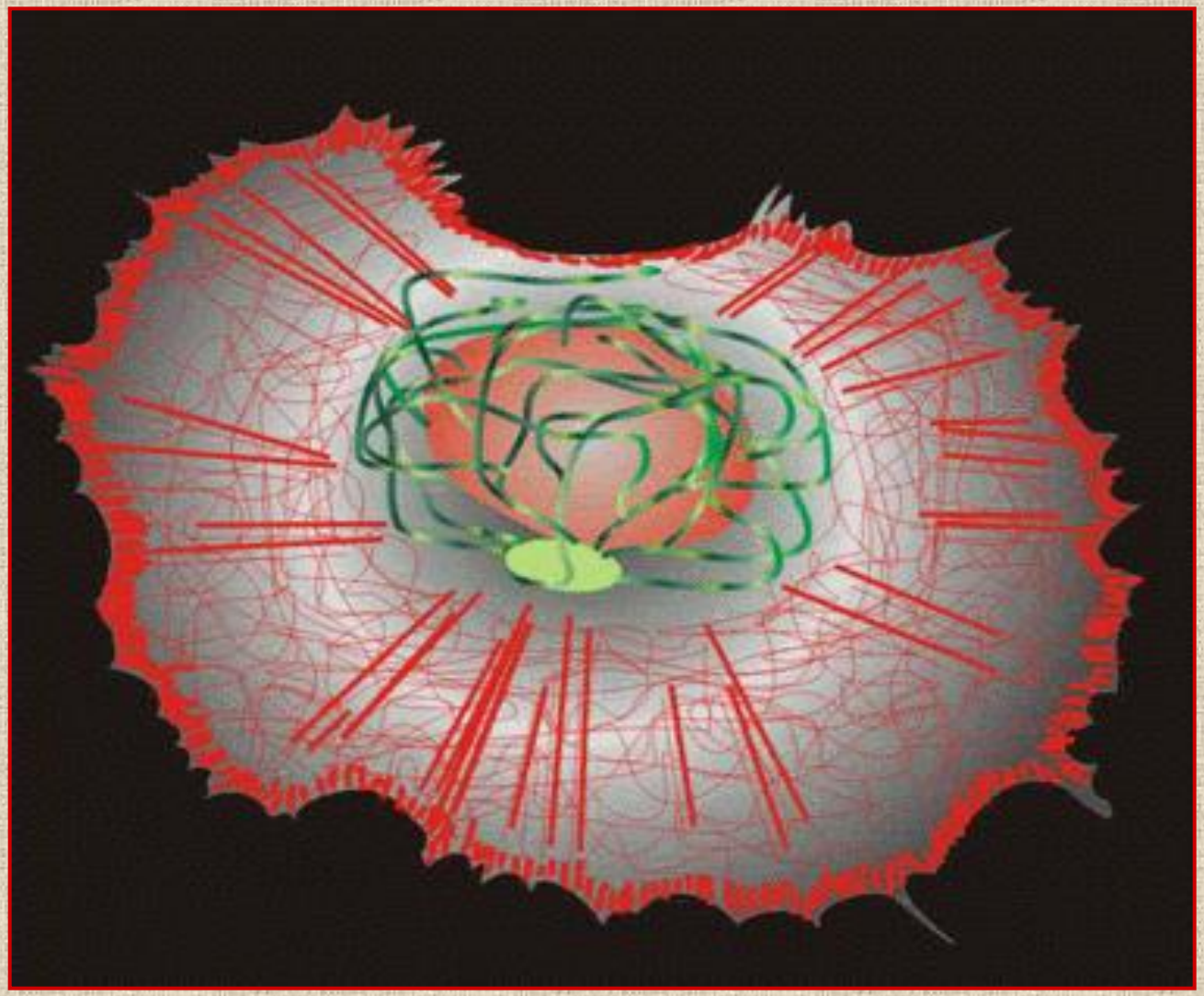
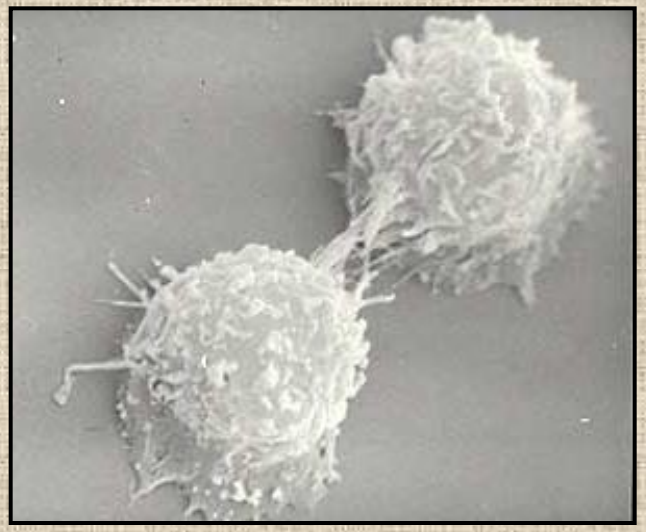
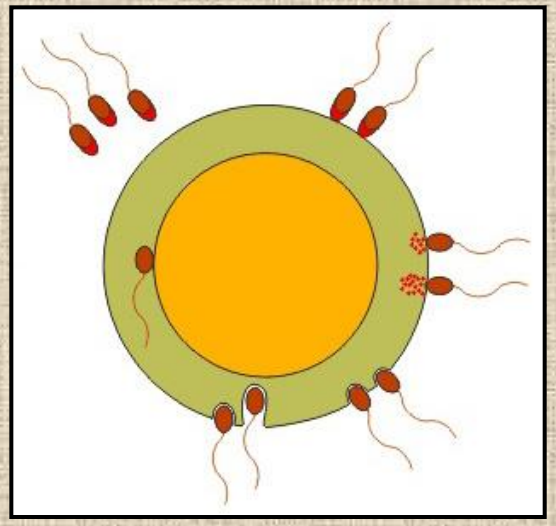
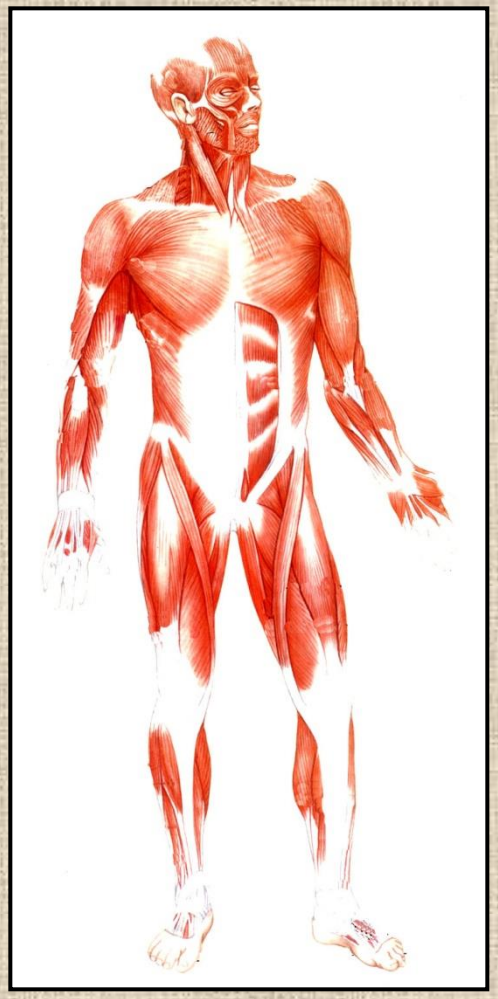
# CYTOSKELETON

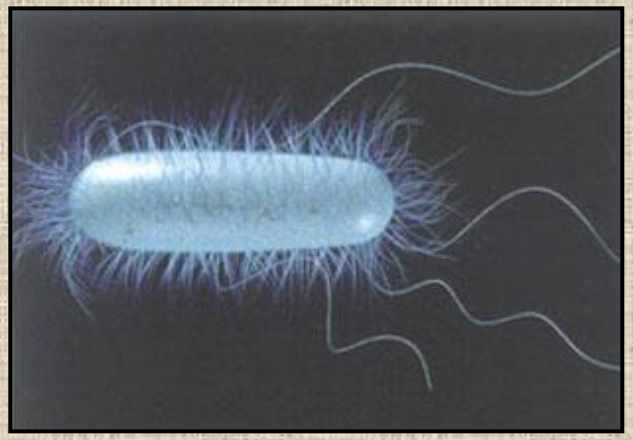


Eukaryotic cells are capable of changing their shape, moving organelles, moving from place to place.
This requires **network of protein filaments** placed in the cytoplasm and known as the **cytoskeleton**.



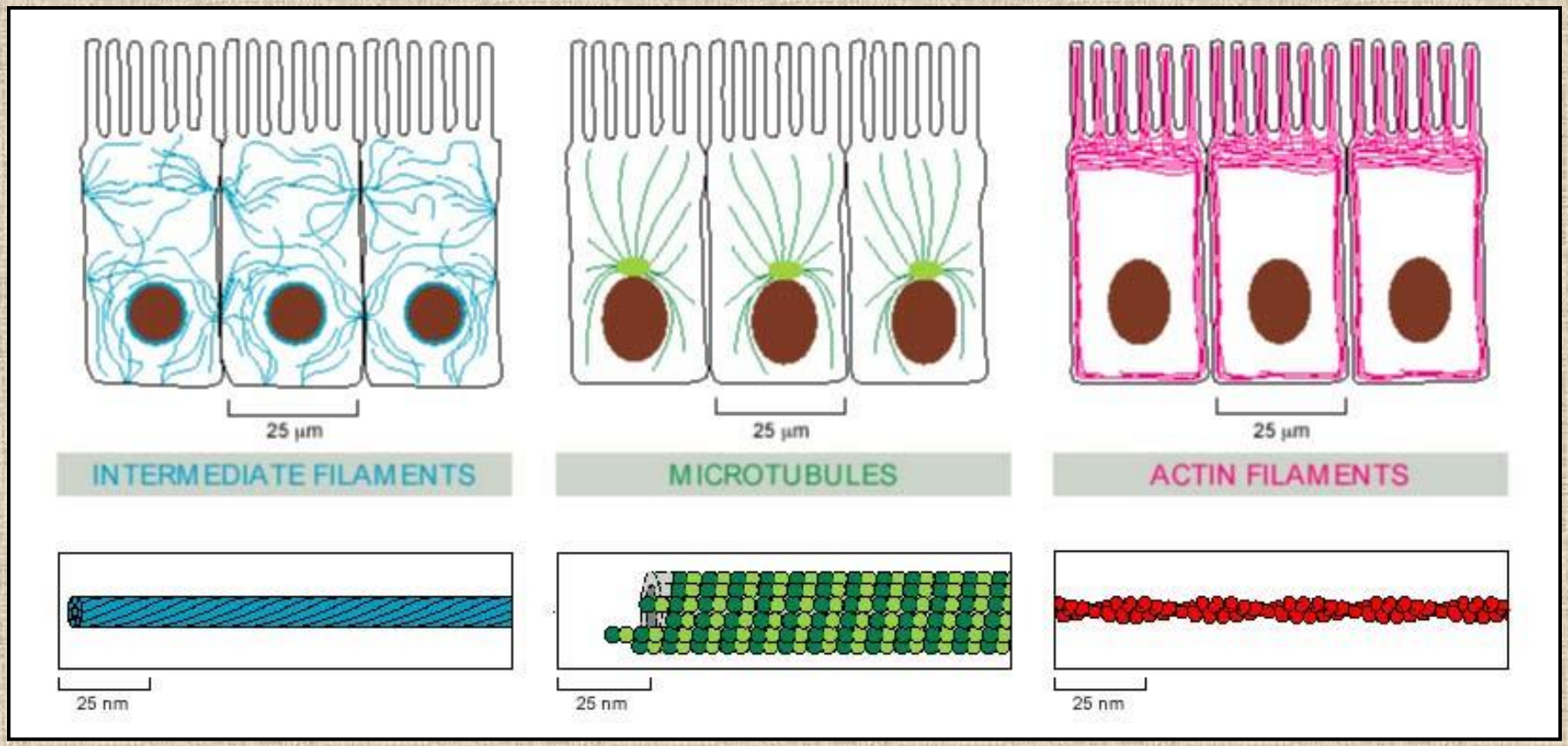






# CYTOSKELETON

- **microtubules**……(**25 nm in diameter**)
- **intermediate filaments**……....(**10 nm**)
- **actin filaments** (microfilaments)….(**7 nm**)



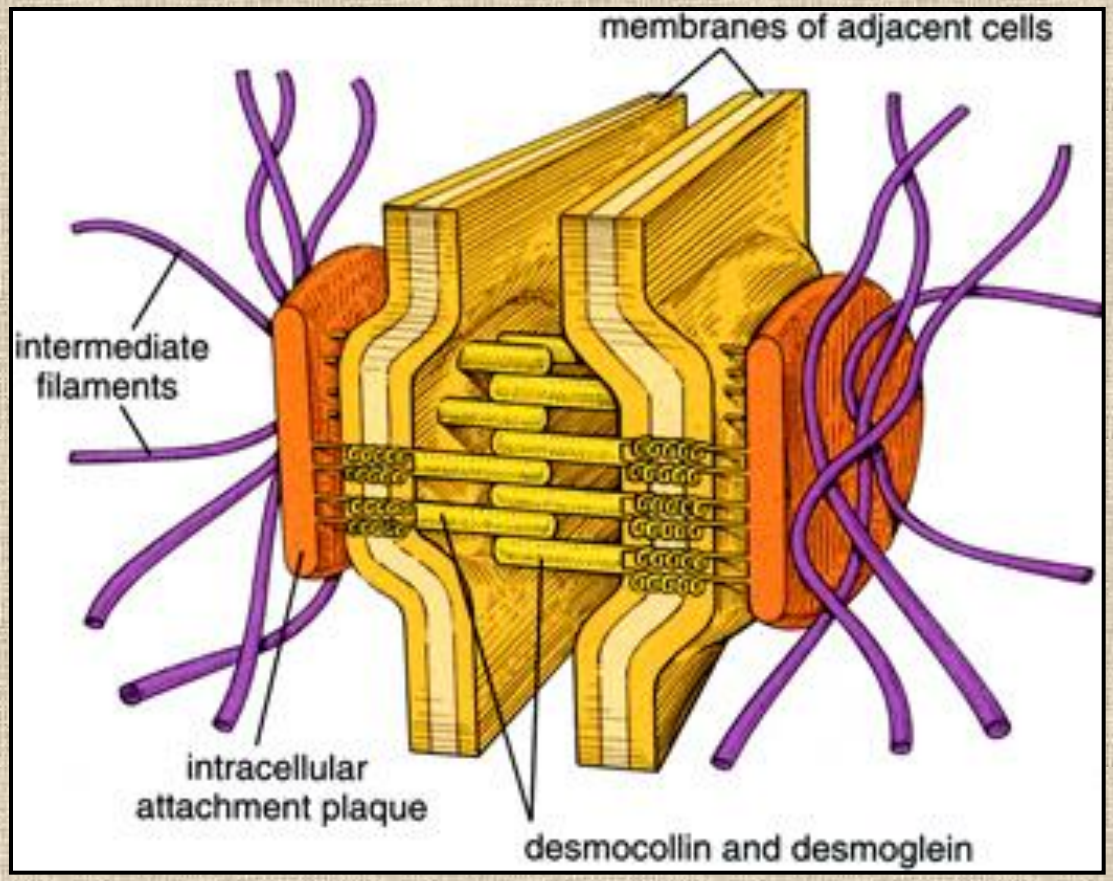
# INTERMEDIATE FILAMENTS

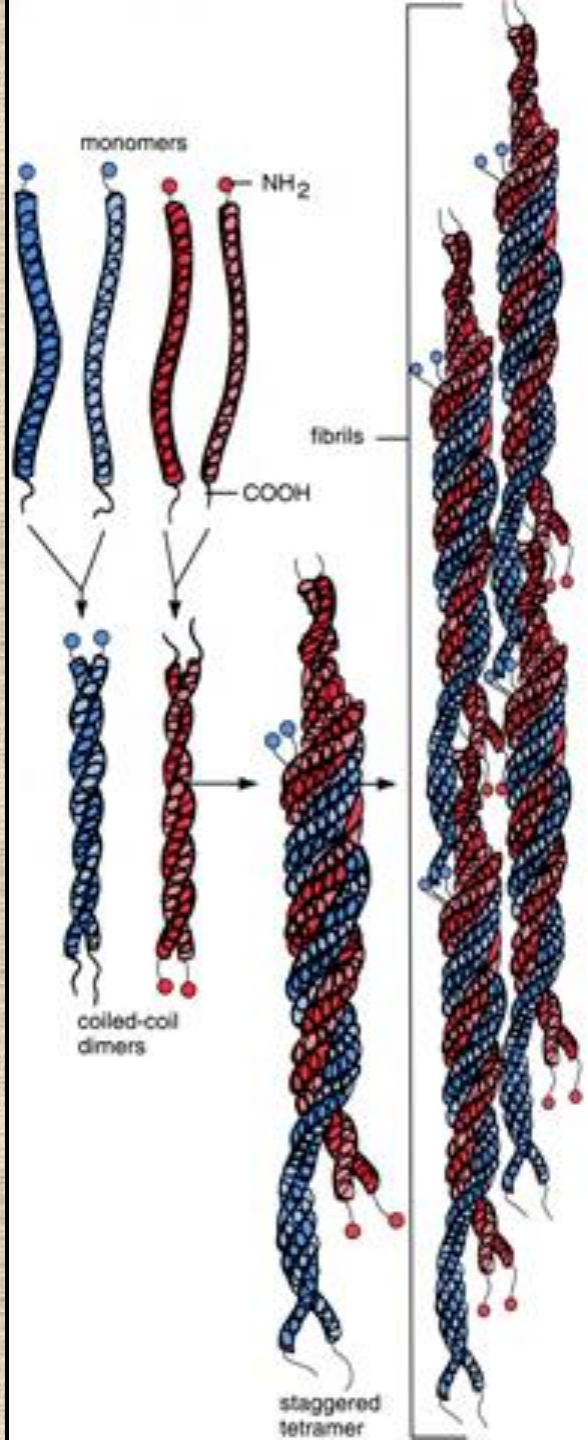
**Size: 10 nm** in diameter, variable length

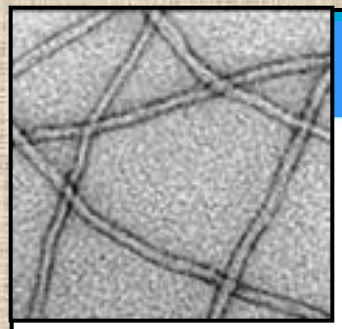
**Structure:** rope-like **fibrous proteins**

**alpha-helical region** - **coile-coil dimer** - **tetramer** - **filament** (8 tetramers)

- least dynamic of cytoskeletal filaments





## Types of intermediate filaments

CYTOPLASMIC

NUCLEAR

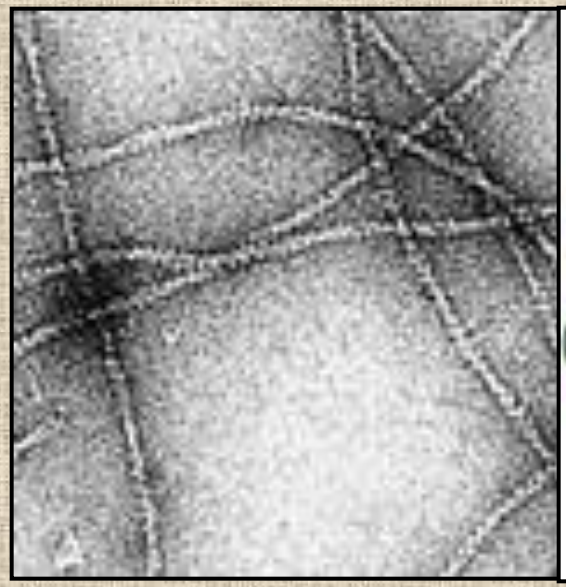
| keratins | vimentin and vimentin-related | neurofilaments | nuclear lamins |
|---|---|---|---|
| in epithelia | in connective tissue, muscle cells, and neuroglial cells | in nerve cells | in all nucleated cells |

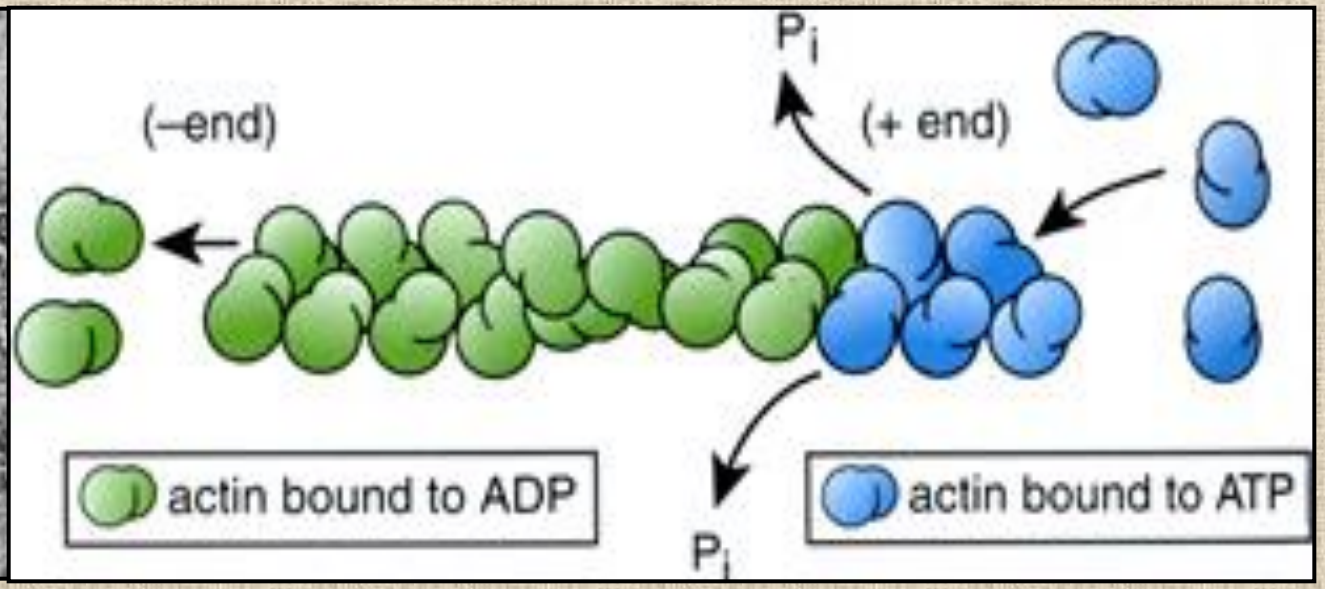
**Fuction:**

- provide cell shape and structural reinforcement (desmosomes)
- anchor organelles
- keep nucleus in place

# ACTIN FILAMENTS

**Size: 7 nm** in diameter, up to several micrometers long
**Structure:** filament made of **globular protein actin** (monomer)
- polymerize to form **dimer** - **trimer**
- **ATP** dependent
- **filament is polar** with fast (+) and slow (-) growing end
- **dynamic instability**



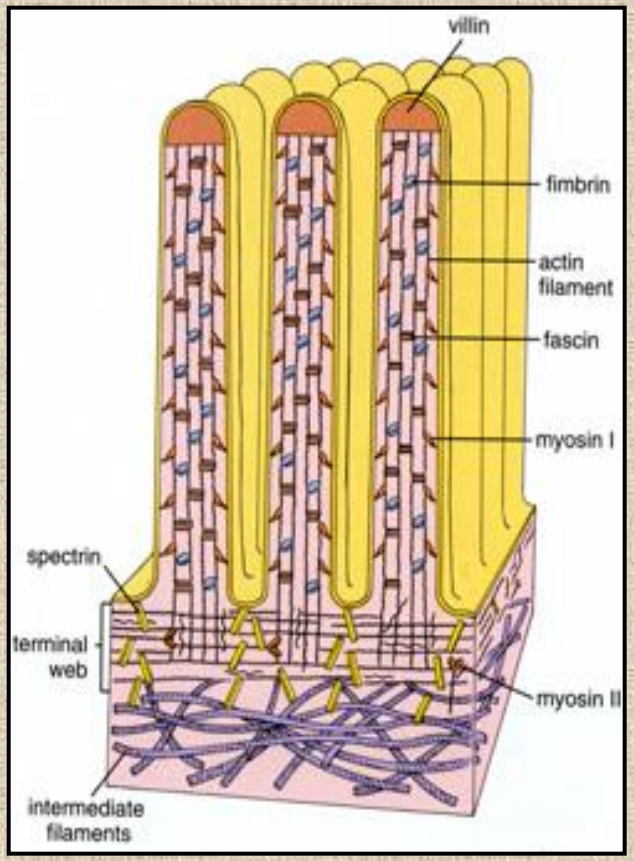


## Molecular motors:

- ✓ myosin I
- ✓ myosin II

## Function:

- ➢ **structural** - projection of the cell (villi of epithelial cell), polymeration of actin in acrosome helps sperm cells to perforate egg during fertilization
- ➢ **movement**
  - amoeboid
  - muscle contraction
- ➢ **mitosis** - contractile ring

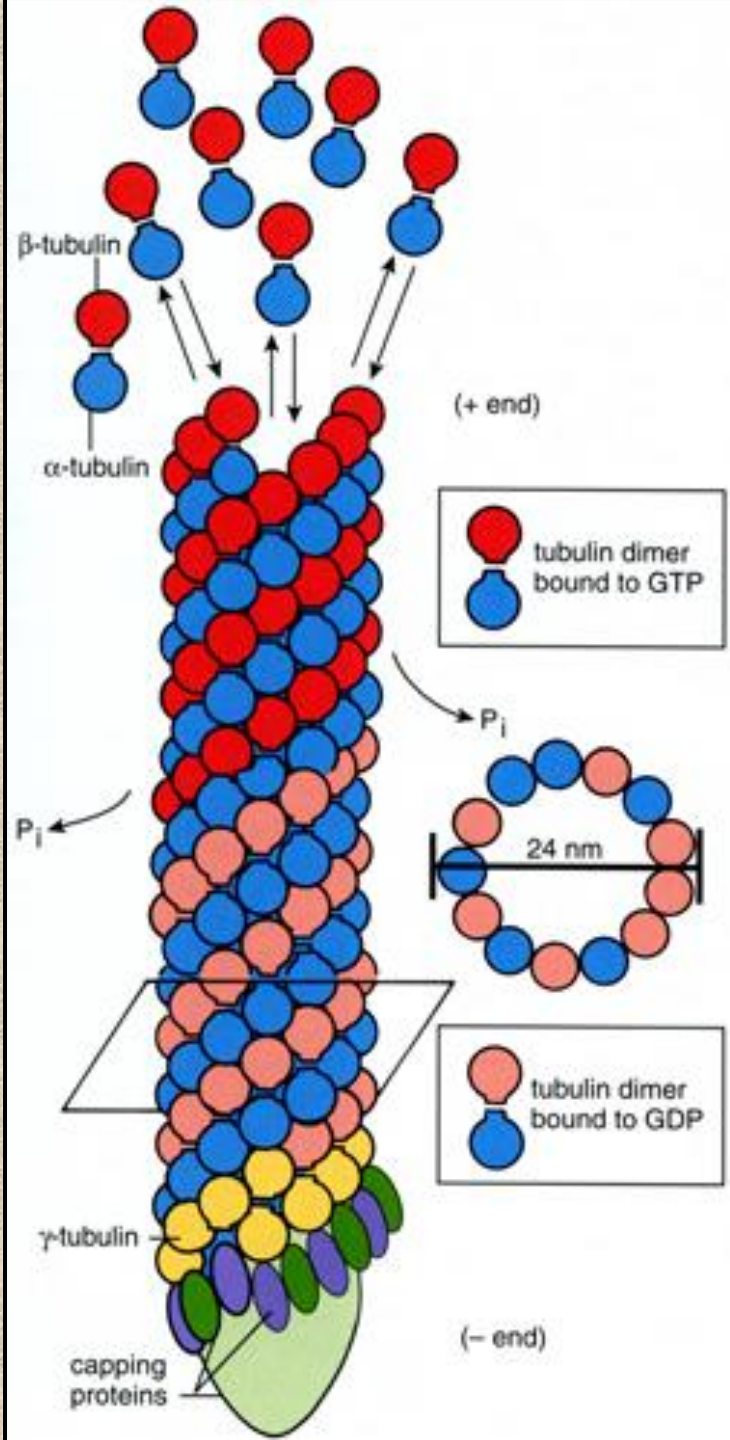


# MICROTUBULES

**Size: 25 nm** in diameter, up to several μm long

**Structure:** tubes of **globular protein tubulin**, heterodimer **subunits α** and **β** (protomer) polymerize into microtubule (13 protomers)

- **GTP** dependent
- **polar**: +end (periphery) polymerizes faster, than -end (center)
- **dynamic instability**



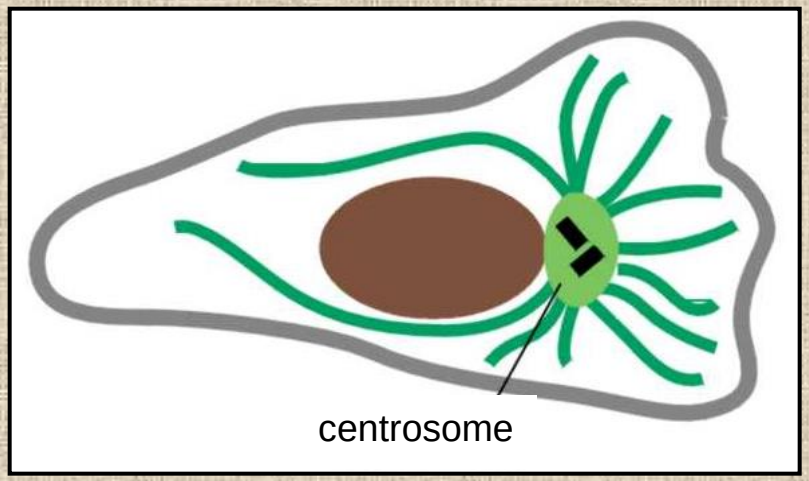

Draw centrosome and mitotic spindle

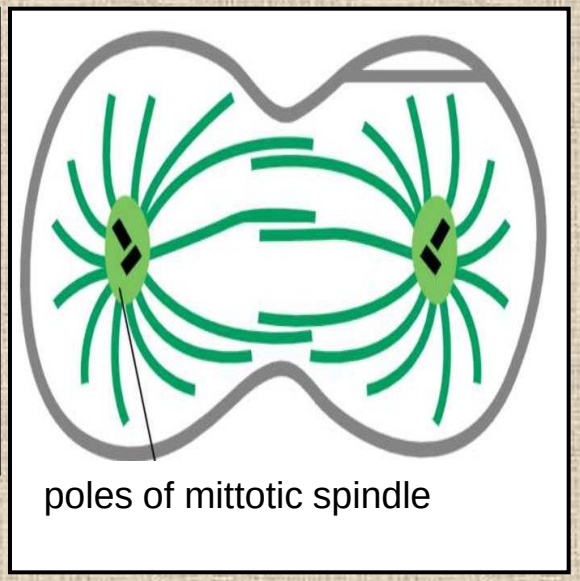
## Microtubule organizing centers:

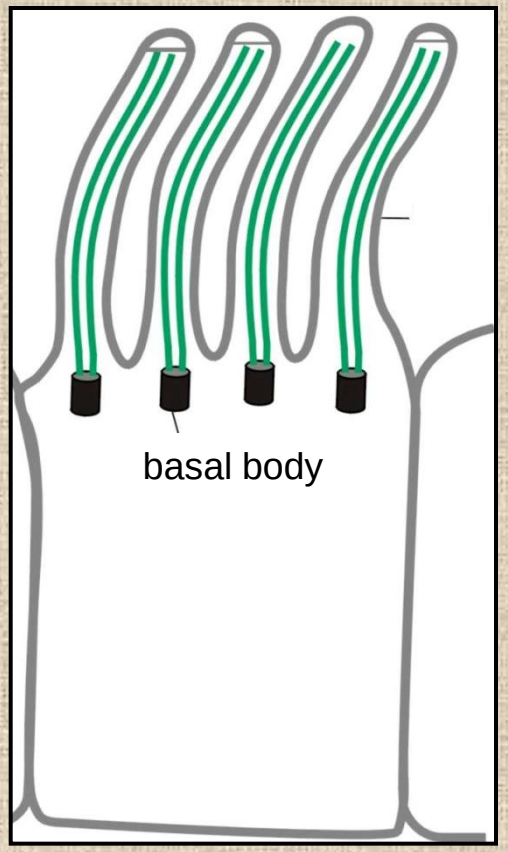
- ✓ **centrosome**
- ✓ **mitotic spindle**
- ✓ **basal body**

## Function:

- ➢ maintain the **cell shape**, anchor organelles
- ➢ **movement** - flagellar, ciliary, intracellular
- ➢ **mitosis** – mitotic spindle



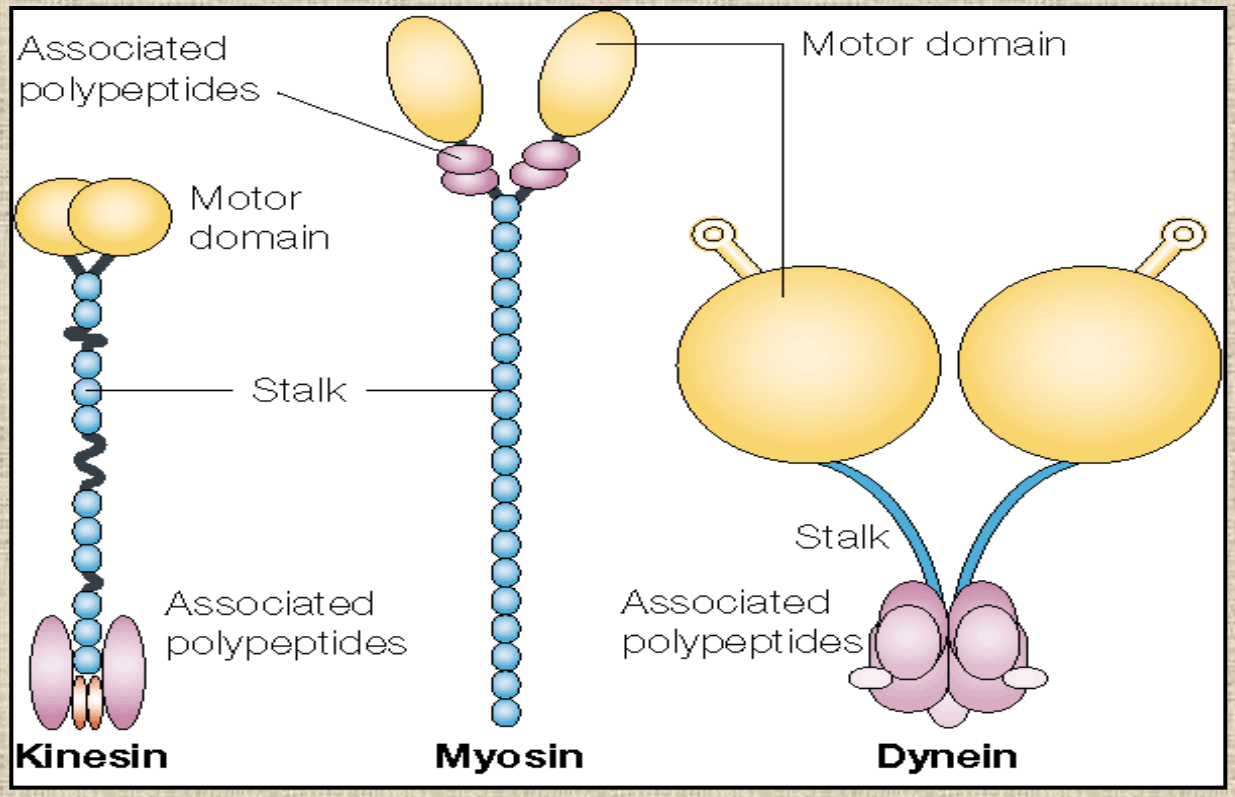
# The princip of movement

- transformation of chemical energy into mechanical

**Molecular motor (motor protein):**

**motor (head) domain** - 1 polypeptid chain, **ATPasa** activity (releases energy by hydrolysis of ATP)

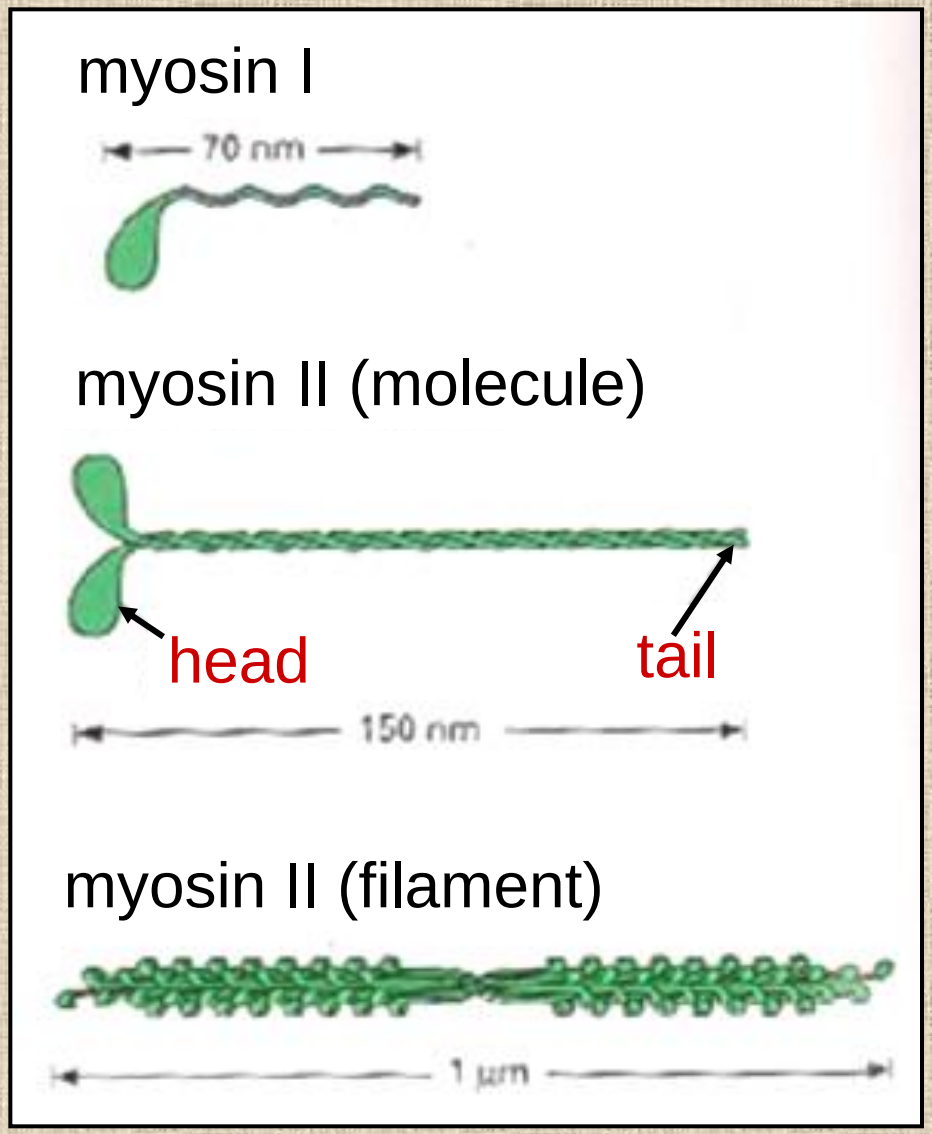
**tail (stalk) domain** - other polypeptid chain, binding site for molecules or cell structures



# Motors associated with actin filaments:

**MYOSIN I**: one motor domain, all types of cells

**MYOSIN II:** two motor domains, filament in muscular cells

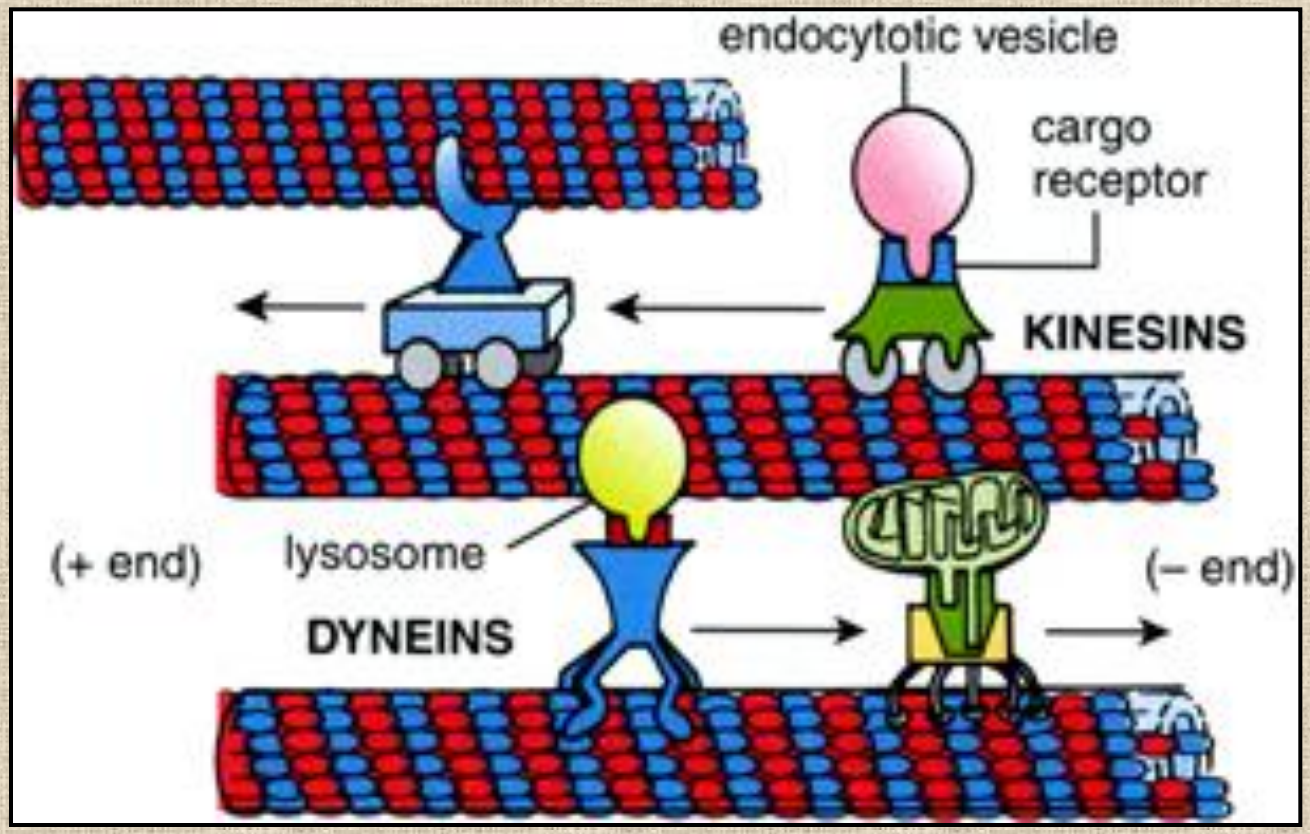


# Motors associated with microtubules:

- microtubules are track for microtubule-based motor proteins that distribute vesicles throughout the cell

**DYNEIN**: **minus-end**-directed

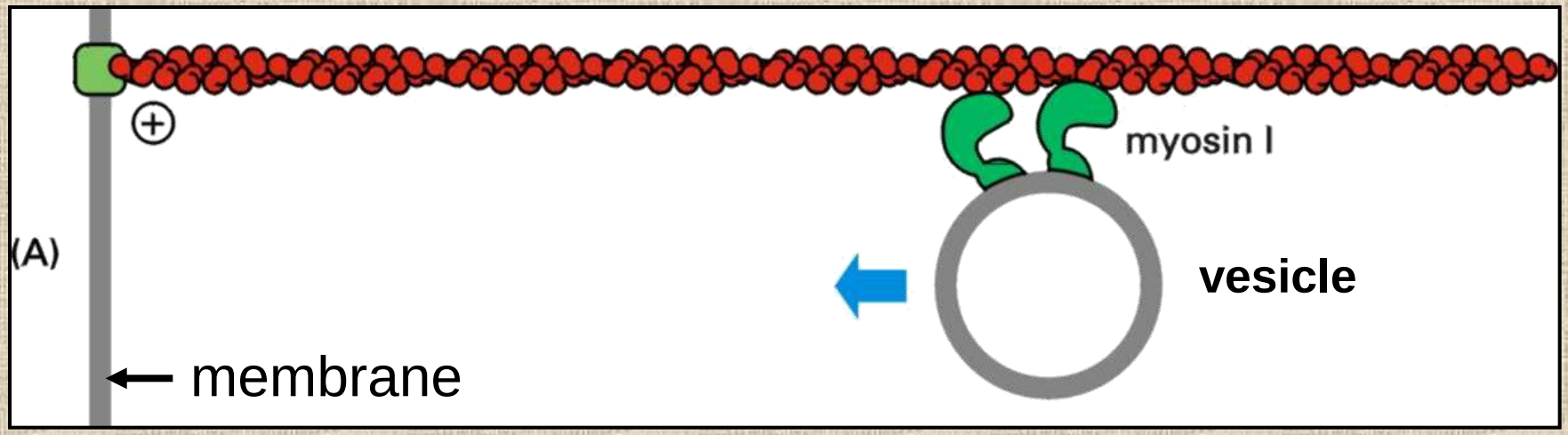
**KINESIN: plus-end**-directed motor



# Types of motor movement

## A) cytoskeletal structure is fixed

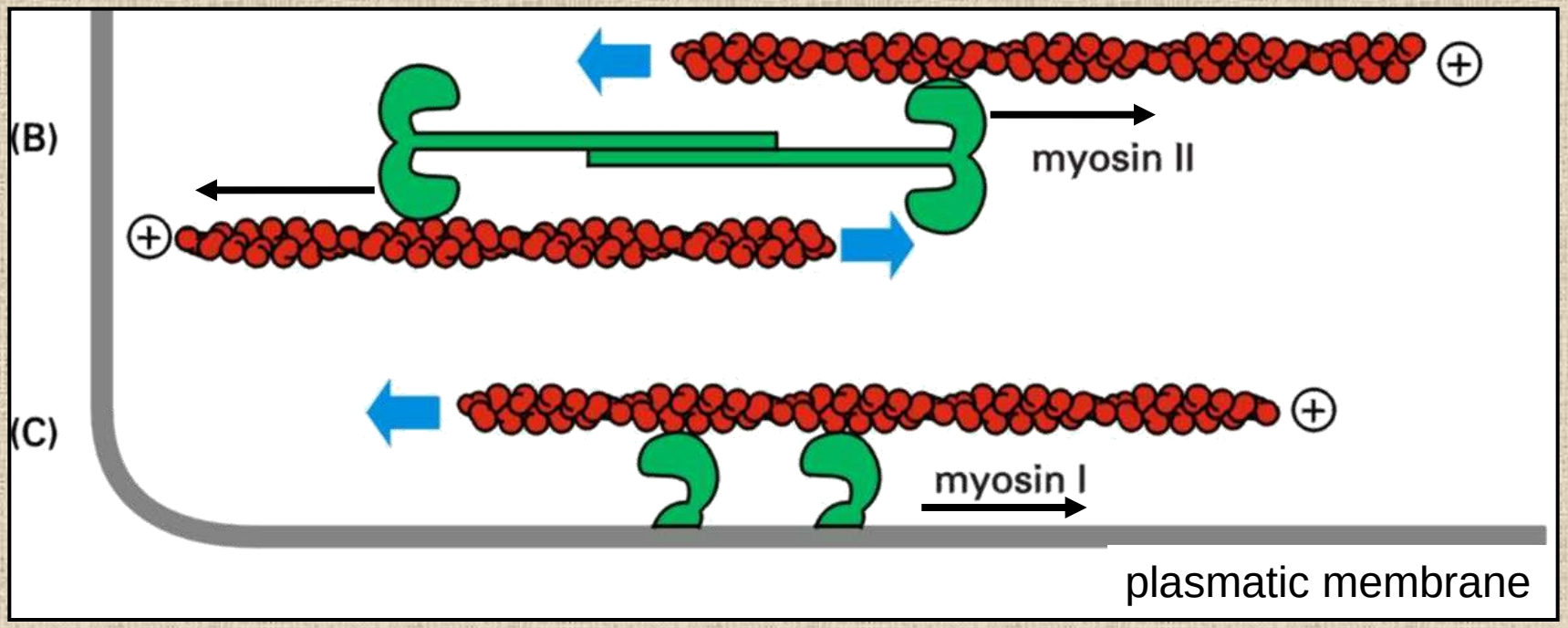
- motor domain binds to cytoskeletal structure that is fixed
- hydrolysis of ATP
- motor domain changes conformation and moves along cytoskeletal structure
- cargo moves together with motor



## B) sliding

- motor is fixed with tail domain to one cytoskeletal structure while its motor domain contacts other cytoskeletal structure ⟶ sliding of cytoskeletal structures

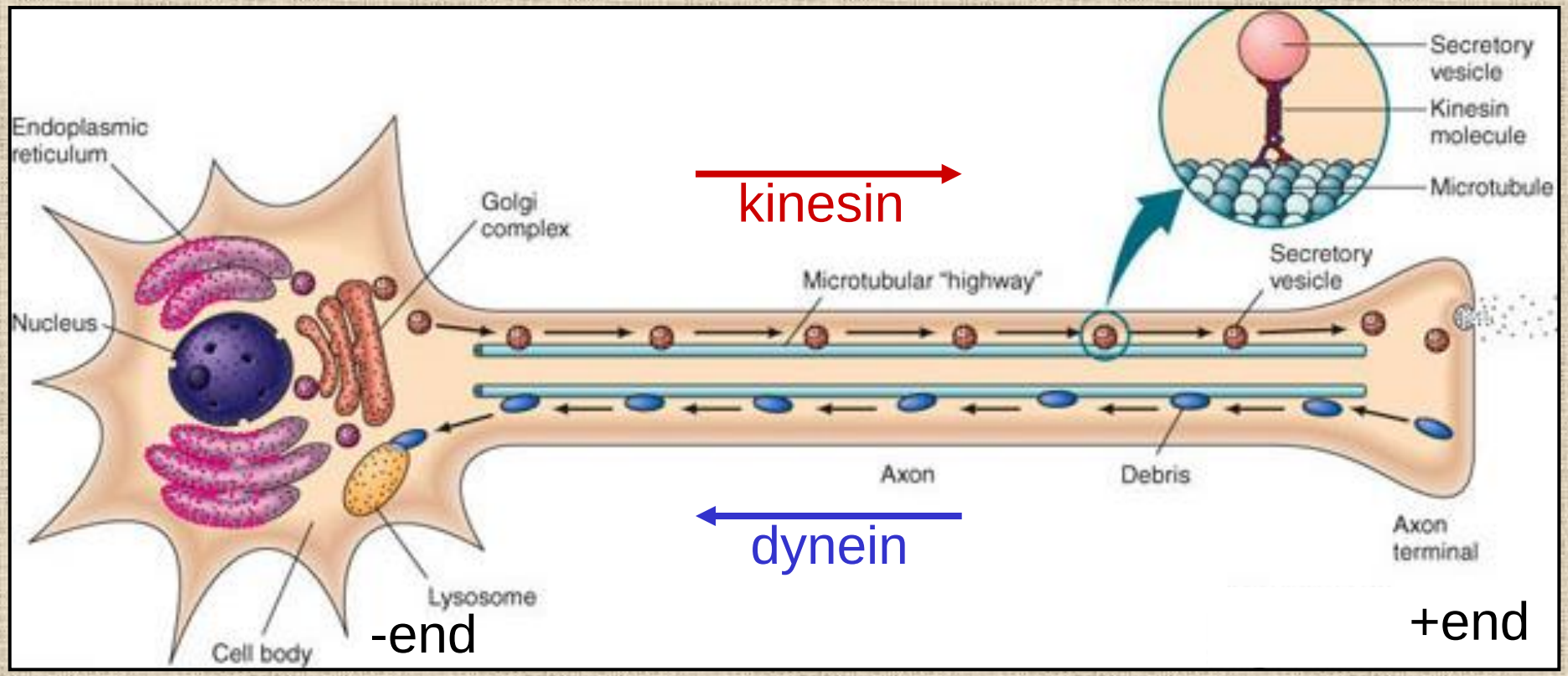
## C) motor is fixed




Animation of molecular motor movement:
http://www.susanahalpine.com/anim/Life/kinesin.htm
http://www.sci.sdsu.edu/movies/actin_myosin_gif.html
Motors in mitosis: http://faculty.plattsburgh.edu/donald.slish/Motors.html

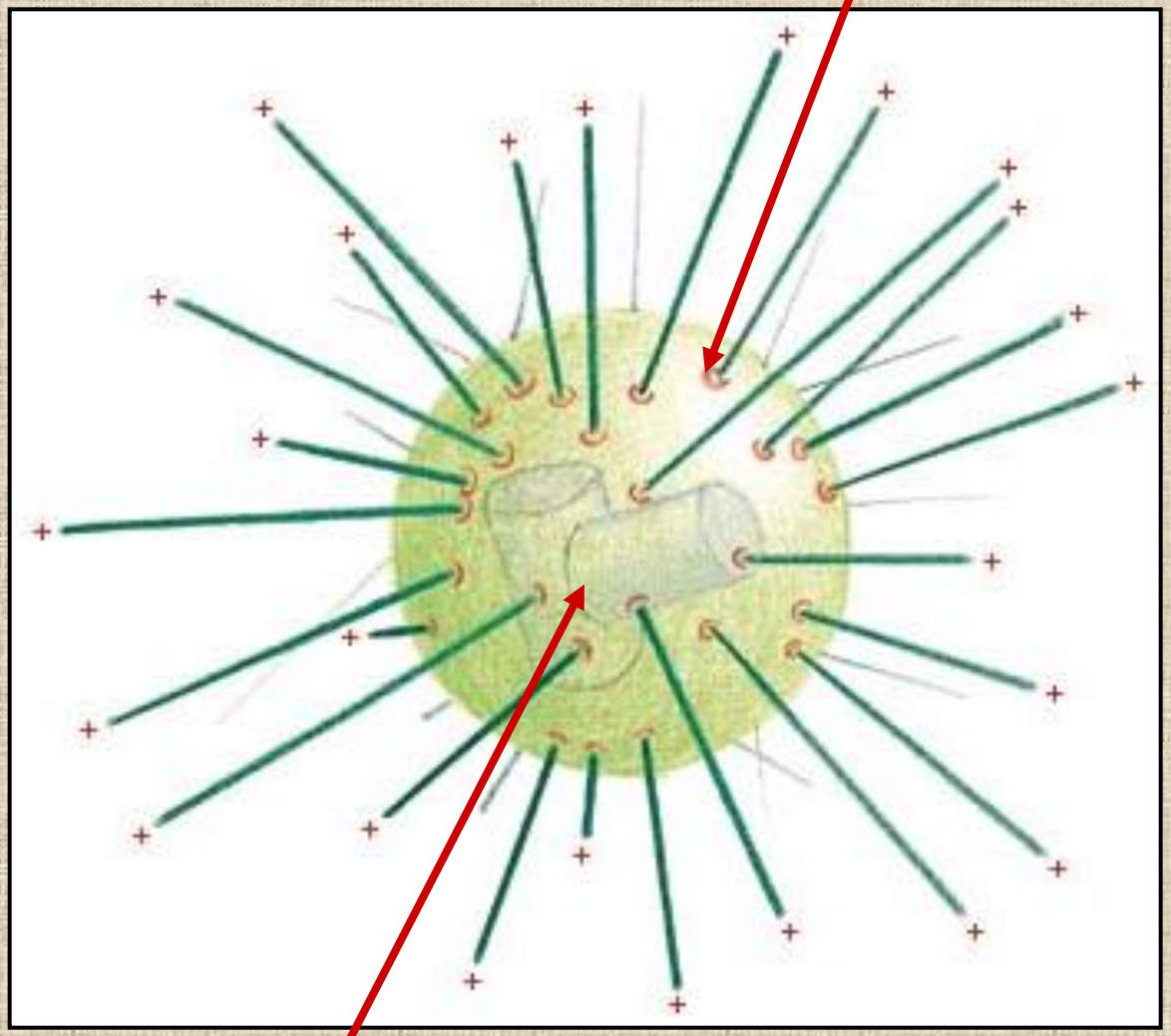
# INTRACELLULAR TRANSPORT

- transport of secretory vesicle by molecular motors (**dynein**, **kinesin**) along **microtubule** „highway“
- secretory pathway, in axons of nerve cells, transport of pigment in melanophores

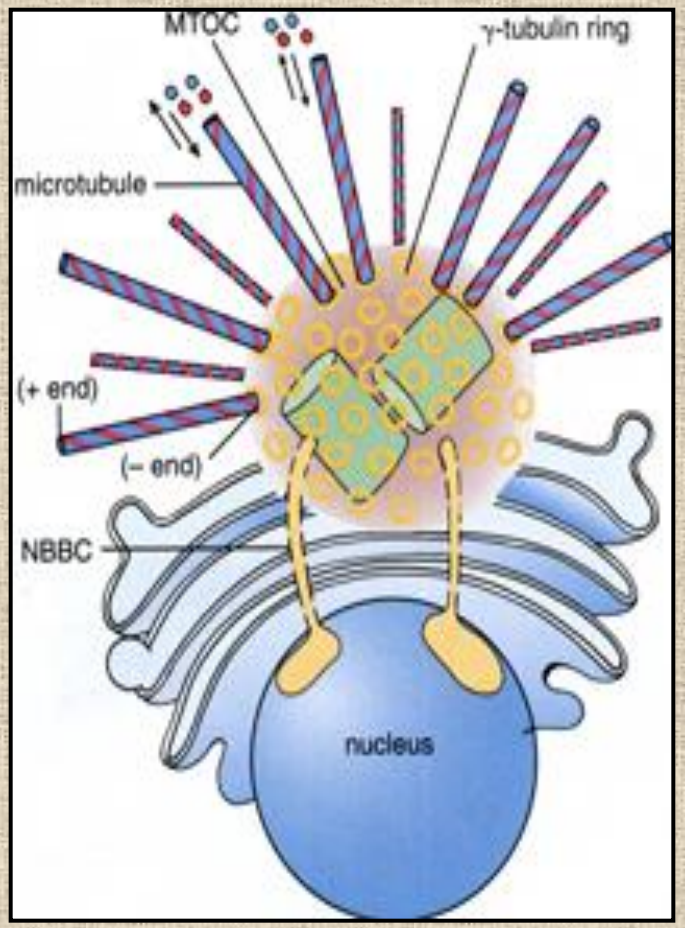


# CENTROSOME

nucleating sites
(rings of **gama-tubulin**)

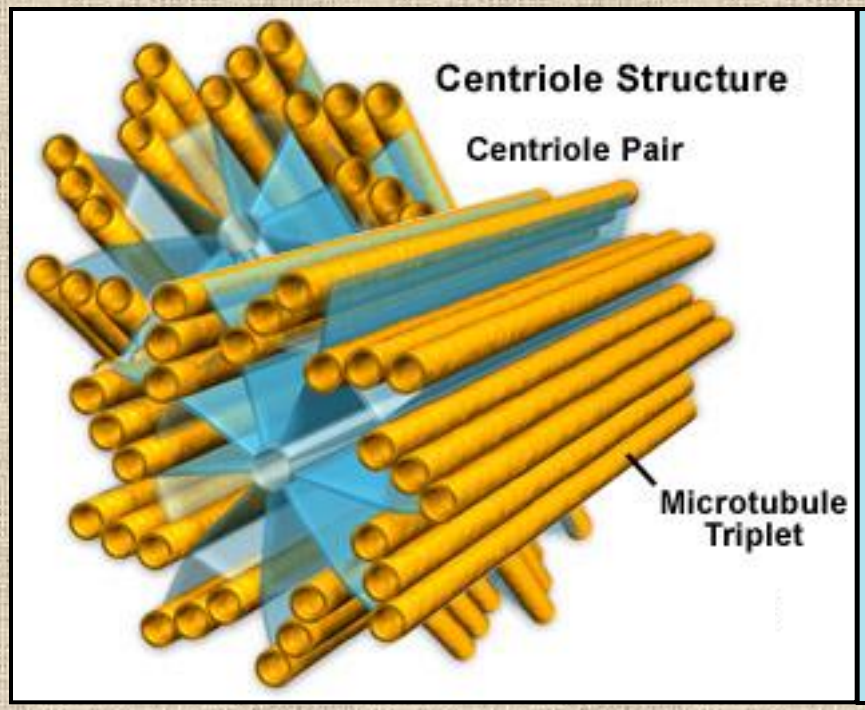


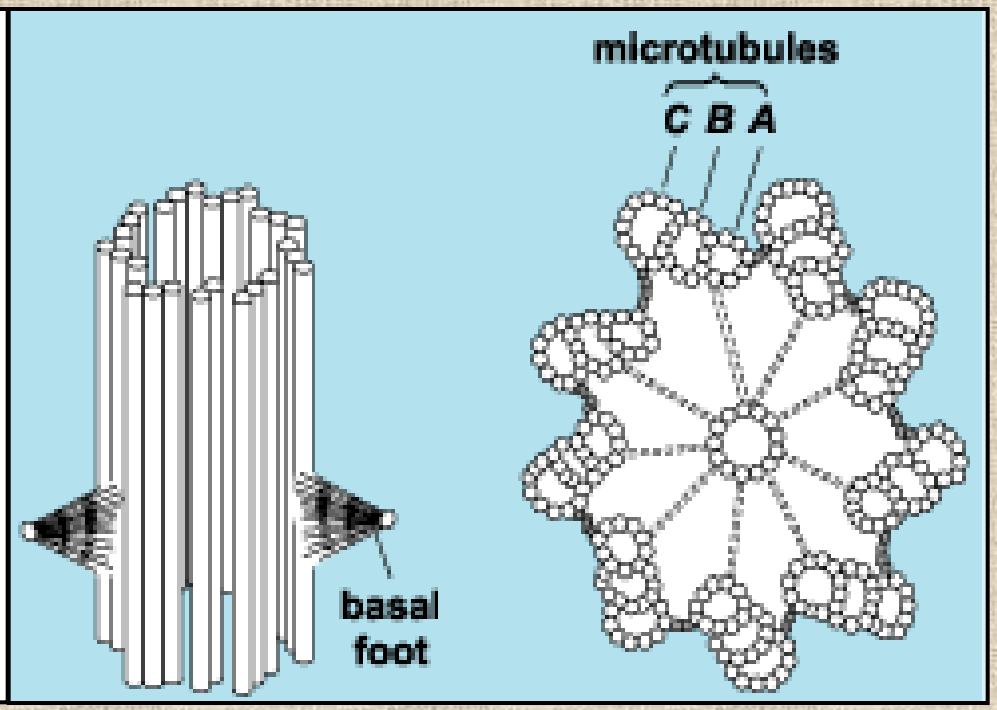
pair of centrioles



# CENTRIOLE

- made of **9 microtubule triplets** arranged in circle and linked laterally
- two centrioles (at 90 degrees) are component of centrosome, found near nucleus of animall cells in interphase
- centrioles duplicate during S phase, migrate to the opposite poles of the cell and form the organizing centers for the mitotic spindles

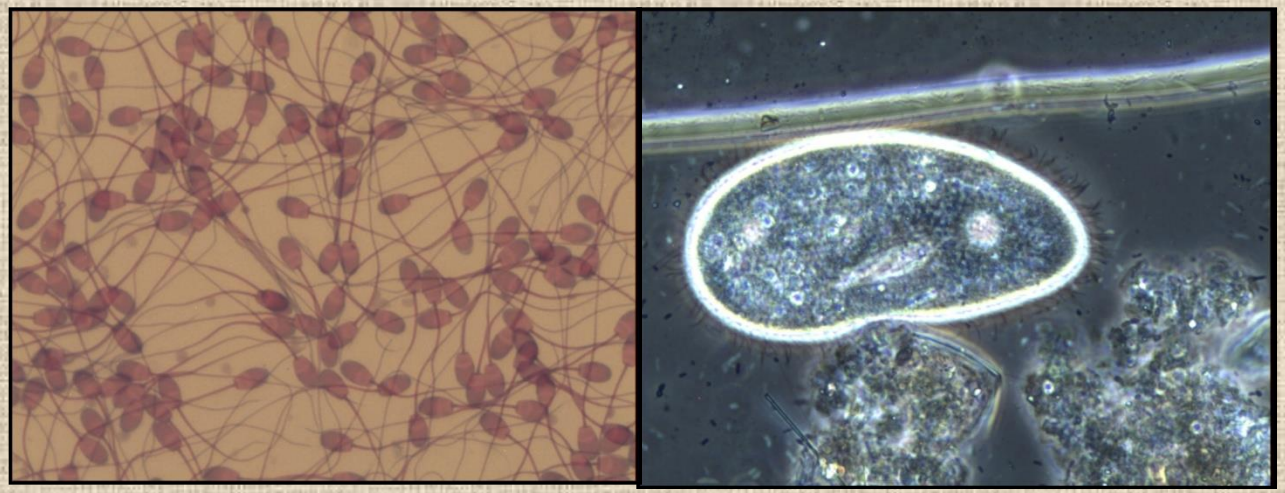


# FLAGELLA AND CILIA MOVEMENT

**Structure:**

- **basal body (base)** - made of modified centriole (procentriole)
- **axoneme** - forms the core, contains:
  - **2 central microtubules (pair)**
  - **9 periferal doublets of microtubule (A and B subunit)** - linked to each other by nexin and to the central sheath by radial spokes
- **dynein**



**flagella** = one per cell, 0.4 μm diameter, 100-200 μm long
**cilia** = many per cell, 0.4 μm diameter, 2-10 μm long

# cilium

Outer doublet microtubules

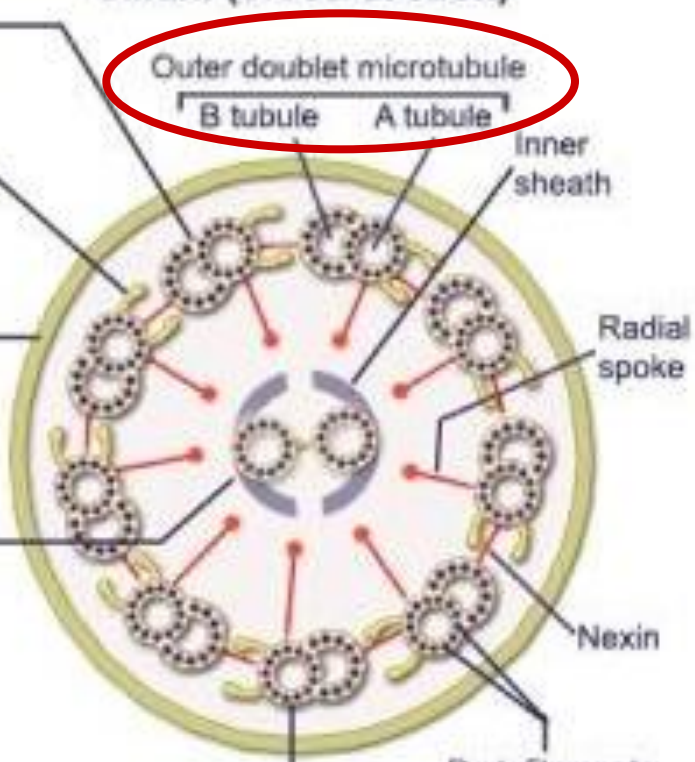
Dynein arms

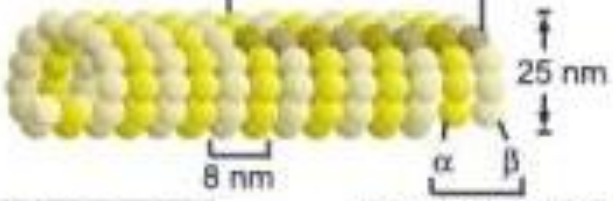
Plasma membrane

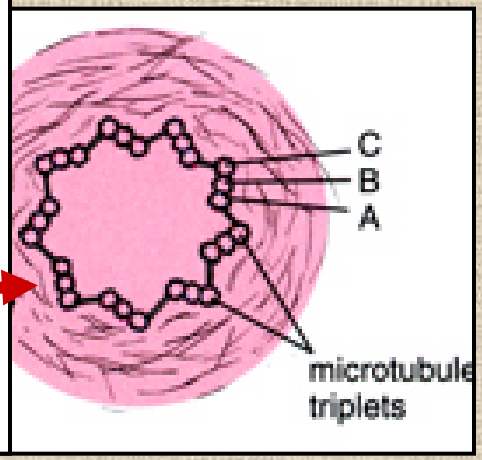
Central microtubules

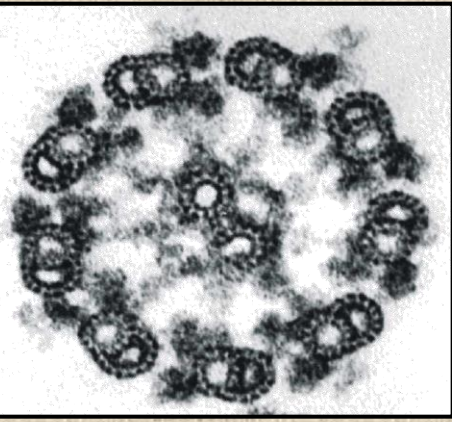
## basal body

(courtesy of D.T. Woodrow and R.W. Linck)



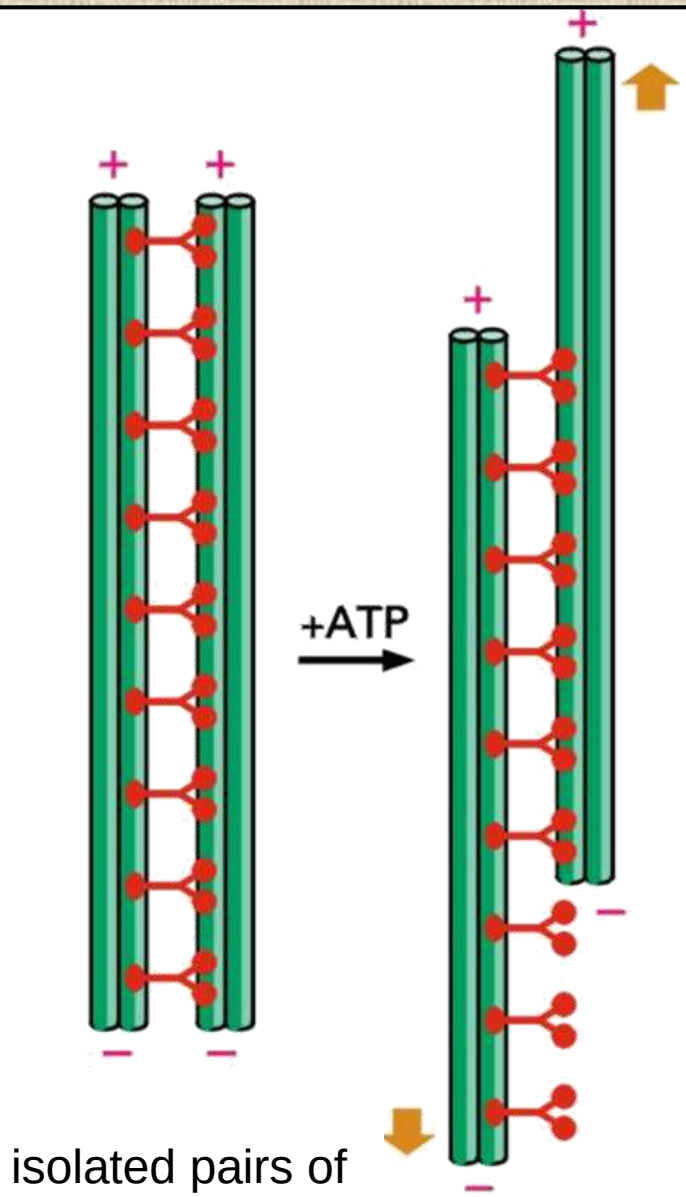



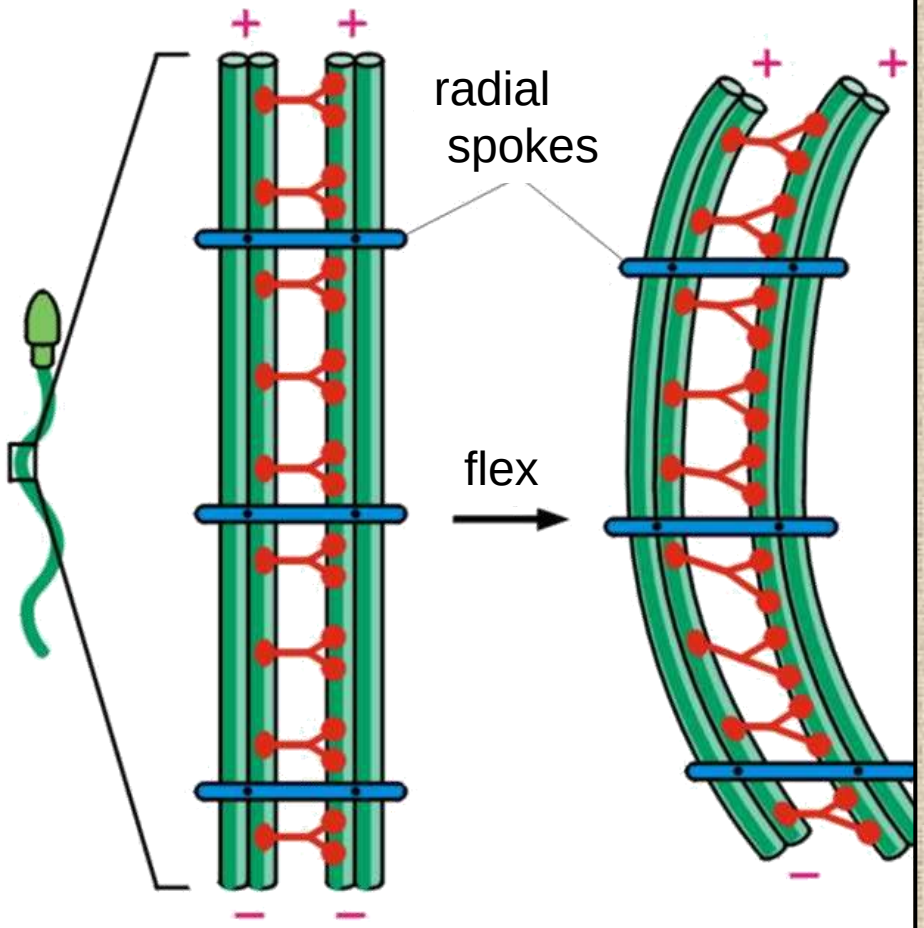



## structure

## 9 + 2

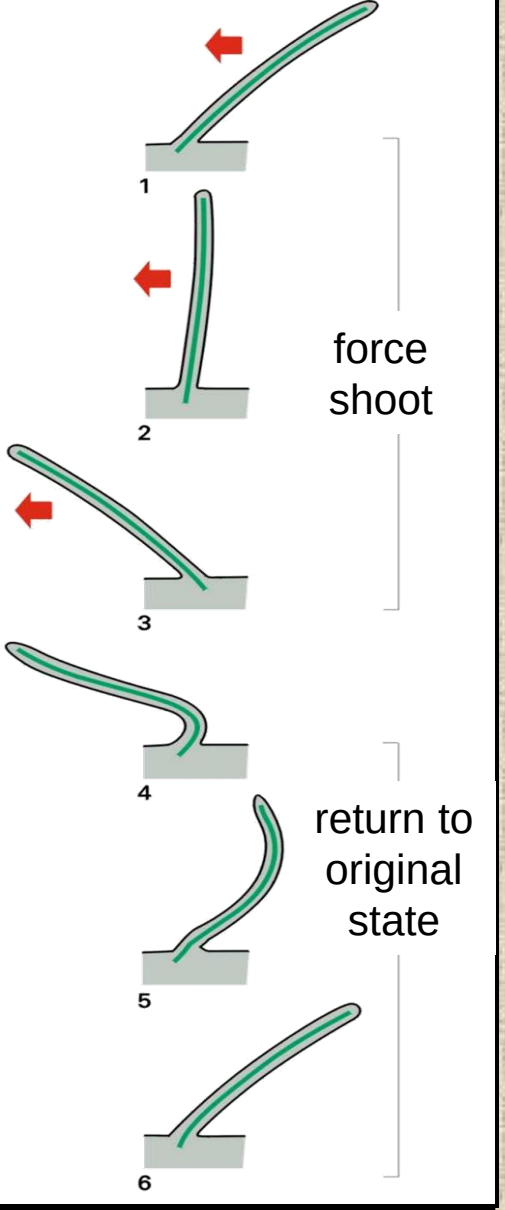
**EUCARYOTIC FLAGELLUM**

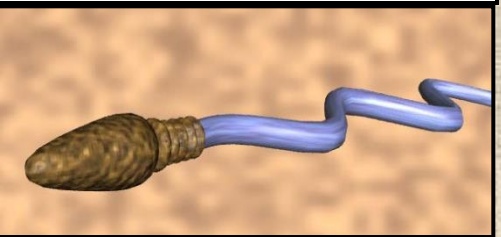



sliding of microtubules

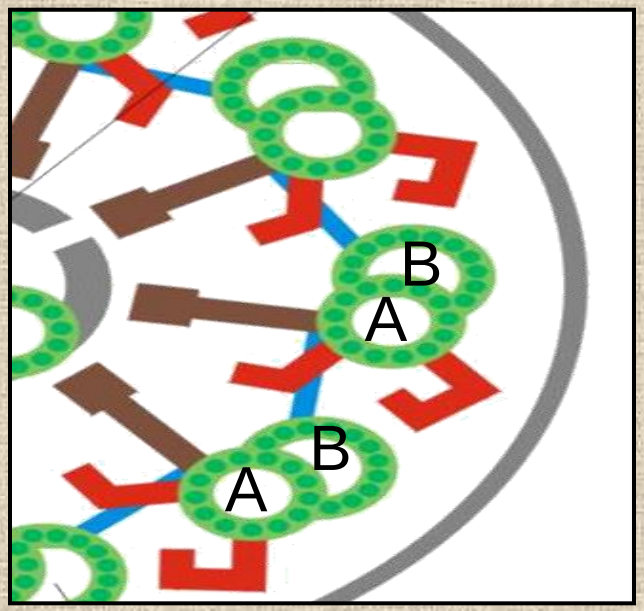
flexion of microtubules



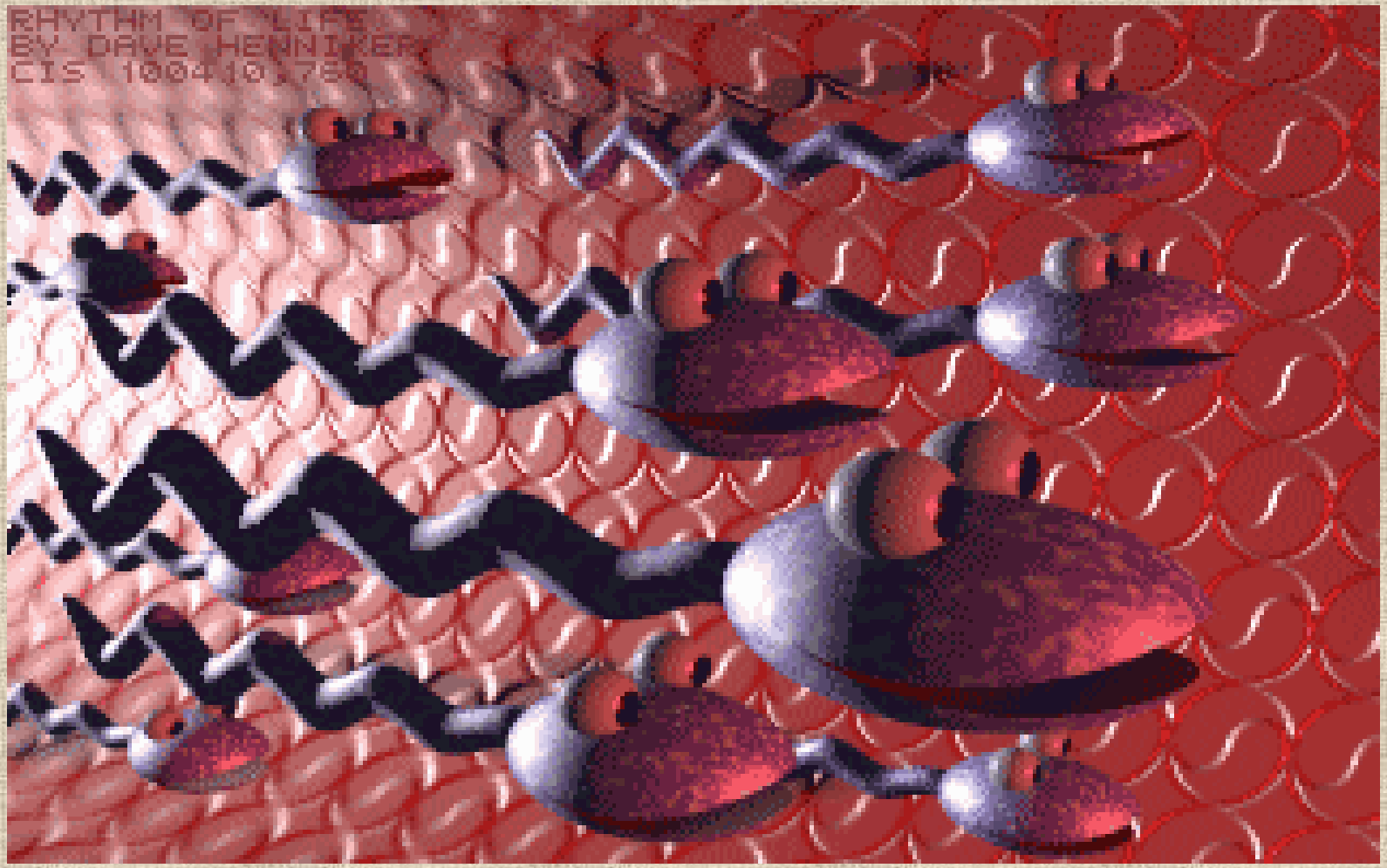
## Princip of flagellar and ciliar movement

- tail domain of **dynein** is fixed to **A** subunit of microtubule doublet
- motor domain of dynein contact **B** subunit of neighbouring microtubule doublet causing **hydrolysis of ATP**
- activated motor domain changes its conformation
- because microtubule doublets are fixed by radial spokes, they will not slide, but flex



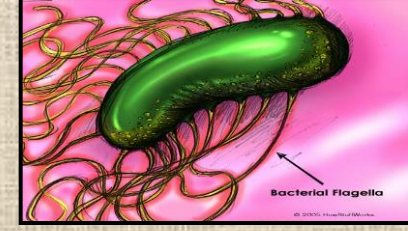
## Function:

- move things along the surface of the cell that lines lumen (respiratory, reproductive tracts)
- used in locomotion (sperm cell)




Animation of flagellar and ciliar movement: *http://programs.northlandcollege.edu/biology/Biology1111/animations/flagellum.html

# BACTERIAL FLAGELLUM




## Composition:

- helical hollow **filament** composed of the protein **flagellin**
- sharp **hook** outside of the outer membrane
- **basal body rings** - 2 in Gram-positive, 4 in Gram-negative

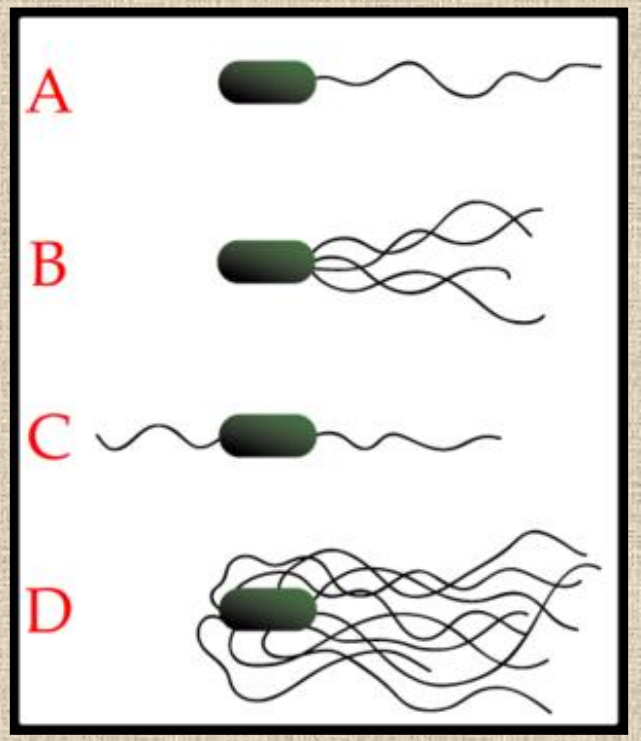
## Types of bacteria:

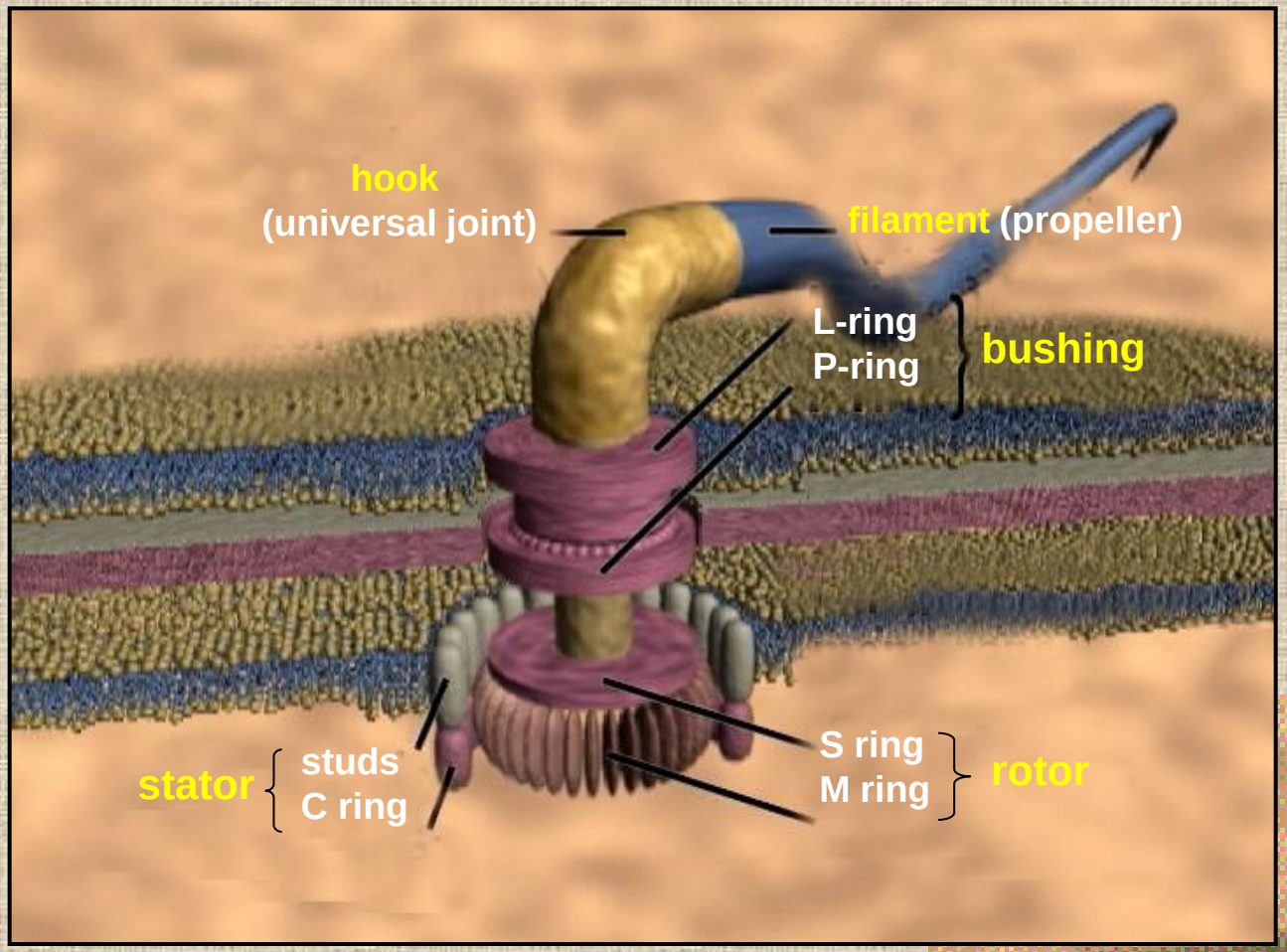
**A) monotrichous** - single flagellum

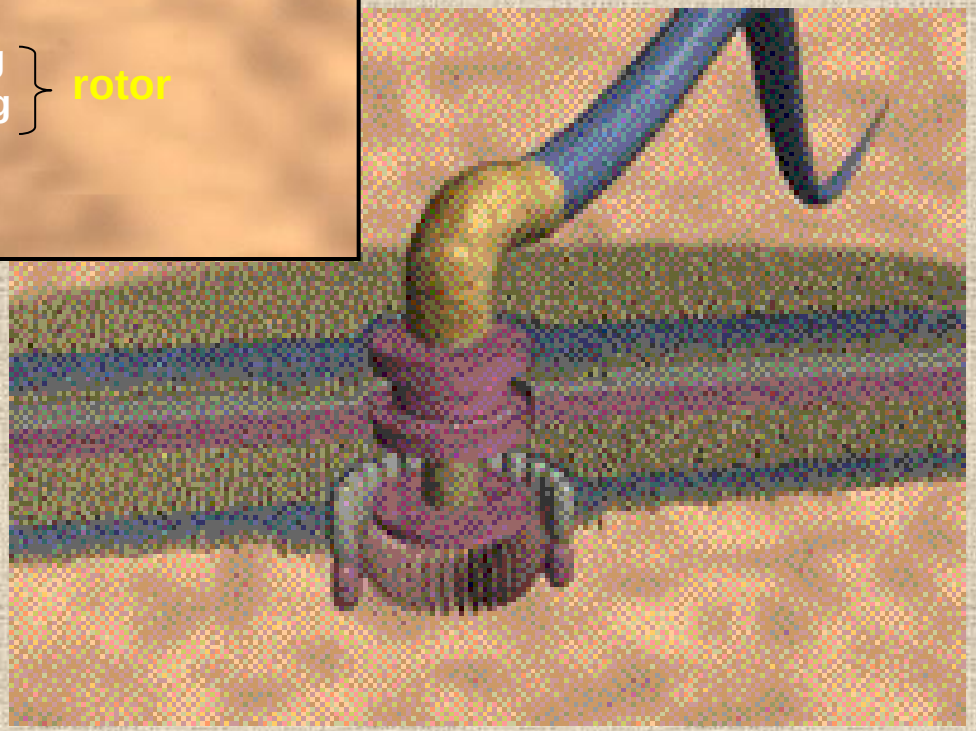
**B) lophotrichous** - multiple flagella at the same spot

**C) amphitrichous** - single flagellum on each of two opposite ends

**D) peritrichous** - flagellas in all directions (*E.coli*)




hook
(universal joint)
filament (propeller)
L-ring
P-ring
bushing
stator
studs
C ring
S ring
M ring
rotor



# BACTERIAL FLAGELLUM

**Princip:**

- flagellum rotate like screws, driven by **flow of protons** ($H^+$ ions, ocassionaly $Na^{2+}$ ions) across bacterial cell membrane due to concent. gradient
- flagellum **rotates independently**
- flagellum is **thick and hollow** tube - flagellin subunits flow up the inside to add at the tip (flagella grow by the **addition of flagellin subunits at the tip**)
- direction of movement is changed due to signals from outside

---

# ARCHAEAL FLAGELLUM

**Princip:**

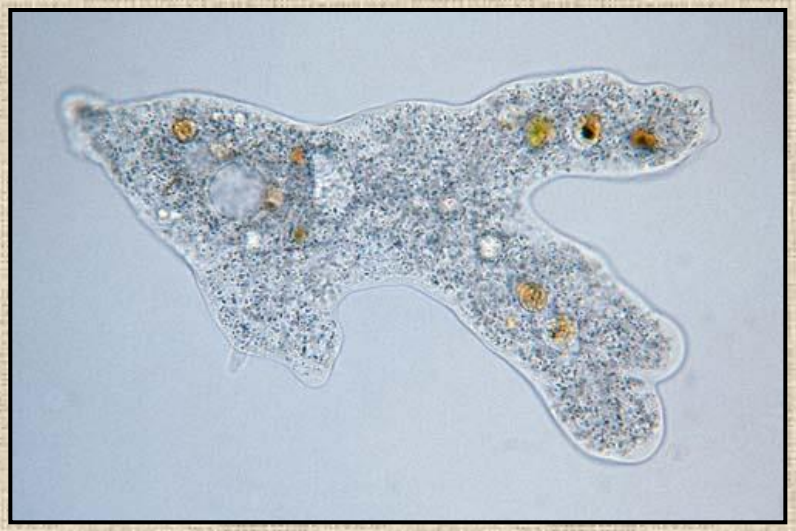
- **powered by ATP**
- many filaments **rotate as a single flagellum**
- flagellum is thin and grows by the **addition of subunits to base**

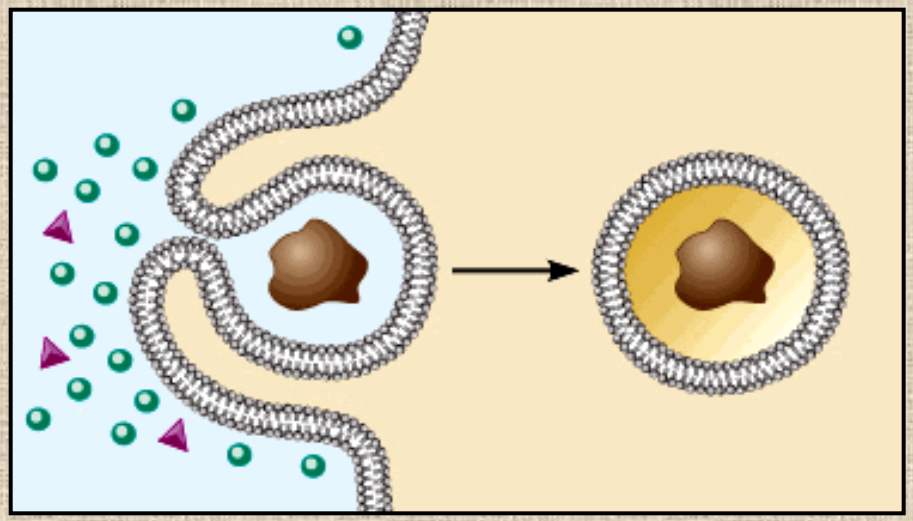
# AMOEBOID MOVEMENT

- “amoeba-like movement” seen in **amoeba**
- in man (**Kupffer cells** in liver, white blood cells e.g.**monocytes** and **neutrophils**, **macrophages**)

**Princip:**

- cells can sense changes in the environment and respond to them by going towards or away from such stimuli
- movement is based on **changing shape of cell by forming pseudopodia** (= false feet produced anywhere on cell with no fixed position, numbers vary from 0 to dozens)





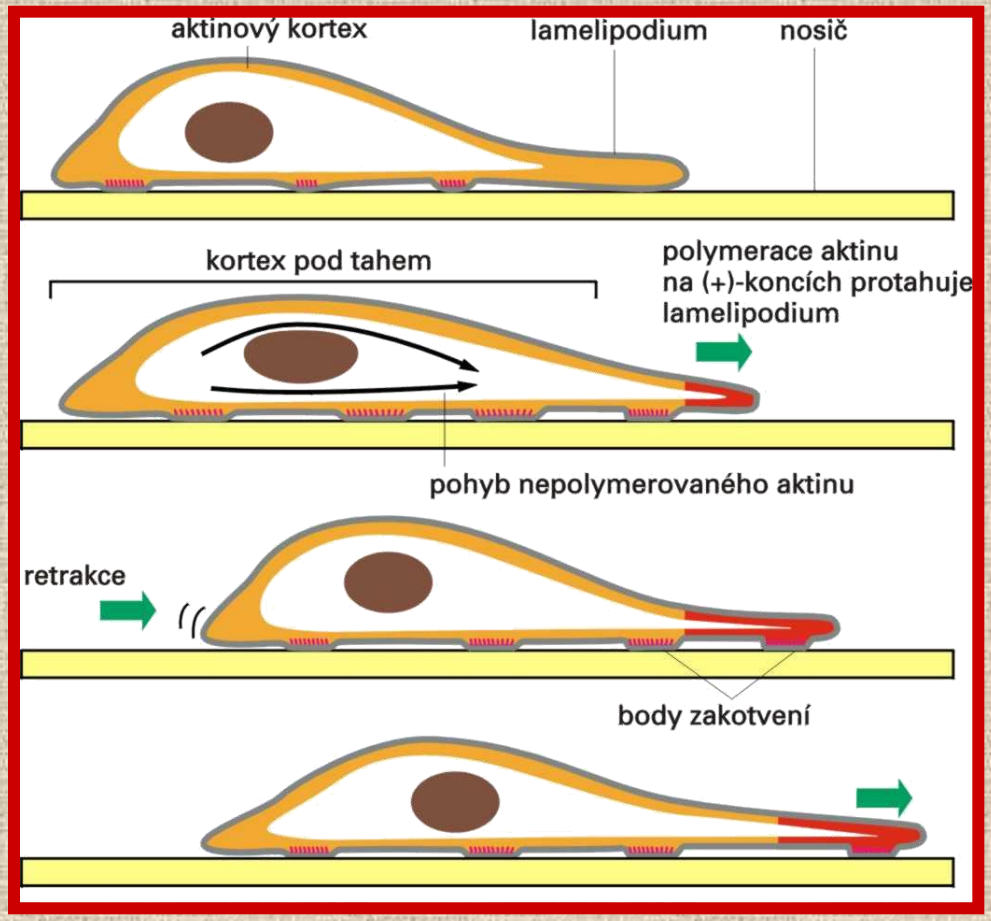
# Amoeboid movement:

**1) protrusion of a pseudopodium** from one end of a cell

**2) pseudopodium is attached** by proteins (**integrin**) in its leading position on a base

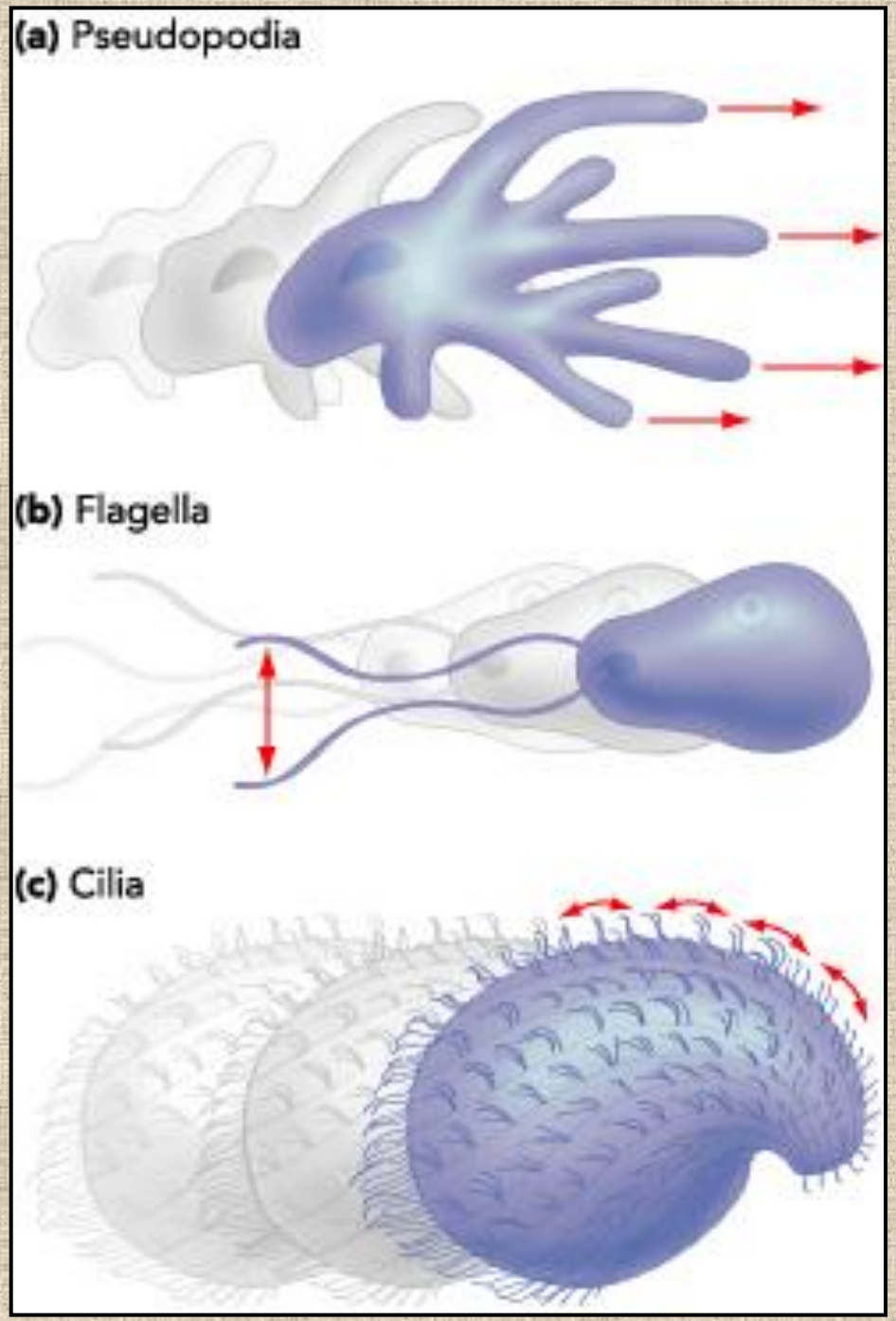
**3) rest of the cell body is pulled** towards the growing pseudopodium

- **actin** polymerizes to form filamentous network
- **MYOSIN I** binds to actin and the network contracts pulling the cell in the direction of pseudopodium (energy from **ATP**).



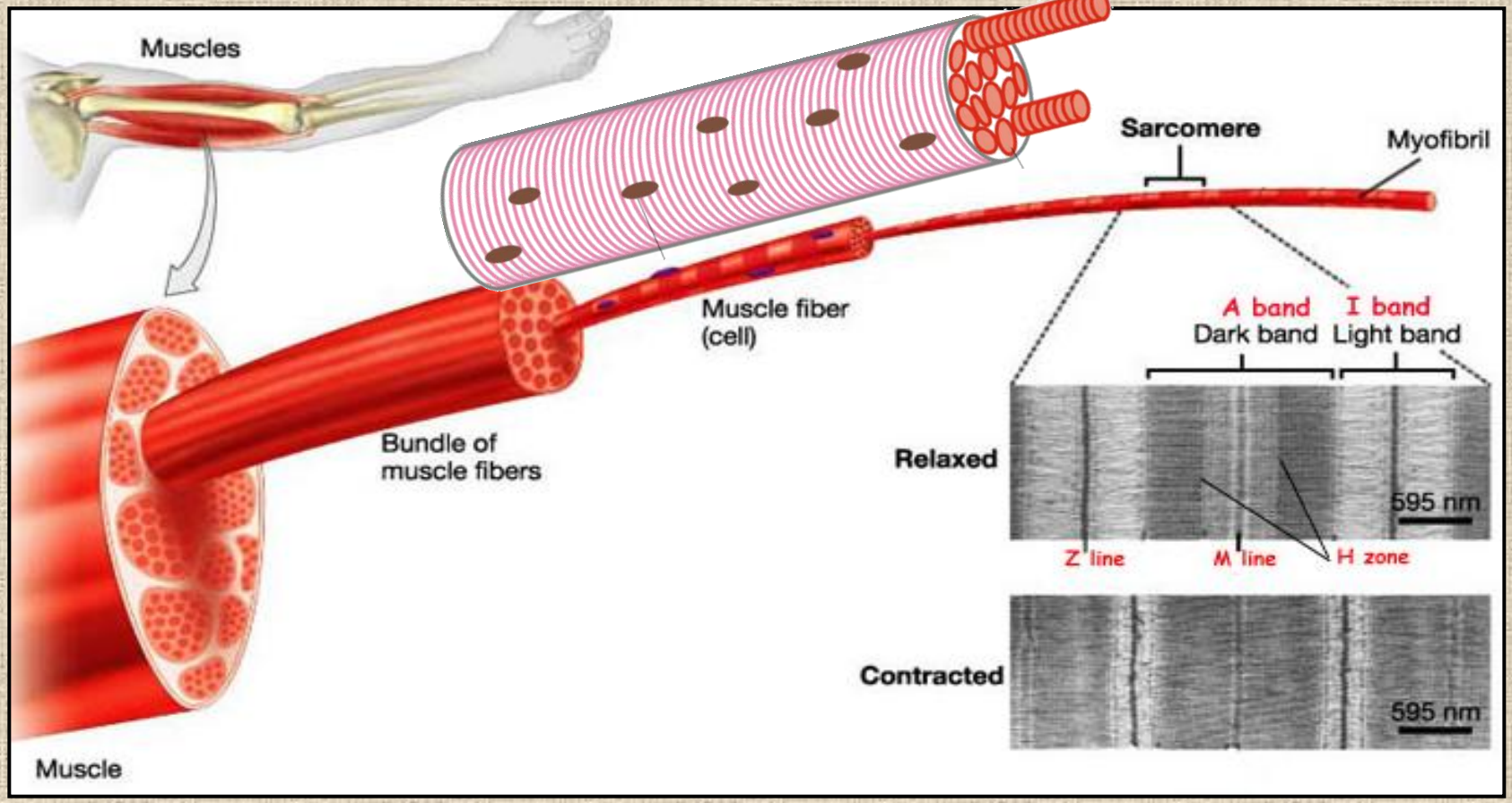

Animation of phagocytose:
http://www.stolaf.edu/people/giannini/flashanimat/cellstructures/phagocitosis.swf

# Comparison



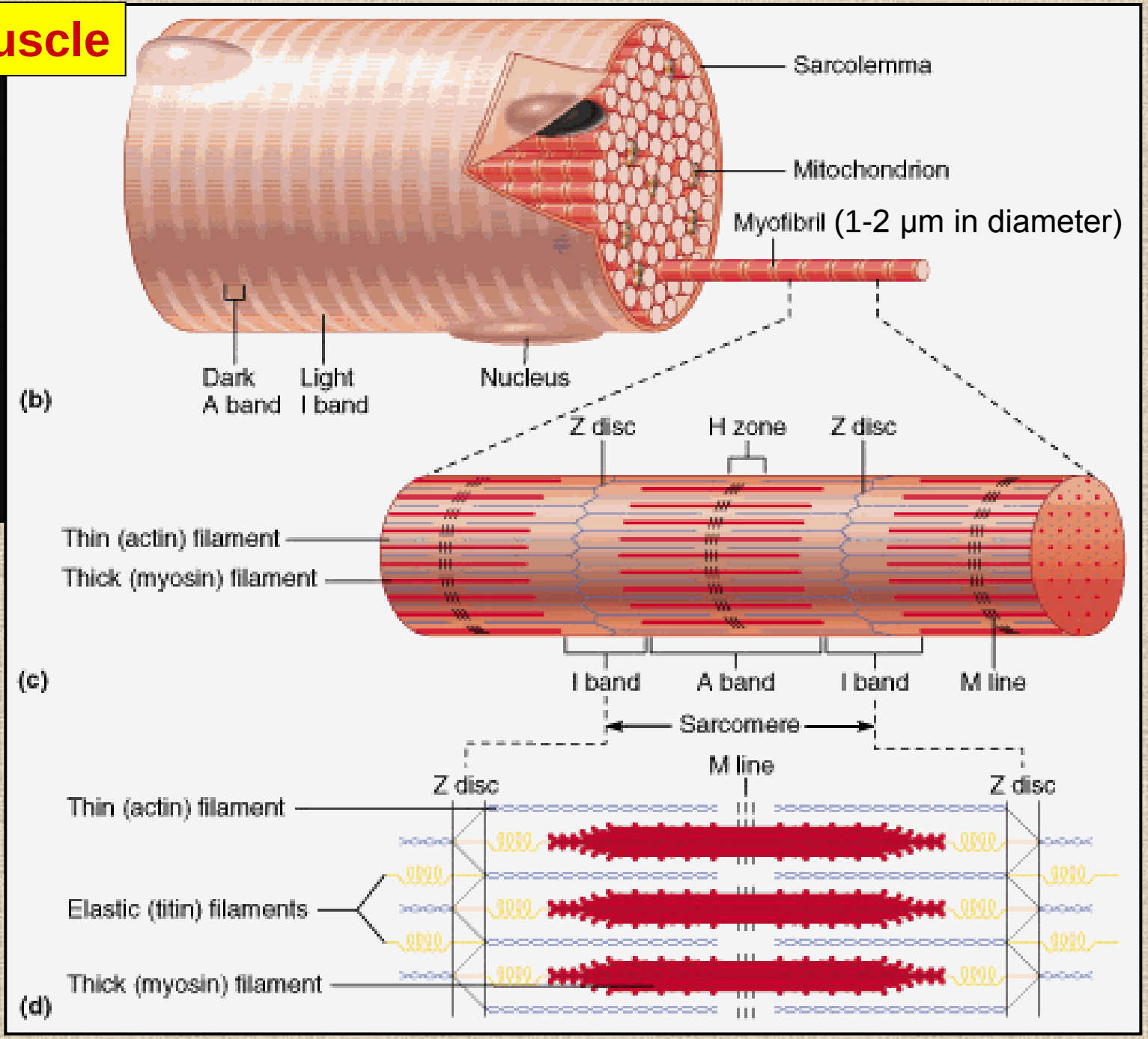
# MUSCLE CONTRACTION

- contraction is controlled by **central nervous system**
  **brain** - controls voluntary muscle contractions (skeletal muscle)
  **spine** - controls involuntary reflexes (heart and smooth muscle)




Draw sarcomere

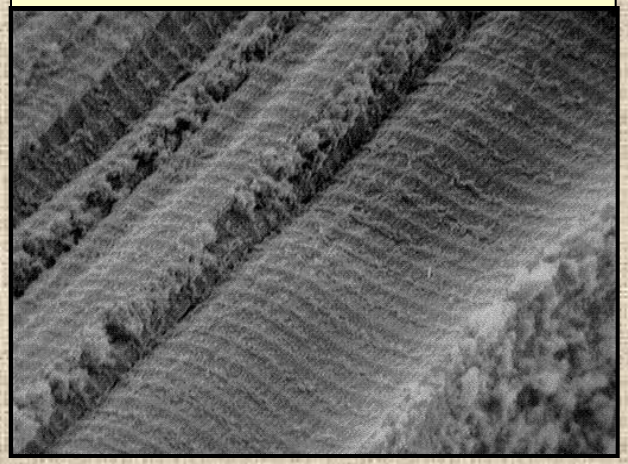
# Skeletal muscle

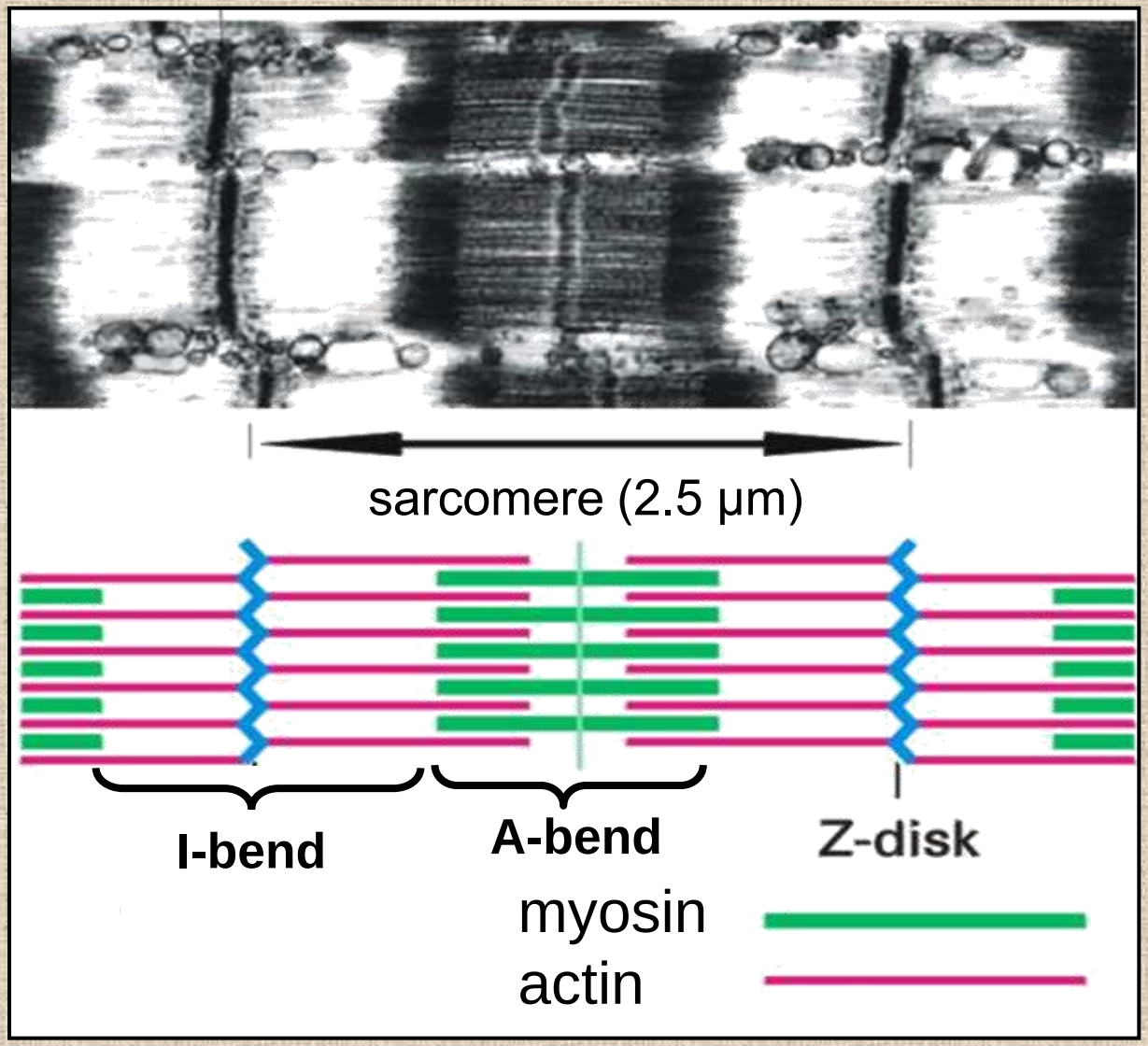


# Sarcomere

**MYOSIN II (thick filament)** - in the middle of sarcomere
**ACTIN (thin filament)** - anchored to Z-disk of sarcomere

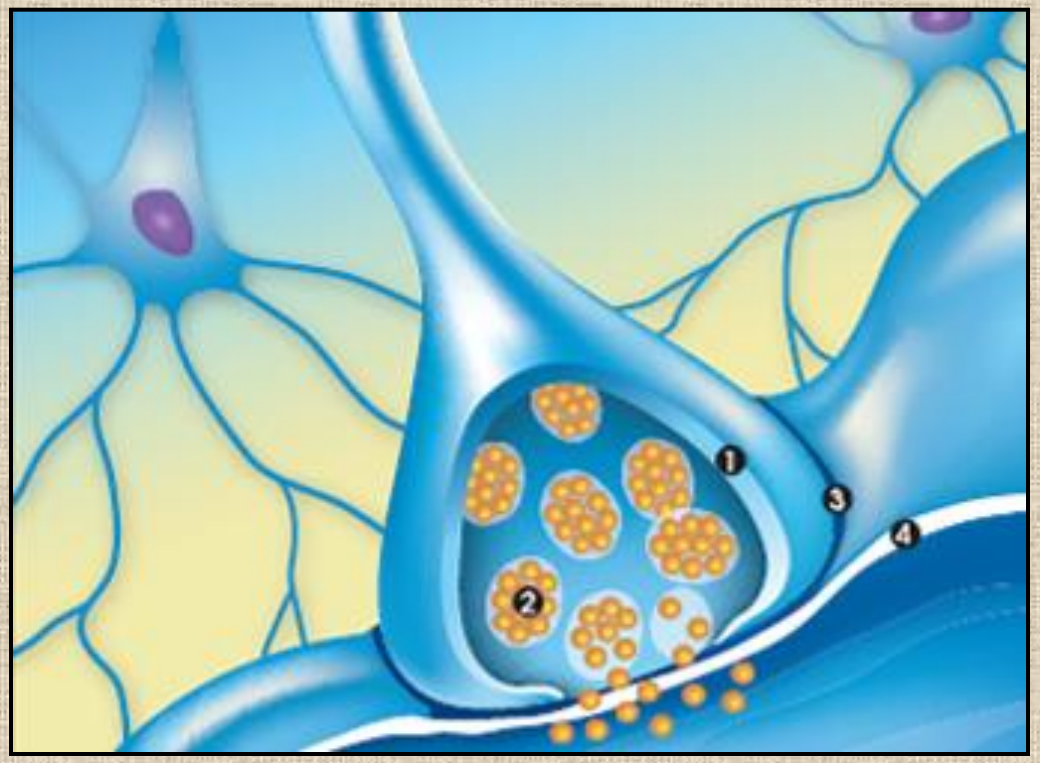
**striated muscle**
- skeletal and cardiac
- striped appearance under microscope due to alternating pattern of **A band** and **I band**

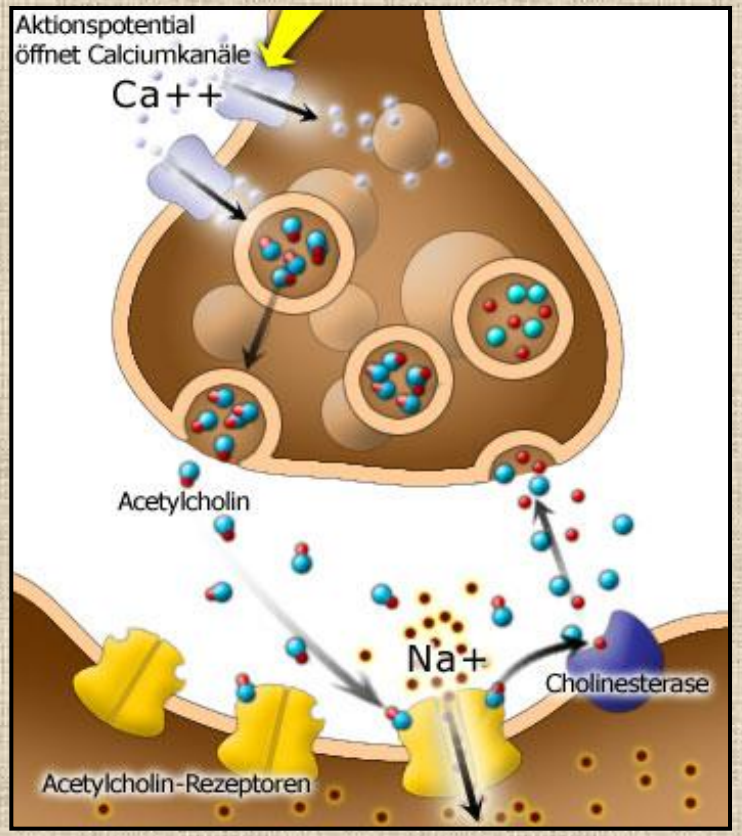




# Muscle contraction

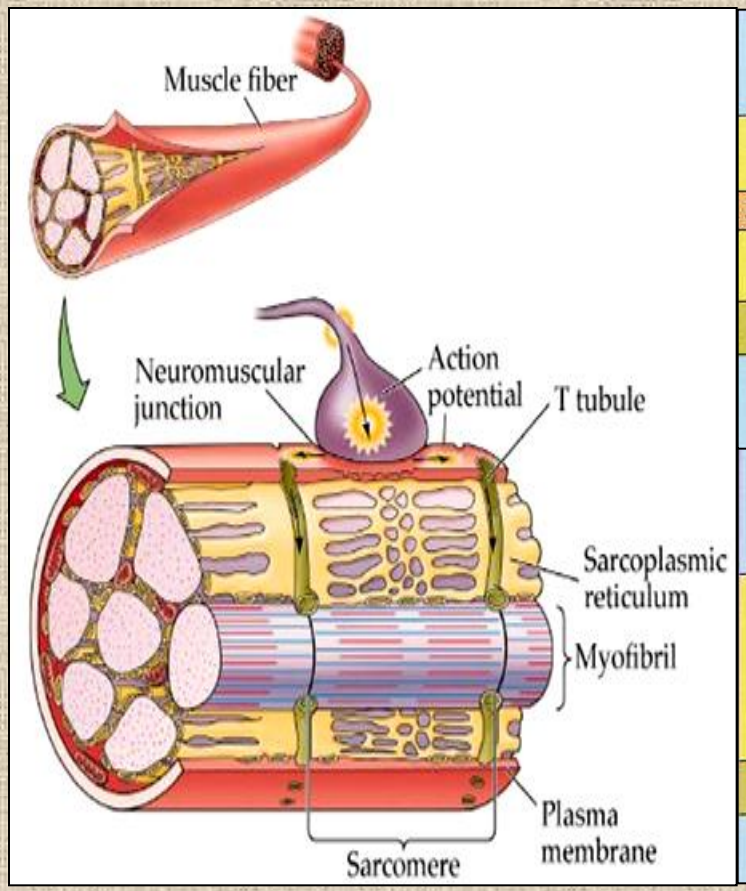
1) **action potencial originated in CNS** transmits action potential down its own axon
2) action potential activates voltage-gated calcium channels on the axon, and **calcium rushes in**
3) calcium causes that vesicles release neurotransmitter **acetylcholine** into synaptic cleft between neuron and skeletal muscle fiber

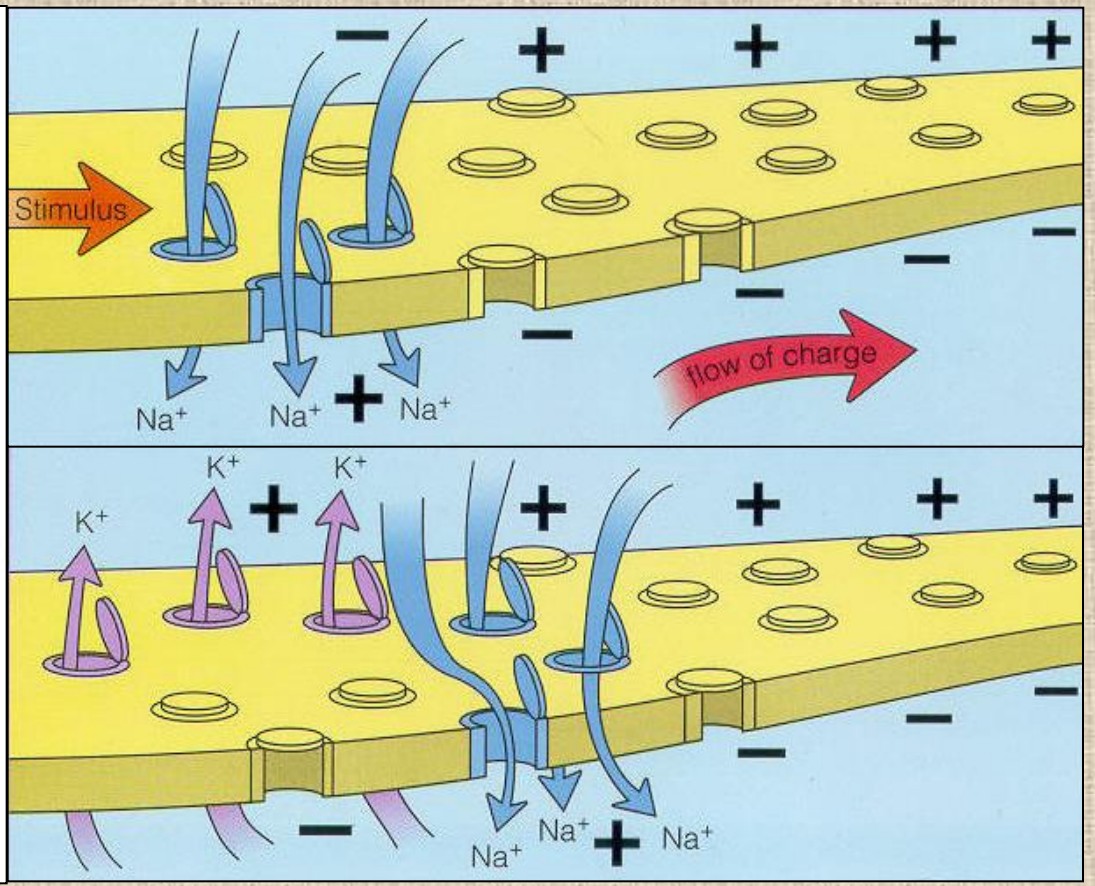


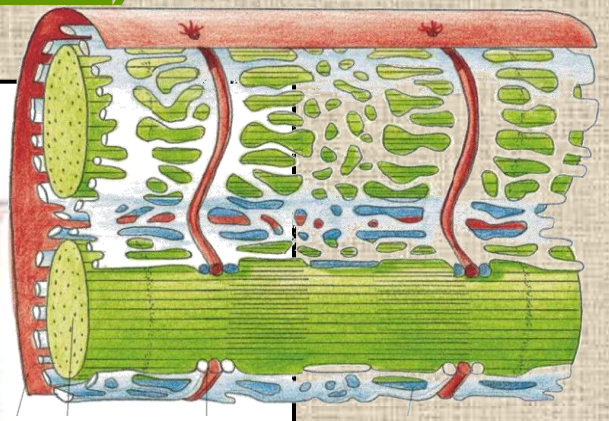
4) acetylcholine diffuses across synapse, activates receptors of muscle resulting in opening of sodium/potassium channel (sodium rush in, potassium rush out)

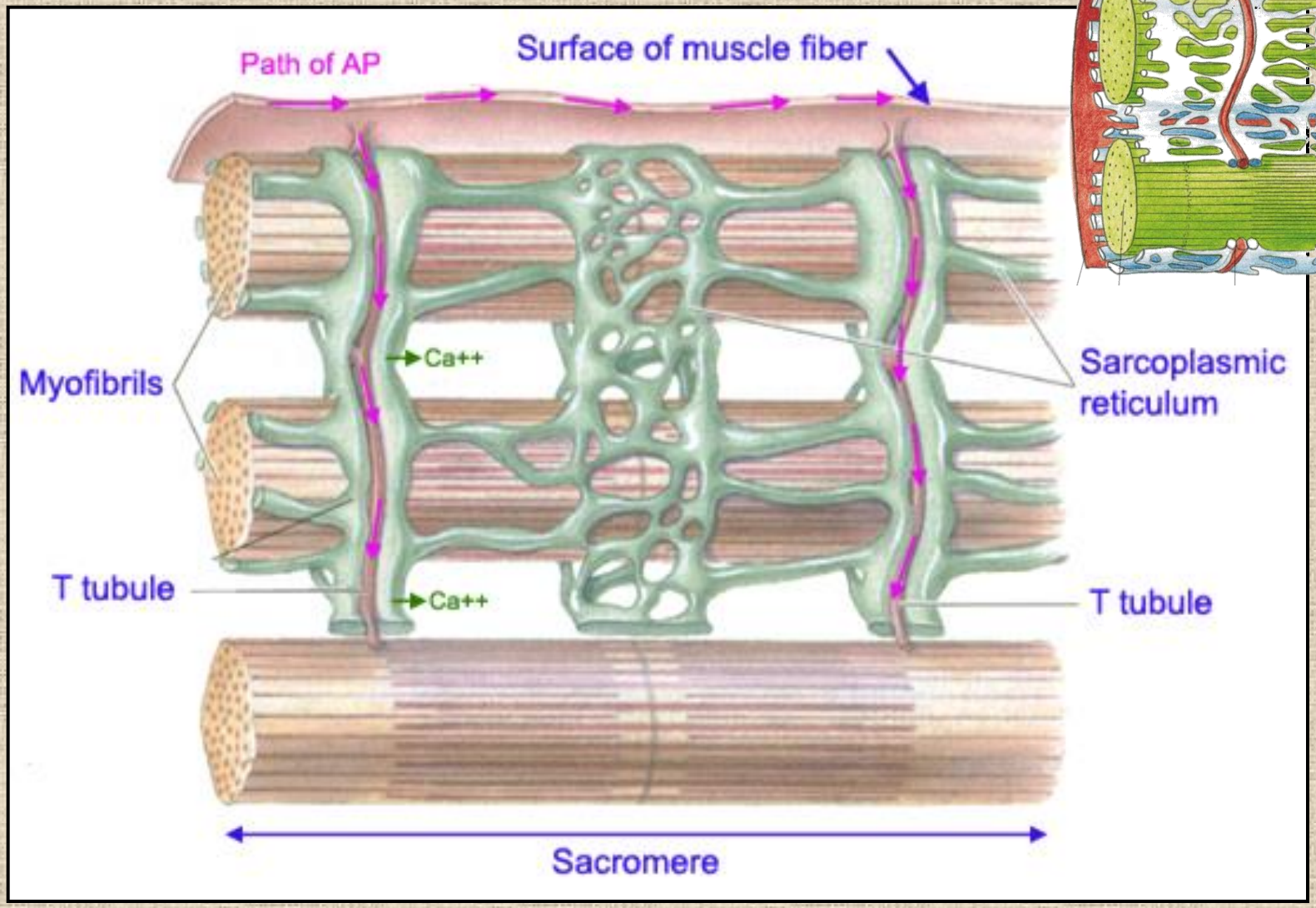
5) **action potential (nerve signal) spreads through muscle** fiber's network of T tubules (invagination of membrane) and depolarizes the inner portion of the muscle fiber



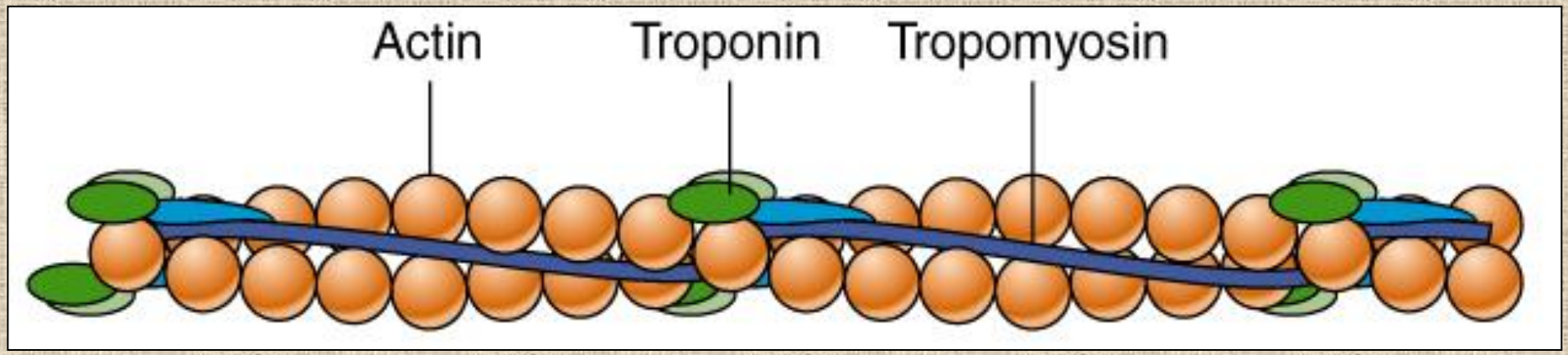



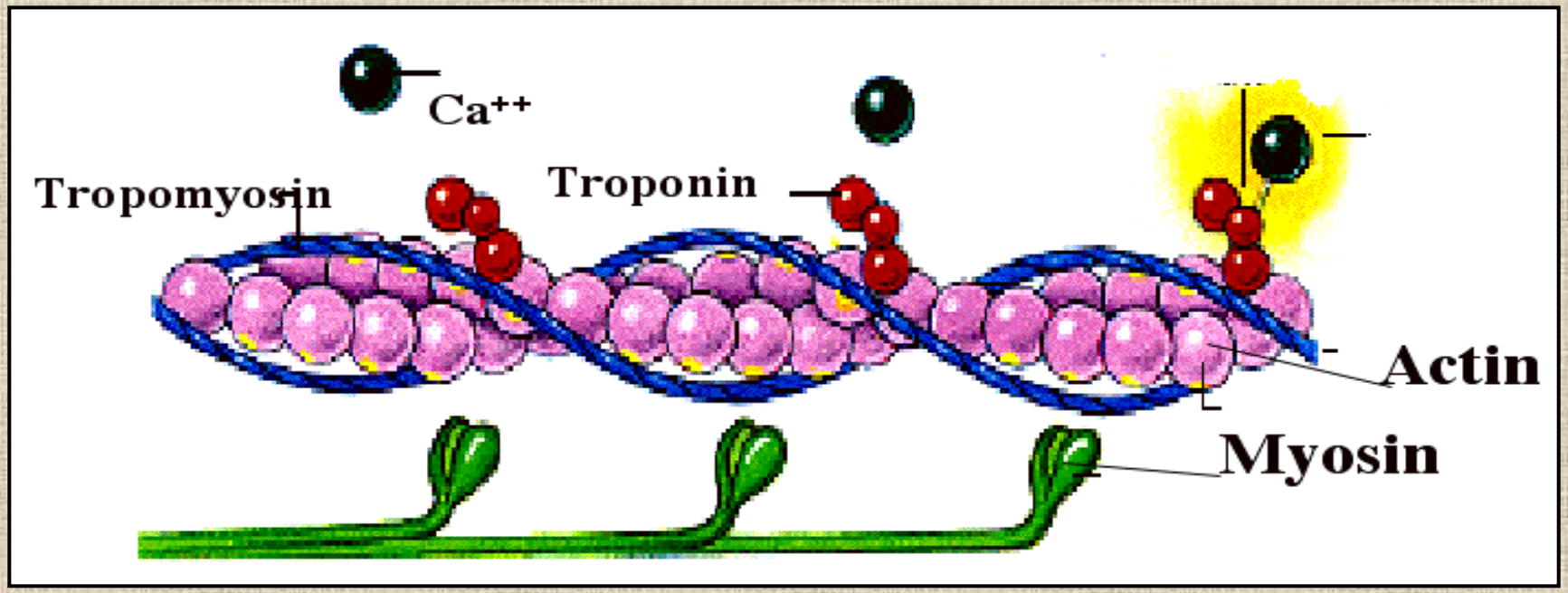
6) depolarization activates voltage-gated calcium channels (in T tubule membrane) that interact with calcium-release channels of **sarcoplasmic reticulum (SR)** to activate them and **release calcium**



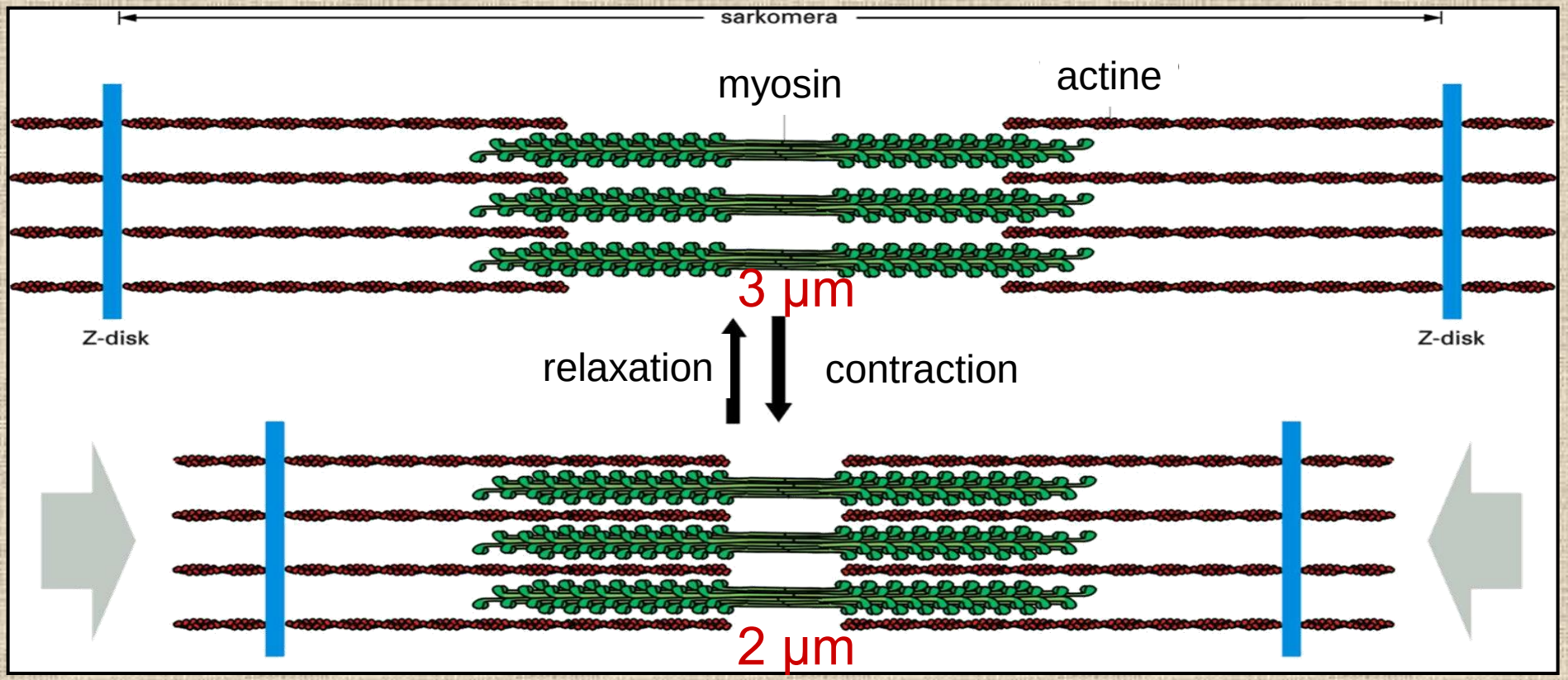


7) **calcium binds to troponin C** (protein), that **modulates tropomyosin** (protein) allowing it to move and unblock the binding sites on actin for myosin



8) **myosin binds** to uncovered binding sites on actin, this pulls Z-disk towards each other and **shortens sarcomere** (myosin hydrolyzes ATP to obtain energy)

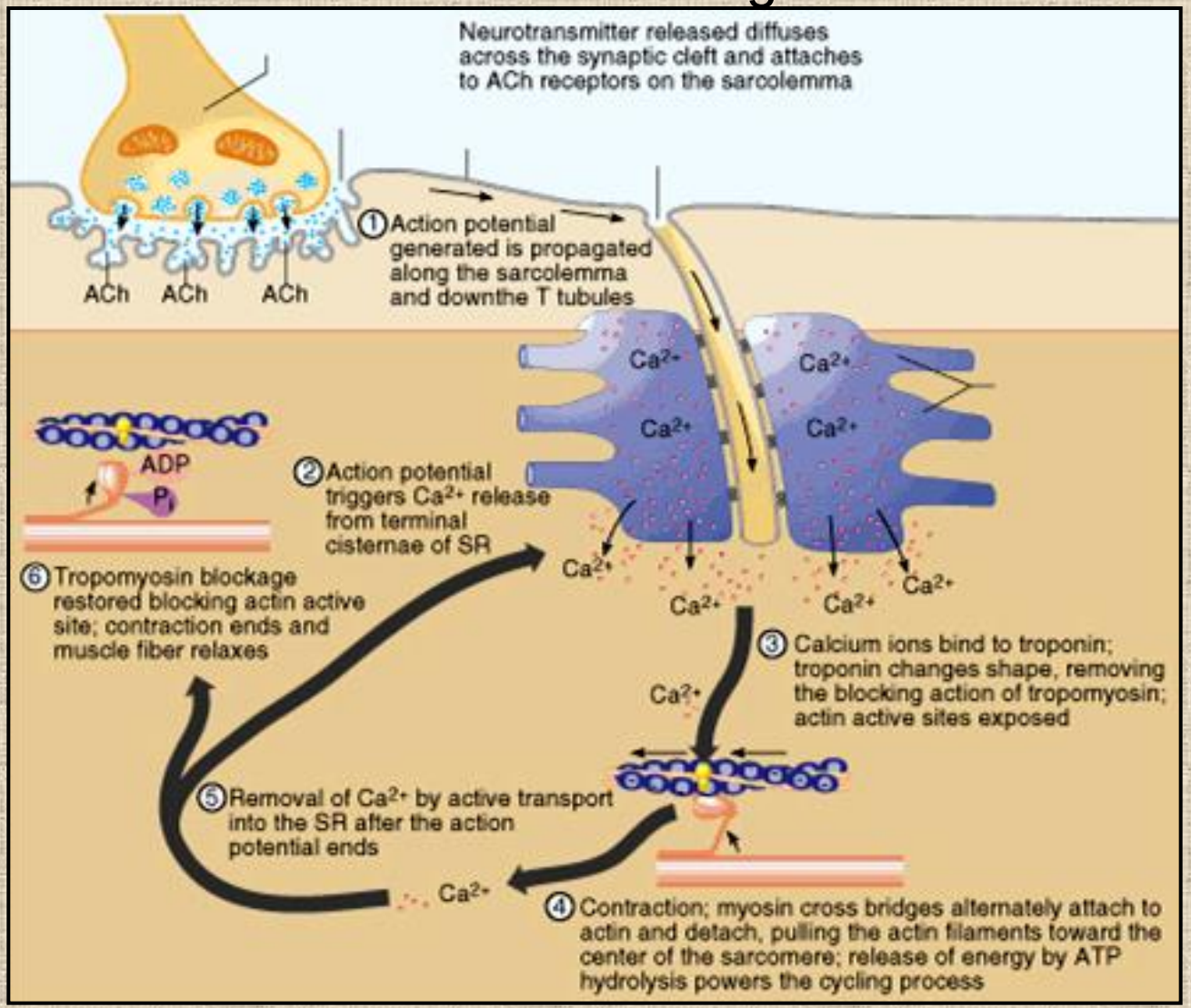



Constriction of sarkomere during **0.1 sec**

- Animation:
  *http://bcs.whfreeman.com/thelifewire/content/chp47/4702001.html
  http://www.sumanasinc.com/webcontent/animations/content/muscle.html
- Neuromuscular junction and Action potential and musscle contraction
  http://highered.mcgraw-hill.com/sites/0072495855/student_view0/chapter10/animation__function_of_the_neuromuscular_junction__quiz_1_.html
- Sarcomere contraction
  http://www.edumedia-sciences.com/a502_l2-muscle-contraction-sarcomere.html

**Muscle relaxation**:

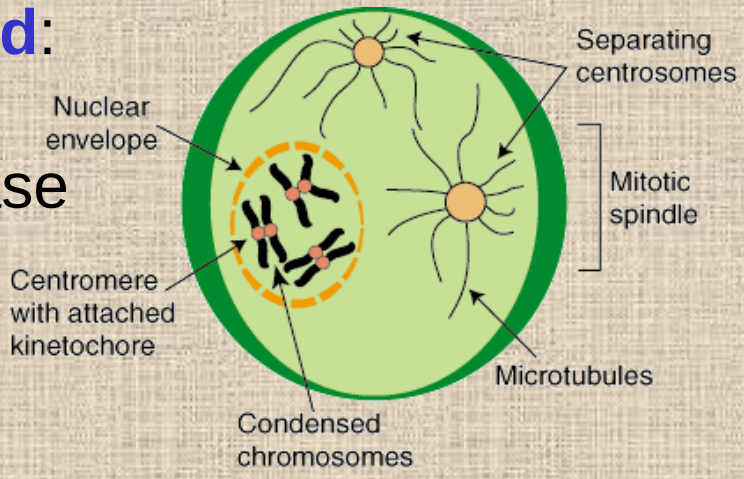
- after no nerve signal, **calcium is pumped to SR** by calcium pump, **tropomyosin changes conformation back to its previous state** to block the binding sites of actin

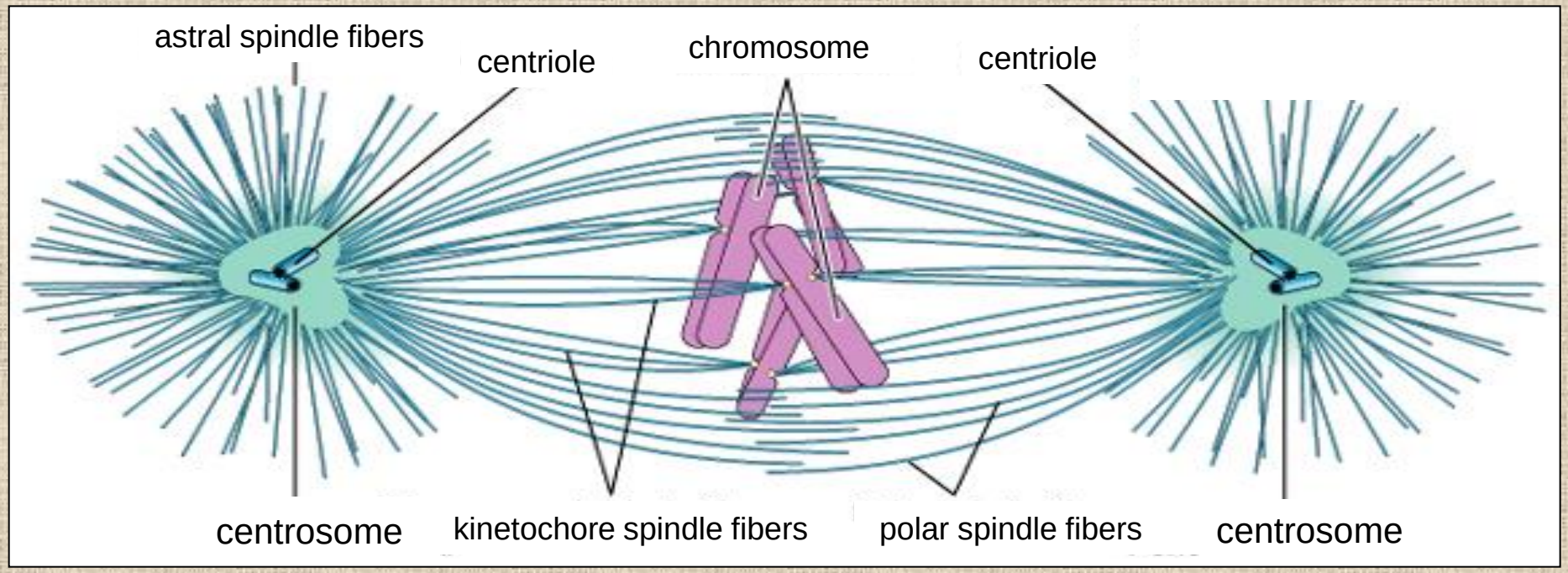


# MITOSIS

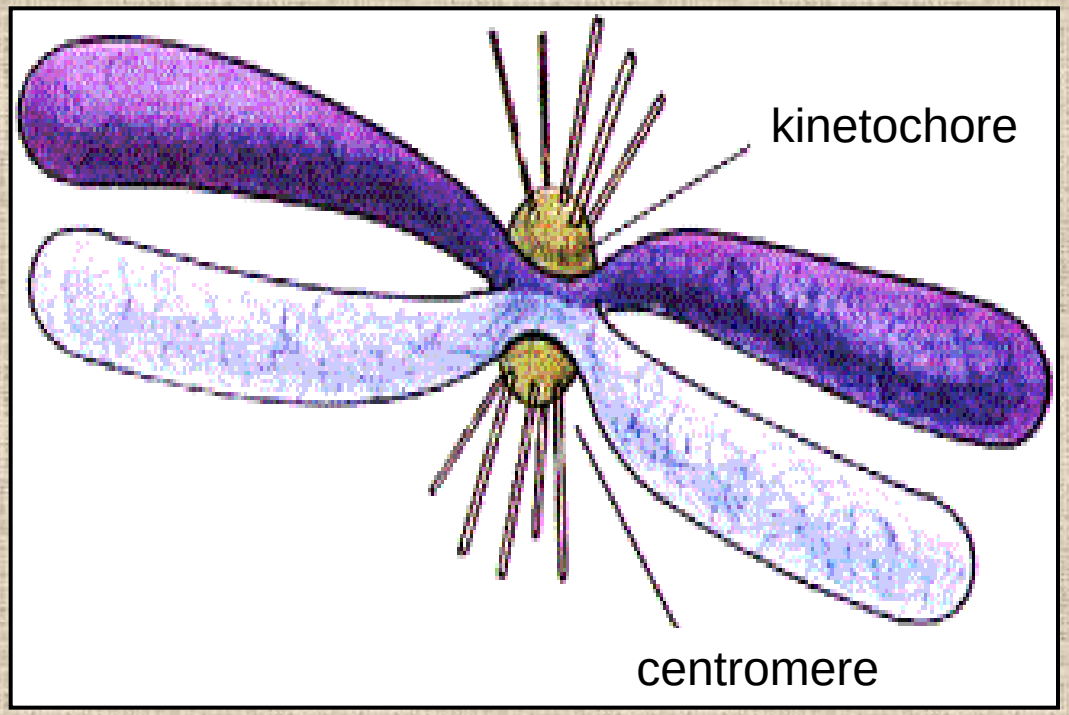
**PROPHASE** - **mitotic spindle is formed**:

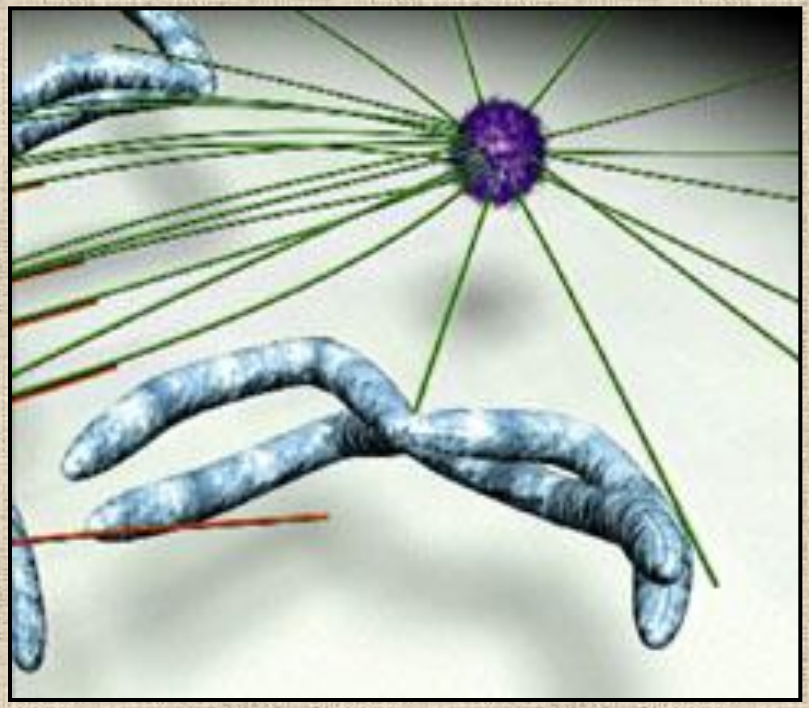
- **centrosome** = organizing centrum for microtubules; duplicated in S phase
- **astral microtubules**
- **polar microtubules**
- **kinetochore microtubule**



**PROMETAPHASE** - kinetochore microtubules attach to each sister chromatid at the **kinetochore** (complex of protein - **dynein**, near centromere)

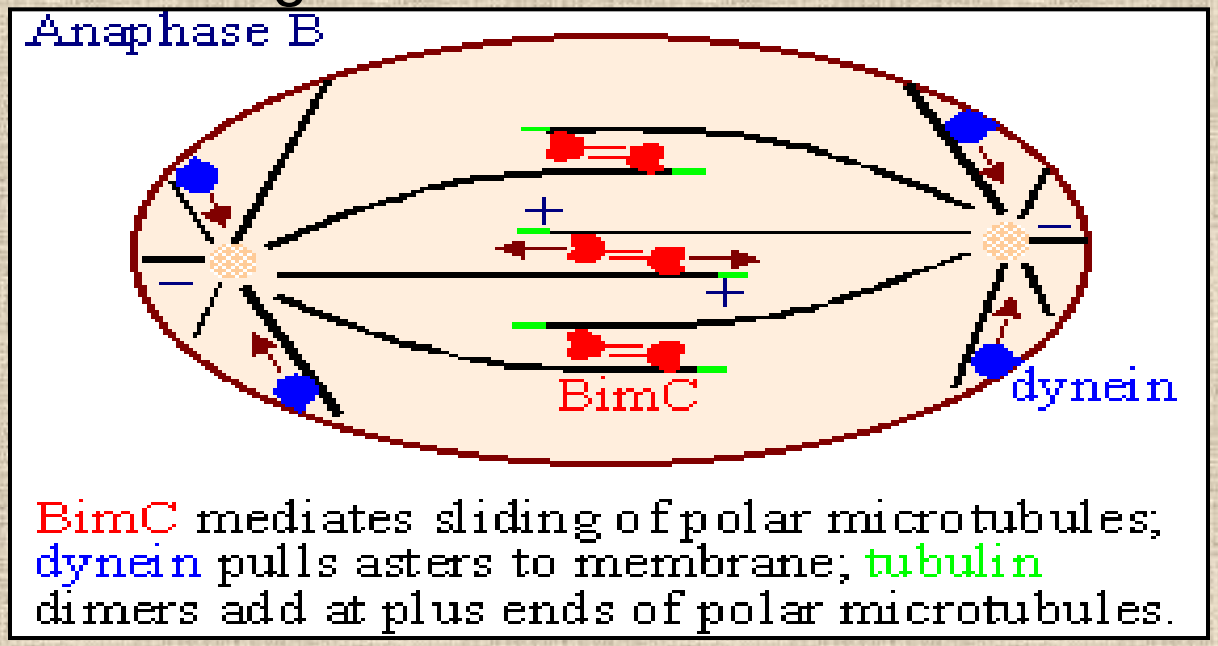


**METAPHASE** - **chromosomes line up** in the middle "equator" of the cell by polymeration and depolymeration of kinetochore microtubules (motors are involved)

# ANAPHASE

- kinetochore microtubules begin to shorten and **DYNEIN pulls chromatids to opposite poles**
- **polar microtubules slide** (with help of **KINESIN**) and polymerate at +end and so the distance between mitotic poles enlarge

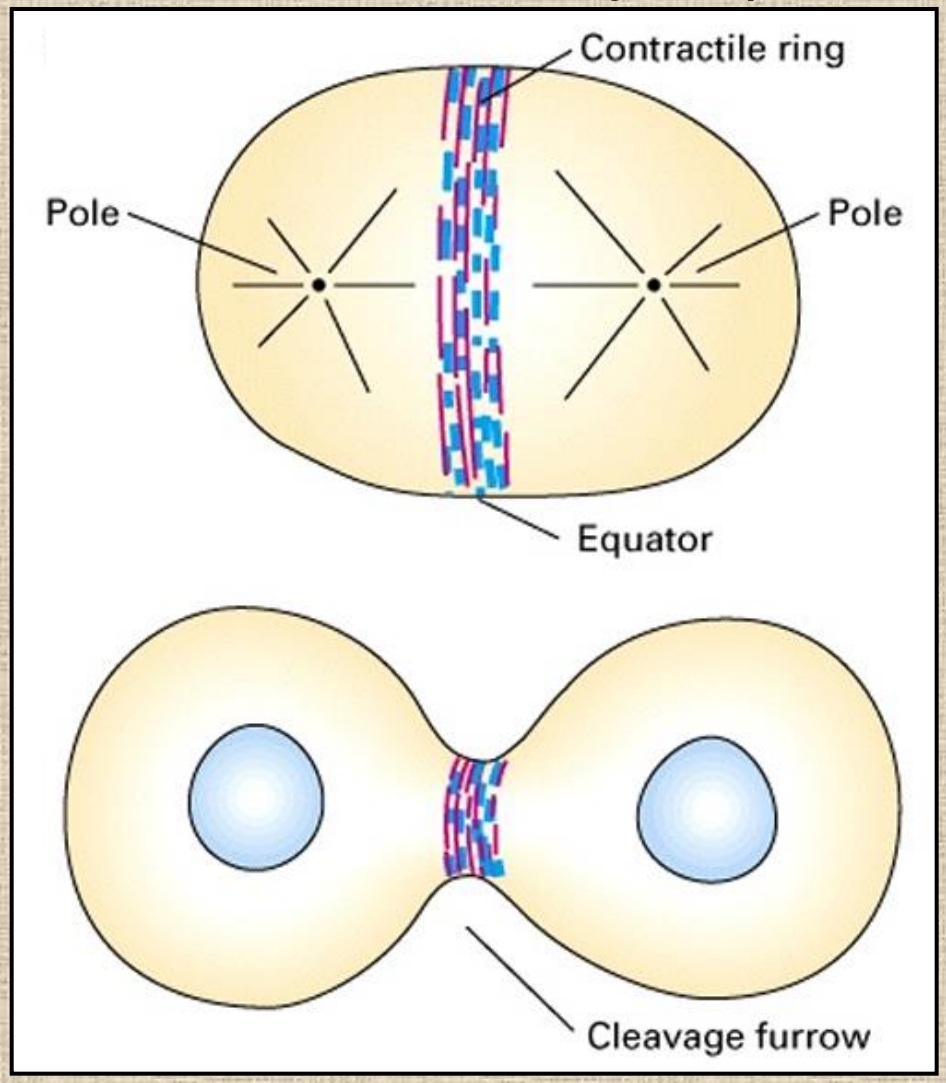


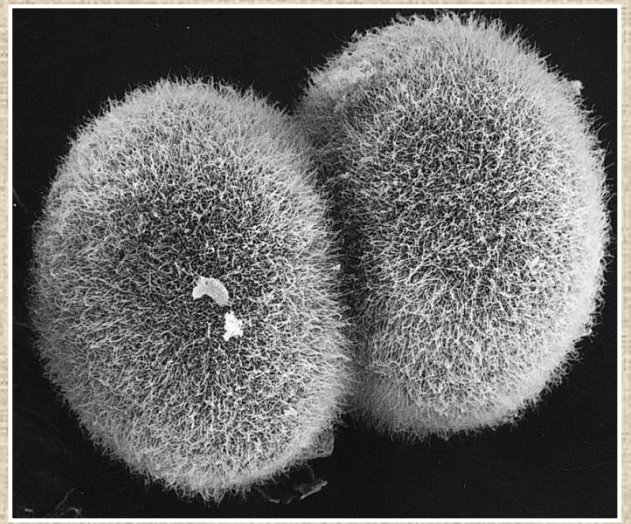

Animation of molecular motors in mitosis:
*http://faculty.plattsburgh.edu/donald.slish/Motors.html

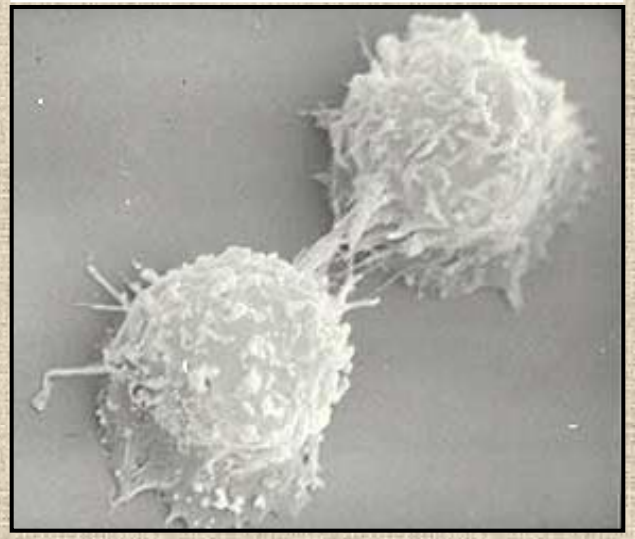
**TELOPHASE** - **kinetochore microtubules disappear**, **polar microtubules still polymerate**

# CYTOKINESIS in animal cell

- by process known as **cleavage**
- **contractile ring is formed** under plasmatic membrane, **actin filaments slide** by help of **MYOSIN II**

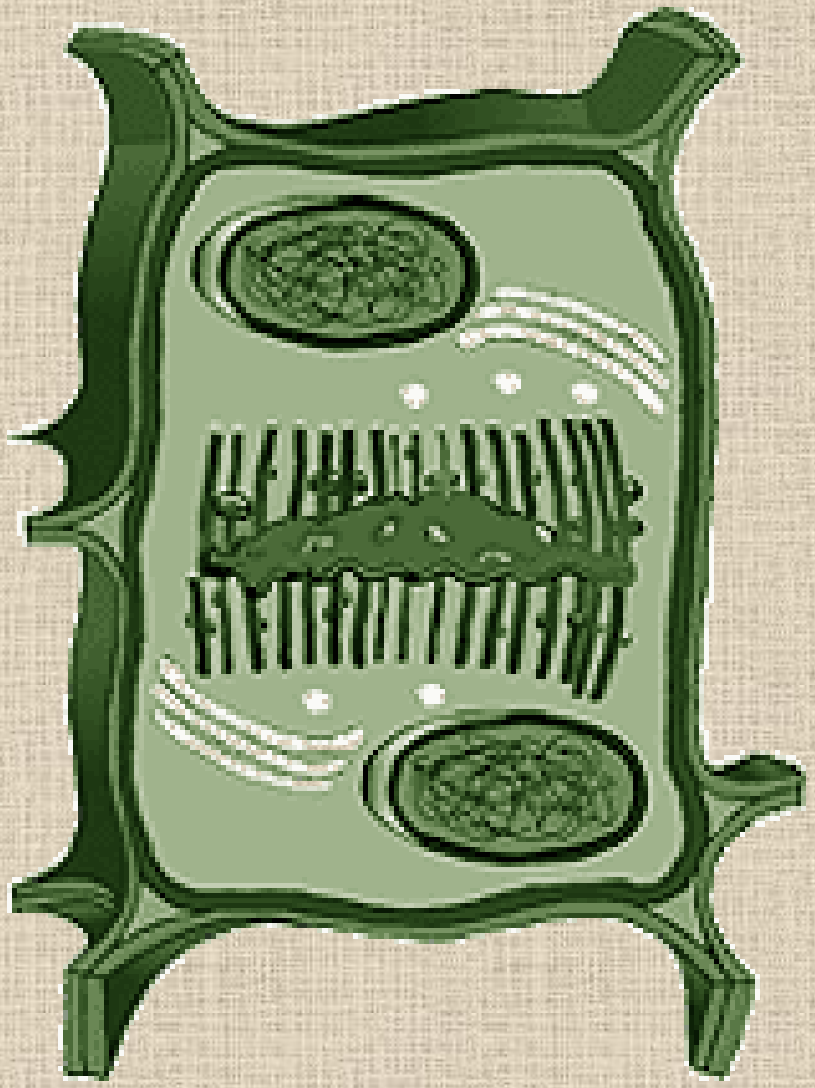


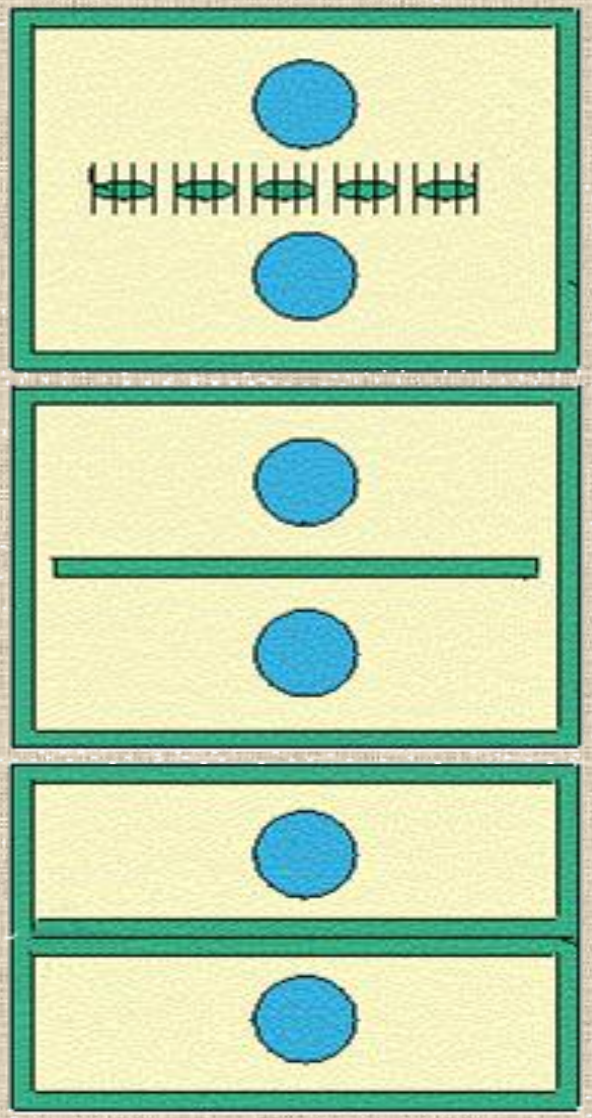





# CYTOKINESIS in plant cell

- vesicles from the Golgi apparatus move along microtubules to the middle of the cell and fuse, producing the **cell plate**





Animation of mitosis:
http://www.johnkyrk.com/mitosis.html

http://highered.mcgraw-hill.com/sites/0072495855/student_view0/chapter2/animation__mitosis_and_cytokinesis.html

# SUMMARY

| cytoskeleton | **microtubules** | **actin filaments** |
|---|---|---|
| molecular motor | kinezin, dynein | myosin I, II |
| movement | - intracellular<br>- flagellar<br>- cilliar<br>- mitosis (mitotic spindle) | - amoeboid<br>- muscular<br>- mitosis (contractile ring in animal cells) |