LS 7A Notes

Lecture Notes

9/28 - Week 0

- Cells are the basic units of life.
    - Need mechanism to separate and regulate internal environment to maintain homeostasis.
    - Need mechanisms to acquire, transform, and use energy.
    - Need some sort of information storage and transfer.
- Human body is mainly made up of carbon, hydrogen, and nitrogen.

10/3 - Week 1

- Different atoms have different numbers of electrons to share.
    - Carbon has 4, Hydrogen has 1, Oxygen has 2, and Nitrogen has 3
    - Determines the number and type of bonds they can form.
- Atoms don't like to share electrons equally
    - Electronegativity is a measure of how an much atom attracts electrons.
- Polar covalent bond is where we don't have an equal distribution of electrons between the two atoms.
    - One of the atoms will have a partial negative charge, and one will have a partial positive charge.
- Nonpolar covalent bonds have electrons that are shared equally between the two atoms.
- In a saturated fatty acid, there is one polar region and one nonpolar region
    - Since it has both regions, it is amphipathic.
    - The carbon chain with attached hydrogens is nonpolar, and the oxygen carbon double bond is the polar part.
- Outside and inside of the cell, we have polar environments, but the membrane is nonpolar.
- Hydrogen bonds happen mostly between water molecules where the partial positive hydrogen atoms get attracted to the negative oxygen.
    - Hydrogen bonds can't form with nonpolar surfaces.
- Molecules are hydrophilic based on its ability to form hydrogen bonds with water.
- Polar means that there is an atom that has a higher electronegativity and hogs the electrons.
- Van der Waals forces are electrostatic interactions between atoms that have a temporary partial charge.
    - It is temporary because the electrons are just moving around so much that at some point, they happen to build up on side of the molecule forming that partial temporary charge.
- Strength of bonds: Covalent, Ionic, Hydrogen, van der Waals forces.

- Bond strength is the amount of energy needed to break a bond.
    - Stronger bonds require more energy to break.
- When you have a negative potential energy on that graph, it refers to the energy required to break the bond.
    - The lower you go on the graph, the lower the potential energy and the stronger the bond.

10/5 - Week 1

- Membranes create barriers between the cell interior and the outside world.
- In eukaryotic cells, all the organelles are each surrounded by lipid bilayers, or membranes.
    - “Bunch of membranes contained within an outer membrane”
- Cells have different shapes, intracellular contents, and organization depending on their function within the body.
    - Red blood cells have a shape with a large surface area so that it can carry blood.
    - Depending on the cell’s function, they could have more or less of a particular organelle.
        - Red blood cells won’t have a lot of mitochondria.
        - Phagocytes eat a lot of material including foreign material, so they would have more lysosomes.
        - In B cells which secrete a lot of antibodies (proteins), you would see more Golgi and ER.
- Fluid mosaic model of cell membranes refers to how proteins are allowed to move within the membrane and how there is a large variety of proteins, lipids, carbohydrates, glycoproteins, etc.
- When you put phospholipid molecules in beakers of liquids, the molecules will always arrange in a bilayer structure, either with the heads pointing outward in water, or pointing inward when in oil.
- Cytoplasm is a location, basically everything except the nucleus. Cytosol just refers to the liquid (not the organelles).
- The phospholipid structure influences the fluidity of the membrane.
    - Phospholipids with saturated fatty acids with straight tails create rigid membranes.
        - Because the tails are so straight and packed, they have a lot of interactions with each other due to the van der Waals forces.
    - Phospholipids with unsaturated fatty acids with kinks in the tails which create fluid membranes because the tails are not packed as tightly together as thus aren’t influenced as much by van der Waals forces.
- As the temperature increases, the mobility of the membrane increases as well.
    - Cholesterol is the exception because it has the fused ring structure. With cholesterol, at low temperature, we will have an increased mobility. At high

temperatures, the mobility will increase, but not as quickly. Cholesterol basically acts as a buffer to make sure that membrane fluidity doesn't change as much.
    - If you want to maintain the fluidity of a membrane, adding cholesterol helps.

10/10 - Week 2

- Membranes are barriers that separate cells from the surrounding environment.
    - Need to be selective about what goes in and goes out.
- Hydrophobic/non polar molecules diffuse freely through a lipid bilayer.
- Cell membranes are permeable to water.
- Rate of diffusion is directly proportional to the concentration gradient.
    - Just know that solutes and solvents always move toward lower concentration when we're talking about passive transport.
        - Or with water, you can think in terms of the solute where the water will go from low solute to high solute concentration.
- Inference is taking a guess about something based on observations.
- Main difference between active and passive transport is that active transport will always use energy (ATP or charge difference) to move things against the concentration gradient while in passive transport it never uses energy and will move with the gradient (high to low).
    - The thing that is driving the secondary active transport (let's say Na+ in the example) will always move down the concentration gradient while also allowing something else (like glucose) to move against its gradient. If that Na+/Glucose transporter stopped working, then you won't have anything to move the glucose against its gradient. Thus the concentration of glucose inside would decrease.
- Want to keep sodium concentrations inside the cell to be lower than that of the external environment and want potassium concentrations inside the cell to be higher than that of the external environment
- The sodium-potassium pump uses ATP to kick out sodium ions from the cell to the outside (even though the concentration is higher outside anyway).
    - Form of primary active transport I believe.

10/12 - Week 2

- All biological processes involve energy transformations and some process require energy and some release it.
- Energy is the capacity to do work.
    - Chemical energy, thermal energy, radiant energy, and mechanical energy.
    - All of the above forms can be characterized as PE (stored energy) or KE.
- Work is the ability for cells to move or change things (aka do some sort of energy transformation).

- PE in biology is the structure/position of a molecule. Can also be stored in the concentration gradient. Molecules like ATP have bonds that store a lot of potential energy.
- KE in biology is just basically motion at the molecular scale.
- 2 Laws of Thermodynamics in biology.
    - Energy is conserved
    - Energy transformation is correlated with an increase in entropy.
- Cells must work against entropy to maintain order.
- Cells require energy to maintain internal order and increase the entropy of their surroundings.
    - When thinking about the entropy of the universe, it is always increasing.
- The overall change in chemical PE depends on the relative PEs of the reactants and the products.
- Gibbs Free Energy is the amount of energy available to do work.
    - Is a function of change in chemical PE, temperature, and change in entropy
- Exergonic reactions release energy, are spontaneous, and have negative delta G
- Endergonic reactions require energy, aren't spontaneous, and have positive delta G

10/17 - Week 3

- No matter if a reaction is catabolic or anabolic, you have to break and form bonds.
    - Breaking bonds requires an input of energy, and forming bonds releases energy.
    - Both of these reactions need that initial input in order to get to that transition state. The key difference is the amount of energy that gets released when you form those new bonds. That difference determines whether the final energy is greater or less than the initial energy, which lets you know whether the reaction is exergonic or endergonic.
- Enzymes decreases the free energy of the transition state, regardless of whether the reaction is endergonic or exergonic.
- For reversible reactions, a high concentration of reactants compared to products, the forward direction is exergonic. The opposite makes the reverse reaction exergonic. The enzyme will speed up the reaction in both directions.
- Changing the active site on the substrate can change the activity of the enzyme.
- Enzyme activity is responsive to environmental conditions like temperature and pH.
- pH is a measure of proton (H+) concentration. pH is inversely proportional to the proton concentration. Lower pH means a high concentration.
- You can think of electron carriers like FAD and NAD as energy management molecules.
- From one molecule of glucose, we can create 32 molecules of ATP. The energy cost of making 1 ATP molecule is 7.3 kcal. We got about 200 kcal in total. This is about 34% of the total energy given off by the cellular respiration equation (686 kcal).
- Glucose oxidizes to CO_2 and oxygen reduces to H_20.
- OIL RIG = Oxidation is loss of electrons and reduction is gain of electrons.

- Generally when a reactant goes from being small to a product that is large, it probably gained electrons and thus got reduced.
- Highly reduced forms of carbon have high PE.
- Strong bonds have low chemical energy and weak bonds have high chemical energy.

10/19 - Week 3

- C-H and C-C bonds have the most PE among single bonds.
- Big picture is that the cell concerts food molecules into CO2, free energy is released, transformed, and used to make ATP.
    - During stage 1 and 2, the fuel molecules are partially broken down to produce ATP and electron carriers.
    - Stage 3 is where the fuel molecules are completely broken down.
    - Stage 4 is where the electron carriers donate electrons to the electron transport chain (This stage is important because it generates a lot of ATP).
- Glycolysis takes place in the cytoplasm.
    - Everything else is in the matrix??
- The first 4 stages of glycolysis require energy, and are thus endergonic.
    - This is made possible by the coupling of some of these reactions to ATP hydrolysis as well as the build of reactants that move the reactions forward.
- Sometimes the final product of a set of reactions can be an allosteric inhibitor for the first reaction, so that the reaction eventually slows down
- The enzyme PFK is affected by 4 things
    - Activated by ADP and AMP
    - Inhibited by citrate and ATP
- NADH is more reduced than NAD+. NAD+ is an oxidizing agent (promoting the oxidation reaction).
- In the citric acid cycle, we get 2 carbons in (Acetyl CoA) and 2 carbons out (2 molecules of CO2).
- Following the citric acid cycle, but before the electron transport chain, most of the energy is stored in FADH.
- During oxidative phosphorylation, the electron complexes are pumping H+ into the intermembrane space creating a gradient. ATP synthase is really important because it takes that gradient and those H+ to create ATP.
- The production of ATP is endergonic and is directly coupled to the movement of those protons (with their concentration gradient) from high to low concentrations.
- Difference between substrate level phosphorylation and oxidative phosphorylation
    - The latter has actual mechanical energy being used to catalyze AP synthesis.

10/24 - Week 4

- Entropy increases in a closed system and yet organisms are able to be highly ordered. We're able to decrease entropy because we take energy in.

- Electrons have the most potential energy in the electron transport chain when it is excited by Photosystem 1. As the electron moves down the electron transport chain, it moves to lower energy state.

10/26 - Week 4

- Central dogma is DNA -> RNA -> protein
    - DNA -> RNA is transcription
    - RNA -> protein is translation
- In Eukaryotes, because we have the nucleus, we have more control over when proteins and translated, and when they are released. Prokaryotes don't have that nucleus and thus all of this must happen in the cell itself.
- When we add heat to a DNA molecule, it denatures and transforms to denatured DNA. When the temperature cools down, it forms back together and reforms into natured DNA.
    - The normal protein is in its native state.
    - Some denaturations are reversible and some are not.
- C-G have 3 hydrogen bonds while A-T has 2 hydrogen bonds. Thus, the C-G bond, you'll need a higher amount of heat to break the molecule.
- Gel electrophoresis is a way to measure the size of DNA
    - Add DNA (which is negatively charged) to the negative side of the gel and run power to it, and the DNA runs to the positive side
    - Separates DNA according to the size of the molecule/fragment.
    - The further down it is, the smaller the fragment is.
- In order for transcription to happen, the enhancer needs to be activated.
- Major groove of DNA is the bigger gap between strands.
- Interacting with the outside of the double helix is less energetically costly than unwinding DNA. RNA will look at this outside structure, and look at what's visible in the base pair bonds.

10/31 - Week 5

- In a eukaryotic cell, you have promoter regions that are a GC box, a CAT box, and some other box.
- Enzyme catalyzing transcription is called RNA polymerase.
    - Main function is make RNA from a DNA template.
    - As the RNA poly moves it is locally unwinding into the template and nontemplate strands.
- rNTP entry channel is where a bunch of nucleotides come in and attach to the correct complement of the DNA. The RNA polymerase is what does the base pairing.
- Basically, for each nucleotide added in base pairing occurs and then the phosphodiester bond forms.

11/2 - Week 5

- mRNA is always shorter than the DNA coding for it. This is because of the intron removal process called splicing.
- 5’ splice site is the side of the intron that is closer to the 5’ end of the mRNA.
- Peptide bond is formed in the A site, but then transferred to the P site.
- One tRNA synthetase for each amino acid.

11/7 - Week 6

- The 5’ cap is important for the recruitment of the small ribosomal subunit.
- Silent mutation is where there is a mutation but no change to the amino acid created.
- Missense mutation is where there is a mutation that changes the amino acid created
- Nonsense mutation is where there is a mutation that creates a stop codon.
- Frameshift mutation is where there is a mutation that deletes one nucleotide thus changing the reading frame for everything afterward.
- In frame insertions are where you add one or more amino acids without changing the reading frame (so multiples of 3 have to be added)
- In frame deletions are where you remove one or more amino acids without changing the reading frame (so multiples of 3 have to be removed)
- Transitions are changing nucleotides to a nucleotide that looks similar
    - A and G, C and T
- Transversions are changing nucleotides to a nucleotide that looks different
    - A and C, G and T
- More likely that transversions cause changes in the amino acids created.
- The initial binding of ribosomes and the mRNA is in the cytosol as the mRNA exits the nucleus.
    - After that initial binding the complex can then move to different areas depending on the type of protein but the beginning of the protein synthesis always STARTS in the cytosol.
    - Once the protein starts getting the synthesized, it outputs a signal sequence that is noticed by the SRP, which binds to the signal sequence, and determines where to take the protein to finish the translation.
        - Location could be in the rough ER or the cytosol. The rest of the protein will get translated in that location.
- All mRNAs exit the nucleus and enter the cytosol immediately.
- The vesicle uses v snares on the outside of the vesicle to determine where to go. It will look for the t snares on the target (such as a Golgi or a lysosome). The correct v snare will look for the correct t snare.
- Two vesicles that come off the ER and go to different places will both have different t snares and different v snares.

11/9 - Week 6

- Secreted proteins need to exit the cell by exocytosis

- If protein needs to be secreted, it is going to be synthesized in the rough ER. We basically need to be put in a vesicle, where it goes to the Golgi and then to the plasma membrane to exit the cell.
- SRP doesn't play a role in translation. It simply moves the move the protein complex to the ER. If there is a defect in the SRP, then the proteins will be translated in the cytosol, rather than the ER or the other locations where the SRP takes it.
- Our body has the same genome, but cells often have different functions because different parts of the genome are expressed at different times.
- Prokaryotic mRNA don't have 5' cap or poly A tail, but a single RNA that can code for multiple proteins. This characteristic is being polycistronic.
    - A polycistronic mRNA that encodes 3 different proteins will have 3 start codons and 3 stop codons. Because for each protein we need somewhere to start the protein translation and somewhere to stop.
- Initiation in prokaryotes is at Shine Dalgarno sequence (performs similar function to 5' cap but since prokaryotes don't have that 5' cap, it needs something else) and basically the site can recruit a ribosome. When the ribosome binds, it will start looking for a start codon.
- B galactosidase is an enzyme that cleaves the bond in lactose (and thus creates glucose and galactose) and catalyzes hydrolysis of lactose because lactose isn't in a form ready to be consumed.
- Negative control: Operon is expressed in the absence of the repressor.
- LacI creates a repressor protein.
- When the repressor (lacI) is present and lactose is absent, no transcription occurs because that repressor protein stops transcription.
- When the repressor (lacI) is present and lactose is present, the lactose will bind to the repressor which keeps the repressor from binding to the DNA, which means that the RNA polymerase can continue with its transcription.
- When no repressor is present, and regardless of whether lactose is present or not, the transcription always happens, because the repressor isn't there.
- A haplotype is telling us what genes are there in that particular bacteria.
- I+O+Z+
    - The plus means it's operating correctly. I is the repressor. O is the operator and Z is lacZ.
    - If all are functioning and no lactose is present and no inducer, then no need to produce B-gal and transcription won't happen.
    - If an inducer is present, then we would get B-gel produced.
- If the repressor gets mutated, then it can't bind.
- If the operator gets mutated, then you'll have continuous expression and a O constitutive mutation. It's an operator that can bind the repressor.
- I+O(C)Z+
    - This means that there is an O constitutive mutation. Thus, it doesn't matter if lactose is present. There is always going to be transcription even if there is no inducer.

- Positive control: Operon is expressed in the absence of the activator.
- Amount of glucose is inversely proportional to cAMP.
- When CAP is binds to DNA, lac operon is activated.
- Repressor acts as an on/off switch (basically creates a roadblock or doesn't) while CRP can adjust the level of transcription.
- First thing to always look for is the repressor because if that roadblock is present, doesn't matter if glucose or CRP is there.
- High glucose + low lactose = No transcription b/c roadblock from repressor
- High glucose + high lactose = Low transcription
- Low glucose + high lactose = High transcription
- When cAMP levels are high, they will bind to CAP and that means low levels of glucose.
- I-O+Z+ or I+O(C)Z+
    - First says no repressor and second says constitutive operator
    - Both will produce B-gal in the absence of lactose.

11/14 - Week 7

- Partial diploid?
- LacI is a trans acting element and the operator is a cis acting element
- Every cell in your body has the same DNA. The only differences between them is the type of gene expression that each one takes part in.
- We can control the transcription of DNA by sliding histones over by acetylating them.
- You can acetylate and deacetylate histones in order to alter its structure.
    - Histone acetylation is generally associated with increased gene expression.
        - Need the histone acetyltransferase protein to make that happen.
    - Histone deacetylation is generally associated with less gene expression.
        - Need the histone deacetylase protein to make that happen.
- Histone acetylation is a modification that happens after the protein is translated.
- The SWI/SNF complex is the molecule that actually moves the histones around. This will make transcription easier. It needs ATP to be functional.
- DNA methylation is associated with transcription inhibition or the silencing of genes.
    - DNMT is the short form
    - Methyl groups are added at cytosines.
    - Some repressors don't completely block gene expression, but DNA methylation contributes to completely shutting off gene expression.

11/16 - Week 7

- Dosage effects are related to the amount of gene expression
- In order to make sure that males and females express the same response from the X chromosome, females need to deactivate the effect of one of those chromosomes.
    - Using Xist is one of the methods
- Xist uses methylation to deactivate the effect of a chromosome.
    - You can expect a decrease of the expression of all the genes in a chromosome.

- There needs to be a bacterial promoter in the vector. It also needs an origin or replication in order to tell the plasmid to replicate. It also needs an antibiotic resistance marker
- Step 1 - Use restriction enzyme to cut at a specific spot
- Restriction enzymes recognize palindromic sequences of DNA.
    - Basically means that it reads the same across as the backward direction on its complementary strand.
- Step 2 - Isolate the insulin gene.
    - The cut in the previous step can result in sticky ends
    - When cutting, we want to have the same overhangs.

11/21 - Week 8

- Since bacteria don't splice introns, if you add a gene that does have a intron in it, then the protein that gets created won't be functional.
    - How can we express a eukaryotic gene in a prokaryote.
- Reverse transcriptase is an enzyme that can synthesize DNA using a RNA template.
    - It makes a cDNA based on the mature RNA, so it won't have any introns.
- Cells cultured with serum divide more frequently than those in plasma.
- https://www.albert.io/blog/g1-g2-phases-cell-cycle/
- For G1, you have a lot of cells with relatively little DNA. Then, during the S phase, we replicate our DNA and thus get more DNA and the number of cells goes down, and then during G2 phase, the number of cells will double because the DNA replication is complete.
    - Cells are found in G1 and fewer are found in the M phase because the cells spend way more time in the G1 phase (10 hours) than in the M phase (2 hours). Thus, at any given time, you'll find more cells in G1.
- DNA polymerase is the central catalyst behind DNA replication.
- DNA polymerase cannot initiate the DNA synthesis on its own, it has to latch onto a preexisting nucleic acid.
    - Needs some sort of primer strand.
- Eukaryotic DNA is linear, while prokaryotic is circular.
    - On prokaryotic chromosomes, you have an origin of replication so that it knows where to start.
    - On eukaryotic, we have multiple origins of replications so that the replication happen faster.

11/28 - Week 9

- Chromosome shortening occurs with an overhang from the parent strand, because the parent strand will have some length x and the daughter strand that gets created will always be less than x (since you have a leading strand end and a lagging strand end).
- Telomerase is an enzyme with an RNA template that can extend DNA strands.

    - Its job is to basically add gibberish to the end of the shortened chromosome so that the length doesn't exponentially decrease through several rounds of replication.
- Cells that need to divide a lot have a lot of that enzyme telomerase.
- PCR has reagents that really mimic what happens in actual DNA replication.
    - Main difference is that the primers are made of DNA instead of RNA.
- PCR gives you exponential growth of the target DNA.
- Human DNA polymerase won't be effective in PCR because the temperatures required in PCR are extremely high, while the optimal temperature for a typical human enzyme is around 30-40 degrees.
- DNA polymerase can't synthesize DNA without a primer.
    - The primer has to be specific to the target sequence.
- The reasons organisms can have the same number of genes but can be so different in terms of complexity
    - Alternative splicing
    - Gene expression
    - Chromosome duplication
- 99.5% of human DNA are the same from person to person
    - STRs and VNTRs are what differ
- STR - short tandem repeats are 2-9 bp repeats
- VNTR - variable number tandem repeats are 10-100 bp repeats
    - Number of repeats varies from individual to individual.

11/30 - Week 9

- You can have different alleles on each chromosome.
- Heterozygous chromosomes are where you have different alleles on each chromosome.
- If DNA polymerase adds a wrong base into a growing strand of DNA, then this mismatch is not a mutation.
    - It isn't a mutation because the mismatch can still be corrected by the proofreading function in DNA polymerase.
    - It becomes a mutation when it the mismatch becomes a permanent heritable change in the DNA. That happens during the next round of replication.
- Mutations can come from radiation, chemicals, and infectious agents.
- Types of DNA damage
    - Depurination, deamination, …
- HIV in particular has a high mutation rate because its RNA polymerase does not have proofreading ability.
- DNA polymerase makes a mistake every 10^7 pairs, but the actual mutation rate is 10^-11 because there are other ways besides the polymerase proofreading function to catch mistakes before they become mutations.
    - DNA repair pathways
        - Mismatch repair

        - Base excision repair - Fixes small problems with single bases
        - Nucleotide excision repair - Fixes bigger problems
- Really dangerous when you have a double stranded DNA backbone break.
- Base excision is different from mismatch repair because mismatch repair is for correcting errors with the DNA polymerase and with DNA replication while base excision is more when there is a damaged base that needs to be removed and corrected.
    - When the mutation is induced by a mutagen, that is a signal for base excision.

12/5 - Week 10

- BRCA1 deficiencies can lead to large chromosomal changes.
    - Weird combination of chromosomes that leads to problems.
- When you see a breast cancer karyotype
- 3 major cell cycle checkpoints
    - G2/M Checkpoint - Do I have the DNA replicates and is all the damage repaired?
    - M Checkpoint - Are all the chromosomes properly attached to the mitotic spindle?
    - G1/S Checkpoint - Is the environment favorable for the actual replication of the DNA?
- Cyclin's expression values will change as the cell moves through its cell cycle.
- 3 different cyclins that correspond to an expression pattern that reflects with they are needed in the cycle
    - G2/M phase cyclin
    - G1/S phase cyclin
    - M phase cyclin
- They each bind to a CDK.
- Homologous chromosomes are found in meiosis when you have one chromosome from the father and one for the father. In mitosis, we just have chromosomes and sister chromatids that get duplicated.
- Cohesin is a molecule that holds the sister chromatids together.
    - During Anaphase, the last cohesin molecules have to be cleaved so that the daughter chromosomes can get pulled apart.
- Microtubules are huge molecules that can grow and shrink.
- As the mitotic spindle pulls the chromatids apart, the microtubules must shorten from one end
    - If the microtubules shorten at the spindle pole end, then the dark mark will move to the left.
    - If they shorten at the kinetochores, then the dark mark will stay at the same place.
- In reality, the microtubules shorten at the kinetochores.

12/7 - Week 10

- Proto-oncogenes are moving the cell cycle forward, while tumor suppressor genes hold it back.

- Mutations leading to cancer
    - Proto-oncogenes can turn into oncogenes and cause the cell cycle to go too fast.
        - Gain of function mutation
    - Tumor suppressor genes can be inactivated and thus there is no opposing force to the cell cycle.
        - Loss of function mutation
- Different karyotypes can have size differences between the set of chromosomes (result of translocations, insertions, deletions) or if you have differences in number of chromosomes, that could be due to separation issues during cell cycle.
- Mitosis cell cycle checkpoint
    - If kinetochores are unattached to the microtubules, then that sends signals causing MAD to bind to CDC20. When **all** the kinetochores are attached, then MAD releases CDC20 and CDC20 binds to APC.
    - Then that complex ubiquitinates Securin which releases Separase from Securin.
        - If Securin has a loss of function, then Separase would be free and thus the cell would go into anaphase prematurely.
        - Separase is a proto-oncogene while Securin is a suppressor.
    - The Separase then goes and attacks the chromosome thus destroying all the cohesin, causing the chromatids to separate.

Textbook Notes

Chapter 2 - Chemistry of Life

2.1 - Properties of Atoms

- Fundamental substances are **elements**.
    - Pure substances that can't be broken down further.
- Elements contain only one type of atom.
- Atoms contain dense nucleus with surrounding electrons.
- Isotopes are atoms of the same element with different number of neutrons.
- Electrically charged atoms are ions
    - Positive charge = **cations**
    - Negative charge = **anions**
- Orbitals are locations an electron is present most of the time.
    - Max # of electrons in any orbital is 2.
- Electrons in orbitals close to nucleus have less energy than those far away.
- Several orbitals can exist at one energy level, or shell.
- After the first shell, the max number of electrons per shell is 8.
- For the first 3 rows of the periodic table, elements in the same row have the same number of shells.
- Elements in each vertical column are called a **group**

2.2 - Molecules and Chemical Bonds

- Atoms combine to form other molecules.
    - The atoms interact through chemical bonds.
- **Valence electrons** are the ones at the outermost orbitals, and are at the highest energy level.
    - Atoms share valence electrons when they form a molecule.
    - Merged orbital is called a molecular orbital.
    - Shared electrons constitute a covalent bond.
- 2 adjacent atoms can share 2 pairs of electrons, forming a double bond.
- Electrons are not shared equally if there is a large difference in electronegativity between the two atoms.
    - Known as polar covalent bond.
- Ionic bonds are between oppositely charged ions.
- Sodium chloride (NaCl) is an ionic bond.
    - When it is placed in water, the salt dissolves into its ions.

2.3 - Water: The Medium of Life

- Water is the most abundant molecule in cells.
- **Hydrogen bonds** are the interactions between a hydrogen atom and an electronegative atom of another molecule (often Oxygen, Nitrogen, or Flourine).
    - Most form between 2 water molecules.
    - Much weaker than covalent bonds.
- Interactions between 2 temporarily polar molecules are referred to as **van der Waals forces**.
    - Not as strong as interactions between charged particles.

Chapter 5 - Cell Organization and Membranes

5.1 - Structure of Cell Membranes

- A **cell** is the smallest, most basic unit of living things.
- Cell theory states that all organisms are made of cells, cells are the fundamental units of life, and cells come from preexisting cells.
- **Membranes** physically separate cells from their external environments.
    - Lipids are the main components of cell membranes.
        - They form barriers in watery environments.
    - Proteins and carbohydrates are also found in cell membranes.
- **Lipids** are hydrophobic molecules.
    - Primary function is long term energy storage.
    - Also used for protection, insulation, and lubrication.
- 4 types of lipids: Triglycerides, phospholipids, steroids, and waxes.
- Lipids are insoluble in water.
- **Triglycerides** include fats and oils.

    - Contain 2 types of molecules: glycerol, 3 fatty acids.
        - Fatty acids have a hydrocarbon chain, a methyl group, and an acid group.
            - Can be saturated (single C-C bonds) or unsaturated (> 1 double blonds)
            - Saturated are mainly solid and unsaturated are liquid.
    - 3 fatty acids + 1 glycerol molecule have to bond together to become a triglyceride through dehydration synthesis.
- **Phospholipids** take glycerol and 2 fatty acids
    - A phosphate group rather than a 3rd fatty acid is attached to the 3rd carbon of glycerol.
    - Phosphate head of molecule is hydrophilic but the tails are hydrophobic.
    - The structure is the major component of the plasma membrane in the cell.
- **Steroids** are composed of 4 fused rings of carbons.
    - Cholesterol is a steroid and also found in the plasma membrane.
- **Waxes** are nonpolar.
    - Found in plants, ears, and honey.
- Molecules with hydrophobic and hydrophilic regions are **amphipathic**.
    - Since phospholipids are amphipathic, they will arrange in structures when the polar heads are outside interacting with water, and the tails point inward.
- Lipids with bulky heads and one tail go into structures called **micelles**.
- Lipids with 2 tails go into structures called **bilayers**.
    - Bilayers can then combine to form **liposomes**.
- Bilayer structures form spontaneously.
    - pH of the solution needs to be similar to that of the cell so that the heads are in their charged form.
- Lipids associate with one another through weak van der Waals forces.
    - This allows cell membranes to be dynamic and fluid.
    - Fluidity of membrane depends on types of lipids (length of tails, presence of double bonds, etc).
- Longer the fatty acid tails -> more surface area -> tighter packing -> reduced lipid mobility.
- Double bonds -> kinks in the tails -> less tightly packed -> enhanced lipid mobility.
- Cell membranes contain other types of lipids besides phospholipids.
    - Cholesterol is another type, it is also amphipathic.
        - Hydrophilic region is a hydroxyl group and hydrophobic region consists of 4 carbon rings. They will interact with the other fatty acid tails.
- Cholesterol decreases membrane fluidity because of its rigid ring structure, which hurts mobility.
    - At low temperatures though, cholesterol increases fluidity because it prevents other phospholipids from packing extremely tightly.
- Specific types of lipids also arrange in lipid rafts.
    - Thus membranes aren't always uniform, but can be composed of discrete components.

- Transfer of a lipid between layers of the bilayer which is called a lipid flip flop is rare.
- Proteins required for structure, function, regulation of tissues and organs.
    - Made up of amino acids attached together in chains.
    - Sequence of amino acids determine a protein's 3-D structure.
- Proteins can be antibodies, enzymes, messengers, structural components, and transport/storage.
- Most membranes contain proteins and lipids.
- Protein functions.
    - Transporter: Move ions/molecules across membrane.
    - Receptors: Allow cell to receive signal from environment.
    - Enzymes: Catalyze chemical reactions.
    - Anchors: Attach to other proteins to maintain shape.
- **Integral membrane proteins** are permanently associated with cell membrane, cannot be separated.
- **Peripheral membrane proteins** are temporarily associated with the lipid bilayer.
    - Mainly associated with either the internal or external side of the membrane.
- **Transmembrane proteins** are integral ones which span the entire lipid bilayer.
- We know proteins move in the membrane because of the Fluorescent recovery after photobleaching (FRAP).

5.2 - The Plasma Membrane and Cell Wall

- The internal environment of the cell needs to operate in a particular pH range and salt concentration while the outside environment is constantly changing.
- Many organisms have a **cell wall** external to the plasma membrane.
    - It keeps the shape of these cells.
- Active maintenance of a constant environment is **homeostasis**.
    - The plasma membrane maintains this because it is selectively permeable b/c it lets some molecules in and out freely under certain conditions.
- Hydrophobic interior of the lipid bilayer prevents ions or polar molecules from diffusing across the membrane, and some molecules are too large to cross on their own.
- Gases, lipids, and small polar molecules can move across the layer.
- The most important thing in determining whether a molecule can pass is the charge. If a molecule is charged it <u>really</u> wants to be in a polar environment.
- Main things to determine whether a molecule can pass through the bilayer, the size/polarity/charge
    - If charged (Ions), they can't get through. If not charged (non polar), they can get through.
    - If polar and large, then it's unlikely but if polar and small, then it's likely
- Protein transporters allow the export and import of molecules, including ions and nutrients that can't cross on their own.
- Simplest form of movement in and out of the cells is **passive transport**.
    - Works by **diffusion**, which is the random movement of molecules.

        - Leads to net movement of a substance to places with lower concentrations.
        - It is an exergonic process.
    - Passive transport only works when the concentration gradient is in the right direction, from higher on the outside to lower on the inside.
- Some molecules diffuse freely across the plasma membrane as a result of differences in concentrations between the inside and outside of the cell.
- Molecules that can't move across the layer directly can move passively toward low concentration regions through protein transporters.
    - When molecules moves across the bilayer through a membrane protein, that is **facilitated diffusion**.
- Membrane transporters can be channels or carriers. When molecules use channels or carriers, they are taking part in facilitated diffusion.
    - **Channels** provide an opening between the inside and outside of the cell within which certain molecules can pass.
        - Some of these channels are gated and open in response to a signal.
    - **Carriers** bind to and then transport certain molecules.
        - One type of carrier is open to one side of the bilayer and the other is open to the other side. When the molecule binds to the protein, it then opens to the other side so that the molecule can go through.
- Even though outer membrane is hydrophobic, water molecules are small enough to pass through by simple diffusion.
- There are specific protein channels called aquaporins for transporting water molecules.
- Net movement of solvents like water across the membrane to a higher solute concentration is called **osmosis**.
- Water will move from regions of lower solute concentration toward regions of higher concentration (or from higher water concentration to lower water concentration).
- The amount of diffusion is influenced by the thermal energy of the environment and the energy transferred from molecular collisions in the cell.
- A **hypertonic** solution is one where there is a high salt concentration compared to another solution.
    - When a red blood cell is placed in a hypertonic solution, water moves from inside to outside the cell by osmosis to equalize the water concentration. The cell also shrinks.
- A **hypotonic** solution has a low concentration of salt compared to another solution.
    - For the red blood cell situation, water would go into the cell and it would increase in size.
- Uphill movement of cells **against** a concentration gradient is active transport and it requires energy.
- Some transport proteins act as pumps that use energy to directly move a substance in or out of a cell.
- **Active transport** uses forms of energy to pump molecules against their concentration gradients.

- Within cells, sodium is kept at concentrations lower than the external environment, and potassium is kept at concentrations higher than external environment.
    - Sodium potassium pump actively moves sodium out of the cell, and potassium into the cell.
        - To power this pump, it uses chemical energy stored in ATP. Active transport that uses energy directly is called primary active transport.
- **Antiporters** are the protein transporters that move sodium and potassium ions in different directions.
- **Symporters** or cotransporters move molecules in the same direction.
- Transport proteins can also build up concentration of a small ion on one side, and this stores potential energy to drive the movement of other substances across the membrane.
    - These proteins can facilitate the movement of protons so that there is a charge difference between the inside and outside of the cell.
- The pump generates a concentration gradient which is also called a chemical gradient.
- In addition to chemical gradients, a difference in charge favors the movement of protons back across the membrane.
    - Difference in charge causes an electrical gradient.
- A gradient that has both charge and chemical components is called an **electrochemical gradient**.
    - This stores energy which can be harnessed to move molecules in or out of the cells.
- When the movement of a molecule is driven by movement of protons and not by ATP, the form is called **secondary active transport**.
- Cells use active transport to maintain equal concentrations inside and outside.
- Human red blood cells avoid shrinking or bursting by maintaining an intracellular environment isotonic with the extracellular environment.
    - For single celled organisms, they have contractile vacuoles that take up excess water from inside that cell, and expel it into the external environment.
- Moving “downhill” means going from regions of high to low concentration. When this happens, it is called passive transport or facilitated diffusion (if a protein is involved) and it doesn’t require energy. Processes that do require energy and thus go against gradients go under active transport.

5.3 - The Internal Organization of Cells

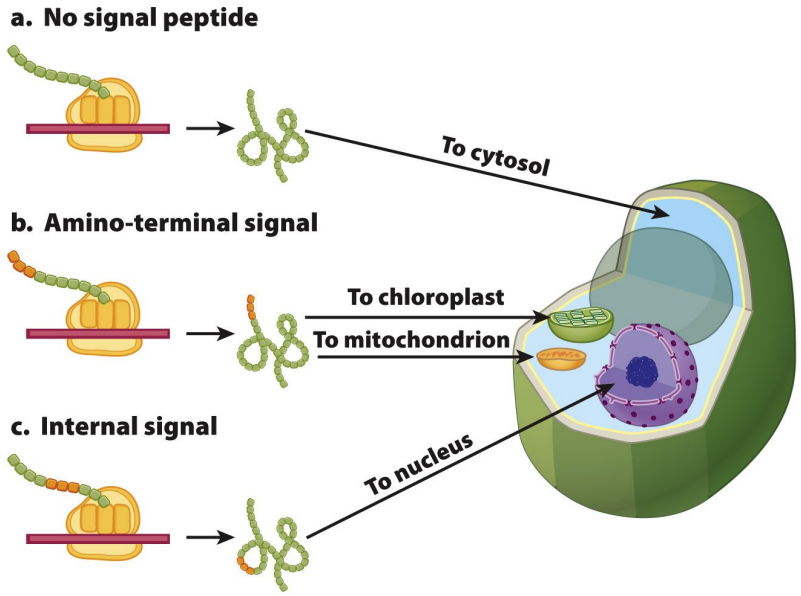
- All cells have a plasma membrane and contain genetic material.
- Prokaryotes don’t have a nucleus, eukaryotes do.
    - Presence of nucleus allows for transcription and translation to be separated -> complex gene expression
- Prokaryotes and eukaryotes also have different lipids in their cell membranes.
    - Prokaryotes synthesize hopanoids, not steroids.
- Since no nucleus in prokaryotes, no physical barrier separating genetic material from the rest of the cell.

    - DNA concentrated in the nucleoid.
    - Bacteria have circular molecules of DNA known as **plasmids**.
        - They are transferred between bacteria through threadlike structures called **pili**.
- Since prokaryotes are small, they have high SA to volume ratio and can thus absorb nutrients from environment.
- Eukaryotes' nucleus holds majority of cell's DNA.
    - Nuclear membrane allows for complex regulation of gene expression.
- DNA transcribed to RNA in nucleus, but RNA molecules carry the message from inside to outside the nucleus.
- Eukaryotes have membranes which define compartments or organelles.
    - The endoplasmic reticulum is where proteins and lipids are synthesized.
    - The Golgi modifies and sorts the proteins and lipids.
    - Lysosomes have enzymes that break down macromolecules.
    - Peroxisomes contain enzymes involved in metabolic reactions.
    - Mitochondria harness energy for the cell.
    - Cytoskeleton provides cell with internal structure.
- Plant cells have cell wall outside of the cell membrane, vacuoles for water uptake, and chloroplasts that convert energy from sunlight to chemical energy.
    - Cytoplasm is all the other stuff in the cell besides the nucleus.
    - Jelly like internal environment that surrounds organelles is cytosol.

5.4 - The Endomembrane System

- The membranes of organelles in a cell are physically connected by membrane bridges or connected by **vesicles** which are small sacs that transport substances within a cell to the exterior.
    - When a vesicle fuses with the plasma membrane, the process is called **exocytosis**. Provides a way for the vesicle to empty its contents to the extracellular space.
    - Vesicles can also bud off from the plasma membrane and bring outside stuff into the cell by **endocytosis**.
- Interconnected membranes make up the endomembrane system.
- Extensive internal membrane common in eukaryotic, but not prokaryotic cells.
- The **nuclear envelope** defines the boundary of the nucleus. Has an inner and outer membrane.
- The 2 membranes are continuous with each other at openings called **nuclear pores**.
    - They are proteins that allow molecules to move in and out of the cell.
    - mRNA moves through these pores.
        - They go outside the nucleus and bind to ribosomes.
- Outer membrane of envelope is continuous with the ER. It produces and transports many of the lipids and proteins used inside and outside the cell.
    - **Rough ER** associated with ribosomes. It is a place where transmembrane proteins are synthesized.

        - Proteins destined for secretion are synthesized by ribosomes in the rough ER.
    - **Smooth ER** lacks ribosomes but is a primary site of lipid synthesis.
- Interior of the ER is continuous and is called the lumen.
- Golgi is the next stop for vesicles that are carrying lipids and proteins and they bud off the ER.
- The flattened membrane sacs in Golgi are called **cisternae** and are surrounded by vesicles.
- Movement of vesicles from the ER to the Golgi to the rest of the cell is a biosynthetic pathway where lipids and proteins are sequentially modified and delivered to final destinations.
- **Lysosomes** are specialized vesicles that degrade damaged or unneeded macromolecules.
    - Contain a variety of enzymes that break down molecules.
        - The enzymes are synthesized in the RER, sorted in the Golgi, and packaged into lysosomes.
    - Derived from the Golgi
- Proteins in the lysosomal membrane include proton pumps that keep the internal environment pH at a level of 5.
    - The enzymes cannot function in normal cell environment with a pH of 7.
- Protein sorting is the process by which proteins end up where they need to be to do their job.
- Proteins produced in two places
    - On free ribosomes in the cytosol.
        - These are sorted after they are translated. The signal sequences they contain will indicate where they should go for sorting.
    - On membrane bound ribosomes on the rough ER.
        - End up in the lumen of the endomembrane system or go out of the cell or are transmembrane proteins.
        - If has no signal sequence, continues into the lumen.
        - If contains a second sequence (signal anchor sequence) then it continues through the lumen and ends up in membrane.
- Nuclear localization signals are for the proteins that need to move through pores in the nuclear envelope.



Chapter 6 - Capturing and Using Energy

6.1 - Metabolism

- All cells use energy in the form of the molecule ATP.
- Metabolism encompasses the set of chemical reactions that convert molecules into other molecules and transfer energy in living organisms.
- Metabolism has 2 branches.
    - **Catabolism** is the set of reactions that **break down** molecules into smaller units, which produces ATP, and they release energy
        - Catabolic reactions normally transfer energy from complex molecules to ATP
        - Exergonic (negative delta G) reaction
        - The smaller units have low PE, but high entropy
    - **Anabolism** is the set of reactions that **build molecules** from smaller units, and require inputs of energy.
        - Anabolic reactions normally transfer energy from ATP to complex molecules.
        - Endergonic (positive delta G) reaction
        - The complex molecules have high PE, but low entropy
    - These reactions inevitably lose energy in the form of heat.

6.2 - Kinetic and Potential Energy

- Energy is defined as the capacity to do work. Two types of energy.
    - **Kinetic**: Energy of motion
    - **Potential**: Stored energy
- Chemical energy is a form of PE held in the chemical bonds between atoms in a molecule.

    - Strong bonds don't have a lot of chemical energy, but weak bonds do. They require a lot of energy to stay intact and thus have a lot of chemical energy. Covalent bonds can be weak.
- Chemical energy in the bonds of ATP are used to drive many cellular processes.
- ATP is composed of adenosine
    - Made up of adenine and 5 carbon sugar ribose that is attached to a triphosphate.
    - Chemical energy of ATP is held in the bonds connecting the phosphate groups.
- Energy is released when new, more stable bond are formed (these would contain less chemical energy).

6.3 - The Laws of Thermodynamics

- First law of thermodynamics is the law of conservation of energy, stating that the universe contains a constant amount of energy.
    - Energy neither created nor destroyed.
- When going from one energy form to another, the energy available to do work decreases because energy is always lost to heat, etc.
- 2nd Law states that the transformation of energy is associated with an increase in disorder/entropy in the universe.

6.4 - Chemical Reactions

- Amount of energy available to do work is called **Gibbs free energy** (G).
- Can compare the free energy of the reactants and products to determine whether the reaction releases energy that is available to do work.
    - Difference between the 2 values is called delta G
        - If the products of a reaction have less free energy than reactants, then delta G is negative and energy is released and available to do work.
- Reactions with negative delta G release energy and proceed spontaneously are called **exergonic**, and reactions with positive delta G require input of energy and are not spontaneous and are called **endergonic**.

Total amount of energy (H) = energy available to do work (G) + energy lost to entropy (TS)

- To see if there is energy available to do work, we compute the following.

$$\Delta G = \Delta H - T\Delta S$$

- Useful for determining if a chemical reactions happens spontaneously, what direction it proceeds in, and whether net energy is released or required.
- Delta G is a function of enthalpy and the change in disorder.
- ATP reacts with water to form ADP and an inorganic phosphate. Hydrolysis of ATP is an exergonic reaction.
    - It is a hydrolysis reaction where a water molecule is split into a proton and a hydroxyl group.

    - Exergonic because there is less free energy in the products than the reactants.
    - ADP is more stable than ATP, thus a negative value of delta H. Also, ATP splits into 2 molecules so there is increase in entropy and thus delta G is negative.
- Delta G for the forward and reverse reactions have the same absolute value but opposite signs.
- **Energetic coupling** is a process in which a spontaneous reaction (exergonic) drives a non-spontaneous reaction (endergonic).
    - Requires net delta G to be negative.
    - Enzymes coordinate these coupled reactions. They interact with the substrates and orient them to allow for the creation of the product.
- In some cases, exergonic reactions can drive the synthesis of ATP by energetic coupling.

6.5 - Enzymes and the Rate of Chemical Reactions

- Rate of chemical reaction defined as the amount of product formed per unit of time.
- **Catalysts** are substances that increase the rate of chemical reactions without being consumed.
- All chemical reactions need initial input of energy.
    - True for exergonic reactions because the energy released is more than that initial required energy.
- Intermediate stage between reactants and products is called the transition state.
    - Highly unstable and has a large amount of free energy.
- Energy input needed to reach the transition state is called the **activation energy**.
    - The lower the energy barrier, the faster the reaction.
- Enzymes are able to lower the activation energy by stabilizing the transition state and decreasing the free energy.
    - They accelerate the reaction by reducing the activation energy but the delta G does not change.
- Enzymes remain unchanged from a chemical reaction.
- In a chemical reaction catalyzed by an enzyme, the reactant is referred to as the substrate.
    - When there is an enzyme present, the substrate forms of complex with the enzyme (ES), the substrate is converted to product (EP), and the complex dissociates, releasing the enzyme and product.

$$S + E \leftrightharpoons ES \leftrightharpoons EP \leftrightharpoons E + P$$

- Enzymes folded into 3-D shapes that bring amino acids into close contact to form an active site which is the location where the enzyme binds to the substrate and catalyzes its conversion to product.
- Enzymes also reduce energy through specific positioning of the substrates.
- Enzymes are specific both for the substrate and the reaction that is catalyzed.

    - Recognize either a unique substrate or a class of substrates.
    - Can be attributed to the structure of their active sites.
- Activity of enzymes can also be influenced by inhibitors and activators.
    - **Inhibitors** decrease the activity of enzymes. They can shut down pathways when they are no longer needed.
        - Irreversible inhibitors form covalent bonds with enzymes and irreversibly inactivate them.
        - Reversible inhibitors form weak bonds with enzymes and easily disassociate them.
        - Work by binding to the active site of the enzyme (or some other part) so that the substrate can't bind to it. Usually changes the shape and activity of the enzyme.
    - **Activators** increase the activity of enzymes
- Enzymes that are regulated by molecules that bind at sites other than their active sites are **allosteric enzymes**.
    - Allosteric site is just somewhere that a activator/inhibitor can bind where it's not the active site.
- Negative feedback is when the final product inhibits the first step of the reaction.

Chapter 7 - Cellular Respiration

7.1 - Overview of Cellular Respiration

- Organisms break down organic molecules through the process of cellular respiration, which releases energy.
- Cellular respiration is a series of chemical reactions that convert chemical energy in fuel molecules into chemical energy of ATP, which can be used by cells.
    - One of the major sets of catabolic reactions in a cell.
        - Fuel molecules like glucose, fatty acids, etc are catabolized into smaller units which releases the energy stored in their bonds.
    - Aka converts chemical potential energy stored in organic molecules to chemical potential energy that is useful to cells.
- Cellular respiration releases a large amount of energy because the sum of the PE in all the chemical bonds of the reactants is higher than that of the products.
- This energy is gradually released in a series of chemical reactions.
- The chemical energy stored in a molecule of glucose is used to produce ATP in 2 different ways.
    - One, a phosphorylated organic molecule directly transfers a phosphate group to ADP. There are two coupled reactions carried out by a single enzyme.
        - Hydrolysis of phosphorylated organic molecule
            - Releases enough free energy to drive the synthesis of ATP
        - Addition of a phosphate group to ADP
          This way of generating ATP is called substrate level phosphorylation.

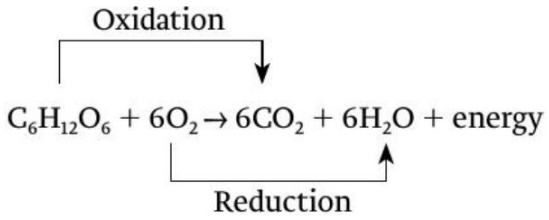
    - Two, chemical energy of organic molecules is transferred first to electron carriers which transport electrons released during the catabolism of organic molecules to the respiratory electron transport chain which transfer them along a series of proteins to a final electron acceptor which harnesses energy to produce ATP. This way of generating ATP is oxidative phosphorylation.
- Chemical reactions in which electrons are transferred from one atom/molecule to another are called oxidation reduction reactions.
    - Oxidation = loss of electrons
    - Reduction = gain of electrons
        - Normally endergonic process because you're going from molecule(s) of smaller structure (low PE) to more complex structures (high PE).
- 2 electron carriers are called FADH and NAD
    - Oxidized form: NAD+ and FAD
    - Reduced form: NADH and FADH2
- Reducing NAD+ to NADH is an endergonic process because you're going from a lower PE in NAD+ to a higher PE in NADH.
- When fuel molecules like glucose are catabolized, some of the steps are oxidation reactions, which are coupled with the reduction of electron carrier molecules.

$$NAD^+ + 2e^- + H^+ \rightarrow NADH$$
$$FAD + 2e^- + 2H^+ \rightarrow FADH_2$$

- In their reduced forms, NADH and FADH2 can donate electrons. The oxidation of those 2 allows electrons to be transferred to the electron transport chain.

$$NADH \rightarrow NAD^+ + 2e^- + H^+$$
$$FADH_2 \rightarrow FAD + 2e^- + 2H^+$$

    - The products can then accept electrons from the breakdown of fuel molecules and start the whole process over.
- Cellular respiration can be a redox reaction.



- You go from the left side reactants which are high PE to the right side where you have low PE for the molecules but also a release of energy.
- The reduced forms of molecules have more chemical energy than their corresponding oxidized forms.

- Oxidation reduction reactions always occur together since the electrons lost in one reaction are gained in the other, and as a result they are useful to transfer electrons and energy in biological systems.
- Cellular respiration happens in 4 stages.
    - Stage 1 - Glucose partially broken down to make pyruvate and energy is transferred to ATP and reduced electron carriers, a process called glycolysis.
    - Stage 2 - Pyruvate is oxidized to another molecule called acetyl-coenzyme A producing reduced electron carriers and releasing carbon dioxide.
    - Stage 3 (The Krebs cycle/Citric acid cycle) - Acetyl group is completely oxidized to carbon dioxide and energy is transferred to ATP and reduced electron carriers.
    - Stage 4- Oxidative phosphorylation is where the reduced electron carriers produced in the first 3 stages donate electrons to the electron transport chain and ATP is produced.
- A basic summary is that glucose is oxidized through a series of chemical reactions, releasing energy in the form of ATP and reduced electron carriers.
- Glycolysis (stage 1) takes place in the cytoplasm while the others take place in the mitochondria. The electron transport chain is located in the plasma membrane.

7.2 - Glycolysis

- Glucose is the starting molecule for glycolysis, which results in the partial oxidation of glucose and the synthesis of a small amount of ATP and reduced electron carriers.
    - Process is anaerobic since no oxygen is consumed.
- Glycolysis uses FAD and NAD+ as molecules that can be reduced while coupling with oxidation reactions of the fuel molecules.
- Glycolysis begins with a molecule of glucose and produces 2 3-carbon molecules of pyruvate and two molecules of ATP (through substrate level phosphorylation) and 2 molecules of NADH.
- First phase of glycolysis adds 2 phosphate groups to glucose. Requires input of energy, endergonic process. Phosphorylation of glucose traps glucose in the cell and destabilizes the molecule so that it can be broken apart in the 2nd phase.
- Second phase is the cleavage phase, where the 6-carbon molecule is split into 2 3-carbon molecules.
- Third phase is called the payoff phase because ATP and NADH are produced.
- In general, the whole process of glycolysis doesn't produce a lot of ATP, but pyruvate has a lot of PE in its bonds.

7.3 - Pyruvate Oxidation

- Pyruvate is first transported into the mitochondria from the cytoplasm.
- For the mitochondria, the space between the inner and outer membranes is called the intermembrane space and the space enclosed by the inner membrane is called the mitochondrial matrix.
- Pyruvate is transported into the mitochondrial matrix where it is converted to acetyl-CoA. Part of the molecule is oxidized and splits off to form carbon dioxide. The lost electrons

are donated to NAD+, which is reduced to NADH. The remaining part of the pyruvate is transferred to coenzyme A (CoA) which carries the acetyl group to the next reactions.

- Synthesis of one molecule of acetyl-CoA from pyruvate (but normally we have 2 molecules as a result of glycolysis) results in the formation of one molecule of CO2 and one molecule of NADH.

7.4 - The Citric Acid Cycle

- This is the stage where the fuel molecules are completely oxidized and electrons are supplied to the electron transport chain.
- Also takes place in the mitochondrial matrix.
- The 2-carbon acetyl group of acetyl-CoA forms of a 6-carbon molecule of citric acid which is oxidized in a series of reactions.
- The cycle results also in the complete oxidation of the acetyl group of acetyl-CoA.
    - These oxidation reactions release carbon dioxide.
- The oxidation reactions are paired with the reductions of NAD+ to NADH.
    - Citric acid cycle produces a large quantity of electron carriers, which donate to the electron transport chain.
- The only substrate level phosphorylation in this cycle is one reaction that generates a molecule of GTP which can transfer its terminal phosphate to a molecule of ADP to form ATP.
- At the end of this cycle, the energy in the original glucose is held in ATP, NADH, and FADH2. Mostly in the electron carriers. Most of the free energy is stored there.

7.5 - The Electron Transport Chain and Oxidative Phosphorylation

- The first 3 stages produced a lot of electron carriers, NADH and FADH2.
    - Energy stored in these carriers is released in redox reactions that occur as electrons pass through proteins in the inner mitochondrial membrane to the final electron acceptor (oxygen) which is reduced to water.
    - Passage of these electrons is coupled to a transfer of protons across inner mitochondrial membrane thus creating a concentration and charge gradient which provides the PE used to drive synthesis of ATP.
- Electrons are moved along 4 large protein complexes that form the electron transport chain. These are embedded in the inner mitochondrial membrane.
- Electrons donated by NADH enter from complex 1 and electrons donated by FADH2 enter from complex 2.
- Within each protein, electrons pass from electron donors to electron acceptors.
    - Each donor and acceptor is a redox couple, consisting of an oxidized and reduced form of a molecule.
- When oxygen accepts the electrons at the end, it is reduced to form water, and the reaction is catalyzed by complex 4.
    - This is why oxygen is needed for cellular respiration.
- Coenzyme Q accepts electrons from complexes 1 and 2, and once CoQH2 forms, it diffuses to complex 3 where the electrons are transferred to cytochrome c and the

protons are released. Then, when it accepts an electron, the cytochrome is reduced, diffuses, and passes the electron to complex 4.

- Inner mitochondrial membrane is selectively permeable.
    - Protons can't passively diffuse across.
- Because we have the movement of electrons through protein complexes is coupled with pumping of protons from matrix to intermembrane space, we create a proton gradient.
- Proton gradient (which is an electrochemical one) has 2 components.
    - Chemical gradient due to difference in concentration and electrical gradient due to difference in charge on both sides of the membrane.
- Protons will have a high concentration in the intermembrane space, and low in the mitochondrial matrix.
    - Since the membrane will block the attempted diffusion from the space to the matrix, this gradient stores potential energy.
- This gradient and the potential energy it brings is the main source of energy used to synthesize ATP.
- First, to release the PE of the gradient, we need an opening in the membrane for protons to flow through and the movement of protons through the enzyme (across the mitochondrial membrane, from the matrix to the intermembrane space) must be coupled with the synthesis of ATP.
    - Coupling made possible by the enzyme ATP synthase which has 2 units.
        - F0 forms the channel in the inner mitochondrial membrane. The protons get rotated when they pass through it.
        - F1 is the catalytic unit that synthesizes ATP. The rotational energy catalyzes the synthesis of ATP from ADP and P.
- Flow of energy in cellular respiration: Chemical PE in the bonds in glucose -> energy released through reactions which generate ATP through substrate level phosphorylation or to electron carriers which pump protons across inner membrane of mitochondria and form an electrochemical gradient -> ATP synthase converts energy of the gradient to rotational energy which drives synthesis of ATP.

7.6 - Anaerobic Metabolism and the Evolution of Cellular Respiration

- In the absence of oxygen, pyruvate can be broken down through fermentation, which does not rely on oxygen or other electron acceptors.
    - These fermentation pathways allow organisms to extract energy from fuel molecules without oxygen.
- Fermentation takes place in the cytoplasm.
- 2 main pathways are lactic acid fermentation and ethanol fermentation.
    - During lactic acid fermentation, electrons from NADH are transferred to pyruvate to produce lactic acid and NAD+.

$$\text{Glucose} + 2\ \text{ADP} + 2\ P_i \rightarrow 2\ \text{lactic acid} + 2\ \text{ATP} + 2H_2O$$

    - During ethanol fermentation, pyruvate releases CO2 to form acetaldehyde.

$$\text{Glucose} + 2\ \text{ADP} + 2\ P_i \rightarrow 2\ \text{ethanol} + 2CO_2 + 2\ \text{ATP} + 2H_2O$$

- In both of the above cases, pyruvate is reduced during fermentation.
- The most important product of the fermentation pathway is NAD+. During fermentation, NAD+ is regenerated from the reduction of pyruvate.
- Anaerobic respiration can also happen where the final electron acceptor is just something other than oxygen, like sulfate and nitrate.

7.7 - Metabolic Integration

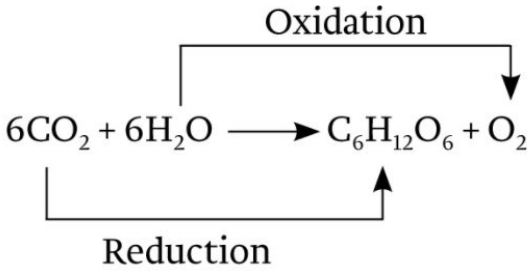
- Excess glucose is stored as glycogen in animals and starch in plants. Both are molecules that are large branched polymers of glucose.
- Basically, the glucose that isn't immediately consumed by glycolysis are linked together to form glycogen in liver and muscle.
- Glycogen provides a source of glucose to feed glycolysis when the blood glucose is low.
- Sugars other than glucose can contribute to glycolysis. Some sugar units are disaccharides (2 sugar units) and some are monosaccharides.
- The disaccharides can hydrolyzed into monosaccharides. Those then can enter glycolysis (not as glucose, but as intermediates of glycolysis).
- Lipids are also good source of energy.
- Fatty acid molecules are rich in C-C and C-H bonds, which carry chemical PE.
- Beta-oxidation is where fatty acids are oxidized and are shortened by a series of reactions that remove carbons from their ends. This doesn't make ATP, but releases a bunch of electron carriers.
- Proteins can also be broken down into amino acids which can enter glycolysis.
- Depending on the levels of ATP within a particular cell, the activity in enzymes throughout the cellular respiration process can either increase or decrease their activity.
    - ADP and AMP are allosteric activators of PFK-1 which is an allosteric enzyme. When those 2 things are abundant, one binds to the enzyme causing its shape to change.
    - PFK-1 is inhibited by ATP and activated by ADP.

Chapter 8 - Photosynthesis

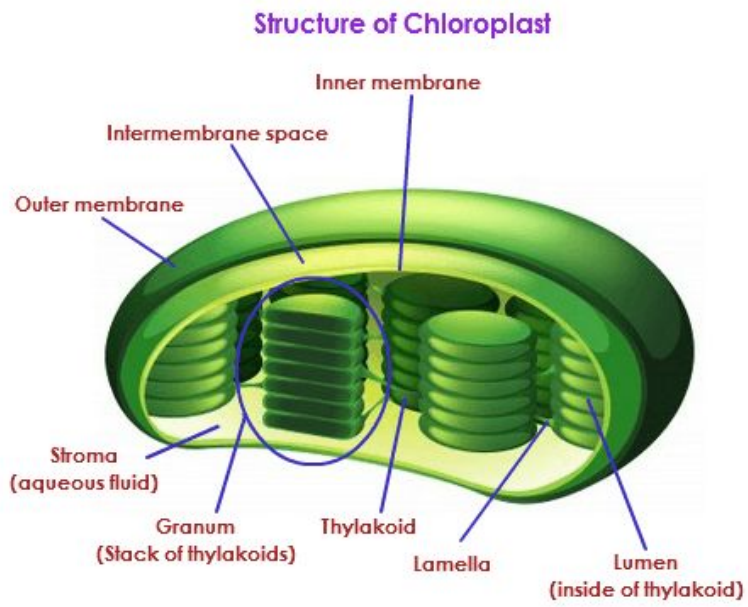
8.1 - Photosynthesis: An Overview

- Process that allows trees to increase mass using material pulled from the air is called **photosynthesis**.
    - It is a biochemical process for building carbohydrates using energy from sunlight and carbon dioxide taken from the air.
- Trees, grasses, and shrubs are examples of photosynthetic organisms.

    - Occurs among prokaryotic and eukaryotic organisms.
- Photosynthesis takes places everywhere where sunlight is available to serve as a source of energy.
    - In the ocean, it occurs in the **photic zone**, where enough sunlight penetrates
    - On land, occurs where the environment is moist and warm.
- Carbohydrates are built from CO2 molecules as well as an input of energy.
- Synthesis of carbohydrates from CO2 and water is a redox reaction.
    - During photosynthesis, CO2 molecules are reduced to form higher energy carbohydrate molecules. This reduction requires
        - input of energy from ATP
        - Transfer of electrons from an electron donor. These electrons will reduce the CO2.
    - Reduction of CO2 is also coupled with the oxidation of water to form oxygen. Water is the electron donor that really gives the CO2 the electrons necessary for that reduction.
- The electrons needed for the above process come from the oxidation of water. Oxygen is formed as a byproduct.
- Equation for photosynthesis



- Oxidation of water linked with the reduction of CO2.
    - Those reactions constitute the photosynthetic electron transport chain.
    - Movement of electrons through this chain is used to produce ATP and NADPH which are the energy sources needed to synthesize carbohydrates through the Calvin cycle.



- The **photosynthetic electron transport chain** is located within the cytoplasm (prokaryotes) or in the chloroplast in the thylakoid membrane (the membrane that surrounds each thylakoid). Region around this membrane is called the **stroma**.
- The sacs inside the membrane are grouped into structures called **grana**. They are stacks of thylakoids.
    - They are connected to each other and enclose a single interconnected compartment (fluid filled) called the **lumen**.
- Carbohydrate synthesis (aka Calvin Cycle) takes place in the stroma and the light reactions which create the energy sources take place in the thylakoid membrane.
- Photosynthetic electron transport chain creates chemical energy from the sunlight
- Photosynthetic organisms are called **autotrophs** because they can form carbohydrates from CO2 (with ATP ofc which is produced in the chloroplast).
- Only carbohydrates, not ATP, are exported from chloroplast to cytosol.

8.2 - The Calvin Cycle

- 15 Chemical reactions that synthesize carbohydrates from CO2. 3 main steps.
    - Carboxylation: CO2 is added to a 5 carbon molecule
    - Reduction: Energy and electrons are transferred to compounds from step 1. Triose phosphate molecules are created from the 3-PGA molecules.
    - Regeneration of the 5 carbon molecule needed for step 1.
- In step 1, CO2 is added to a 5 carbon sugar called ribulose 1,5-bisphosphate (**RuBP**) and is catalyzed by the enzyme **rubisco**.
    - Enzymes that add CO2 to other molecules are called a carboxylase.
- CO2 and rubisco both need to be in the active site. Once there, the reaction proceeds spontaneously.
- Product is a 6 carbon compound that breaks into 2 molecules of **3-phosphoglycerate (3-PGA)**.

- Those carbon products need to be reduced for a reducing agent like NADPH which transfers electrons that allows carbohydrates to be synthesized.
- Reduction of 3-PGA
    - ATP donates a phosphate group to 3-PGA
    - NADPH transfers 2 electrons and 1 proton to the phosphorylated compound, which releases one phosphate group.
    - One molecule of CO2 -> 2 3-PGA and thus 2 ATP and 2 NADPH required.
    - NADPH provides most of the energy incorporated in the bonds of the carbohydrate molecules produced.
    - Product is the formation of 3-carbon carbohydrate molecules called triose phosphates (G3P).
- The triose phosphates that are created are the principal form in which carbohydrates are exported. However, we can't just export them rn, we need to use it to regenerate RuBP.
    - For every 6 produced, only one can be withdrawn from the Calvin cycle.
- 5 3-carbon triose phosphates need to be turned into 3 5-carbon RuBP molecules.
    - ATP is required for this regeneration. (+1 to the ATP needed count)
- Calvin cycle can't operate without energy input from NADPH (this is the major input of energy) and ATP, which are supplied by the photosynthetic electron transport chain (which gets its chemical energy from the sunlight).
- Excess carbohydrates are converted to starch which is a storage form. They aren't soluble and thus osmosis does not occur.
- This Calvin Cycle process can shut down if we don't have ATP or NADPH or CO2.

8.3 - Capturing Sunlight into Chemical Forms

- To power the Calvin cycle (aka to create ATP and NADPH), cell has to use light energy.
    - The light energy drives the flow of electrons through the chain, which leads to formation of NADPH and ATP.
- Each point on the EM spectrum has a different energy level and a corresponding wavelength. Visible light is the portion that includes the range of wavelengths used in photosynthesis.
- Pigments are molecules that absorb some wavelengths of visible light.
- **Chlorophyll** is the major photosynthetic pigment.
    - Has a large, light absorbing head that contains a magnesium atom at its center, and a hydrocarbon tail.
    - Has a large number of alternating single and double bonds in the head region.
- Those molecules are bound by their tail region to the integral membrane proteins in the thylakoid membrane.
    - The protein pigment complexes are referred to as photosystems and are the units that absorb light energy and use it to drive electron transport.
- The photosystems also contain other pigments (such as carotenoids) which can absorb other types of light.
- When chlorophyll molecules take in light energy, one of its electrons is elevated to a higher energy state, and then the energy is rapidly released in the form of heat (most of

it is heat) or re-emitted as light through fluorescence. This allows the electron to go back to its ground energy state.
    - For molecules within an intact chloroplast, energy can be transferred to other chlorophyll molecules. Most of the molecules in the thylakoid membrane function as antennas where the absorbed energy is transferred until it reaches the reaction center.
    - The actual electrons aren't getting transferred, but the energy released from the high energy state of the electron.
- The **photosynthetic reaction center** is different from the antenna chlorophylls and it becomes oxidized when it passes its excited electron (from the light energy) to the electron acceptor which is thus reduced. This reaction center is where light energy is converted to chemical energy
    - Then, the reaction must obtain an electron from an electron donor so that it can absorb additional energy.
- While there is a lot of water in cells, it takes a great deal of energy to pull electrons from water. So, the absorption of light energy by the photosystem 2 allows electrons to pulled from water (water is split at this point), and those electrons go to the electron transport chain. The movement of those electrons is being powered by light. A second input of light energy by the 2nd photosystem produces the electron donor molecules.
- In summary, to get water -> NADPH
    - Photosystem 2 supplies electrons to the beginning of the transport chain. When it loses its electron (gets oxidized), it pulls electrons from water. Photosystem 1 gains that electron which gets energized with the light energy which you can do reduce NADP+.
    - Water basically donates electrons to one end of the chain, and NADP+ accepts electrons at the other end.
- The two photosystems need each other because photosystem 1 when oxidized is not strong enough to split water, and photosystem 2 is not strong enough of a reducer to form NADPH.
    - In other words, "one photosystem is needed to pull electrons from water, and a second one is needed to raise the energy of these electrons enough that they can reduce NADP+"
- Photosystem 2 basically has chlorophylls that take in light energy and pass that energy to the reaction center where the center is oxidized and gives away its electron. Because it now needs an electron, it uses its energy to pull electrons from water. Now, photosystem 1 takes the electron that was given away from photosystem 2 and then hits it with more light energy so that the electron can be used to reduce NADP+ to NADPH.
- The major protein complexes in the chain include two photosystems and a cyt complex between them (this is where electrons pass between the two photosystems).
    - Pp compounds convey electrons between 2 and the cyt complex and Pc compounds convey them from cyt to 1.
- NADPH is formed when electrons are passed from photosystem 1 to a membrane associated protein called ferredoxin.

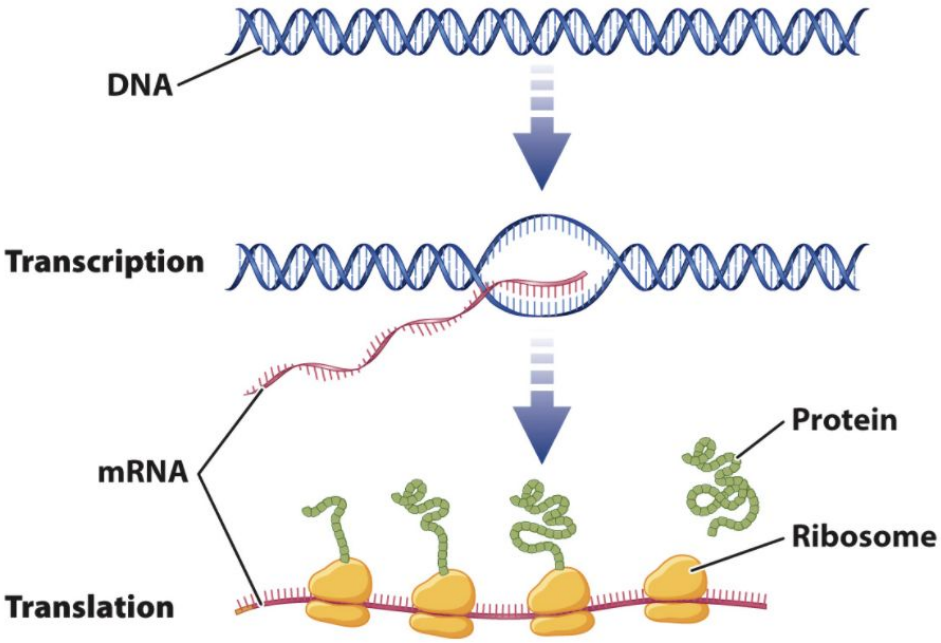
$$NADP^+ + 2e^- + H^+ \rightarrow NADPH$$

- One other effect of the electron transport chain (besides creating NADPH) is creating the buildup of protons in the thylakoid lumen. As the electrons are moving through the chain, protons are being pumped from the outside (stroma) into the inside of the thylakoid membrane. Remember that the membrane separates the stroma (outside the thylakoid) and the lumen (inside the thylakoid).
    - The oxidation of water releases protons and O2 into the lumen.
    - The cyt complex and plastoquinone function as a proton pump.
        - Transport of 2 electrons and 2 protons from the stroma side of photosystem 2 to the lumen side of cyt complex.
        - Transfer of electrons within the cyt complex to a different molecule of plastoquinone which results in protons being picked up from stroma and then released into the lumen.
- This accumulation of protons on one side of the thylakoid membrane can be used to power the synthesis of ATP by oxidative phosphorylation. This is like the opposite of cellular respiration, because now we have a higher concentration of H+ on the inside, and thus the ATP synthase will take in those protons and release ATP out the other side.

Chapter 3 - Nucleic Acids and Transcription

3.1 - Major Biological Functions of DNA

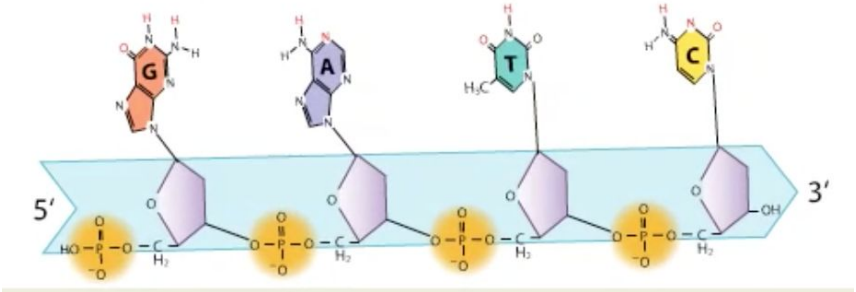
- DNA stores genetic information in the form of genes. These genes are turned on/off in process called **gene expression**.
- Molecules transfer genetic information from one organism to another.
- DNA uses replication to get info from one DNA molecule to get transferred to another.
    - An unrepaired error in DNA replication results in a mutation.
- In a cell, DNA acts indirectly by specifying the sequence of amino acid subunits of which each protein is composed and determines the 3-D shape of the protein.
    - In specifying this sequence, DNA acts through RNA.
- The flow of info from DNA to RNA to protein is the central dogma of molecular biology.



- First process is transcription, where the DNA is used as template to create RNA (you can think of it as a complementary strand). Second step is translation where RNA is used as a code for the sequence of amino acids. Next is protein synthesis which takes place in the cytoplasm. It takes place on ribosomes. When the ribosome is finished it releases the protein into the cytoplasm.

3.2 - Chemical Composition and Structure of DNA

- DNA is made of subunits called **nucleotides**, which are composed of a 5 carbon sugar, a base, and one or more phosphate groups.
    - Sugars and phosphate group from backbone, each sugar linked to the phosphate group of the next nucleotide. The sugar is also linked to a base which juts out from the sugar.



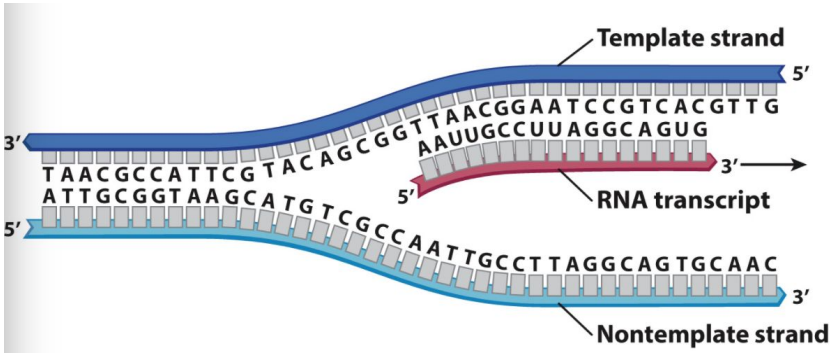
- Nucleotides consists of one of 4 bases
    - Adenine (Purine - double ring)
    - Guanine (Purine - double ring)
    - Thymine (Pyrimidines - single ring)
    - Cytosine (Pyrimidines - single ring)

- A and T are complementary, and G and C are complementary.
    - The reason is because 2 hydrogen bonds form between A and T, while 3 are needed for C and G.
- Combination of a sugar and a base is known as a nucleoside.
    - Nucleoside with one or more phosphate groups is a nucleotide.
- Nucleotides are linked together by covalent bonds, specifically by the 3' - 5' **phosphodiester bond**, which gives polarity.
- The end with a free phosphate attached to the 5' carbon atom is known as the 5' end of the molecule.
- The nucleotide with a free hydroxyl group attached to the 3' carbon atom is known as the 3' end.
- The Watson-Crick structure is known as the double helix and it describes the structure of DNA.
    - There are two DNA strands with the sugar phosphate backbones winding around the outside of the molecule and the bases pointing inward.
    - The outside contours of the twisted strands form an uneven pair of grooves, called the **major** and **minor groove**.
    - Individual DNA strands in the helix are antiparallel. The 3' end of one strand is opposite the 5' end of the other.
- Each base pair consists of a purine and a pyrimidine.
- Base stacking is a stabilizing force in the double helix and it refers to the interactions between bases in the same strand.
    - Van der Waals forces are also in play.
    - The bases are hydrophobic so that like to stick together in the surrounding
- Hydrogen bonds like the pairs of bases in the 2 strands.
- Many DNA molecules in prokaryotic cells are circular and form supercoils where the molecule coils upon itself. This is caused by enzymes called topoisomerases.
- In eukaryotic cells, most DNA molecules in nucleus are linear, and each molecule forms one chromosome.
- DNA molecules are packaged with proteins called **histones**, and they form a complex referred to as **chromatin**.
    - Histone proteins interact with all DNA because they are evolutionarily conserved and thus have very similar sequence from one organism to the next.

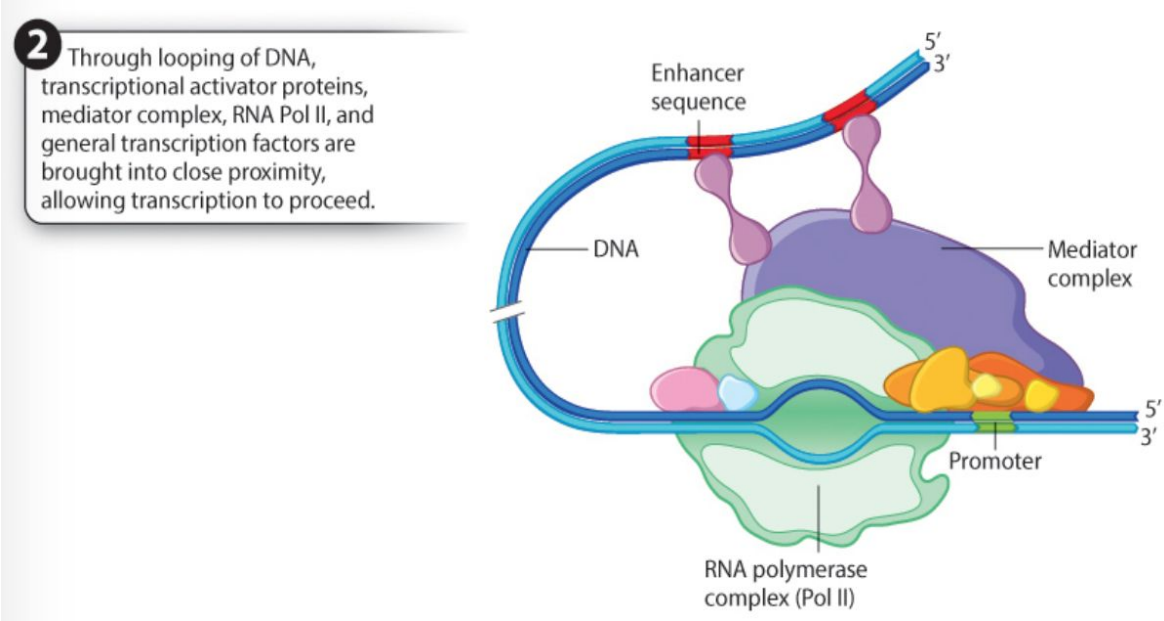
3.3 Retrieval of Genetic Information Stored in DNA: Transcription

- Proteins are synthesized on particles called ribosomes.
- RNA is polymer of nucleotides linked by phosphodiester bonds.
- Each strand has a polarity depending on which end of the chain carries the 3' hydroxyl and which carries the 5' phosphate.
- Differences between RNA and DNA
    - Sugar in RNA is ribose which carries a hydroxyl group on the 2' carbon.
    - The base uracil replaces thymine.

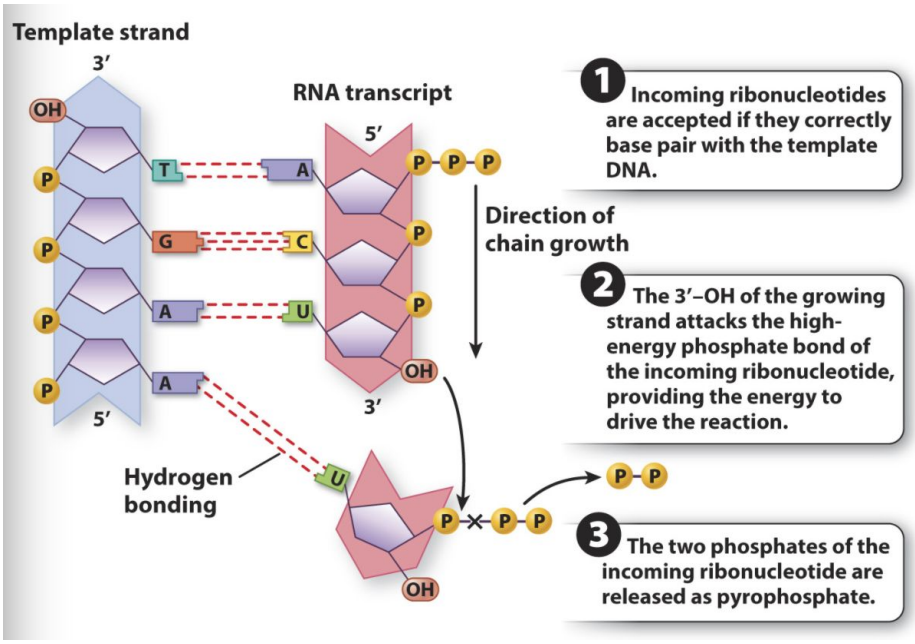
    - While the 5’ end of a DNA strand is a monophosphate, the 5’ end of an RNA molecule is typically a triphosphate.
    - RNA molecules are shorter and are single stranded.
- For the process of transcription, the DNA will unwind, and one strand is used as the template (the other is unused) for the synthesis of an RNA transcript which is complementary to the strand.
    - Transcript is produced by the polymerization of ribonucleoside triphosphates and is carried out by the RNA polymerase enzyme.



- RNA grows in the 5’ to 3’ direction and the DNA template runs in the opposite direction from the RNA.
- Transcription is initiated at a promoter sequence and ends at a terminator sequence.
- In bacteria, promoter recognition is mediated by a protein called a sigma factor which associates with RNA polymerase and facilitates its binding to specific promoters.
- Transcription in eukaryotes requires a lot and is called transcriptional initiation.
    - 6 proteins known as general transcription factors that assemble at the promoter of the gene
        - These will bind to the promoters
    - Need a transcriptional activator protein, each of which binds to a DNA sequence called an enhancer.
        - These will bind to the enhancers.
    - Once that is done, they attract a mediator complex of proteins which attracts a RNA polymerase complex to the promoter.
    - Once the mediator complex and Pol II complex are there, the transcription begins.



- A transcription bubble forms and transcription takes place inside the polymerase.
- After transcriptional initiation, ribonucleotides are added to the transcript. This is known as elongation.
    - They are accepted if they correctly pair with the template DNA.



- The RNA polymerase has structure that allows an RNA-DNA duplex to form, elongate the transcript, release the finished transcript, and restore the original DNA helix.
    - It also always reads the DNA strand from 3’ to 5’.
- The Southern Blot is used to verify the presence or absence of a specific nucleotide sequence in the DNA.
    - DNA is isolated from each source and digested with a specific enzyme.

    - Fragments are placed in a gel and are separated.
- To summarize:
    - RNA grows in its 5' to 3' direction and thus the DNA template's 3' to 5' direction.
    - RNA polymerase reads the DNA template strand from the DNA's 3' to 5'.
    - Direction of the RNA polymerase tells you what the template strand is.
- When you have an RNA poly problem
    - 1) Determine direction of the RNA polymerase. The direction of the poly will go with the direction of the DNA template's 3' to 5'
    - 3) Transcribe what the RNA is seeing (from the DNA's 3' to 5').
    - 4) Think about the complementary pairs (Instead of T's, replace with U's)
    - 5) Label each side with 3' as the growing end and 5' as the start end (And so RNA grows from 5' to 3')

3.4 Fate of the RNA Primary Transport

- RNA transcript that comes off the template DNA strand is the **primary transcript**, which contains genetic info of the gene that was transcribed.
- An RNA molecule called mRNA will combine with the ribosome to direct protein synthesis.
- In prokaryotes, the primary transcript is the mRNA.
    - Ribosomes will bind starting with the 5' end, and will start protein synthesis.
    - Transcription and translation are thus coupled.
- In prokaryotes, primary transcripts will often contain genetic info for the synthesis of two or more proteins. Molecules of mRNA that code for multiple proteins are **polycistronic mRNA**. Eukaryotes don't have these type of mRNA molecules.
- In eukaryotes, the transcription takes place in the nucleus and translation in the cytoplasm. Separation allows for a chemical modification of the primary transcript, which is known as **RNA processing** which converts the primary transcript into finished mRNA which can then be translated by ribosomes.
    - RNA processing happens in the nucleus.
- RNA processing has 3 types of chemical modification (which happen in this order)
    - 5' end is modified by the addition of a special nucleotide (5' cap) which consists of 7-methylguanosine and is attached backwards. The cap is linked to the RNA transcript by a triphosphate bridge between the 5' carbons of both ribose sugars. The 5' cap is important because the that is what the ribosome uses to recognize mRNA. The cap is added after the RNA polymerase begins transcription.
    - Polyadenylation is the addition of a string of 250 consecutive A-bearing ribonucleotides to the 3' end, forming a poly(A) tail. Plays a role in the export of the mRNA into the cytoplasm. This probably takes place right after the RNA polymerase has finished its processing.
        - Specific parts of that 3' end are bound by cleavage factors. The poly A polymerase will then bind to the end at will cleave it. The polymerase will then synthesize the tail until it is told to stop adding adenines.

    - Removal of noncoding introns and this is called RNA splicing, and catalyzed by a protein called spliceosome.
        - The spliceosome will bring the sequence within the intron close to the 5' end of the intron (place known as 5' splice site) and this causes a reaction that cuts the RNA at the side, causing it to connect back on itself and form a loop called a lariat. Then the cleaved 5' splice site attacked the 3' splice site and the lariat breaks down into nucleotides, while the spliced exons are now next to each other.
- The first 2 modifications protect the ends of the transcript and increase the stability and are used during translation to make sure that the mRNA is fully intact.
- mRNA processing events (5' cap addition, excision of introns, poly-A tail addition, and splicing of exons) all occur in the nucleus of human cells.
- Transcripts often contain protein coding regions called exons and noncoding regions called introns. Only eukaryotes have those introns.
- Presence of multiple introns in most genes allows for **alternative splicing**, where primary transcripts can be spliced in different ways to yield different mRNAs and different protein products.
    - While all introns are spliced, some exons can also be spliced. Sometimes they will be and sometimes they won't be. That's what causing the alternative splicing.
    - The different forms of the protein encodings are called isoforms.
- Structure of a mature and processed mRNA is 5' cap, 5' UTR, coding region (only exons), 3' UTR, and 3' polyA tail.
- If the primary transcripts aren't processed into mRNA, some RNA transcripts will undergo other preprocessing and become
    - Ribosomal RNA - Found in ribosomes that aid in translation. Found in the nucleolus.
    - Transfer RNA - Carries individual amino acids for use in translation
    - Small nuclear RNA - Involved in splicing, polyadenylation, etc
    - Regulatory RNA like microRNA and small interfering RNA.
    - Messenger RNA - Carries information for making a single type of protein
- The most abundant transcripts are those for ribosomal RNA and some for transfer RNA.

Chapter 4 - Translation and Protein Structure

4.1 - Molecular Structure of Proteins

- Amino acids are like letters, and proteins are like words.
    - Order of the amino acids in a protein determines the protein's shape and function.
- Amino acids always have a central carbon atom called alpha carbon, connected by covalent bonds with 4 groups, an amino group, a carboxyl group, a hydrogen atom, and a R group.

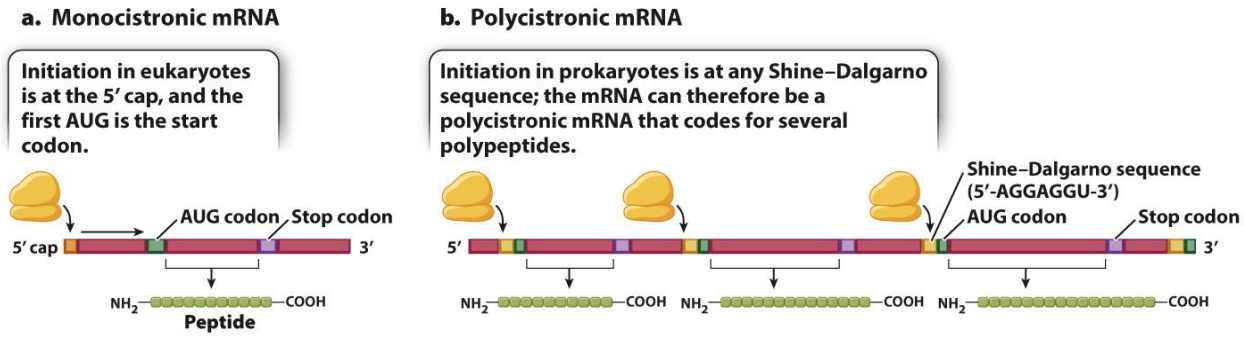
- Amino acids are linked together to form proteins. Bond between two amino acids is a peptide bond.
    - The carboxyl group of one amino acid reacts with the amino group of the next amino acid and a molecule of water is released.
    - The C-O group in the peptide bond is the carbonyl group and the N-H group is the amide group. That C-O acts like a double bond because of its greater electronegativity.
- Polymers of amino acids have ends that are chemically distinct. One end has a free carboxyl group and the other has a free amino group.
- A polymer connected by peptide bonds is known as a polypeptide. Protein is called used as a synonym for polypeptide.
- Amino acids incorporated into a protein are often referred to as amino acid residues.
- Sequence of amino acids in a protein is its primary structure. This sequence determines how the protein will fold. You can't really see the sequence when looking at the protein as a 3-D shape.
- Interactions between stretches of acids within the protein form local secondary structures.
- Longer range interactions between secondary structures form tertiary structures is the entire 3-D structure, and proteins with several individual polypeptides make up the quaternary structure.
    - Alpha helix and beta sheet are two types of secondary structure and both are stabilized by hydrogen bonding along the polypeptide backbone. Primary force holding together secondary structures is hydrogen bonding.
    - Altering the secondary structure of a protein like GFP will likely change the tertiary structure and will thus affect the overall function.
        - You can do this by changing the primary sequence or breaking the hydrogen bonds.
            - Breaking the hydrogen bonds will affect the secondary structure, but not the primary structure.
- When folded, the proteins can have specific structures to allow it to do its specialized job such as some proteins have pockets with charged chains at the right positions to trap small molecules.
- Amino acids in a protein are listed in order from left to right, starting at the amino end and proceeding to the carboxyl end.
- Tertiary structure of a protein is the 3-D conformation of a single polypeptide chain, made up of several secondary structure elements.
    - Shape is largely defined by interactions between amino acid R groups.
    - Formation of secondary structures, though, is determined by interactions in the polypeptide backbone and is independent of the R groups.
- The tertiary structure determine the function of the protein since it is the 3-D shape of the molecule, which determines the function.
- Proteins can be unfolded or denatured by chemical treatment or high temperature.

- Other larger proteins are composed of two or more polypeptide chains with a tertiary structure that come together to form a higher order quaternary structure.
- The primary structure of a protein determines how it forms secondary, tertiary, and quaternary structures.
- Slow folding of proteins can be dangerous since there are hydrophobic effects and van der Waals forces that could mess up the folding. Therefore, cells have proteins called chaperones that help protect the slow folding or denatured proteins until they get the proper 3-D structure.
    - They bind with hydrophobic groups and nonpolar R groups.

4.2- Translation: How Proteins are Synthesized

- In translation, the sequence of bases in an mRNA molecule is used to specify the order in which successive amino acids are added to a new polypeptide chain.
- To do translation the cell needs ribosomes which bind with mRNA and are the site of translation. Ribosome has a small subunit and a large subunit which have proteins.
- The large subunit of the ribosome has 3 binding sites for molecules of tRNA.
    - A site, P site, and E (exit) site.
- Goal of the ribosome is to make sure that the mRNA is getting read in successive, non-overlapping groups of 3.
- Each group of 3 nucleotides is called a codon.
- Different ways of parsing the string into 3 letter words are called reading frames.
- Once the reading frame is established, the translation of each codon to amino acid is carried out by tRNA. The tRNA have anticodons which are 3 nucleotides that undergo base pairing with the corresponding codon.
- tRNA always has sequence CCA at its 3' end and the 3' hydroxyl of A is the attachment site for the amino acid.
- Enzymes called aminoacyl tRNA synthetases connect amino acids to specific tRNA molecules.
    - Normally, we have one synthetase for each amino acid.
    - Each tRNA synthetase binds to one uncharged tRNA and its corresponding amino acid.
        - The tRNA has to have the anticodon corresponding to that amino acid.
- The binding pockets in the aminoacyl tRNA synthetase waits to become occupied with the correct amino acid and the corresponding tRNA. A covalent bond is made between the amino acid and one end of the tRNA. This is a process creates charged tRNA, which is set free afterwards.
    - Correct translation of mRNA into protein needs a full set of charged tRNA molecules.
    - 20 of these synthetase enzymes must be present in cells to properly synthesize proteins.
- We need a full set of these charged tRNA molecules so that they can attach to the codons that we'll see in the mRNA.
- Codons specify an amino acid according to a genetic code.

- Codon at which translation begins is called the initiation codon. The small subunit of the ribosome looks for that codon. When it finds it, a tRNA with the right anticodon will attach and then the large subunit will come by. The polypeptide is synthesized from the amino end to the carboxyl end. Once the amino end is created, the downstream codons are read one by one in groups of 3 bases. A ribosome will bind to a tRNA with an anticodon that can base pair with the codon. When a stop codon is read, the polypeptide is finished and is released into the cytosol.
- The first tRNA that comes by will occupy the P site, and the second will occupy the A site.
- The large subunit of the ribosome contains 3 binding sites for tRNA molecules. When the first(rightmost) codon has attracted the corresponding charged tRNA, the growing polypeptide chain is attached to the incoming amino acid.
- During translation, the amino end of the new polypeptide is the end that protrudes from the large subunit of the ribosome.
- The initiation codon is always coded by AUG. This specifies the Met amino acid.
    - Polypeptide is synthesized from amino end to the carboxyl end.
- The stop codon is always coded by UAA, UAG, or UGA.
- Translation is divided into 3 separate processes.
    - Initiation - The initiator codon is recognized and the amino acid is established as the first in the polypeptide chain. Requires a number of protein initiation factors that bind to the mRNA. They initially bind near the 5' cap and they recruit a small unit of the ribosome and bring up a tRNA to move along the mRNA. The complex will move along the mRNA until it finds an AUG, which at that point, the large ribosomal subunit joins, the initiation factors are released, and a tRNA binds with the A site (there's already one for the AUG codon at the P site).
    - Elongation - Successive amino acids are added one by one to the growing chain. Ribosome movement along the mRNA and formation of the peptide bonds require energy, which is obtained through the help of proteins called elongation factors. As elongation continues, the growing peptide is continually transferred to the A site tRNA, the ribosome moves along the mRNA, and new tRNAs enter.
    - Termination - Addition of amino acids stop and the completed chain is released from the ribosome. Stop codons cause termination because they do not have corresponding tRNA molecules. A release factor will enter the A site and cause the bond connecting the polypeptide to the tRNA to break.
- Synthesis of transcription factors occur in the cytoplasm and they bind in the promoter region of the DNA. Transcription starts at the 3' end of the template strand.
- When AUG codon is encountered, a large ribosomal subunit joins the complex, the initiation factors are released, and the next tRNA is ready to join the ribosome.
- When you're trying to figure out the anticodon for a specific codon,
    - 1) Find the complementary base pairs for that codon. Just go from left and right and translate across
    - 2) Reverse the 5' and 3' ends.
- Translation is different in prokaryotes and eukaryotes



4.3 - Protein Evolution and the Origin of New Proteins

- Proteins can be grouped into 25,000 protein families.
    - It's a group of structurally and functionally related proteins as a result of shared evolutionary history.
- Region of a protein that folds in a similar way relatively independently of the rest of the protein is known as a folding domain.
    - TIM barrel consists of alternating alpha helices and parallel beta sheets connected by loops.
- Number of known folding domains is about 2500.
- A mutation is a change in the sequence of a gene.
    - Some affect the amino acid sequence, some affect the level of protein expression, some change the time in development of the cell.
- Random mutations are retained or eliminated through the process of selection among individuals based on who survives.
    - Mutations may actually persist in a population if the organism is better off with it.

Chapter 19 -

19.3 - Transcriptional Regulation in Prokaryotes

- Regulation of gene expression occurs through a hierarchy of regulatory mechanisms acting at different levels.
- Transcription can be positively or negatively regulated. In positive regulation, a protein must bind to DNA at a site near the gene in order for transcription to take place. Same with negative regulation and preventing the transcription.
- Promoters contain sequences that help recruit proteins needed for the transcription.
    - Called -10 and -35 sequence motifs
- In positive regulation, the activator protein has to interact with its binding site in the DNA and then the RNA polymerase comes and the transcription starts.
    - Basically the RNA polymerase can bind to the promoter only if an activator protein binds to a site near the promoter.
    - Without the activator protein, the transcription can’t happen.

- The activator protein can have allosteric effects where it changes shape after interaction with a small molecule and it affects its binding affinity for DNA.
- Negative regulation has similar processes except it has a protein called a repressor. If the repressor binds to the DNA, RNA polymerase won't do the transcription.
- A molecule that interacts with the repressor and prevents it from binding DNA is called an inducer.
- Product of one gene can regulate the transcription of other genes.
- lacZ is the coding sequence or gene for the enzyme beta galactosidase. Its job is to cleave the lactose molecule into glucose and galactose constituents.
- lacY is the coding sequence or gene for the protein lactose permease, which transports lactose from the external medium into the cell.
    - They are structural genes because they code for the primary structure of proteins.
    - They are transcribed when lactose is present and the glucose levels are low.
- A functional form of both of those genes is necessary for the utilization of lactose and for cell growth.
- The structural genes are controlled by the product of another gene, called lacI.
    - LacI encodes a repressor protein.
    - The lactose operon is negatively regulated by this protein.
        - The repressor protein will bind with the operator, lacO, and the then the RNA polymerase is not recruited and thus no transcription.
        - The repressor protein can only bind when there is no lactose present in the cell.
        - If the protein doesn't bind, then the polymerase is recruited and the mRNA is produced.
- lacP is a promoter that recruits the RNA polymerase and initiates transcription.
- lacO is the binding site for the repressor. This is called an operator.
- The two structural genes share a promoter and are transcribed together into a single molecule of mRNA. These molecules of mRNA are polycistronic.
- Region of DNA consisting of the promoter, the operator, and the coding sequence for the structural genes is called an operon.
- Phenotype of a cell carrying a mutation that represses a protein is constitutive for the production of proteins.
    - Most common constitutive phenotype resulted from the mutation in the lacI gene that produces a defective repressor protein.
- Even when one out of two copies has a working repressor, it can repress transcription for both proteins.
- Repressor can bind with the operator in the absence of lactose or with the inducer in the presence of lactose.
- The CRP-cAMP complex is a positive regulator of the lactose operon. Function is to provide another level of control of transcription that is more sensitive to the nutritional needs of the cell.

- In the absence of large amounts of glucose, cAMP levels are high and the CRP–cAMP complex binds to a site near the promoter, where it activates transcription. (b) In the presence of large amounts of glucose, cAMP levels are low and the CRP–cAMP complex does not bind, so transcription is not induced to high levels, even in the presence of lactose.
    - For the lactose operon, the CRP-cAMP is an activator
- Concentration of cAMP is a signal about the nutritional state of the cell.
- For the lactose operon, lactose is an inducer.

19.1 - Chromatin to Messenger RNA in Eukaryotes

- Different cells express different genes. Cells basically share the same genome but express different genes.
- Gene regulation can take place in the chromosome before the transcription takes place.
- DNA in eukaryotes is packaged as chromatin.
    - When in its coiled state, DNA is not accessible to the proteins that carry out the transcription.
    - Thus chromatin remodeling has to be done to expose the stretches of DNA to the nuclear environment.
        - Modifications can be to the histone tails, which protrude from the histone proteins.
        - The pattern of modifications constitutes a histone code that affects chromatin structure and gene transcription.
- Gene expression also affected by chemical modification of certain bases in the DNA
    - Common modification is adding a methyl group to the base cytosine.
    - Occurs in cytosine bases that are adjacent to guanosine bases on a DNA strand.
    - Occurs in CpG islands which are clusters of adjacent CG nucleotides located near the promoter of the gene.
        - Heavy methylation is associated with the repression of the gene that is near the island and thus proper transcription may not occur.
- Epigenetic mechanisms of gene regulation involve changes to the way that DNA is packaged, not changes to the actual DNA itself.
- Sex specific silencing of gene expression is known as imprinting.
- Regulation of genes in X chromosome is different in females and in males. The differential regulation is called dosage compensation.
    - For flies, males would need to double the transcription of the single X chromosome to achieve equal expression compared to females.
    - You can also have inactivation of one X chromosome in each cell in females. The process is known as X-inactivation.
- Process of X-inactivation
    - There is small region in the X chromosome called the X-chromosome inactivation center (XIC) which has a gene called Xist. When a chromosome becomes inactive, the gene's transcription increases. The transcript undergoes splicing but doesn't produce a protein. The Xist RNA coats the XIC region, and eventually the

whole chromosome, which recruits the factors that promote DNA methylation, histone modification, and other changes with transcriptional repression.
    - Xist gene is necessary and sufficient for X-inactivation.
- Even after the template DNA goes through the chromatin remodeling and the histone modification, the mechanisms that regulate whether or not transcription occurs are known as transcriptional regulation.
- Transcription can be regulated in the recruitment of the general transcription factors and components of the RNA polymerase.
    - Recruitment is controlled by regulatory transcription factors.
        - Main job is to recruit the general factors.
        - Some will recruit chromatin remodeling proteins.
        - Others have two binding sites, one of which binds with a particular DNA sequence near a gene called an enhancer and the other recruits factors to the promoter region.
        - Can also bind with DNA sequences known as silencers which repress transcription.
    - Transcription doesn't occur without that recruitment.
- Transcription only takes place when you have the proper combination of regulatory transcription factors.
    - Type of regulation is called combinatorial control.
- Some RNA molecules can be a substrate for enzymes that modify particular bases in the RNA. This is called RNA editing.

19.2 - Messenger RNA to Phenotype in Eukaryotes

- In eukaryotes, the processed mRNA exits the nucleus and will then proceed with the translation in the cytoplasm. Once it is in the cytoplasm, there are opportunities for gene regulation at the levels of mRNA stability, translation, and protein activity.
- Translation of mRNA into protein is another place where gene expression can be controlled.
- All mRNA molecules will have a 5' cap, a 5' UTR, an open reading frame, a 3' UTR, and a poly A tail.
    - Proteins can bind to the 5' UT to transport it to different locations in the cell.
- Translation initiation is when an initiation complex moves along the 5' UTR and looks for an AUG codon.
    - Efficiency of initiation is increased when there is a protein that binds the poly A tail and also binds the 5' cap.
    - This translation initiation is the principal mode of translational regulation.
- After translation is complete, the resulting protein can alter the phenotype of the cell by affecting metabolism, signaling, gene expression, or cell structure.
    - Processes are called posttranslational modification.
        - Can help regulate protein activity.
    - Some proteins need help folding after they come off the ribosomes from chaperones.

Chapter 13 -

13.3 - Gene Number, Genome Size, and Organismal Complexity

- Humans have same number of protein coding genes as other organisms with smaller genomes.
    - We have 100 million times the number of cells that worms do but we have the same number of protein coding genes.
- Differential gene expression allows the same protein coding genes to be deployed in different combinations to get different cell types.
    - A single gene can also yield multiple proteins because of alternative splicing or posttranslational modification.
- This shows that evolutionary changes can happen not only with the acquisition of new genes, but by modifying existing genes.
- No relationship between the size of the genome and the complexity of the organism.
    - This disconnect is called the C value paradox.
        - Only applies to eukaryotes.
- Genomes are measured in the number of base pairs.
- Eukaryotic genomes can be large because of polyploidy which is the characteristic of having more than two sets of chromosomes in the genome.
    - But the main reason for the large size is that genomes contain a large amount of DNA that does not code for proteins.
- Human genome has about 25,000 genes and 2.5% of the human genome codes for proteins.
- Transposons are non coding DNA sequences that can replicate and insert themselves into new positions in the genome.
    - DNA transposons replicate and transpose by DNA replication and repair.
    - Retrotransposons transpose by means of an RNA intermediate.
        - RNA is used as a template to synthesize complementary strands of DNA. It reverses that usual flow of genetic information.
- Method for making copies of a piece of DNA is the polymerase chain reaction (PCR) which allows a targeted region of a DNA molecule to be replicated into as many copies as possible.
- Amplification entails denaturation where the DNA is heated to separate the strands, then annealing where the temperature gets lowered and two primers anneal to their complementary sequences, then extension where DNA polymerase synthesizes new DNA strands by extending the primers 5’ to 3’.
    - Taq polymerase is used in particular in PCR because it is taken from bacteria that live in high temperatures so that it can stay active during the denaturation steps of the reaction.
- Temperatures at each stage of PCR
    - Denaturation - High temp (90 degrees)

    - Annealing - Low temp (55 degrees)
    - Extension (or elongation) - Medium temp (70 degrees)
- In order to start PCR, we need template DNA, DNA polymerase, all the bases, and 2 primers.
- One caveat is that you have to add new DNA polymerase after every cycle since the enzymes get rendered unusable afterwards.
- At different locations in the genome, there are some sequences that are repeated and the location and number can differ from chromosome to chromosome.
    - This variation is called a variable number tandem repeat or VNTR.
        - Specifically it is the location of a repeated sequence.
- Differences among people with respect to these VNTRs are the basis of DNA typing.
- The possible number of VNTR alleles in a population is number of times a short noncoding sequence is repeated in tandem along the DNA.

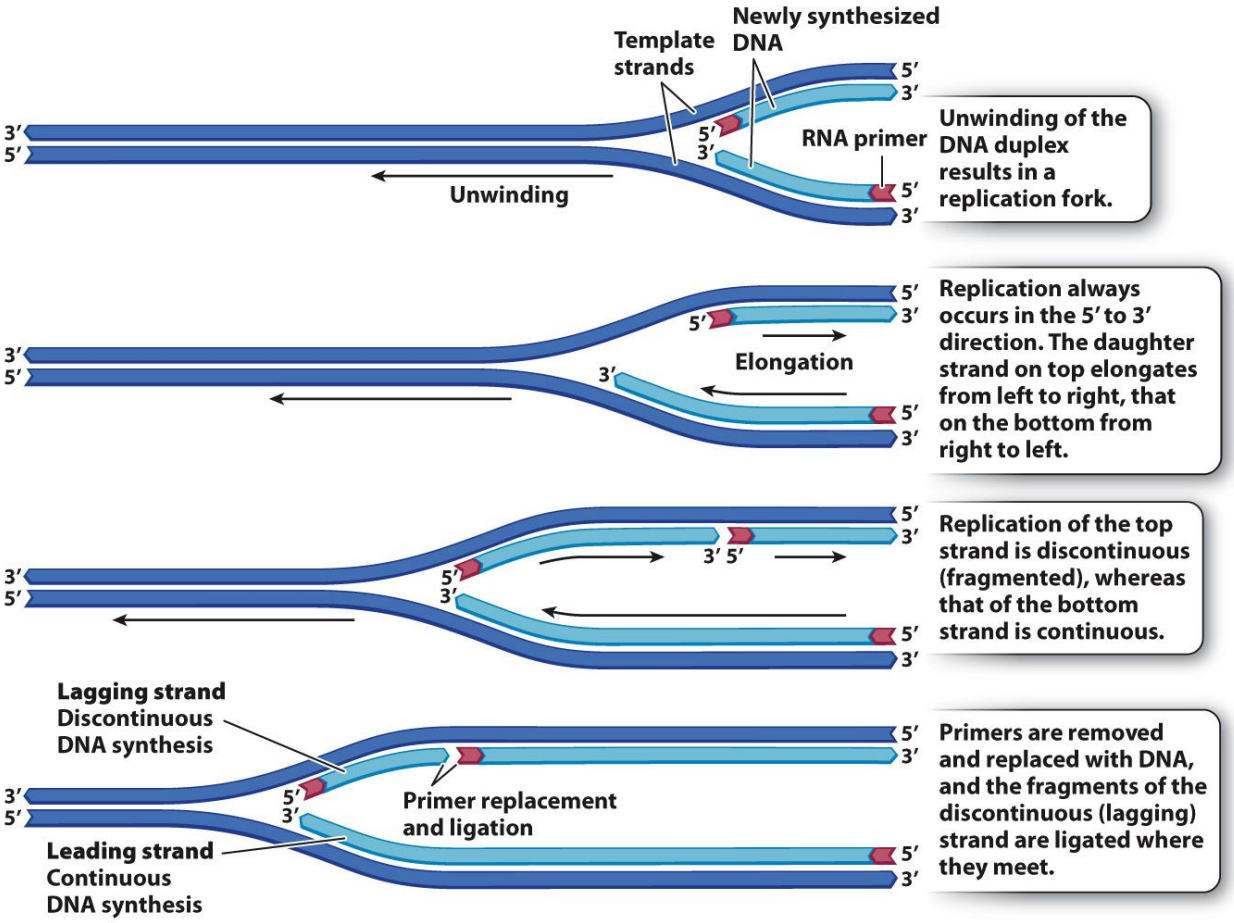
13.4 - Organization of Genomes

- Genomes of all organisms are large relative to the size of the cell.
    - Therefore there is a need to be able to package DNA into a form that will fit inside the cell.
- DNA in bacteria is underwound, which means it takes fewer turns in going around the circle than would allow every base in one strand to pair with its partner.
    - This is caused by the topoisomerase enzyme that breaks the double helix.
    - The strain it creates is relieved by the formation of supercoils where the DNA coils on itself.
- Supercoils resulting from underwinding are called negative supercoils. (More often)
- Supercoils resulting from overwinding are called positive supercoils.
- Supercoils of DNA form a structure with multiple loops called a nucleoid.
    - Held together by proteins
- In eukaryotic DNA, the DNA is linear and forms a single chromosome, which is packed with proteins to form a protein-DNA complex called chromatin.
    - Basically the DNA gets wrapped twice around histone proteins called a nucleosome. Then chromosome condensation happens, then the histones are removed, and then the DNA spreads out in loops around a structure called the chromosome scaffold.
- Size of eukaryotic chromosome is vastly greater than the size of the bacterial nucleoid.
- Pairs of chromosomes that match in size and appearance are called homologous chromosomes.
- Chromosomes are isolated in the metaphase of mitosis.
- Humans have 23 pairs of chromosomes.
- Each type of organelle in a cell has its own DNA. Each cell has a nuclear genome consisting of the DNA in the chromosomes.
    - Cells with mitochondria/chloroplast also have genomes for those organelles.
- Smallest to largest: gene, DNA strand, chromosome, genome

Chapter 12 -

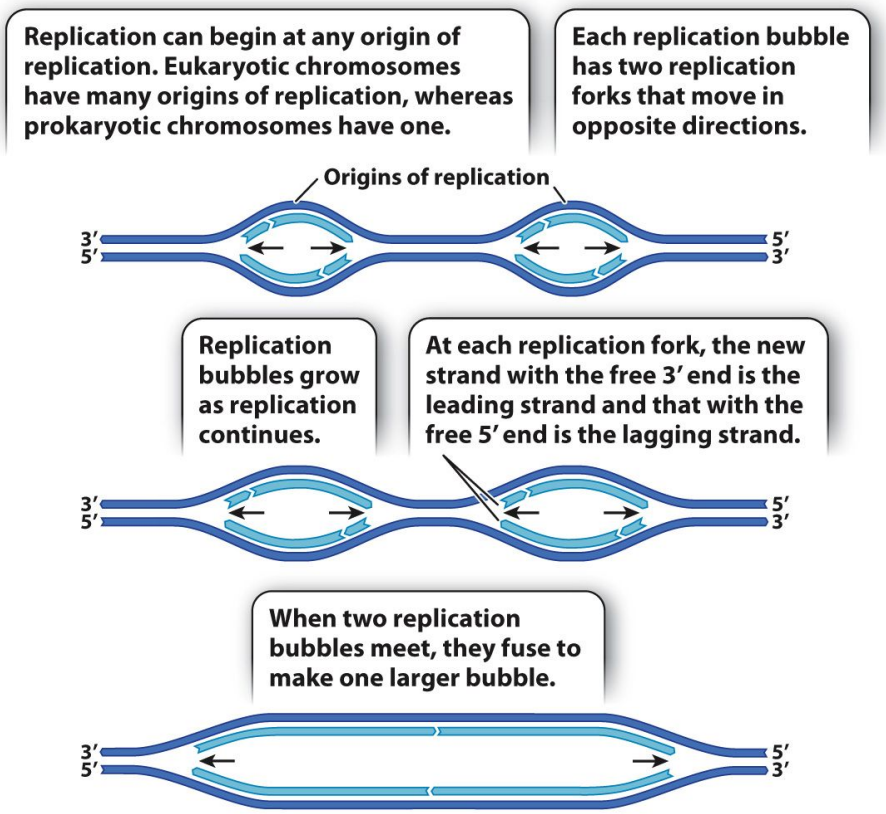
12.1 - DNA Replication

- When DNA unwinds, each strand serves as a template strand for the synthesis of a daughter strand.
- The sequence 5' - ATGC - 3' specifies 3' - TACG - 5' in the daughter strand.
- Semiconservative replication is where after replication, each new DNA duplex consists of one strand that was part of the parent duplex and one newly synthesized strand.
    - Alternate is conservative replication where original DNA duplex is intact and the daughter DNA is completely new.
- DNA replicates semiconservatively.
- The site where the parental strands separate is called the replication fork.
- DNA polymerization occurs in the 5' to 3' direction. The DNA polymerase, however, reads the DNA template strand in the 3' to 5' direction.
    - Thus a new DNA strand can be elongated only at the 3' end.
- The 3' OH off the growing daughter strand attacks the high energy phosphate bond of the incoming nucleotide to power the synthesis reaction.
- Polymerization reaction is catalyzed by the enzyme DNA polymerase.
    - Their job is to synthesize a new DNA strand from an existing template.
- The leading strand is the daughter strand that has its 3' end pointing to the replication fork.
- Replication of the non leading strand is discontinuous while the replication for the leading strand is continuous.
    - For the non leading strand, as the helix unwinds, a new piece of DNA will be synthesized so the whole process happens in short and discontinuous pieces.
    - The non leading strand is also called the lagging strand.
    - The short fragments are called Okazaki fragments.
- Each new DNA must begin with a short stretch of RNA that serves as a primer. This is needed for the DNA polymerase to start.
    - Primer is made up of an RNA polymerase called RNA primase.
    - After primer gets created, the DNA polymerase will start adding nucleotides to it and will eventually remove that RNA primer.
- Thus, all strands of DNA have a short stretch of RNA at their 5' ends.
    - For the lagging strand, you have a lot of primers since you have so many short and discontinuous pieces.
    - Adjacent fragments are joined by an enzyme called DNA ligase.
- Other proteins like topoisomerase II work upstream from the replication fork to relieve the stress on the double helix from all of the unwinding.
- Helicase separates the strands of the parent helix at the replication fork.
- Single strand binding proteins bind to the single stranded regions so that they don't join back together.
- Synthesis of both strands is coordinated with each other in terms of speed.

- DNA polymerases can correct their own errors in a process called proofreading.
    - Mispairing between a base in the parental strand a newly added base in the daughter strand activates a DNA cleavage function of DNA polymerase that removes the incorrect nucleotide and adds the correct one.
    - When you have an improper pairing, the exonuclease activity of the DNA polymerase gets activated, and this cleaves and removes the incorrect one.
- 

12.2 - Replication of Chromosomes

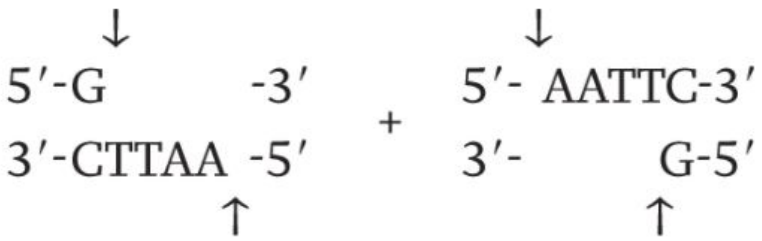
- Each point at which DNA synthesis is initiated is called an origin of replication.
- Opening of the helix creates a replication bubble with a fork on each side.



- To fix the problem of shortened ends that could come with the lagging strand on the daughter strands, each eukaryotic chromosome is capped by a repeating sequence called the telomere.
    - Basically, the shortened end will be restored by an enzyme called telomerase.
- There is high activity of telomerase in germ cells and stem cells, but inactive in almost every other cell.
- In contrast to linear DNA replication, circular DNA replication has a single origin of replication.

12.3 -

- Cutting DNA allows whole genomes to be broken up into smaller pieces for further analysis.
- Restriction enzymes are those that can recognize nucleotide sequences and can cleave the DNA at those sites.
    - The sequences they cleave are called restriction sites.
        - 4 to 6 bases long
- The cleaved double stranded molecules will terminate with overhangs.



- There can be 5', 3', and no overhangs.
- The base pairing of complementary single stranded nucleic acids is known as renaturation.
    - Denatured DNA strands from one source can renature with DNA strands from another source if their sequences are mostly complementary.
    - The degree of base pairing between two sequences affects the temperature at which they nature.
- A small DNA fragment called a probe can be used to determine whether or not a sample of double stranded DNA contains sequences that are complementary to it.
- Using Southern Blot will tell you the sizes and the number of copies a particular DNA sequence present in the starting sample.

12.4 - Genetic Engineering

- Isolating genes from one species and introducing them in another is called recombinant DNA technology.
    - Involves cutting DNA by restriction enzymes, isolating them by gel electrophoresis, and ligating them with enzymes used in DNA replication.
- To insert a gene into a loop of bacterial DNA (called a plasmid), you need
    - A double stranded DNA as the donor
    - Vector sequence into who the donor fragment is to be inserted.
    - Restriction enzymes
- In order to make sure the donor DNA can be fused with the vector DNA, both pieces are cut with the same restriction enzyme so they both have the same overhangs.
- Basically, the donor and vector DNA are cleaved with the same enzyme, the fragments are mixed and joined together by DNA ligase, the genomic and vector fragments have complementary ends, and the recombinant DNA molecule is finally introduced into a bacterial cell, where they multiply.
- Using fragments with overhangs is preferred over those without overhangs because the former allows greater control over what can bind to it. Restriction fragments created by BamHI can combine only with other fragments produced by BamHI. In contrasts, any blunt end (fragments without overhangs) can be attached to any other blunt end.
- Genetically engineering organisms are known as transgenic organisms.

Chapter 11 -

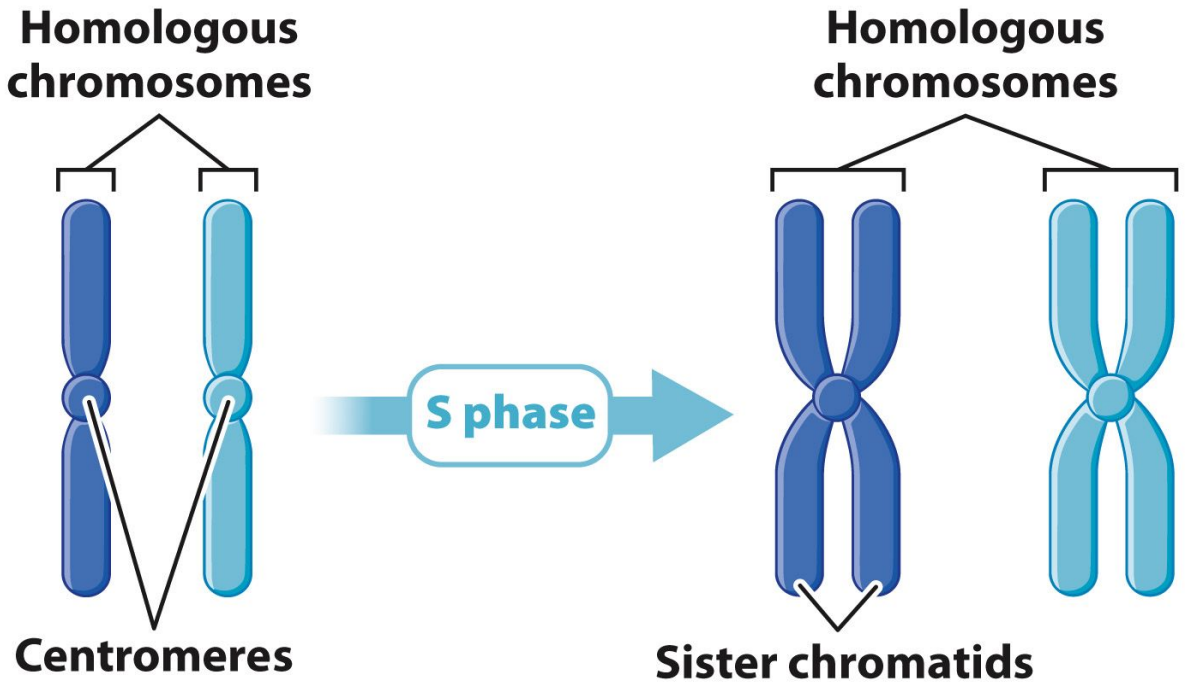
11.1 - Cell Division

- Cell division is the process by which a single cell becomes two daughter cells.
    - Both must receive full complement of DNA in the parent cell.
    - Parent must be large enough to divide into 2.
        - Cellular components should be duplicated before the actual division.
- Prokaryotic cells divide by binary fission, and eukaryotic cells divide the nucleus by mitosis and then divide the cytoplasm by cytokinesis.
- Binary fission consists of a cell replicating its DNA, increasing in size, and dividing into 2 daughter cells.
    - DNA replication happens at the origin of replication
    - Two DNA molecules get created, which are attached to the plasma membrane at different sites.
    - The two sites move apart and cells split apart with new membrane and wall at the midpoint.
    - The gene FtsZ is one that plays a key role in the actual division.
        - If it gene is mutated, then cell division will get blocked.
- Since the genome of eukaryotes is much larger and is organized into one or more linear chromosomes, it's more complicated.
- Cell cycle consists of M phase and interphase.
    - In M phase, the parent splits into 2 daughter cells. It consists of mitosis (separation of chromosomes into two nuclei) and cytokinesis where the division of the cell happens.
        - Cytokinesis can start before mitosis is complete.
    - Interphase is the other stage where cell makes preparations for division.
        - This is where replication of the DNA happens (S phase).
        - Before that though, is the G1 phase and after is the G2 phase. In these phases, regulatory proteins are made and activated. The proteins promote the activity of enzymes that synthesize DNA.
        - G1 preps for S phase and G2 preps for M phase.
- Cells can pause between the M and S phases for long periods, and is characterized by the G0 phase. It's different from G1 because you have the absence of preparations for DNA synthesis.

11.2- Mitotic Cell Division

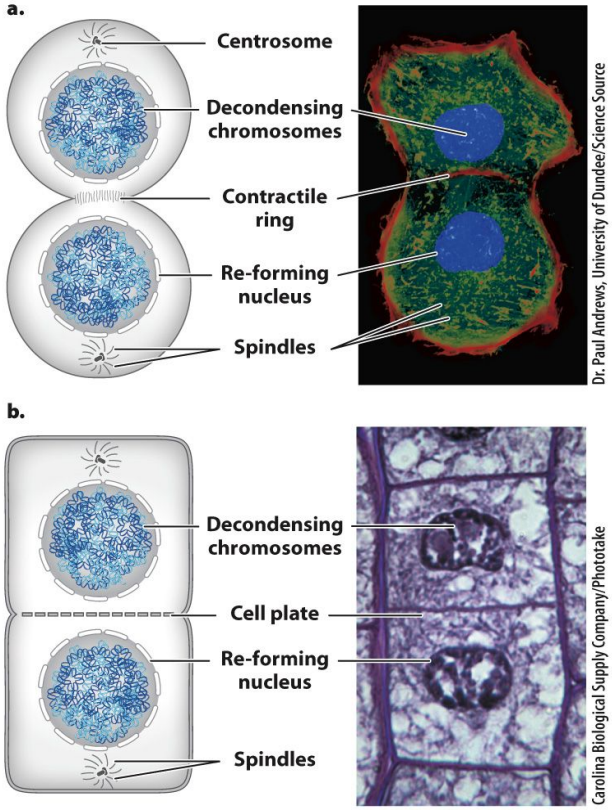
- Mitotic cell division (mitosis + cytokinesis) is the normal mode of asexual reproduction in unicellular eukaryotes.
- One of the main concerns is making sure daughter cells get an equal and complete set of chromosomes.
- In eukaryotic cells, DNA is packaged with histones and other proteins in chromatin which forms chromosomes.
- Every species is characteriszed by a specific number of chromosomes and each chromosome contains a single molecule of DNA with a specific set of genes.

    - The portrait formed by the number and shape of chromosomes representative of a species is called its karyotype.
- Most cells in the human body contain 46 chromosomes.
    - In human karyotypes, the 46 chromosomes can be arranged into 23 pairs.
        - Homologous chromosomes contains the same set of genes. One chromosome is taken from the father and one from the mother. The DNA sequences of the two are similar but not identical.
    - 22 pairs are homologous and 1 pair is the sex chromosome.
- A cell with one complete set of chromosomes is a haploid.
- A cell with two complete sets is a diploid.
- When each chromosome is duplicated so that each daughter cell gets a set of chromosomes, the identical copies called the sister chromatids don't separate and are held together at the centromere.
    - Sister chromatids will have identical DNA sequences.



- Stages of mitosis
    - Prophase - Chromosomes condense and the centrosomes migrate to opposite poles. Mitotic spindle assembles, spindle is a structure made of microtubules that pull the chromosomes into separate cells. Centrosome is the structure from which the spindles radiate.
    - Prometaphase - Nuclear envelope breaks down and microtubules of the mitotic spindle attach to chromosomes at their centromeres. There are proteins called kinetochores that are associated with each of the sister chromatids and they each from a site of attachment.

    - Metaphase - Once each chromosome is attached to the spindles from both poles of the cell, the spindle length will adjust so that chromosomes align in center of cell.
    - Anaphase - Sister chromatids separate as the centromere holding the pair splits and travel to opposite poles. The spindle microtubules also shorten.
    - Telophase - Nuclear envelope reforms and the chromosomes decondense.
- In animal cells, cytokinesis begins when a ring of filaments called the contractile ring forms between the two cells and uses motor proteins (myosin) to pinch the cytoplasm and divide the cell in 2.
- In plant cells, the dividing cells with form a structure called a phragmoplast in the middle of the cell. It is called the cell plate during anaphase and telophase and once it is large enough it fuses with the original cell wall at the perimeter of the cell.



- If a cell went through mitosis but not cytokinesis, then you would have a single cell with two nuclei and thus twice the amount of data. It is a multinucleate cell.

11.4- Regulation of the Cell Cycle

- Cells need signals on when to divide.
- Animal embryos undergo many rapid mitotic cell divisions.
    - In the cycle of rapid S and M phases, proteins appear and disappear in a cyclical fashion.
    - Several enzymes (kinases) become active and inactive in cycles.

- Regulatory proteins for cells are cyclins because their levels rise and fall as a function of the current phase of the cell cycle. They also activate kinases.
    - New cyclin proteins appear in the cytoplasm through protein synthesis.
- Kinases (or CDKs) are activated by cyclins and they are complexes that trigger required cell events. They activate only when they are bound to the appropriate cyclin.
    - They carry out their function by adding phosphate groups to the target proteins.
- 3 types of cyclin-CDK complexes that help the cell at several stages.
    - G1/S cyclin-CDK is active at the end of the G1 phase and is necessary to enter to S phase and prepares the cell for DNA replication by promoting the expression of histone proteins.
    - S cyclin-CDK helps initiate DNA synthesis. It is necessary and activates enzymes and proteins necessary for DNA replication.
    - M cyclin-CDK helps prepare the cell for mitosis. It phosphorylates structural proteins in the nucleus, which triggers the breakdown of the nuclear envelope in prometaphase.
- If something goes wrong in the cell cycle, then there are mechanisms to block cyclin-CDK activity. Mechanisms are called checkpoints.
- 3 checkpoints
    - Presence of damaged DNA stops the cell at the end of G1 before DNA synthesis.
        - Damage needs to be repaired so that it is not inherited by the daughter cells.
        - A specific protein kinase gets activated that phosphorylates a protein called p53. It activates a gene that blocks the activity of the G1/S cyclin-CDK complex and thus blocks the cell at the G1/S transition.
    - Presence of unreplicated DNA arrests the cell at the end of G2 before mitosis.
    - Abnormalities in chromosome attachment stop the cell in early mitosis.

11.5- Genes Involved in Cancer

- Viruses are assemblages of protein surrounding a core of RNA or DNA.
    - They only carry a handful of genes but can multiply rapidly.
- The v-src gene is an example of an oncogene which is a cancer causing gene that is found in viruses.
- There are also normal cellular genes called proto-oncogenes which are involved in cell division but don't cause cancer.
- Human proto-oncogenes can turn into oncogenes by environmental agents.
- Every protein that performs a step in a signaling cascade that promotes cell division can be the product of a proto-oncogene.
- When the p53 protein is mutated, then the cell can divide before the DNA damage is repaired. It is an example of a tumor suppressor which are proteins whose normal activities inhibit cell division.
    - They act in opposition to proto-oncogenes.
- Oncogenes cause cancer by producing an excess of protein activity that causes the cell to divide while tumor suppressors prevent cell division.

- Most cancers need more than the overactivation of one oncogene or the inactivation of one tumor suppressor.
- Malignant cancers are ones that grow rapidly and invade the surrounding tissue.
- A gene associated with promoting normal cell division is a proto-oncogene. Tumor suppressor doesn't fall under that category.

Chapter 14 -

14.1 - The Rate and Nature of Mutations

- Mutations can occur from mistakes in DNA replication, unrepaired damage to DNA (from chemicals, radiation), insertion/jumping of DNA sequences in a gene, and incorrectly repaired chromosome breaks caused by reactive chemicals or radiation.
- Mutations are mostly spontaneous and random.
- Mutation rates are not related to genome size.
- There can be sites in the genome that are especially mutable and they are called hotspots.
- Rate of mutation per nucleotide per replication is greater in somatic cells that in germ cells.
- You can measure mutation based on the rate of mutation per nucleotide per replication or by the rate per genome per generation.
- Mutations in eggs and sperm are called germ line mutations and those in nonreproductive cells are called somatic mutations.
    - The former may be transmitted to future generations and uses rate per genome per generation while the latter won't and uses rate of mutation per nucleotide per replication.
- For cancer to develop, mutations must occur in the same cell lineage.
- Most cancers come from mutations in somatic cells.
- Any mutation that increases the risk of disease in an individual is known as a genetic risk factor for that disease.

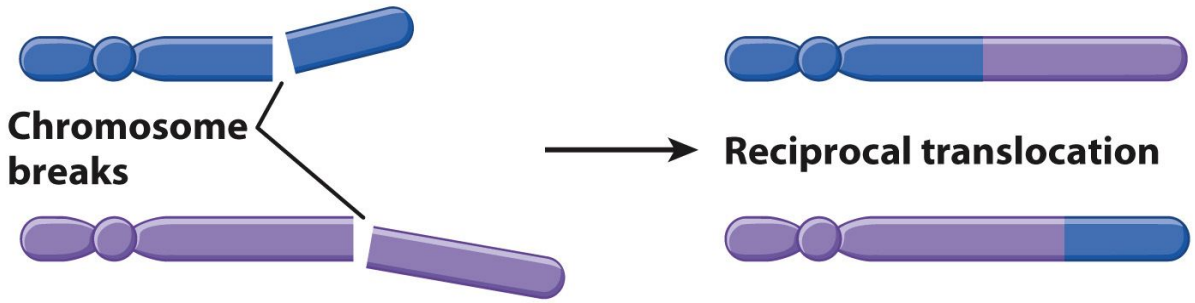
14.2 - Small Scale Mutations

- A mutation is a change in the nucleotide sequence of a genome.
- Most DNA damage or errors in replication are immediately removed by enzymes in the cell.
    - Proofreading function of the DNA polymerase is a prime example.
- If a mutation doesn't get caught by the proofreading function, then the mutation will just stay in the cell lineage for the subsequent replication rounds.
- Source of new mutations is many organisms is the insertion of movable DNA sequences into or near a gene. Sequences are called transposons.
    - They can move from one position to another in the genome.
    - Transposition of these elements occur differently depending on the type of transposon.

    - Cut and paste mechanisms are used where an enzyme called transposase cleaves the transposon and inserts it into a different position.
    - You can also undergo transposition through an RNA intermediate. This where you have a reverse transcriptase and an integrase

14.3 - Chromosomal Mutations

- Chromosomal mutations can affect large regions that extend over hundreds of thousands of nucleotides.
    - They can delete or duplicate regions of the chromosome containing several or many genes.
- Common chromosome abnormality is when a segment of the chromosome is either present in two copies or is missing altogether.
    - The chromosome where the region is present twice contains a duplication.
    - The other chromosome where the region is missing contains a deletion.
- The deletion can persist harmlessly if the chromosome is present along with a normal chromosome.
- Dosage is the number of copies of a gene.
- There are rarely any deletions or duplications that include the centromere which is the site where there is the attachment of the spindle fibers that move the chromosome during cell division.
- Process of creating new genes from duplicates of old ones is called duplication and divergence.
- Group of genes created from ^ is known as a gene family.
- Chromosomes where the normal order of a block of genes is reversed contain an inversion.
- Reciprocal translocation occurs when two different chromosomes undergo an exchange of parts.



- These don't affect the survival of organisms since they only change the arrangement of genes and not their number.
- Translocation is when a chromosomal segment breaks off and attaches to another chromosome.

14.4 - DNA Damage and Repair

- Mutagens are agents that increase the probability of mutation.

- Mutagens can cause breaks in the sugar phosphate backbone and can cause breaks in the strands of DNA.
    - You can also lose a base in one of the sugars.
- Repair of breaks in the backbone is handled by DNA ligase which is an enzyme that uses ATP energy to join the 3’ and 5’ ends.
    - One type of ligase seals the single stranded breaks and a different type seals the double stranded breaks.
- There is also a postreplication mismatch repair which helps to catch mutations. MutS is a protein that will bind to the site of the mismatch, will recruit MutL and MutH, MutL will determine which backbone to cleave, and then MutH will break the backbone, an exonuclease will remove the nucleotides, and then a DNA polymerase will fill in the missing correct ones, and then the DNA ligase will join the backbones.
- Base excision repair corrects abnormal or damaged bases. An enzyme that recognizes the incorrect base will cleave it from the backbone, and then a different enzyme called AP endonuclease will recognize the site and cleaves the backbone so that there is a gap, and then other proteins come to add the correct base.
- Mismatch repair is a backup for the DNA polymerase proofreading function