BIO 1140 Midterm 1 Notes

Organization of Matter

- *Matter*: composed of elements
- *Elements*: pure substances composed of one type of atom
    - 94 natural (pure elements)
    - 6 major elements: C H N O P S (98% body weight)
        - Usu incorporated into organic mlcs
- *Molecule*: 2+ atoms bond together to form mlc (NaCl, H_2, CH_4 …)
- *Compound*: mlc composed of 2+ elements (H_2 is not one)
- Electrons in outer valence shell determine chem reactivity of atoms

Cell Chemistry

- Atoms might lose / accept / share e^-s in valence shell
- *Octet rule*: atoms with 2+ shells, chem. rxn completes number of electrons to 8
- Chem bonds:
    - *Ionic*: complete loss / gain of 1 e^-s, –ve or +ve ions
        - More EN atom attracts e^- to steal from other element
        - Anion = –ve charge
        - Cation = +ve charge
        - Salt: organized crystals when saturated / at ↓T
        - E.g. NaCl
    - *Covalent*: e^-s are shared b/w 2 atoms
        - 2 atoms complete valence shells by sharing e^-s
        - E.g. methane
            - Non polar, tetrahedral
            - 109°
- Carbon
    - Electronic config in C makes it possible to form covalent bonds with other ele
    - Bonding with another C
        - Linear, branched, cyclic C, backbone chains
        - Result in almost unlimited diversity of mlcs
    - Covalent bonding with ele other than C
        - Results in important fnal grps → see p. F-18
    - Hydroxyl R – OH Alcohols, sugars
    - Carboxyl R – COOH Amino acids, fatty acids
    - Carbonyl R – CO – R Ketones, aldehydes
    - Amino R – NH_2 Amino acids, proteins
    - Sulfhydryl R – SH Amino acid Cysteine, proteins
    - Phosphate R – PO_4H_2 ATP, nucleic acids
- Water
    - Most abundant mlc on Earth → 3/4 globe's surface covered

- 70-90% of total cell mass
- *Water mlc*: O_2's 2 valence e^-s shared with 1 e^- of 2 H
- Planar, tetrahedral, asymmetric, unequal e^- distribution
- 2 pairs of unshared e^-s
- O is very EN → polar mlc
- Properties of water
    - *Hydrogen bond*: Strong polarity → H bonds (~5 kcal/mol)
        - δ+ and δ- dipoles attracted
    - *Aqueous phase*: Avg ~3.4 H bonds / mlc
        - Forms interconnected water lattice, less ordered
    - *Solid phase*: 4 H bonds / mlc, rigid bonding + ordered
        - Intermlclar spacing ↑ (V expansion) → lower density → floating → spaced out but less moveable
        - If ice sinks → oceans solidify bottom-up → no more life
    - *Water as solvent*: for polar mlcs + mlcs with ionic bonds (*hydrophilic*)
        - Dipole-dipole interactions form *hydration shell* around hydrophilic mlcs
        - Dispersion of ions + mlcs in water = dissolving
    - *Biological importance*:
        - Water bridge in macromlcs stabilizes struc
        - All biochem rxns req aq enviro
    - *Cohesion*: H bonding b/w water mlcs holds then together
    - *Adhesion*: H bonding with other mlcs makes water stick to other objects
        - Both cause water rise in xylem fibrils in plants → resist gravity → continuous flow
    - *High heat capacity*: 1 cal/g ↑T 1°, almost 2x as others
        - High boiling T (100°C)
        - Resists fast evaporation + cells can resist drying in wide T span

Biological Molecules

- *Macromolecules*: large molecules make up cells / carry out cellular vital processes (~26% of bacterial cell mass)
    - Structural / fnal roles
    - Bacterial cell

- Dynamically synthesized + replenished → have life span → constant supply need
- Constructed by linking together dis/similar subunits (monomers) → basic unit
- Role in cell
  - *Informational*: order of monomeric units not random + req for proper fn
    - I.e. proteins, nucleic acids → have meaning / cannot change sequence
  - *Structural/storage*: single repeating / alternating subunits; order does not carry info
    - E.g. polysaccharides (cellulose, starch, glycogen)
    - Back-up for E
    - Doesn't matter for # of repeating subunits
    - Proteins, RNA can be these
- Recycling of macromlcls controlled *enzymatically* through biochem pathways → catalysts + analysts
- Synthesized from monomers by *dehydration* rxns
  - Condensation of monomers by *losing* water mlc
- Degraded by *hydrolysis* rxns
  - *Addition* of water mlc

- Carbohydrates → ose = sugar
  - Include simple sugars + polysaccharides → polyalcohol = many OH grps
  - *Monosaccharides*: simple sugars, major nutrients of cells → $(CH_2O)_n$
  - Glucose ($C_6H_{12}O_6$) important → provides principal source of cellular E → glycolytic pathway
  - Ribose ($C_5H_{10}O_5$) + deoxyribose (OH at C2) are 5C sugars found in RNA + DNA
  - Monosaccharides
    - $(CH_2O)_n$ = 3 to 7, n=5 or 6 most common
    - Aldose sugars (fnal grp is aldehyde –CHO at end of mlc)
    - Ketose sugars (ketone C=O as fnal group in middle of mlc)
  - Stereoisomerism
    - *Isomers*: same formula but struc diff
    - E.g. glucose (aldohexoses), galactose and fructose (ketohexoses) → all $C_6H_{12}O_6$ but struc diff
    - *Stereoisomers*: differ only in spatial arrangement
      - E.g. OH and H swapped around on same C atom
    - *Enantiomers*: optical isomers, same form, same grps bonding with C skeleton, but grps on *asymmetric carbon* are *mirror images* of each other
      - Symmetric C = α-carbon → non rotate-able
      - E.g. glyceraldehydes
      - D-form (Dexter = right) –OH facing to right
      - L-form (Laevus = left) –OH facing left of backbone
      - D + L are mirror imgs + cannot be superimposed
      - *D-form* important for cells → fits to active sites b/c evolution
    - Triose sugars ($C_3H_6O_3$)

        - Glyceraldehyde + dihydroxyacetone are 3C aldo-trioses + keto-trioses that result from breakdown of glucose in glycolytic pathway
        - All cells do this b/c of evo
    - Linear vs. Ring → KNOW DRAWINGS
        - C5 and C6 sugars exist in linear + ring forms
        - Glucose, C1=O bonds with OH grp on C5
        - Pentose, C1=O bonds with OH grp on C4
        - α/β configuration isomers may form
            - Important for polysaccharide formation
        - Diff with respect to pos of –OH relative to plane of ring
        - α isomer –OH below plane
        - β isomer –OH above plane
    - *Glycosidic* bond
        - Dehydration rxn joins 2 simple sugars by glycosidic bond
        - α 1→4 bond or β 1→4 (C1 + C4) common → alternates αβαβ
            - Main polysaccharide *backbone*
        - α 1→6 (C1 + C6) causes *branches* in chain
    - Polysaccharides
        - Short term E storage mlcs
            - Plants store glucose as *starch (amylopectin)*
            - Animals store as *glycogen* in liver → makes granules
        - Structural role
            - Cellulose in plants
            - Chitin in arthropod exoskeleton
            - Thicker + thicker to be struc support → thicker = stronger
        - Glycogen + starch composed entirely of glucose mlcs in α config
            - Struc same but diff cell type
            - Both branching
        - *Cellulose* most abundant polysacc → plant cell wall
            - Unbranched glucose polymer with β1→4 glycosidic bond
            - H-bonding b/w many parallel polymers forms strong fibres → spontaneous
        - *Chitin*: polymer of N-acetylglucoseamine with β1→4 linkage; polysacc that forms exoskeleton of crabs, lobsters, insects

- Lipids
    - Diverse grp of macromlcls that are insoluble in water
    - Mostly struc use, E source
    - Includes
        - *Fats + oils* → used for E storage + other purpose → excess glucose
        - *Phospholipids* → components of membranes that surround cells
        - *Glycolipids* → 2 hydrocarbon chains linked to polar head grps that contain carbohydrates

        - *Steroids* → diff struc from most lipids, used as hormones + other purposes → signal transduction pathways
    - 3 main roles
        - E storage
        - Major component of cell membranes
        - Important in cell signalling: steroid hormones + messenger mlcs
    - Triglycerides (Fats)
        - Van der Waals forces
            - Unsaturated = liquid → usu at C9 and C10 of fatty acid chain
            - Saturated = solid
            - Have lower boiling point
        - Contain 2 subunits
            - Glycerol → polyalcohol with 3 polar –OH grps
            - Fatty acids long hydrocarbon chains (16/18 Cs) with carboxyl grp ($-COO^-$) at one end
        - *Triglyceride*: dehydration rxns adds fatty acids to –OH grps of glycerol + broken down by hydrolysis rxns
            - Insoluble in water; accumulate as fat droplets in cytoplasm
            - When req, break down for use in E-yielding rxns
            - Produce twice as much E as sugars per unit weight
    - Phospholipids → KNOW DRAWINGS
        - Principal components of cell membs
        - *Glycerol phospholipids*: 2 fatty acids bound to C in glycerol
            - 3^{rd} C of glycerol bound to phosphate grp (fnal grp)
        - Mlc *hydrophilic* at phosphate end + *hydrophobic* at fatty acid tails
        - Called *amphipathic* property important for formation of bilayer biological membs
        - Polar grps added to phosphate to make mlc more polar → (All 1 WORD)
            - Ethanolamine (phosphatidyl ethanolamine) → cell membs
            - Choline (phosphatidyl choline) → struc
            - Serine (phosphatidyl serine) → a.a.s
            - Inositol (phosphatidyl serine) → sugar mlc, secondary msger
    - Glycolipids
        - In cell membs
        - Amphipathic
        - 2 FA, serine instead of glycerol + 1+ sugar instead of phosphate
        - Fn as cell surface markers used for cell recog → immune cells use these
    - Cholesterol
        - Amphipathic 4-ringed hydrocarbon
            - Abundant in eukaryotic membs
            - Increases memb fluidity
            - Planar struc seeps into phospholipids + interrupts interaction b/w phospholipids
        - Derivatives include
            - Estrogen + testosterone
            - Steroid hormonal messengers

        - Corticosteroids

- Nucleic acids
    - Informational macromlcl → monomers have meaning to order and amounts → disturbance = change meaning
    - Deoxyribonucleic acid (DNA) genetic material
        - Linear for eukaryotic
        - Circular for prokaryotic
    - Ribonucleic acid (RNA)
        - Messenger RNA (mRNA) carries info from DNA to ribosomes / cytoplasm → code delineated by ribosomes
    - RNAs involved in regulation of gene expression + processing + transport of RNAs and proteins
        - MicroRNA pathways to silence genes, regulate proteins, respond to invaders (defence)
    - DNA and RNA are polymers of *nucleotides*
    - Nucleotides consist of
        - Purines and pyrimidine bases DRAW THESE
            - Purines: adenine (A) and guanine (G) → 2 ringed
            - Pyrimidines: cytosine (C) and thymine (T) → 1 ring
        - 5C sugar (5' phosphorylated)
            - D-Ribose (RNA)
            - D-2'deoxyribose (DNA)
        - Phosphate: 1-3 phosphate at 5'C of sugar (CH_2OH)
    - Base pairing: H-bonding
        - N-H H-bonds with O or N
        - ↑ bonding str so melt at higher T
        - A-T → 2 H-bonds
        - G-C → 3 H-bonds
    - Backbone
        - Sugar phosphodiester forms backbone
        - Ribose for RNA
        - 2'-deoxyribose for DNA
        - *Nucleoside*: covalent bonding of C1 of sugar and base
            - Guanosine, Adenosine, Cytidine, Thymidine, Uridine
        - *Nucleotide*: Nucleoside + 5'phosphate (1-3)
            - Adenosine monophosphate (AMP)
            - Adenosine diphosphate (ADP)
            - Adenosine triphosphate (ATP)
    - Phosphodiester bond formation
        - DNA polymerases catalyze rxn
        - Use complementary dNTPs
        - Dehydration rxn covalent bond = phosphodiester bond
            - 3'-OH of new strand

        - 5'-phosphate of incoming dNTP
        - synthesis is 5'→3'
    - Always 5'-phosphate and 3'-OH that gives DNA polar sense
    - Complementary strands are *antiparallel*
- DNA
    - Geometry of bases + special arrangement to form H-bond causes helix structure of dDNA
    - In B-form → right handed dDNA
    - Pairing bases stack in centre
    - Backbone intertwined
    - Creates minor + major grooves
        - Enzymes usu recog side with major grooves then search for DNA sequence
    - 0.34 nm (3.4 Angstroms) rise per base pair
    - 1 full helix turn = 10 nucleotides
    - 1 period = 34 A
    - Width = 20 A
    - *Oligonucleotide*: artificially assembled DNA (3'→5')
- Central dogma
    - Complementary base pairing allows 1 strand of DNA to act as template for synthesis of complementary DNA / RNA strand
    - DNA *transcribed* to pass genetic info to RNA
    - Info in RNA present in *triplet code* where every 3 bases stands for 1 of 20 amino acids
    - *Translation*: mRNA codes for proteins
        - tRNA matches a.a.s with mRNA
    - Flow of info from DNA to protein called *central dogma*
    - Info flow: DNA → mRNA → Protein
- Mutations
    - DNA contains instructions for sequence of a.a.s in each protein
    - Order of a.a.s in protein determine its shape + fn
    - Errors / changes (*mutations*) in DNA can change a.a. sequence + fn of encoded protein
        - May be conserved
        - May show nothing
        - Variation → diversity → ↑ genomic plasticity → prevent genetic death
        - Some error prone polymerases kick in for mutations
            - 3 error prone
            - Activated by UV rays, pollution, outer enviro stresses
            - ↑ mutation rate with more stress
            - Can = dysfnal metabolic pathways → cells die → 1 individual dies → natural selection
    - Sickle cell anaemia due to 1 nucleotide change affecting haemoglobin → reduced O_2 carrying capacity

- Proteins
  - Most diverse of all macromlcls
  - Each cell has several thousand diff proteins
  - Proteins direct virtually all activities of cell
  - Fns include
    - Enzymes Struc components (e.g. keratin, collagen → rigidity)
    - Motility (e.g. actin) Regulatory (e.g. transcription factors)
    - Hormones (e.g. insulin) Transport (e.g. Na^+, K^+, ATPase)
    - Receptor (e.g. insulin receptors) Xport+storage of small mlcs (e.g.O_2)
    - Defence against infection (antibodies) Transmit info b/w cells (protein hormones)
  - .Amino acids
    - Polymers of 20 diff amino acids
    - 20 types with same orientation

      as this:

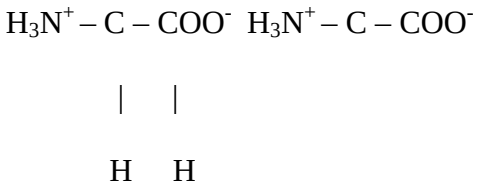
    - Grouped based on char of side chains
      - Nonpolar
      - Polar
      - Basic (charged)
      - Acidic (usu carboxyl grps)
  - Non polar amino acids
    - 10 a.a.s have non polar R-grps (hydrophobic)
    - Simplest is *Glycine* (R = H)
    - 2 contain S + 2 gave cyclic side chains
    - Nonpolar a.a.s tend to be buried in hydrophobic core of proteins
    - Glycine (Gly) G Alanine (Ala) A

$H_3N^+ - C - COO^-$ $H_3N^+ - C - COO^-$

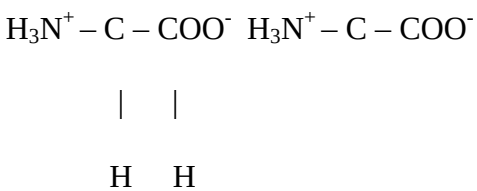
| |

H H

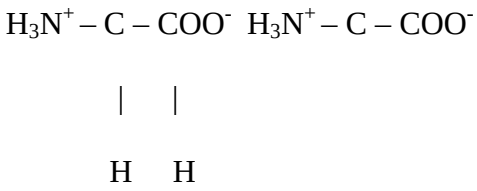
    - Valine (Val) V Leucine (Leu) L



- Isoleucine (Ile) I Proline (Pro) P



- Cysteine (Cys) C Methionine (Met) M



- Phenylalanine (Phe) F Tryptophan (Trp) W

- Polar amino acids
  - 5 a.a.s have polar R-grps → either –OH or NH_2 (hydrophilic)
  - Partial charge → H-bond formation with water
  - Polar a.a.s tend to appear on surface of proteins
  - Serine (Ser) S Threonine (Thr) T

$H_3N^+ – C – COO^-$ $H_3N^+ – C – COO^-$

| |

H H

- Tyrosine (Tyr) Y Asparagine (Asn) N

$H_3N^+ – C – COO^-$ $H_3N^+ – C – COO^-$

| |

H H

- Glutamine (Gln) Q

$H_3N^+ – C – COO^-$ $H_3N^+ – C – COO^-$

| |

H H

- Charged amino acids
  - 3 a.a.s have +vely charged NH_2 grps (basic)
  - Full charge → H-bond + ionic bond
  - Like polar a.a.s tend to appear on surface of proteins
  - Might take part in catalytic core of enzymes
  - Folding of protein on active site
  - Lysine (Lys) K Arginine (Arg) R

$H_3N^+ – C – COO^-$ $H_3N^+ – C – COO^-$

| |

H H

- Histidine (His) H
- 2 a.a.s have –vely charged $–COO^-$ grp (acidic)
- Full charge → H-bond + ionic bond
- Tend to appear on surface of proteins / enzyme catalytic core
- Aspartic acid (Asp) D Glutamic acid (Glu) E

$H_3N^+ – C – COO^-$ $H_3N^+ – C – COO^-$

| |

H H

- Peptide bond formation
  - *Polypeptides*: chains of a.a.s joined by peptide bonds → *dehydration* rxn
  - Number of a.a.s varied
    - Oxytocin – 9 a.a.
    - Insulin – 51 a.a.
    - Titin (connectin) – 34 350 a.a.s
    - Avg / protein – 400-500 a.a.s
  - 1 end polypeptide terminates in α amino grp (*N terminus*)
  - Other end is α carboxyl grp (*C terminus*)

- Protein structure
  - *Primary* struc → chain of a.a.s
    - Sequence of a.a.s in protein determined by order of nucleotide bases in gene
    - Can deduce a.a.s sequence from mRNA / nucleotide bases in gene
    - 3D config critical to proteins fn → must be 3D to work

        - Combined effect of a.a. side chains and folding that determines 3D shape + reactivity → interactive b/w a.a.s of same protein
        - Christian Anfinsen denatured *robonuclease* (RNase → degrades RNA to nucleotides) by heat treatment → breaks H-bonds
        - If treatment was *mild*, proteins would return to their normal shape at room T
        - Would mean that info for folding protein is in its primary sequence
            - Test by boiling → eventually reforms
            - Denature → lose fn → degrade RNA
            - Fold it → folds back to normal to be fnal
            - Sufficient to fn
- *Secondary* struc → spirals + sheets
    - Regular arrangement of a.a.s w/i localized regions
    - 2 types of secondary structure
        - Coil in *spiral helix* shape
        - Fold to form *β-pleated sheet* (parallel / antiparallel)
        - Both held together by H-bonds b/w CO and NH grps of peptide bonds

- *Tertiary* struc → spirals + sheets joined together → polypeptide chain
    - Folding of secondary struc elements to form 3D arrangement
    - 2° elements connected by loops + less ordered a.a.s
        - *Disulfide bonds* form
        - Can bond with itself (bridge can form on same strand)
    - Interactions b/w side chains of a.a.s in diff regions of protein stabilizes 3° struc
        - Covalent bonds (S-S bridge)
        - Hydrophobic interactions
        - Ionic bonds
        - H-bonds
    - Forms *domains*
        - Basic unit of tertiary struc
        - Def: Stable units of protein struc that can fold autonomously, compact struc, fn and evo, folding, exist independently
        - 2 proteins can be diff but make domain that has same fn as other enzymes
    - Similarly disrupting disulfide bonds (S-S) using chem. *denaturing* agents (e.g. β-mercaptoethanol) denatures proteins (-SH reforms)
        - Continues to fold again until reforms
    - Incubation under O_2 refolded RNase back to its fnal conformation
        - i.e. enzyme gained capacity to degrade RNA
        - Indicates that higher lvl of struc important for fn relies on covalent S-S bridge

        - Quaternary struc → many polypeptide chains joined
            - Interactions b/w diff polypeptide chains
            - Multimeric protein
            - Same bonds + forces for 3^{e} struc hold polypeptides together
            - In multi-subunit enzymes
            - Hemoglobin, for example, composed of 4 polypeptide chains

Cell Theory

- Basic properties of cells
    - Order + complexity
        - Surrounded + caused by *semi-permeable memb*, similar in comp + fn
        - Similar chem. composition: similarity of structural patterns of macromlcs + their fns
        - Similar biochem + regulatory processes
            - E.g. glucose, entry degradation, metabolism shared in all animals…
            - Indication of homology
            - See diversification of orgs
        - Hierarchal complexity from mlcs to supramlclar strucs
    - Self replication by division
        - All cells come from existing cells by division, process of division shares similarities among cells
        - Diff is division type → e.g. binary fission, mitotic division
    - Cells are small
        - Varied b/w ~0.5 to a few 100 µm (e.g. E. coli ~ 0.6 µm, RBC ~ 8 µm …)
        - Why cells small?
            - Limiting factor: SA to V ratio
            - Surface is portal of supply for V of cell (must be proportional to supply enough nutrients)
            - ↑ Reaches point where SA does not give means for cell to work (provide support) → results in division
        - SA / V ratio
            - V increases more quickly as sides get longer
            - Eukaryotes have ↑ S/V by extending internal memb surfaces
        - Bacteria vs. Eukaryotes
            - Genetic capacity is much more in human cell than E. coli
            - E. coli never uses space for nothing while humans have lots of junk space → comes from viruses
            - More proteins than bact are made but size isn't much larger → SA ↑ internally → e.g. Golgi, ER, mitochondria
            - Prokaryotes survive on their own
    - Properties
        - Interaction with enviro → anything out of cell → fluids, other cells…
        - All have transmittable genetic program
            - *DNA*: similar struc + fn, being inherited

        - *Genes*: units of genetic fn, basic similarity
        - *Evolution*: plasticity of genetic info, fnal divergence in response to environmental cues
    - Energy biogenesis
        - Similarity of metabolic pathways → e.g. glycolytic pathway, Krebs cycle, adaptive *variations* on general *theme*
        - Use of ATP as universal cellular E currency
        - Chemical + mechanical activity
            - Similar enzymes carry out metabolic rxns → *chem*. E/work
            - Use of E to do *mechanical* work
- Cell theory (discovery)
    - Robert Hooke (1665): 1st to observe unit struc in cork, called then "cells"
    - Anton Van Leeuwenhoek: discovers microbial single "cells"
    - Matthias Schleiden + Theodor Schwann (1839): independently concurred that all living organisms consist of unit struc called "cells" → cell theory
    - Rudolf Virchow (1855): expanded cell theory → each cell is result of div of previous cells
    - Modern cell bio addresses "how cells work" at mlclar lvl
        - To understand mlclar basis of cellular processes

- Cell diversity and beyond cells
    - Domains of life: cells are organized in 3 domains
    - *Prokaryotes*: Eubacteria, Archaebacteria
    - *Eukaryotes*: Protista, Fungi, Plants, Animals
    - Similarities
        - Cell memb
        - E metabolism
        - Genetic code
    - Differences
        - Prokaryotes are unicellular
        - Except for many membs of protista, rest of euk are multicellular
        - Size: prokaryotes ~ 0.5-5 μm, eukaryotes 5-500 μm
        - Memb bound organelles present in eukaryotes but not prokaryotes
        - Repro: cell fission in pro vs. mitosis in euk

Prokaryotes

- Structure
    - Bacterial cytoplasm surrounded by cell memb, cell wall, *polysaccharide capsule* for some
    - Typically 1-5 um (limited size)
    - *Cell wall*: used for protection, shape, rigidity → Arch have diff comp than Eubact

        - Composed of peptidoglycan → Eubacteria, polysaccharide of alternating *acetylated muramic acid* and *glucoseamine* (N-acetylglucoseamine)
        - β→4 glycosidic bond b/w sugars
        - Cross-linked by short oligopeptide
    - 2 types of bact based on cell wall
        - *Gram positive*: multiple layers of peptidoglycan
        - *Gram negative*: few layers of peptidoglycan
            - Periplasmic space
            - Many enzymes to synthesize PepG
    - Plasma memb
        - Beneath cell wall is plasma memb
            - Phospholipid bilayer with associated proteins
        - Steroid like mlcs instead of cholesterol
            - Fluidity for memb
    - Cytoplasm: lacks memb bound organelles
        - Ribosomes
            - 70S → 3 rRNA + 55 proteins (in 2 subunits 50S and 30S)
            - Protein synthesis
        - *Chromosomes*: single supercoiled chrom that resides in resin called *nucleoid* (not memb bound) → find chrom in area
            - Coiled usu in middle of cell → euk
            - Coiled by enzymes flipping into superhelical struc → pro
- Appendages
    - Have appendages with specific fns
    - *Flagella*
        - Composed of *flagellin* → helps bact move
        - Embedded motor made of 30-40 proteins → form rings
        - Some sense diff in periplasmic space + enviro → gradient
        - *PMF* (proto motor force) generated → released E → motor turns on → flagellum rotates → either clock or counter clockwise
    - *Fimbriae*
        - Small bristle-like fibres that allow bact to attach themselves to surfaces
        - Interact with enviro → help colonize to infect
    - *Sex pili* (F pilus)
        - Can mobilize whole chrom
        - Used for *conjugation* to xfer DNA from 1 bact to another
            - Gram +ve d more frequently than Gram –ve → carry genes for resistance
            - Genes coding for F-pilus (coating) are on F plasmid
            - Makes corridor to xfer DNA
- Reproduction
    - *Fission* instead of mitosis
    - Chromosomal DNA replication
        - Not in middle of cell
        - Passes through memb which does coding
        - Now embedded in it → wall forms afterwards

- Cytokinesis
  - Formation of *divisome* (several proteins that tag centre of cell)
    - Identifies exact centre
    - Build blocks (peptidoglycan)
    - Meet in middle = division
  - Cell wall peptidoglycan deposited
  - Chromosomes separated
    - 2^{nd} set starts before div
    - Multiple copies can occur
    - Theoretically 1 copy only
    - Makes recombination possible

Eukaryotic Organelles → REVIEW ALL VIDEOS FOR THIS SECTION

- Diversity
  - Protista: free-living marine unicell, some photosynthetic
  - Multicell orgs → fungi, plants, animals → not yeast
  - Differentiation occurs in cells
- Structure
  - Usu 10-30 μm
  - Separation of DNA + cytoplasm by nuclear envelope
  - Presence of memb-bound compartments with specialized fns: mitochondria, chloroplasts, ER, Golgi complex
  - Highly specialized motor proteins → also in prok
  - Nuclear envelope + internal membs
    - Originated from cell memb
    - Differentiated + acquired special fns
- Nucleus
  - 5-10 μm diameter: largest organelle → contains linear DNA mlcs → chromatin
  - Nucleus separated +limited from cytoplasm by nuclear membrane
    - *Nuclear pores*: import + export of RNA + proteins
    - *Nucleolus*: sites of rRNA synthesis + ribosomal subunits maturation → rRNA stored + preped
    - Nucleus contains *chromatin* w/i semifluid *nucleoplasm*
    - *Chromatin*: composed of DNA, protein, some RNA, usu network of fine strands
    - Condense during cell div to form visible *chromosomes*

  - *Nuclear envelope* separates nuclear content from cytoplasm
    - Selective traffic of proteins + RNA
    - Critical in regulating euk gene expression

- Consists of 2 memb, under both membs is nuclear lamina, nuclear pore complexes
  - *Outer* memb: continuous with ER, has memb proteins that bind cytoskeleton
  - *Inner* memb: has proteins that anchor nuclear lamina
- Each nuclear memb is phospholipid bilayer permeable only to small nonpolar mlcs
  - Nucleopores mediate interaction with nucleus + cytoplasm

- Nuclear lamina
  - Fibrous mesh that provides structural support
  - Made of proteins called *lamins*
  - Mammalian cells have 3 lamin genes (A, B, C)
  - 2 lamins interact to form dimer in which α-helical regions of 2 polypeptide chains wind around ea other to form *coiled coil*
    - *Dimer*: mlc made up of 2 simpler identical mlcs
    - 2 ends that interact head-tail
  - Lamin dimers associate with ea other to form nuclear lamina

- Nuclear pore complex
  - 2 mechanisms
    - *Passive transport*: small mlcs pass freely in either direction
    - *Active transport*: macromlcls (proteins + RNAs), E-dependent
  - *Nuclear pore*: Sole channels for small polar mlcs, ions, proteins, RNA to pass through nuclear envelope
  - *Nuclear pore complexes*: composed of ~30 diff pore proteins (*nucleoporins*)
    - Inner → anchoring proteins with network of lamin → organized distribution inside cells
    - Pore → not charged, not polar, not small → H_2O passes well so E not needed
  - *Pore complex* consists of 8 spokes connected to rings at nuclear + cytoplasmic surfaces → makes hydrophobic core
    - Proteins, RNA, RNPs (ribonuclear proteins) pass
  - *Spoke-ring* assembly surrounds central channel
  - Protein filaments extend from rings, forming basketlike struc on nuclear side
  - Proteins that must enter nucleus have aa sequences called *nuclear localization signals*
    - Need polymerases + helicases to get into nucleus → ribosomes transport to nucleus from cytoplasm
  - Recog by *nuclear transport receptors* (*importins*)

- Nuclear importation
  - Importin binds to NLS of cargo protein
    - *Cargo protein*: attached protein + signal aas → short segment of aas that are basic
  - Complex binds to cytoplasmic filaments of pore complex
  - Xport proceeds through pore complex by binding to *nucleoporins* (nuclear pore proteins)
  - Once inside, cargo/importin complex is disrupted by binding of Ran/GTP
    - Ran = Ras-related Nuclear proteins
    - Ras family domains regulate division + nuclear xport → GTP + GAP
  - Conformation change in importin releases cargo into nucleus
  - Importin-Ran/GTP complex exported back to cytoplasm where GTP hydrolyzed to GDP w/ *Ran GAP* (GTPase activating protein → in cytoplasm only)
    - Cannot bind with importin like this
  - Importin released back into cytoplasm + can participate in another round of xport
  - Ran/GDP xported back to nucleus by its own import receptor (*NTF2*)
    - *Ran GEF* (Guanine nucleotide exchange factor → in nucleus only) stimulates Ran/GDP to release GDP + pick up GTP → goes back to catalyzing importin
  - Ran/GTP gradient
    - Since Ran/GDP in cytoplasm + Ran/GTP in nucleus, gradient forms
    - Determines direction of transport
- Nuclear export
  - Proteins targeted for export from nucleus by specific aa sequences called *nuclear export signals*
  - Signals recog by receptors in nucleus (*exports*) which direct protein xpot to cytoplasm
  - Ran req for nuclear export and import
    - Needs exportin to make cargo protein + RAN bind
    - Cargo sent back → everything recycled
  - Ran/GTP promotes binding of exportins + cargo proteins, but *dissociates* complexes b/w importins + cargos
  - Exportins recycled through nuclear complex for reuse

- Ribosomes
  - Ribosomes subunits, 1 large + 1 small, are assembled in cytoplasm + used to *make proteins*
  - Found in both prok + euk

        - Nucleoprotein complex
        - Assembled in 2 subunits
        - Euk: 80S (60S+40S subunits)
        - Prok: 70S (50S+30S)
    - In euk cells, ribosomes can be found in diff locations + forms
        - Single free-ribosomes in cytoplasm
            - Grouped into polyribosomes, make cytosolic proteins
        - Attached to ER (rough ER), make proteins targeted to membs + organelles → goes to Golgi after
        - mRNA produced in nucleus
            - Many can attach to traveling ribosomes
        - Xported to cytoplasm by carrier proteins, pass through nuclear pore
    - *Translation*: tRNA into *A site* → into *P site* to get bonded → aa attaches to previous one with peptide bond in P site → leaves at *E site* → stops with release factor at stop codon
    - Proteins in ER for modification → in cytosol 1st
        - Same time as ribosome synthesizes them → *cotranslationally*
        - Signal sequence is translated ~ 70 aas → at N-terminus
        - *SRP* (signal recognition particle) binds to protein during translation
        - Binds protein + ribosome to *SRP receptor* in memb of ER → stalls translation doing this
        - GTP binds to SRP + SRP receptor to trigger transfer of signal sequence to *translocon*
        - Hydrolysis of GTP to GDP leads to dissociation of SRP from receptor + ribosome
        - Xfer to translocon + interacts to move its plug away
        - *Signal peptidase* cuts signal peptide to leave protein mlc in ER
    - Proteins xlated on RER
    - Post-translationally mod in Golgi → e.g. glycosylation
    - Targeted to other organelles → cell memb, lysosomes, Golgi, secretion

- ER and Golgi complex
    - *ER*: tubular membs + cisternae
        - *SER*: lipid + steroid synthesis
        - *RER*: protein synthesis → together with Golgi from cellular secretory machinery
    - *Golgi*: flattened memb sacs
        - Chem. modifies proteins from RER, sorts finished proteins to their destiny
        - Site of lipid synthesis + site of synthesis of some polysacc that compose cell wall (in plant cells)
    - Processing proteins VIDEO

        - From ER travel in transport vesicles to ER-Golgi intermediate compartment (*ERGIC*)
        - Xported + fuse to *cis* Golgi network → protein mod begins
        - Move into Golgi stack while ERGIC vesicles make new compartments in its place (stacks)
        - Compartments mature + become medial, *trans* compartments of Golgi stack, where further protein mod takes place
        - Cis-to-trans direction, carrying contents with them
        - Trans directs mlcs to final destinations
        - Compartment disappears + replaced by maturing compartment behind it
        - Vesicles w/i Golgi return Golgi proteins to earlier compartments for reuse

- Lysosomes
    - Golgi derived memb-bound vesicles (0.1-1.5 μm)
    - Formed in ER at first
    - Found in most euk
    - Involved in intracellular digestion + recycling of macromlcs
    - Contain about 40 acid *hydrolases* (hydrolytic enzymes)
        - Proteases, nucleases, phospholipases, amylases
        - Proteins + enzymes → breakdown by adding 1 H_2O mlc
        - **Active ~5.0 pH**
        - Rupture = not dead cell since cytoplasm pH ≠ 5.0
    - Maintain acidic enviro by pumping protons into their interior (pH 5.0)
    - Digest worn out cellular mlcs, engulfed bact + viruses
        - Fuse with *phagosomes* (e.g. engulfed bact) to form phagolysosomes
    - Lysosomes also participate in *apoptosis* → programmed cell death
    - *Tay-Sachs*: lysosomal dysfnal, autosomal recessive
        - Defective hydrolase involved in breakdown of phospholipids
        - Accumulation of lipids in neurons, infantile death
    - Process
        - When bact/virus ingested with phagocytosis
        - Fuses with particle containing vesicle (phagosome) + delivers hydrolytic enzymes
        - Can also fuse with organelles like old mito, to recycle material

- Peroxisomes
    - Assembly begins on RER, where 2 peroxins, Pex3 + Pex19 initially localize
        - Interaction causes Pex3/Pex19-containing vesicles to bud off ER

        - .Vesicles may then fuse w/ pre-existing Peroxisomes / w/ 1 another to form new peroxisomes
        - Peroxins synthesized on free ribosomes + imported
        - Protein import + addition of lipids from rough ER results in peroxisome growth + division
        - Enzyme content + metabolic activities of peroxisomes changes as they mature
    - *Zellweger syndrome*
        - Pex1, 2, 3, 5, 6, 12, 14, + 26 mutated (1:50 000)
        - Inefficient peroxisomal protein import
        - Long fatty acid chains accumulate in liver + neuron → leads to many neurological disorders
        - Neurological disorders, glaucoma, hepatic enlargement, mental incapacity, seizure, loss of muscular tone (↓ mobility), facial (skeletal) deformities + Jaundice
        - Lethal w/i few yrs → 6 months normally

- Proteasomes + protein degradation
    - After release, some vesicles deliver contents to lysosomes while others deliver to cell memb
    - Quality assurance mechanism
        - Unfolded / misfolded proteins are secreted into cytosol, targeted for destruction by *ubiquitin* polypeptides
        - Proteasomes destroy targeted proteins
    - Process VIDEO
        - Selective degradation of cytosolic + nuclear proteins
        - Use *ubiquitin* → 76 aa polypeptide
        - Attached in chains, targets proteins for rapid proteolysis
        - Attached with C-terminus aa (*glycine*) to aa grp of *lysine* in protein that needs to be degraded → attaches to next ubiquitin w/ a lysine
        - Several enzymes (*E1, E2, E3*) add ubiquitin to proteins
        - Proteasome recogs ubiquinated protein + degrades it
        - E1 (ubi-activating enzyme) activates C-terminus of ubi with rxn with ATP
        - Ubiquitin-AMP splits in rxn attaching ubi to E1, move N-terminus
        - E2 (ubi-conjugating enzyme) + E3 (ubiquitin ligase) add ubiquitin to proteins → many types of E2 + tons of E3, 1 type of E1,
        - Diff E3s recog diff substrate proteins → specificity
        - E3 recog + binds to protein and E2, ubi xferred from E1 to E2 to target protein → occurs multiple times to build chain
        - Proteins w/ ubi chain recog + degraded by proteasome → large multi subunit protease complex
        - Req E to unfold (ATP hydrolysis) → degrades into small peptides
        - Ubiquitin released to be used again

- *Stability* of proteins depends on whether they are ubiquinated

Plasma membrane

- Defines boundary of cell
- Selective interface: determines composition of cytoplasm → mediates interactions with enviro
- Fundamental struc: *phospholipid bilayer* (PB)
  - Proteins embedded in PB carry out specific fn
- Experimental evidence
  - Bilayer property
  - Scanned using e- micrograph → see distinct layers of memb
  - *Gorter & Grendel* (1925) → monolayer of extracted memb lipids of known # of RBC spread on water produced 2x SA → simple, no nucleus, rupturable
- Membrane contains proteins
  - Chem. comp → 50% protein + 50% lipid → (1 protein/~100 lipids)
  - Asymmetric distribution found by *freeze-fracture* followed by e- microscopy
  - Cut where weakest point is (intermemb space) → some proteins stuck in outer layer, some in inner layer → integrated

- *Fluid mosaic model*
  - Proteins + lipids show dynamic lateral movt in memb
    - Naturally reassembles b/c of phospholipid arrangement
  - Phospholipids + proteins freely diffuse laterally
  - Memb proteins of ½ of bilayer are structurally + fnally distinct from other half
    - Bilayers are *viscous fluids*, not solid → dual fn of cholesterol + amnt of unsat fatty acids
  - Proper fluidity maintained over broad range of T (cholesterol)
  - Frye + Edidin (1970) expt evidence
    - Fuse human + mouse cells, then analyzed for memb proteins using fluorescent antibodies
    - Red for human, green for mouse → cell fusion → hybrid human/mouse cell → memb proteins mixed +merged over hybrid cell surface → intermingled → shows yellow signal after
  - *Singer + Nicolson (1972)*: fluid mosaic model, accepted for all bio membs
  - *Desaturases*: produce unsat FAs → xform sat acids to unsat acids
    - Unsat FAs make kinks in chain → keep them from packing together
    - Regulation of desaturases controls amnts of unsat FAs → adjusts memb fluidity

- Molecular organization of membranes
  - Memb lipids

        - Asymmetric distribution of lipids
        - Phosphatidyl*choline*, glycolipids, *sphingomyelin* on *outer leaflet*
        - Phosphatidyl*serine*, phosphatidyl*inositol*, phosphatidyl*ethanoamine* on inner leaflet, -vely charged head grps *facing cytosol*
    - Cholesterol
        - Distributed equally in both layers
        - Polar –OH end aligned w/ hydrophilic heads+ hydrophobic ends w/ lipid tails
        - 2 roles → both resist ΔT
            - High temp: interferes w/ mobility of lipids preventing melt up + reduce permeability → closer tails
            - Low temp: ↓ lipid tails interactions + maintains fluidity + prevents memb freezing
        - *Lipid rafts*
            - *Cluster* of cholesterol, sphingomyelin + glycolipids
            - Highly-ordered than most of PB
            - Glycolipid (GPI) anchored proteins → C-terminus attachment of protein to cell memb
            - GPI = G lcosyl P hosphatidyl I nositol (glycosylphosphatidylinsitol)

- Membrane proteins
    - Receive signals + xport nutrients / waste
    - Glycolipid attached on memb to outside
    - Membrane integral proteins, traverse memb, N / C termini on either side of memb → i.e. in width of memb
    - *Peripheral proteins*
        - Loosely attached to 1 side by protein-protein interactions
            - i.e. not spanning whole memb
        - Signal xfer → cascades numerous protein interactions
        - Protein-protein interactions involve ionic bonds
        - Can be disrupted by polar reagents (salts / extreme pH) Glycolipid attached on memb to outside
        - Proteins dissociate from memb
    - *Transmembrane proteins*
        - Span whole memb
        - Contain hydrophobic transmemb domains (1+)
            - *Domain*: can be folded independently from rest of protein, region of a protein / polypeptide whose 3D config enables it to interact specifically w/ particular receptors / enzymes / other proteins
        - Detergents, *amphipathic* mlcs can solubilize these proteins
            - Surround around protein of hydrophobic → challenges interaction
            - Surround + expose hydrophilic to outside to be soluble
            - Not necessarily same struc when frozen

- *Protein movements*
    - Restricted where protein w/ *cytoskeleton* → sit in place + less mobile
    - Lateral diffusion of proteins is restricted for some → not flipping b/c of enzymes
    - Association w/ cytoskeleton / w/ other memb proteins
    - Proteins on adjacent cells / w/ extracellular matrix
        - Attach C-terminus end of protein to *GPI-anchorage protein*
    - Local lipid composition, GPI-anchored proteins *localize to lipid rafts*
    - *Polarized cells*: apical + basolateral memb domains
        - Apical layer not free movt
        - *Microvilli* absorb nutrients → travel pass cell → excrete into blood
        - Limitations based on tissue struc

- *Integral proteins*
    - Memb-spanning portions are usu α helices of 20-25 hydrophobic aas
        - Inserted into ER memb during synthesis
    - Carb grps added in ER + Golgi
    - Decision to integrate happens at time to production
        - Simplest mode of insertion involves proteins w/ *N-terminal signal* sequence
        - Translocation halfs at *stop-transfer sequence*
        - Stops some from going in → rest gets left out into cytosol
        - Protein exits translocon laterally
    - Becomes anchored in ER memb
    - *Internal signal sequences*
        - Anchored in ER memb by internal signal sequences that are not cleaved by signal peptidase
            - Usu short outside, long inside
        - No stop xfer sequence
            - Integral to memb anyways
            - Signal is hydrophobic + internal instead
            - Protein xferred in → N-terminus outside
        - If C-terminus outside
            - Internal signal in opp direction → longer chain outside then
            - Gives cell option to do both
        - Internal signal sequences act as transmemb α helices
        - Exit translocon + anchor proteins in ER memb in *either orientation*
    - *Multipass proteins*
        - Proteins that span memb multiples times are thought to be inserted by alternating series of *internal signal* sequences + transmemb *stop-transfer* sequences
        - More than 1 stop-transfer sequence
            - Xfers to memb → prepares gate → have more passes in cell memb

- *β barrel Transmembrane Domains*
  - *Porins*: transmemb proteins in outer memb of some bacteria
  - Cross memb as β barrels
  - Make outer memb highly permeable to ions + small polar mlcs
- *Glycocalyx*
  - Def: Carb portion of glycolipids, glycosylated proteins on outer face of plasma memb form *carbohydrate coat*
  - Polarity + thickness interferes w/ antibiotic interactions
    - Used by pathogens to recog where to attack
  - Protects cell from ionic + mechanical stress + is barrier to invading microorganisms
  - Oligosaccharides of glycocalyx participate in cell-cell interactions
  - WBCs (leukocytes) adhere to endothelial cells of BVs
    - Slows leukocytes to land + flatten
    - Lets open to attack inside
  - Involves transmembrane proteins *selectins*
  - Allows them to leave circulatory sys + mediate inflammatory responses
  - *Diapedesis*: seep into memb to address protein

- Transport of Small Molecules
  - Hydrophobic mlcs, small uncharged polar mlcs diffuse easily through cell
  - Large uncharged polar mlcs, ions reflected
  - Internal composition of cell is maintained b/c plasma memb selectively permeable to small mlcs
  - Mechanisms of transport
    - *Passive transport*
      - No chem. E req
      - Mlcs diffuse *down [ ] gradient* until eqbm reached
      - Simple diffusion: O_2, CO_2, H_2O, hydrophobic, small mlcs dissolve in memb → slow rate
    - *Facilitated diffusion*
      - Allows polar + charged mlcs (carbs, aas, nucleosides, ions) to cross plasma memb
      - No chem. E spent
      - Mediated by memb protein
        - Either direction
      - *Carrier protein* → integral protein
        - Bind mlcs on 1 side of memb (↑[ ])
        - Undergo conformational changes that allow mlc to pass through memb
        - Released on other side
      - *Channel proteins*
        - Form open pores through memb
        - Allow free diffusion of any mlc of correct size + charge

        - *Aquaporins* (plant + animal cells)
            - Allow H_2O mlcs to cross memb much more rapidly than they can diffuse through phospholipid bilayer
            - Impermeable to charged ion
- *Ion channels*
    - Well studied in nerve + muscle cells
    - Co-transport
    - Opening + closing of channels → xmission of electric signals
        - Xport through ion channels is extremely rapid: more than 1 000 000 ions/s
        - Most have "gates" that open in response to specific stimuli
    - Ion channels highly selective → specific channel proteins allow passage of Na^+, K^+, Ca^{2+} and Cl^-
        - *Voltage-gated channels* open in response to changes in electric potential across plasma memb → spread through
        - Voltage-gated Na^+, and K^+ channels are selective
        - Na^+ (0.95 Å) smaller than K^+ (1.33 Å) + it is thought that Na^+ channel pore is too narrow for K^+ / larger ions
    - 3D struc of K^+ channels determined by X-ray crystallography
        - Part of channel pore is lined w/ carbonyl oxygen (C=O) atoms from polypeptide backbone → i.e. **-CO- does not react** → water does not move
    - Displace water to which K^+ bound, + K^+ ion passes through
    - Na^+ too small to interact + remains bound to water
    - *Ligand-gated channels*
        - Ligand = anything attached to surface of memb
        - Open in response to binding of NTs / other signalling mlcs
        - NTs released into *synapse*, bind to receptors on another nerve cell to open ligand-gated ion channels
        - Pore blocked by side chains of hydrophobic aas
        - Binding of acetylcholine induces conformational change, hydrophobic side chains shift out of channel, which opens pore for +ve ions

- *Active transport*
    - Mlcs *transported against [ ] gradient*
    - Sodium-potassium pump (ion pumps)
        - Coupled rxn to ATP hydrolysis → need E
        - Na^+–K^+ pump (/ Na^+–K^+ ATPase) uses E from ATP hydrolysis to xport Na and K against electrochemical gradients
        - Pump operates by ATP-driven conformational changes
        - 3 Na xported out of cell + 2 K xported into cell for every ATP used
            - ATP → ADP for ea time carrier protein moves ions
            - P_i binds → releases outside → changes shape
            - K inside → moved outside when P unbinds → changes back
        - Can be driven by gradient too

- *Symport*: solutes move in *same direction* (Na^+/glucose)
    - Simultaneous binding to same transmembrane protein on same side
- *Antiport*: solutes move in *opp directions* (Na^+/Ca^{2+} antiporter)
    - Simultaneous binding to protein on opp side → counter-transport
- Flow of Na down electrochem gradient provides E for xport glucose against [gradient]
- ABC transporters
    - Chars by conserved ATP-binding domains
    - Called ATP-Binding Cassettes
    - >100 of this fam have been identified in prok + euk cells
    - Use E from ATP hydrolysis to xport mlcs in 1 direction
    - In bact, xport nutrient mlcs into cell including ions, sugars + aas
    - Euk cells: xport toxic substances out of cells
        - Solute not released until ATP hydrolyzed

- *Endocytosis*
    - Allows cells to take up macromlcs, fluids, + large particles like bact
    - These processes for bigger mlcs
    - Material surrounded by A of plasma memb which buds off inside cell to form vesicle containing ingested material
        - *Phagocytosis*: engulf bact into vesicle → fuses w/ lysosome + degrades bact
        - *Pinocytosis*: engulfing liquid like phagocytosis
- *Receptor mediated endocytosis*
    - 1st in studies of patients w/ familial hypercholesterolemia
    - Cholesterol xported in bloodstream in form of *low-density lipoprotein (LDL)*
        - Deliver to cells where cholesterol is taken out
        - Protein + phospholipid layer surrounds cholesterol mlcs
    - Macromlcs bind to cell surface receptors in specialized regions called *clathrin-coated pits* → protein recog by receptors
    - Internalization signals bind to LDL receptor, which in turn bind to clathrin
        - *Adaptin* = adaptor mlc → signal to cell
        - Adaptin recruits clathrin mlcs which coat memb
    - *Clathrin* assembles causing bend into basketlike struc that forms invaginated pits
        - Pits bud from memb to form small *clathrin-coated vesicles*
        - *Triskelion* structure (3-legged in Δ shape)
    - *Dynamin* forms rings around necks of pits, eventually leading to release of coated vesicles inside cell
    - After internalization, fuse w/ early endosomes → vesicles w/ tubular extensions at cell periphery → low pH causes LDL receptors to release LDL
        - Receptors recycled to plasma memb w/ budding vesicles
    - LDL remain in early endosomes as they mature to late endosomes + lysosomes for degradation
        - Frees cholesterol + aas b/c LDL digested too
    - Memb degraded → cholesterol released into cytosol for cell use

Mitochondria and Cellular Respiration

- Mitochondria
  - Structure
    - Generate E from lipids + carbs
    - Surrounded by double-memb sys
    - Inner memb has numerous folds (*cristae*), which extend into interior (*matrix*)
      - *Outer memb*: comm. from outside mito → permeable to small mlcs
      - Matrix couples rxns in *intermemb space w/ cristae*
        - Can be almost touching for transport reasons
        - Similar composition to cytosol
      - *Inner memb*: oxidative phosphorylation → impermeable to most ions + small mlcs → helps maintain proton gradient
      - Any ↑ permeability breaks down fn of mito
      - Memb completely diff from other organelles
    - *Porins*: channels in outer memb → allows free diffusion of small mlcs
    - Mito positioned near locations of ↑ E use
      - E.g. synapses in nerve cells, muscle cells
      - Need more vesicles for nerve + contraction for muscle
    - Continually fusing + dividing, remodels network of mito in cell + affects fn + morphology
    - Accommodates → dynamic, same size as bact + wiggle like bact
  - *Endosymbiotic origin*
    - Mito thought to have evolved from bact that began living inside larger cells (*endosymbiosis*)
    - Living orgs that have genomes most similar to mito genome are free-living α-proteobacteria
    - Double memb similar to Gram –ve bact
    - Genes xferred/shared from mito to chrom
      - Labour split
      - Now integrated into part of genome
  - Mitochondrial genome
    - Circular DNA 16 kbp → multiple copies
    - Almost size + similar composition of plasmids
    - Maternal inheritance
    - *D-loop*: origin of replication → comp like bact
    - Code for rRNAs, tRNAs, own ribosomes
    - Encode 13 proteins essential for oxidative phosphorylation
      - Electron transfer chain (ETC) complexes → I, III, IV, + V
    - Genetic code diff from universal code
      - Mito genetic code diff from universal code
      - U in tRNA anticodon can pair w/ any of 4 bases in 3rd codon position of mRNA → 4 codons are recog by single tRNA
      - Some codons specify diff aas in mito than in universal code
      - Trp on UGA happens on prok ~25%, euk 2-5% → error

        - Coding not specific in mito → shows that original prok cell had ability to translate UGA

- Transport + assembly of matrix proteins
    - Mito proteins
        - Contain 1000 – 1500 proteins but nearly half of them remain unidentified
        - Mito from diff tissues contain diff proteins
        - Some genes xferred to nucleus from original prokaryotic ancestor of mito
        - Cytosolic protein synthesis → mito transport
        - All Krebs enzymes in here → have xcription / xlation too
        - Double memb diff from cell memb
        - Xport req complex transport mechanism
        - Gene xlated to mito from outer ribosome → porin
        - Proteins in ER coded outside and inside
    - Matrix proteins
        - *Presequence*: N-terminal 25-35 positively charged aa targets proteins to matrix
            - Partially unfolded by Hsp70
            - Folding not quick + protect hydrophobic w/ other proteins
            - Hsp70 leave after → just assist xport + prevent bending
            - Prevent aggregation + solidifying as emerging from free ribosomes
        - Sequential ATP *hydrolysis*
            - ADP pulls protein inward
            - ATP replaces → lose P_i + pulled in further → repeat until in
        - Bind to receptors on *Tom* protein complex (*Translocase* of *Outer* Membrane)
            - Tom20 on pore → Tom 5 on Tom 20 → import pore → Tom40 → pass through → presequence binds to intermemb tail of Tom22
            - Intermemb space shrinks + both membs get closer
        - Bind *Tim* complex (*Translocase* of *Inner* Membrane)
            - Bind Tim21/50 of Tim 23 complex → into pore → into matrix
        - Once in matrix
            - Reversible binding w/ short hydrophobic aas
            - Powers binding + dissociation of Hsp70 w/i matrix
                - Protein integrated to memb or pulled into matrix
            - *Matrix processing peptidase (MPP)* cleaves presequence
            - Hsp70 binding assists proper folding

- Mitochondrial function
    - Oxidative catabolism of glucose + fatty acids
    - Matrix contains genetic sys + enzymes for oxidative metabolism

- *Pyruvate* (from glycolysis) is xported to mito → complete oxidation to CO_2 yields bulk of usable E (ATP) obtained from glucose metabolism
  - 3C structure pyruvate to 2C struc
  - Glycolytic enzymes catalyze glucose into 2 pyruvate
  - E stored w/ NADH, $FADH_2$

- *Glycolysis* – VIDEO
  - Universal pathway
  - Glucose starting substrate
  - Sequentially broken down to 2 pyruvate
  - 10 steps (cytosolic enzymes)
    - Early steps use 2 mlcs of ATP
    - Later steps: 4 ATP made
      - 2 NAD^+ reduced to NADH
    - 1. Consumed E → converts ATP to ADP → *hexokinase* uses ATP to phosphorylate glucose → *glucose-6-phosphate*
    - 2. Rearranged to *fructose-6-phosphate*
    - 3. *Phosphofructokinase* attaches 1 phosphate → *fructose-1,6-bisphosphate*
    - Phosphorylation rxn are irreversible + important regulatory points
    - Phosphofructokinase inhibited by ↑ lvls of ATP → inhibition results in accumulation of fructose-6-phosphate
      - Fructose-6-phosphate + glucose-6-phosphate reversible (eqbm) → block makes glucose-6-phosphate too
      - ↑ Glucose-6-phosphate inhibits hexokinase → glucose breakdown will be inhibited
      - Need low enough lvls of ATP to proceed w/ rxn
    - 4. Cleavage of fructose-1,6-bisphosphate into open chain →
    - 5. 2 mlcs of 3C sugar *glyceraldehydes-3-phosphate* (1 was something else after cleaving chain)
    - 6. Gets oxidized to 1,3-bisphosphoglycerate
      - 2 NADH produced here (2 NAD^+ used)
      - Mlc ↑ free E for hydrolysis → drives conversion of ADP to ATP
    - 7. Results is *3-phosphoglycerate*
    - 8. Converted to *2-phosphoglycertae*
    - 9. Then *phosphoenolpyruvate* → 2nd high E intermediate in glycolysis
    - 10. Hydrolysis → converts ADP to ATP → now *pyruvate*
  - Net yield: 2 ATP (4-2), 2 NADH, 2 pyruvate
    - NADH serves as donor for other oxidation-reduction rxns in cell
  - Provides substrates for Krebs cycle

- *Krebs cycle* – VIDEO
  - Euk cells, glycolysis takes place in cytosol

- Pyruvate then xported into mito where completely oxidized
- Pyruvate undergoes *oxidative decarboxylation* in presence of *coenzyme A* (*CoA-SH*), forming acetyl CoA → common carrier of acetyl grp
- Acetyl CoA enters *citric acid cycle* / Krebs cycle
  - 2C acetyl grp combines w/ *oxaloacetate* (4C) to yield *citrate* (6C)
- Remaining rxns, 2C of citrate completely oxidized to CO_2 + oxaloacetate regen
- Completes oxidation of glucose to 6 mlcs of CO_2
- Yields 1 GTP, 3 NADH, 1 reduced *flavin adenine dinucleotide* ($FADH_2$) another e- carrier

- Electron Transport Chain (ETC) – VIDEO
  - High E e-s from NADH + $FADH_2$ xferred through series of carriers in mito memb
  - E- carriers organized in ET complexes I, II, III, IV
  - Low E e-s from IV carried on O_2 + $2H^+$ to form H_2O
  - E from ETC used to pump protons to intermemb space
  - e- reduced slowly → step-wise → use e- gradually released from e- xfer
    - Each e- gets more protons pumped through to make E
  - Same arrangement of bacteria + periplasmic space
  - e- from FADH2 are xferred through complex II
  - Carried by Coenzyme Q to complex III + IV

  - NADH dehydrogenase, Coenzyme Q (ubiquinone) , Cytochrome b-c_1, Cytochrome C (e- 1 at a time), Cytochrome oxidase, ATP synthase
    - 2 e- from NADH xferred through NADH dehydrogenase complex
    - Next e- xferred to ubiquinone (mobile xfer mlc) to cytochrome b-c_1 complex
    - To cytochrome c which takes e- 1 at a time (1 H^+ pumped per e-)
    - Cytochrome oxidase needs 4 e- that interact w/ O_2 + 8 H^+ here → 4 pumped across, 2 make water
      - O_2 is final acceptor
      - Final e- is in lowest redox rxn
    - Xfer of protons across memb generates proton motive force (PMF) across memb of mito
    - Brought to ATP synthase to pump H^+ back into cell to make ATP
      - Formation of ATP is referred to as oxidative phosphorylation
    - We adapt to oxygen but others adapt to something else
    - Anaerobic use diff acceptors, N_2, NO_3, SO_4 …
  - Proton gradient established across inner memb
    - Leaks = big problem since no more gradient
  - Chemiosmotic coupling: E stored in H+ gradient is coupled to ATP synthesis
    - Does with ATP Synthase (Complex V)

- Oxidative phosphorylation – VIDEO
    - Protons cross memb only through protein channel (complex V)
    - Complex V (ATP Synthase) has 2 units (F_0 and F_1) linked by slender stalk
        - F_0 spans inner memb + forms channel through which protons move → regulated
            - Protons drive rotor in memb → bind to rotor subunits → chem. rotational E made → almost complete full circle → exit channel allows leaving to other side of memb → down shaft
        - F_1catalyzes synthesis of ATP
    - Flow of protons through F0 drives rotation of part of F1, which acts as rotary motor to drive ATP synthesis
        - Can reverse direction to catalyze ATP + pump out H^+
        - High ATP and low H^+ will reverse + hydrolyze ATP to pump protons out
    - 4 protons req to synthesize 1 ATP
    - Oxidation of 1 NADH yields 3 ATP → $FADH_2$ yields 2 ATP
    - Krebs + glycolysis: total 38 ATP/mlc of glucose (i.e. 2 pyruvate)

Cytoskeleton

- Structure + organization
    - Network of protein filaments + tubules
    - Extends from nucleus to plasma memb
    - Structural framework
        - Cell shape
        - Localize organelles
        - General organization of cytoplasm
    - Movt
        - Cell movt → helps single + multicell organisms move
        - Internal xport of organelles
        - Muscle contraction
    - Dynamic struc → continually reorganized → de/polymerizing
    - 3 main types of protein filaments
        - Actin filaments (7 nm)
        - Intermediate filaments (8-11 nm)
        - Microtubules (25 nm)

- Actin + actin filaments
    - *Actin filaments* (*microfilaments*):
        - Polymer of actin (7nm) → several μm in length
        - Actin 1st isolated from muscle cells 1942

        - Abundant in all types of euk cells
        - Mammals have 6 actin genes: 4 expressed in muscle cells, 2 non-muscle
        - Highly conserved → sequence identity → 95% identical b/w yeast actin + human actin
        - Prok ancestor is MreB (gene makes MreB protein)
            - Prok have cytoskel → specially localized under memb
            - Stretch inside of cell
        - Rod vs. Spherical
            - Shape b/c of cytoskel in prok
            - Protein is MreB makes rod, deformed MreB = spherical
            - Rods have it but spheres do not
            - Put protein in sphere + sphere becomes rod
    - 3D struc determined 1990
        - *Globular* actin (*G-actin*): 375 aa (43 kD (kiloDaltons))
            - Barbed + pointed ends
            - Binds head-tail to nucleate a *trimer* (3 G-actins)
        - *Filamentous* actin (*F-actin*): monomers added to both ends
        - Filament is polar → pointed end vs. barbed end

- Actin filament polymerization – VIDEO for up to end of proteins
    - Rate at which monomers added to filaments proportional to [ ]
    - G-actin monomers polymerize to form actin filaments
        - Assembly begins w/ nucleation of 3 monomers to form *trimer* (*nucleation*)
        - Nucleation is rate-limiting step of formation…
        - Happens of both ends making filaments → F-actin
        - High [ ] of subunits → filament growth → *faster at barbed end*
    - ATP bound actin binds to barbed end w/ high affinity
    - ADP-actin has low affinity to pointed ends
        - Barbed end grows 5-10 x faster than pointed end
    - When ATP hydrolyzes to ADP
        - ADP-actin dissociates from filaments more readily than ATP-actin
        - Critical [ ] of actin monomers is higher for addition to pointed end than to barbed end of actin filaments
    - Polymerization is reversible
        - Critical [ ] needed to depolymerise → eqbm
        - Dissociation + association rates equal at both ends
        - Also attached to ATP, ADP
    - *Treadmilling*
        - F-actin growth in 1 direction → gives directionality + formation
            - Dynamic growth
        - When concentration of monomers falls below certain lvl

        - Equivalent gain + loss of monomers
        - Barbed end will ↑ in length
        - Pointed end will ↓ in length
    - ATP bound to barbed end → adds to end of filament
        - ATP hydrolyzed to ADP soon after
        - Zone of ADP-bound monomers towards pointed end of filament
        - ADP-bound actin dissociates more than ATP-bound actin
        - ATP-actin associates more readily than ADP-actin
    - Pointed → barbed direction

- *Actin binding proteins* (ABP) → remodelling
    - Modulate assembly + disassembly of actin filaments → affects stability
    - Actin-binding proteins can
        - Bind along length of actin filaments, stabilizing them / cross-inking them to one another → form larger filaments
        - Stabilize by capping ends + preventing dissociation → permanent
        - Promote dissociation
        - Regulate exchange of ATP for ADP
        - Remodel / modify existing filaments
    - Actin binding domain (ABD) / Actin interaction has diverse fnality
    - Contribute to cellular role of actin filaments
    - *ADF / cofilin* (filament *severing* / depolymerization)
        - Bind to actin filaments + enhance rate of dissociation of ADP-actin monomers from pointed end
        - Remains bound to prevent reincorporation
        - Severs when 2 attach to monomers in middle of filament
        - Makes new barbed + pointed ends
    - *Profiling* (monomer *binding* / polymerization)
        - Reverse of cofilin → exchange actin-bound ADP for ATP (replaces cofilin)
        - Resulting in formation of actin/ATP monomers → can assemble
    - *Formin* (determines whether *filaments* form)
        - Binds to monomers to facilitate nucleation process (trimer)
        - Nucleate long unbranched actin filaments → e.g. muscles
    - *Arp2/3 complex* (determines whether *branches* form)
        - Actin Related Protein complex
        - Scaffold for branch nucleation
        - Bind near barbed ends of filaments
        - Promotes remodelling of actin cytoskeleton (like other proteins) req for movt + changes in cell shape

- Actin filament higher order assemblies

- Mediated w/ cross-linking proteins
- *Actin bundles*: cross-linked into closely packed parallel arrays
- *Actin networks*: cross-linked in arrays that form 3D meshworks
- Bundles
    - Parallel filaments cross-linked by actin-bundling proteins
    - 2 domains to bind actin + align filaments
    - 2 types of actin bundles
        - Non-contractile
            - Filaments (14nm apart) aligned in *parallel*, *same polarity*, barbed ends adjacent to plasma memb
            - *Fimbrin*: 68 kD protein, cross-links by its 2 ABD (domain)
            - Packed b/c fimbrin is small
            - Take part in variety of cell surface protrusions
                - Cell movt, phagocytosis, absorption of nutrients
            - Intestinal *microvilli*
                - Memb projections ↑ SA in epithelial lining of cells
                - Closely packed parallel bundles of 20-30 actin filaments
                - Relatively permanent → cytoskel affects it
                - Cross-linked by *fimbrin* + *villin*
                - Actin bundles attached to plasma memb by calcium-binding protein *calmodulin* w/ *myosin I*
                - At base → attach to actin cortex
            - Membrane protrusions
                - Other surface protrusions transient + form in response to environmental stimuli
                - *Pseudopodia*: phagocytosis / movt of amoebas
                - *Lamellipodia*: board, sheet-like extensions at leading edge of fibroblasts (web)
                - *Filopodia*: thin projections of plasma memb in migrating cells (web threads)
        - Contractile bundles
            - Widely-spaced filaments (40 nm)
            - Cross-linked by α-actinin
            - *α-actinin*: 102 kD protein w/ single ABD + *α-helical* spacer
            - Interacts w/ actin as dimer
                - Formed apart from each other
            - Increased spacing allows actin interaction w/ *myosin II* (motor protein)
            - Important in muscle fibre contraction
- Networks
    - Filamin: 280 kD form flexible cross-links
    - Filamin dimer: flexible V-shaped mlc
        - Actin-binding domains at end of each arm
        - Dimerization domain
        - β-sheet spacer

        - Binds actin perpendicularly
        - Forms 3D network beneath plasma memb
        - Network (*cell cortex*) determines cell shape + cell movt
        - RBCs as mode → no microtubules / intermediate fils
            - Lack other cytoskel components → cortical cytoskel principal determinant of cell shape
            - *Spectrin*: major actin-binding cortex protein
            - Tetramer of 2 polypeptide chains (α or β chains)
            - Ends of spectrin tetramers bind actin filaments resulting in spectrin-actin network
            - *Ankyrin*: links spectrin-actin network + plasma memb
            - Protein 4.1 is another link that binds spectrin-actin junctions + transmemb protein glycophorin

- Actin filaments w/ motor proteins + muscle contraction
    - Brings higher lvl of fnal complexity to cells
        - Cellular / organismal movt
        - Intracellular cargo xportation, cell div
    - Association w/ motor protein myosin
        - *Myosin*: mlclar motor → converts chem. E (ATP) to mechanical E → force + movt
        - *Muscle contraction*: model for understanding actin-myosin interactions + motor activity of myosin mlcs
    - *Muscle fibres* (myofibers): large cells (50 μm in diameter + up to several cm in length) → 1 giant cell of sarcomeres, 1 plasma memb surrounds them
        - Many nuclei
        - Made of actin + myosin
    - Cytoplasm consists of *myofibrils* → myosin fils 9 thin actin fils
    - *Sarcomeres*: myofibril units of skeletal + cardiac muscle → repeated
    - Actin fils attached at barbed ends to Z disc
        - Overlap of actin + myosin occurs here
    - *Titin*: extremely large protein → extends from M line to Z disc → 3000 aas
        - Keeps myosin II fils centered in sarcomere
        - Maintain resting tension that allows muscle to snap back if overextended
        - Scaffold for myosin
            - Myosin has polarity: 2 diff directions → interaction still same
            - Actin attached to Z disc → pointy end → 2 point in opp directions
    - *Nebulin*: associated w/ actin + regulate assembly of actin fils

- Sliding filament model
    - Proposed in 1954
    - Myosin slides on actin filament

- Sarcomere shortens, bringing Z discs closer
- No change in width of A band
- I band + H zone almost disappear
- Actin + myosin
    - Myosin crawls on actin
    - Max at 2 unit
    - Move closer → contract

- Thin + thick fils
    - Core of thin fil is actin fil → based on subunits (G-actin), appears helical
    - Globule site has myosin site but w/o nerve impulse, *tropomyosin* lies blocking sites + bound to actin fils + troponin
    - *Troponin complex* (TnI, TnC, TnT) that receives Ca^{2+} → can move tropomyosin + unblock sites
    - Nerve impulse stimulates release of Ca^{2+} from *sarcoplasmic reticulum*
    - Ca^{2+} binds troponin C → shifts complex → movt
    - Allows myosin binding to actin → shifts around to reveal sites
    - Initiates muscle contraction
    - Once Ca lvls ↓, shift everything back + cause muscles to relax
- Myosin II (in muscle): large protein w/ 2 heavy chains + 2 pairs of light chains
    - Heavy chains have globular head region + long α-helical tail
    - Tail twists around in coiled-coil
    - Globular heads bind actin
    - Myosin moves head grps along actin filament in direction of barbed end
- ATP hydrolysis req
    - Binding of ATP dissociates myosin from actin
    - ATP hydrolysis induces conformational change that displaces myosin head grp
    - Myosin head binds to new position on actin filament + P_i released
    - "Power stroke" → myosin head returns to original conformation → drives actin filament sliding + ADP released

- Actin and myosin in cell division
    - *Cytokinesis*: division of cell following mitosis
    - Contractile ring of actin + myosin II assembled underneath plasma memb
    - Contraction of ring pinches cell in 2
    - Myosin I: much smaller than myosin II → contains globular head grp + acts as mlclar motor
    - Short tails bind to other strucs
    - Movt of myosin I along actin filament
    - Transport cargo (e.g. vesicle)
        - Actin stretched from nucleus to nucleus of 2 cells

        - Contact w/ vesicular memb
        - Delivers vesicles needed
        - Head binds 1 by 1 → sliding motion of vesicle
- *Intermediate filaments*
    - Size: 50-200 kD (8-11 nm)
    - In vertebrates only
    - Supporting scaffold: for organelles + cytoskel
        - Provide mechanical / tensile str → not dynamic
        - In cells that req str
        - Not directly involved in cell movts
    - Diverse fibrous proteins → specific expression
        - Network + mesh work resists mechanical stress → connect *desmosomes*
        - Nuclear lamins: meshwork of nuclear memb to inner memb
    - Not polar → very stable
    - Regulated by phosphorylation
    - 6 types I-VI
        - Can have diff types in cell
        - I and II: *keratins*, small size (40-50 kD), epithelial cells
        - III: in many cells
            - *Vimentin* (54 kD): forms network from nucleus to cell periphery
            - *Desmin* (53 kD): connects Z-discs in muscle + stabilize actin-myosin
        - IV: *Neurofilaments* → variable size 67, 150, 200 kD (light, medium, heavy → NF-L, NF-M, NF-H) expressed in mature neurons
        - V: Nuclear lamins
        - VI: *Nestins* → stem cells, during embryonic dev
    - .Structure + assembly
        - α-helical rod domain of ~ 310-350 aa
        - Globular heal + tail domains, variable size
        - Head + tail domains determine specific fns
        - Folding patterns + interactions specific → based on C-terminus
        - Assembly
            - Orders of complexity built on simplicity
            - Relies on signal system of kinases → falls apart eventually
            - *Dimer*: central rod form coiled coil of 2 monomers
            - *Tetramer*: staggered antiparallel → opposite orientations
                - N and C grp opposite
            - *Protofilament*: tetramers assemble end to end → as long as needed
                - Hollow core w/ fils stacked around
            - *Filament*: 8 interwound protofilaments → force resistant → stretch
            - Ends are similar → stable
    - Interaction w/ cytoskeleton
        - Network in most cells
            - Extending from ring surrounding nucleus to plasma memb
        - Associates w/ cytoskel element → scaffold to organize internal struc
        - *Plectin* binds actin fils + microtubules

        - Plectin is 500 kD protein
    - N-terminus domain binds Keratin (intermediate fil)
    - C-domain binds cytoskel
    - Bridges them to int. fils + areas of cell to cell connections
    - Increases mechanical stability of cell
    - Binds like web / ladder
- Cellular role + disease
    - Elain Fuchs
        - Known that mechanical stress triggers dev of int. fils
        - Transgenic mice expressing mutant Keratin gene
        - Severe skin abnormalities
        - Rubbing skin causes oozing material + blistering
    - Epidermolysis Bullosa Simplex (EBS)
        - Keratin gene mutation (like mouse)
        - Patients dev skin blisters after minor trauma
    - Amyotrophic Lateral Sclerosis (ALS) → Stephen Hawking
        - Abnormalities of neurofils → dysfnal
        - Involving progressive loss of motor neurons
        - Muscle atrophy + paralysis
        - Wrong effect caused by motor neurons
        - Gene therapy is soln

- *Microtubules* (MT)
    - Rigid hollow rods (25 nm)
    - Dynamic strucs → undergo continual dis/assembly
    - Fn: cell movts + determining cell shape, organelle xport, mitosis
    - *Tubulin*: globular protein is monomer
        - α-tubulin and β-tubulin dimers make up microtubules
        - *γ-tubulin* in centrosome plays critical role in initiating assembly
    - Assembly
        - Consisting of 13 *protofilaments* assembled around hollow core
        - Protofilaments composed of head-to-tail arrays of tubulin dimers arranged in parallel
        - 2 distinct ends: fast growing + end and slow growing – end
        - Tubulin dimers polymerize to form microtubules
            - Polymerize at + end of MT → bound w/ GTP
            - Form sheet that zips up into mature MT behind region of growth → tip bound w/ GTP too
            - Rest bound w/ GDP → GTP hydrolyzed shortly after addition
            - Hydrolysis follows growth of MT
        - Can undergo treadmilling
            - Dimers w/ GTP bound to β-tubulin associate w/ growing end
            - GTP hydrolyzed → tubulin less stable → – end dimers disassociate
        - Dynamic instability (alternate b/w growth + shrinkage)

        - ↓ [tubulin-GTP], GTP at + end hydrolyzed + dimers *lost* → hydrolysis catches up to end → presence of GDP at end leads to disassembly + shrinkage (less binding affinity than GTP)
        - ↑ [tubulin-GTP] → dimers added more rapidly than GTP hydrolyzed → microtubule *grows*
- Centrosome + MAPs
    - Centrosome: microtubule organizing centre (MTOC) → amorphous proteins
    - Initiate microtubule growth
    - γ-tubulin as part of γ-tubulin ring complex in centrioles req → need for γ-protein
        - For animals → plants have centres, not centrioles
    - Microtubules extend to cell cortex
    - Might be stabilized locally
    - Microtubule-associated proteins (MAPs)
        - Like actin → capping, severing, dis/assembling rates, end tracking
        - Modulation of fn + stability
        - Post-translational mod of tubulin e.g. phosphorylation
        - Neurons
            - MT organized diff
            - Axon – to +
            - Dendrites: both direction → distribute any way
            - Not many ribosomes → use special proteins

- Association w/ motor proteins → myosin I, II
    - Head w/ 2 conformation state → mod w/ ATP/GTP hydrolysis
    - Kinesins + Dyneins
        - Responsible for powering movts involving microtubules
        - *Kinesin*: 2 heavy chains, 2 light chains, 40 types
            - Moves organelles toward + end → subgrp has N-terminus
            - Ends interact w/ stationary / mobile struc → organelles
            - Looks like giant feet walking (pulling big organelle)
        - *Dynein*: 2-3 heavy chains, number of light + intermediate chains
            - Move toward – end
            - Can walk in both directions (+/-)
        - Can randomly swap direction b/c protein N-terminus in diff direction
            - Can pass cargo to each other

- Higher order molecular machines
    - Cilia + flagella microtubule-based projections of plasma memb → cell movt
        - Covered by memb at base
        - Many cilia, few flagella → diff is size
    - Anchor stationary struc next to other 1
    - Cilia beat in coordinated back-and-forth motion

        - Unified beat (like windshield wipers)
    - Either moves cell through fluid / moves fluid over surface of cell
        - E.g. swimming w/ paramecium, mucus w/ respiratory sys
    - *Axoneme* of cilia + flagella consists of microtubules in:
        - *"9 + 2" pattern*: central pair surrounded by 9 outer doublets
    - Each doublet has complete A tubule + B tubule w/ 10-11 protofilaments, fused to A tubule → B gives surface for interaction → e.g. w/ dynein
    - *Nexin* links MT, 2 arms of dynein attached to each A tubule
        - Dynein arms are motor → make cilia move w/ lots of activity
    - – ends of MTs anchored in *basal body*
        - Contains 9 triples of microtubules
        - Basal bodies serve to initiate growth of axonermal MTs + to anchor cilia + flagella to surface of cell
        - – to + polymerization → push memb out
    - Movt of cilia + flagella: sliding of outer MT doublets relative to 1 another → powered by motor activity of axonemal dyneins
    - Dynein anchors bind to A tubules, while head grps lands on surface to B tubules of adjacent doublets
    - ATP/GTP surge
        - Confirmation change generates force xfer to tubule
        - Movement of dynein head grp towards – end of B tubule = bend
        - Accumulates + bends to 1 side

- Mitotic microtubules
    - MTs reorg during mitosis
    - Interphase MT disassembles
    - Free tubulin subunits reassembled into *mitotic spindle*
        - Divide, fall apart → wind chromosome + envelope dissolves
        - Micro + intermediate fils dissolve
        - Cell has no skel
        - Organelles released → distribute [ ] all over cell
        - 50/50 chance organelle in cell
        - Microtubules reform into spindle cords → separate chrom
    - Centrosome duplicates → MTOC forms at opposite poles of mitotic spindle
    - Process
        - *Kinetochore* MTs attach to condensed chrom at centromeres
        - *Chromosomal* MTs connect to ends of chrom via chromokinesin
            - Kinesin (walking) in opp direction
        - *Polar* MTs stablized by overlapping w/ each other in center of cell
            - Push away each other eventually
        - *Astral* MTs extend outward towards memb
    - Force generated to pull chrom apart → from centromeres → opens up
    - Anaphase A
        - Chrom moves towards spindle poles

- Anaphase B
    - Kinesins cross-link polar MTs + move them towards + end
    - Cytoplasmic dynein moves along astral MTs in – end direction
    - Poles kept apart
- –ve tracking
    - Tubules disassembled
    - Move to sides
    - Force pulls centrosome to either side
    - Everything reforms w/ disassembly