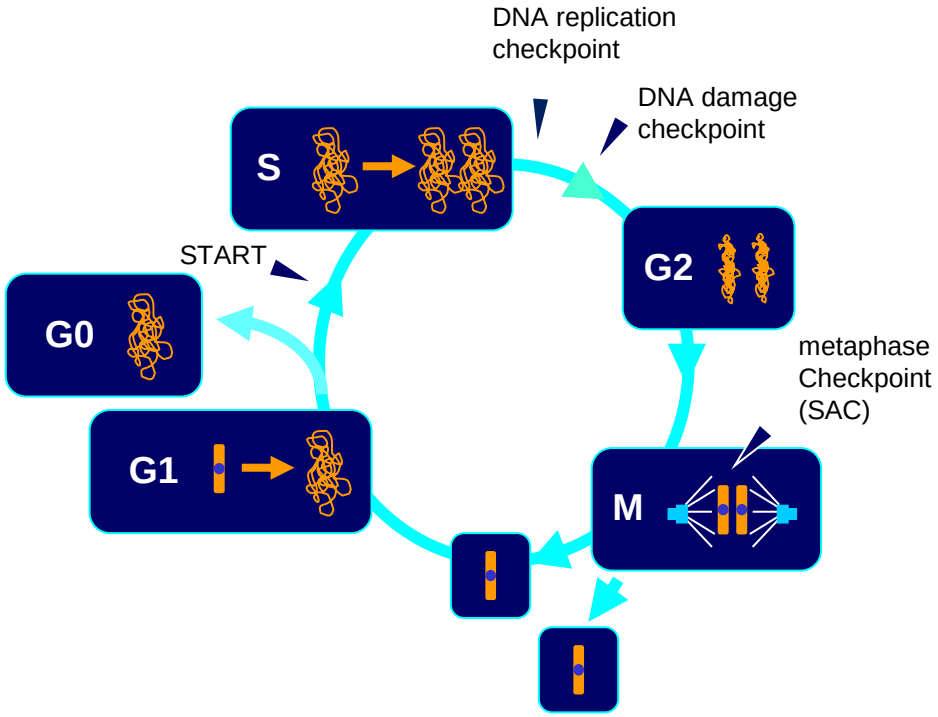
Cell division 5

Basic concept 2

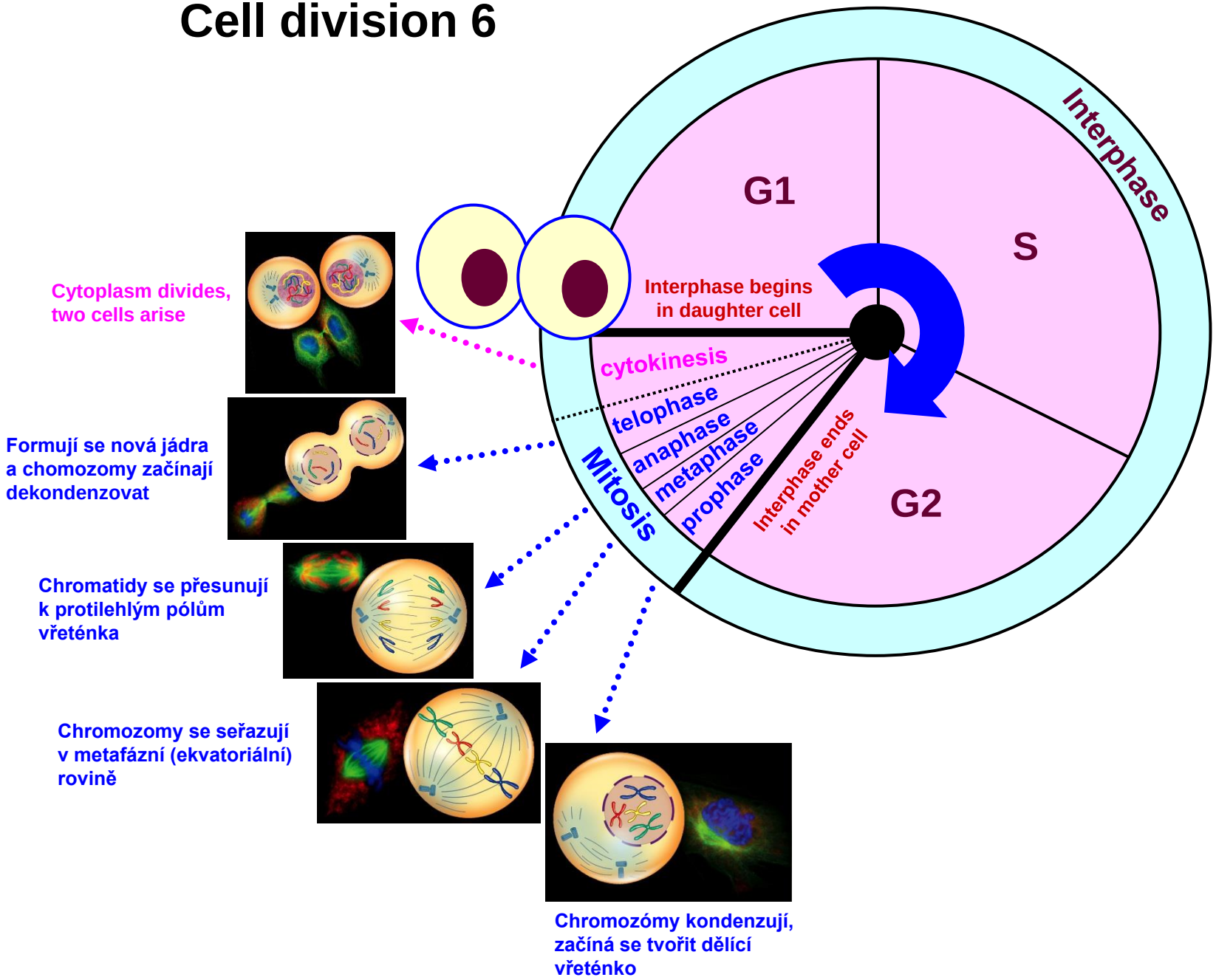
MITOSIS and CYTOKINESIS are parts of cell cycle

CELL CYCLE

- semi-modular character
- equipped with checkpoints
- among cells it is coordinated by signalling molecules

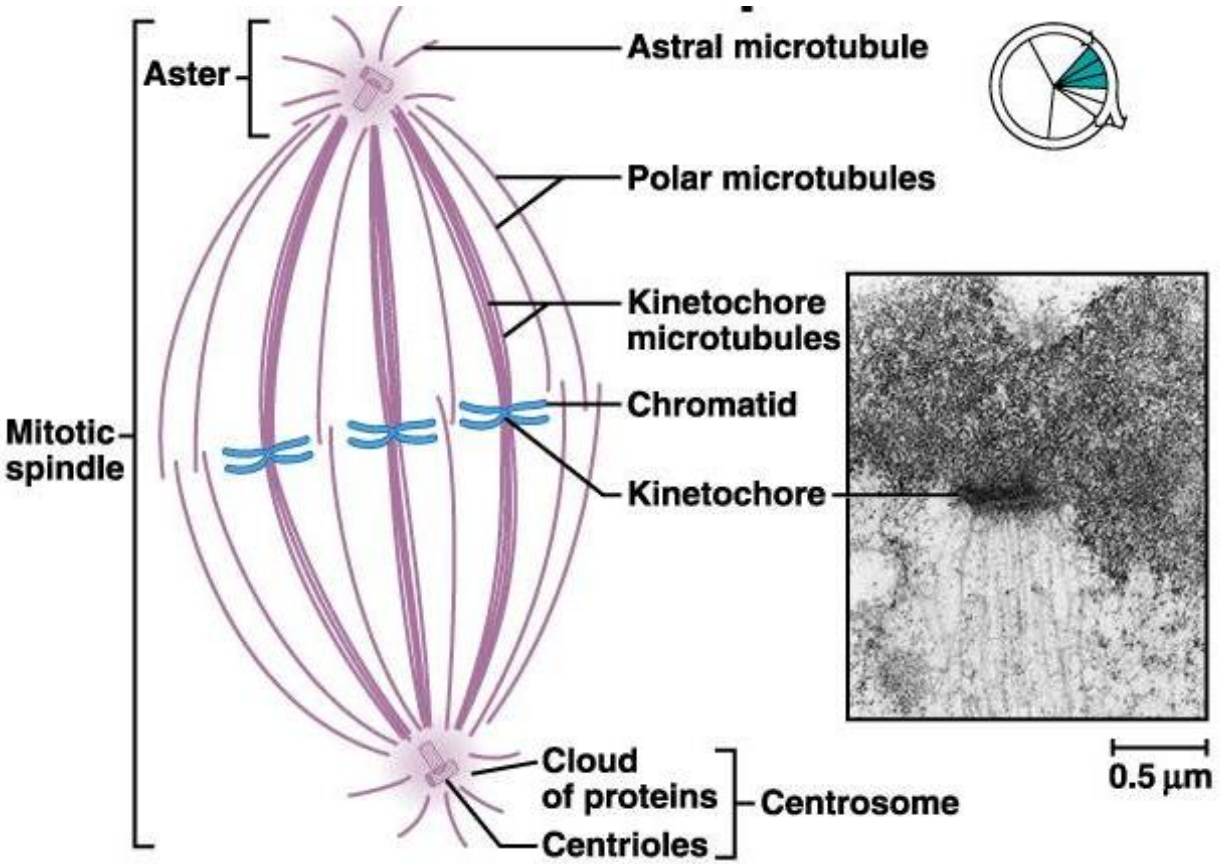


Cell division 6



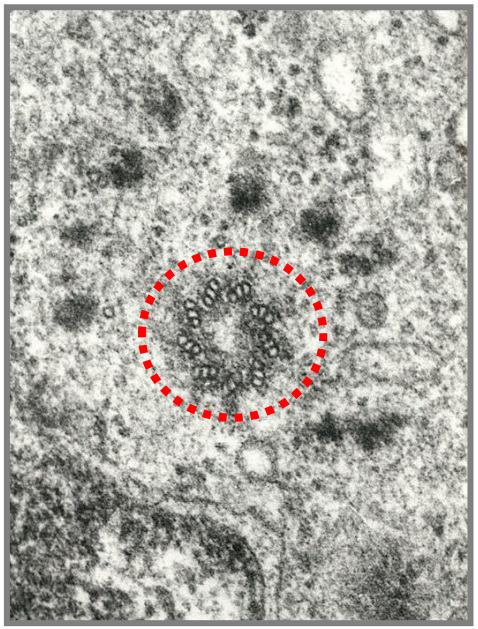
Cell division 7

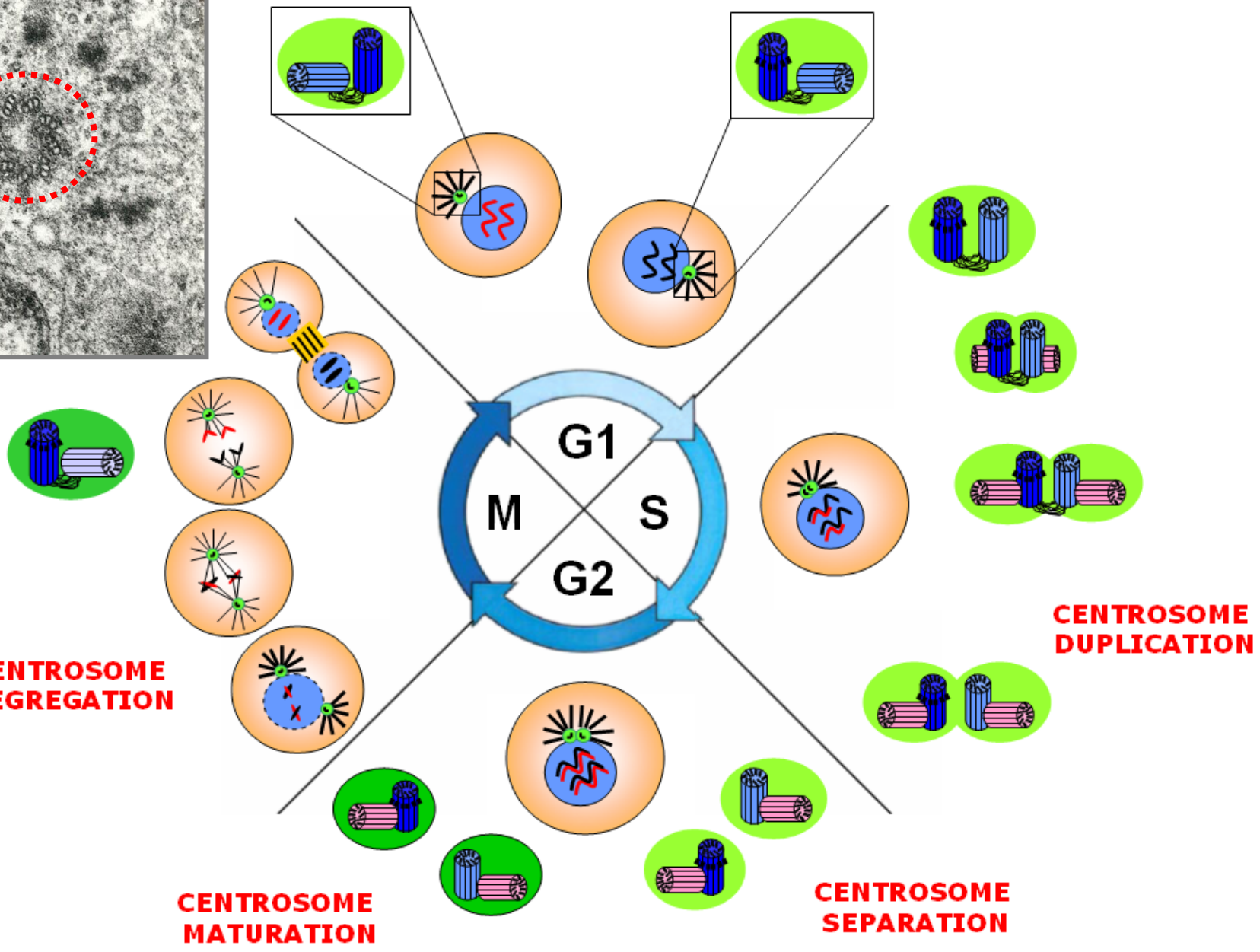
Mitotic spindle



Cell division 8

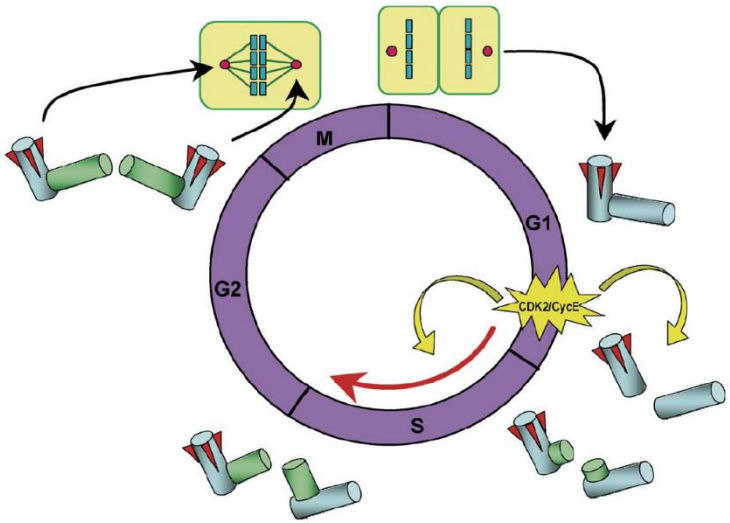
Centrosomal metabolism
Semiconservative duplication

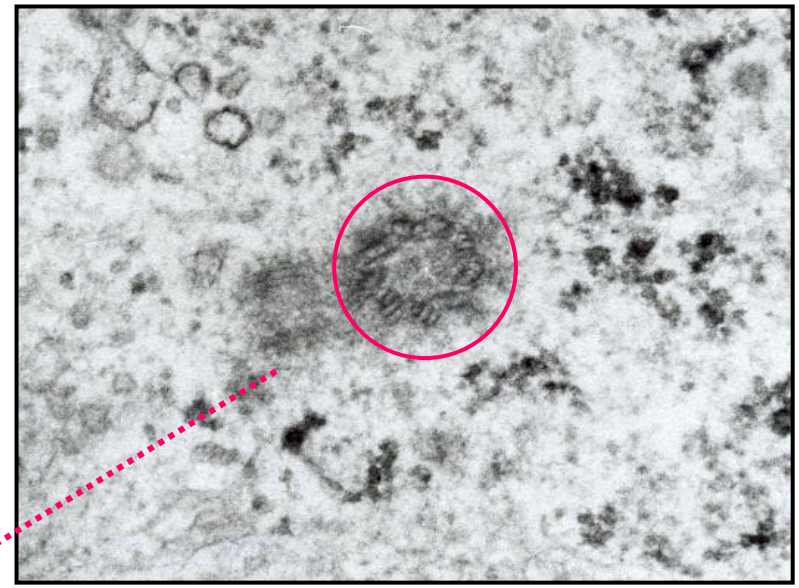


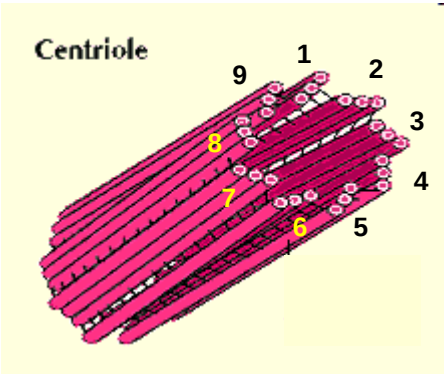


Cell division 9

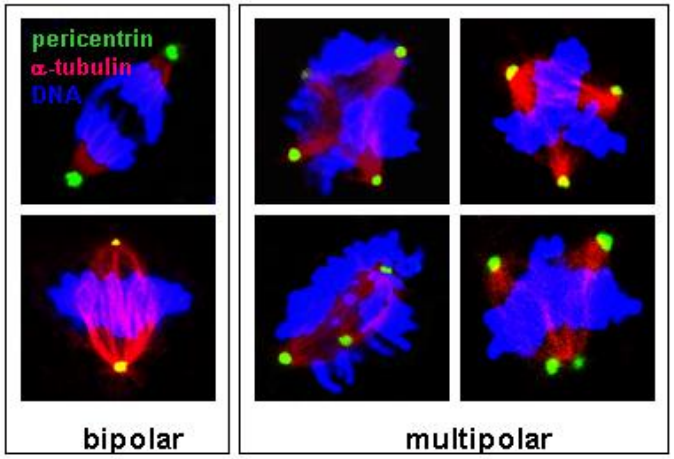
Centrosome structure







Diameter – 0.2 μm
Length - 0.5 μm



Cell division 10

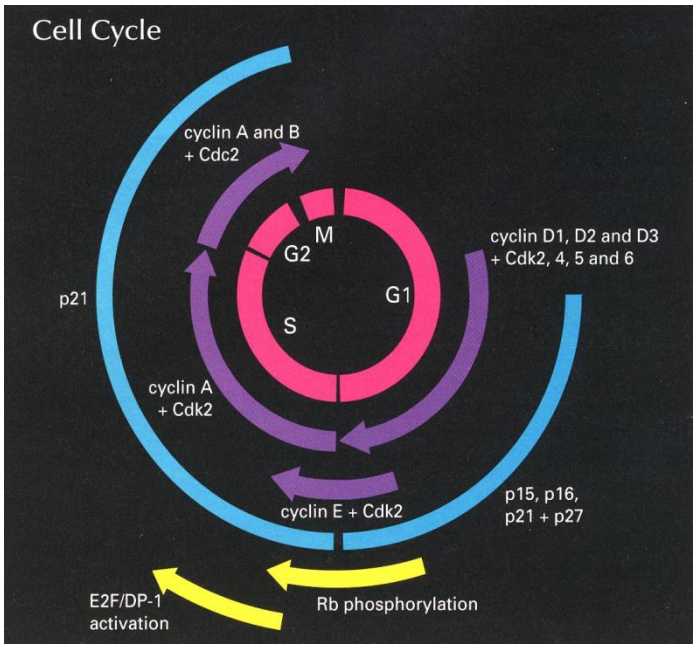
Regulation – Cyklin-Dependent Kinases (CDK) + Cyklins

Cdks and Related Proteins

kinase	PSTAIRE motif	regulatory subunits	putative substrates
Cdc2 p34	PSTAIRE	cyclin A & B	Rb, NF, histone H1
Cdk2	PSTAIRE	cyclin A, E & D	Rb, p27
Cdk3	PSTAIRE	cyclin E	E2F-1/DP-1
Cdk4	PV/ISTVRE	cyclin D1, D2, & D3	Rb
Cdk5	PISSLRE	p35	NF, Tau
Cdk6	PLSTIRE	cyclin D1, D2, & D3	Rb
Cdk7	NRTALRE	cyclin H	Cdc2, Cdk4/6
Cdk8	SACRE	cyclin C	RNA Pol II
Cdk9	PITALRE	cyclin T	Rb, MBP

Major Cyclin-Cdk Cell Cycle Complexes

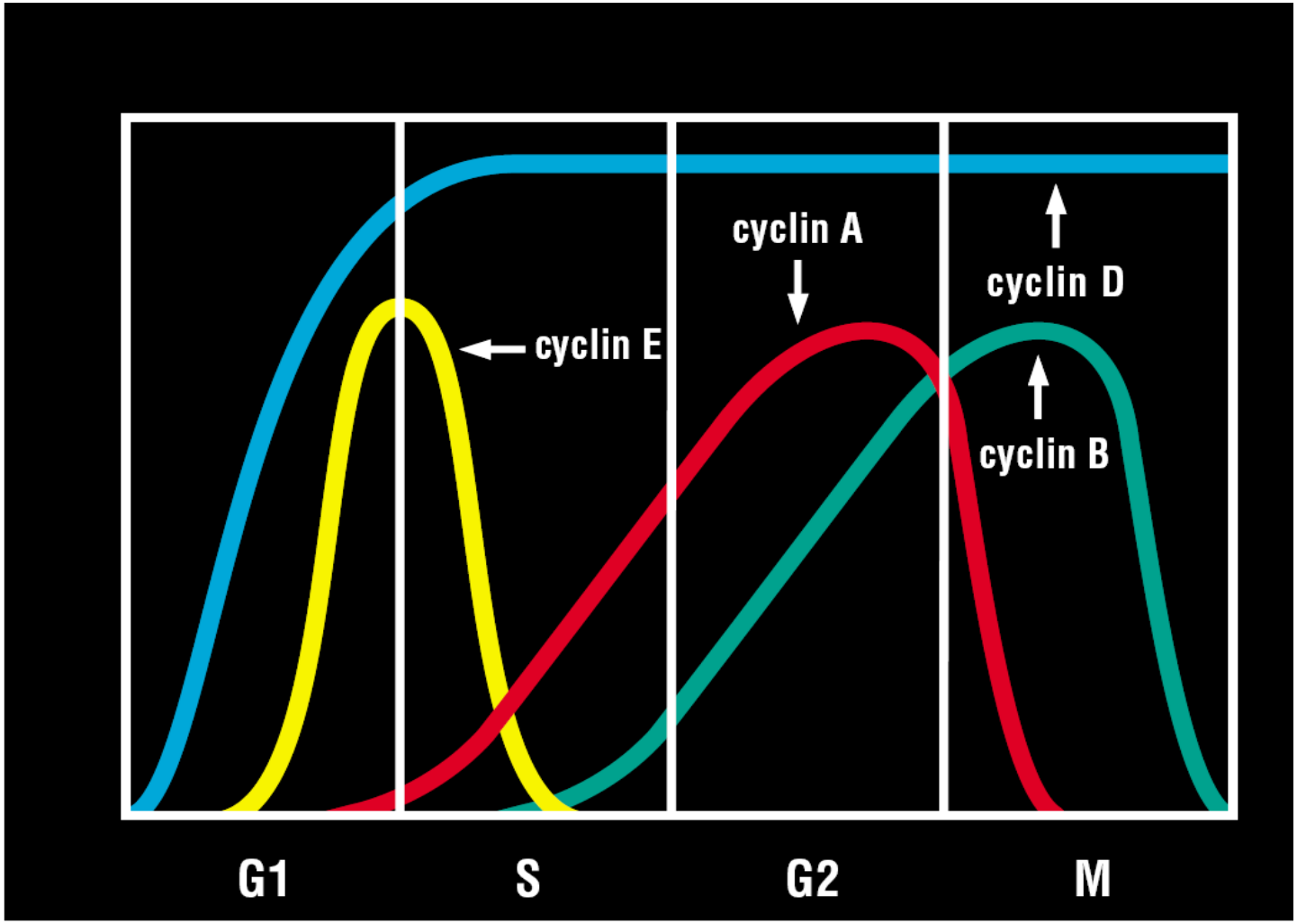
cell cycle stage	cyclin-Cdk complexes	inhibitors						
		p15	p16	p18	p19	p21	p27	p57
G1	cyclin D-Cdk4/6	+	+	+	+	+	+/–	+/–
G1/S	cyclin E-Cdk2	–	–	–	–	+	+	+
S	cyclin A-Cdk2	–	–	–	–	+	–	+
G2/M	cyclin B-Cdc2	–	–	–	–	+	–	–



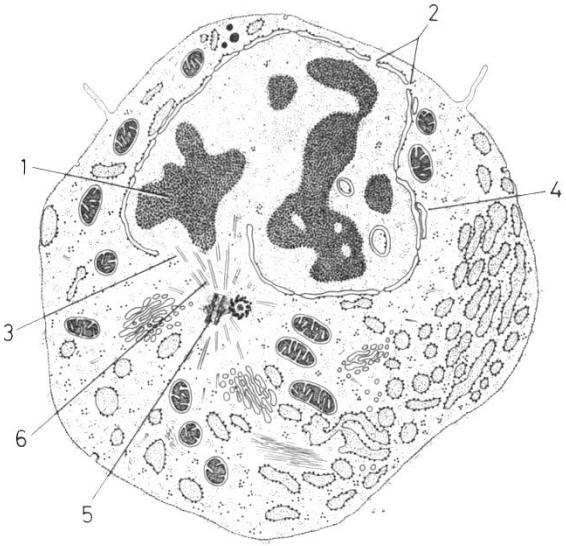
Modified from the catalogue ofSanta Cruz Biomedicals, USA

Cell division 11

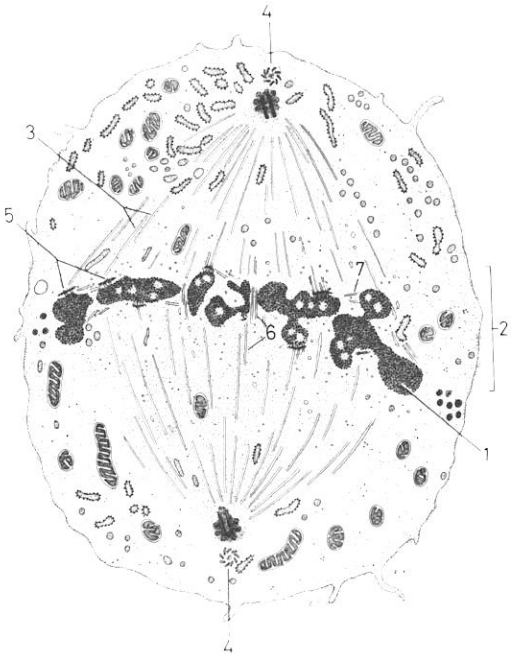
Periodicity of cyclin expression



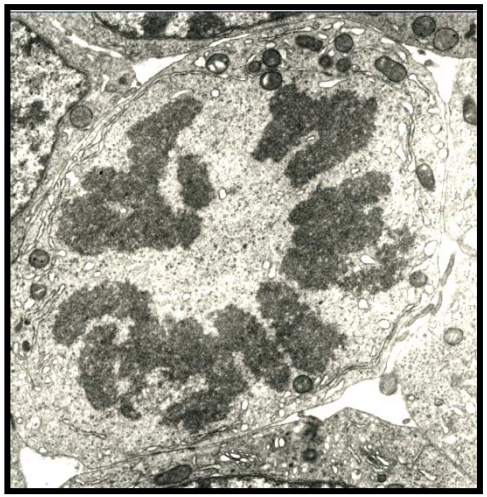
Cell division 12



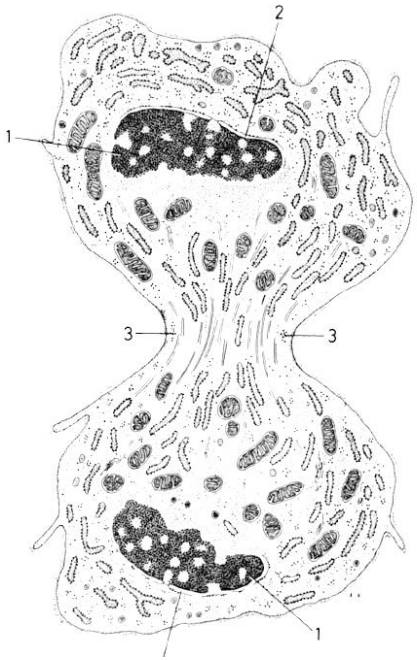
prophase



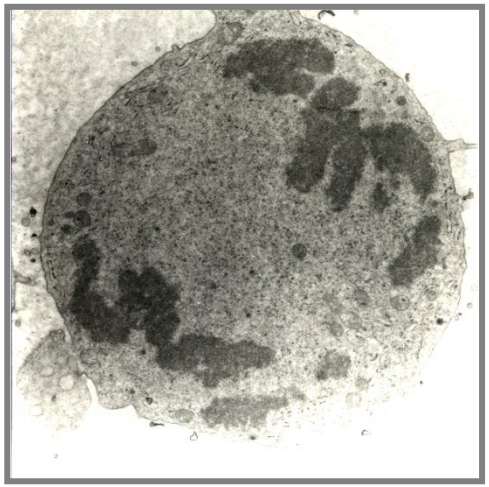
metaphase

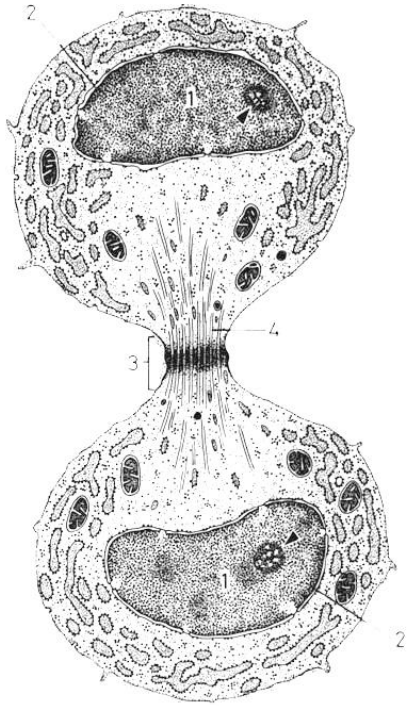


Cell division 13



anaphase - telophase





telophase

Thank you for your attention !