Biochemistry

Wikibooks.org

March 28, 2013

Contents

Contents

1 Biochemistry/Print version

This is the print version[1] of Biochemistry[2]

You won't see this message or any elements not part of the book's content when you print or preview[3] this page.

---

1   http://en.wikibooks.org/wiki/Help:Print_versions
2   http://en.wikibooks.org/wiki/Biochemistry
3   http://en.wikibooks.org//en.wikibooks.org/w/index.php?title=Biochemistry/Print_version&action=purge&printable=yes

2 Introduction

2.0.1 Intro: What Is Biochemistry?

Biochemistry is the study of the chemistry of, and relating to, biological organisms. It forms a bridge between biology and chemistry by studying how complex chemical reactions and chemical structures give rise to life and life's processes. Biochemistry is sometimes viewed as a hybrid branch of organic chemistry which specializes in the chemical processes and chemical transformations that take place inside of living organisms, but the truth is that the study of biochemistry should generally be considered neither fully "biology" nor fully "chemistry" in nature. Biochemistry incorporates everything in size between a molecule and a cell and all the interactions between them. The aim of biochemists is to describe in molecular terms the structures, mechanisms and chemical processes shared by all organisms, providing organizing principles that underlie life in all its diverse forms. Biochemistry essentially remains the study of the structure and function of cellular components (such as enzymes and cellular organelles) and the processes carried out both on and by organic macromolecules - especially proteins, but also carbohydrates, lipids, nucleic acids, and other biomolecules. All life forms alive today are generally believed to have descended from a single proto-biotic ancestor, which could explain why all known living things naturally have similar biochemistries. Even when it comes to matters which could appear to be arbitrary - such as the genetic code and meanings of codons, or the "handedness" of various biomolecules - it is irrefutable fact that all marine and terrestrial living things demonstrate certain unchanging patterns throughout every level of organization, from family and phylum to kingdom and clade. Biochemistry is, most simply put, the chemistry of life.

2.1 Thermodynamics

2.2 Why Do Substances React?

Chemical (and thus, biochemical) reactions only occur to a significant extent if they are energetically favorable. If the products are more stable than the reactants, then in general the reaction will, over time, tend to go forward. Ashes are more stable than wood, so once the energy of activation is supplied (e.g., by a match), the wood will burn. There are plenty of exceptions to the rule, of course, but as a rule of thumb it's pretty safe to say that if the products of a reaction represent a more stable state, then that reaction will go in the forward direction. There are two factors that determine whether or not reactions changing reactants into products are considered to be *favorable*: these two factors are simply called **enthalpy** and **entropy**.

2.2.1 Enthalpy

Simply put, **enthalpy** is the heat content of a substance (*H*). Most people have an intuitive understanding of what heat is... we learn as children not to touch the burners on the stove when they are glowing orange. Enthalpy is **not** the same as that kind of heat. Enthalpy is the sum of all the internal energy of a substance's matter plus its pressure times its volume. Enthalpy is therefore defined by the following equation:

$$H = U + PV$$

where (all units given in SI)

- *H* is the enthalpy
- *U* is the internal energy, (joules)
- *P* is the pressure of the system, (Pascals)
- and *V* is the volume, (cubic meters)

If the enthalpy of the reactants while being converted to products ends up decreasing (ΔH < 0), that means that the products have less enthalpy than the reactants and energy is released to the environment. This reaction type is termed *exothermic.* In the course of most biochemical processes there is little change in pressure or volume, so the change in enthalpy accompanying a reaction generally reflects the change in the internal energy of the system. Thus, exothermic reactions in biochemistry are processes in which the products are lower in energy than the starting materials. As an example, consider the reaction of glucose with oxygen to give carbon dioxide and water. Strong bonds form in the products, reducing the internal energy of the system relative to the reactants. This is a highly exothermic reaction, releasing 2805 kJ of energy per mole of glucose that burns (ΔH = -2805 kJ/mol). That energy is given off as heat.

ΔH	reactants/products	environment	favorable
< 0	releases heat	heats up	yes
> 0	gains heat	cools down	no

2.2.2 Entropy

Entropy (symbol *S*) is the measure of randomness in something. It represents the most likely of statistical possibilities of a system, so the concept has extremely broad applications. In chemistry of all types, entropy is generally considered important in determining whether or not a reaction goes forward based on the principle that a less-ordered system is more statistically probable than a more-ordered system. What does that mean, really? Well, if the volcano Mt. Vesuvius erupted next to a Roman-Empire era Mediterranean city, would the volcano be more likely to destroy the city, or build a couple of skyscrapers there? It's pretty obvious what would happen (or, rather, what *did* happen) because it makes sense to us that natural occurrences favor randomness (destruction) over order (construction, or in this case,

skyscrapers). Entropy is just a mathematical way of expressing these essential differences. When it comes to chemistry, there are three major concepts based on the concept of entropy:

1. *Intramolecular* states (*Degrees of freedom*)

   The more degrees of freedom (how much the molecules can move in space) a molecule has, the greater the degree of randomness, and thus, the greater the entropy.There are three ways molecules can move in space, and each has a name: rotation = movement around an axis, vibration = intramolecular movement of two bonded atoms in relation to each other, and translation = a molecule moving from place to place.

2. *Intermolecular* structures

   When molecules can interact with each other by forming non-covalent bonds a structure is often created.This tends to reduce randomness (and thus entropy) since any such association between molecules stabilizes the motion of both and decreases the possibilities for a random distribution.

3. Number of possibilities

   The more molecules present, the more ways of distributing the molecules in space - which because of statistical probabilities means more potential for randomness.Also, if there is more space available to distribute the molecules within, the randomness increases for precisely the same reason *solid matter (least entropy) << liquids << gases (most entropy)*

Changes in entropy are denoted as ΔS. For the reasons stated above (in the volcano situation), the increase of entropy (ΔS > 0) is considered to be favorable as far as the Universe in general is concerned. A decrease in entropy is generally not considered favorable unless an energetic component in the reaction system can make up for the decrease in entropy (see free energy below).

ΔS	**entropy**	**favorable**
> 0	increases	yes
< 0	decreases	no

2.2.3 Gibbs Free Energy

Changes of both **enthalpy** (ΔH) and **entropy** (ΔS) combined decide how favorable a reaction is. For instance, burning a piece of wood releases energy (*exothermic*, favorable) and results in a substance with less structure (CO_2 and H_2O gas, both of which are less 'ordered' than solid wood). Thus, one could predict that once a piece of wood was set on fire, it would continue to burn until it was gone. The fact that it does so is ascribed to the change in its **Gibbs Free Energy**. The overall favorability of a reaction was first described by the prominent chemist Josiah Willard Gibbs[1], who defined the *free energy* of a reaction as

---

1   http://en.wikibooks.org//en.wikipedia.org/wiki/Willard_Gibbs

-

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

where T is the temperature on the Kelvin temperature scale. The formula above assumes that pressure and temperature are constant during the reaction, which is almost always the case for biochemical reactions, and so this book makes the same assumption throughout. The unit of ΔG (for *Gibbs*) is the "joule" in SI systems, but the unit of "calorie" is also often used because of its convenient relation to the properties of water. This book will use both terms as convenient, but the preference should really be for the SI notation.

2.2.4 What Does ΔG Really Mean?

If $\Delta G < 0$ then the reactants should convert into products (signifying a forward reaction)... eventually. (Gibbs free energy says nothing about a reaction's *rate*, only its *probability*.) Likewise, for a given reaction if $\Delta G > 0$ then it is known that the reverse reaction is favored to take place. A state where $\Delta G = 0$ is called **equilibrium**, and this is the state where the reaction in both the forward and reverse directions take place at the same rate, thus not changing the **net** effect on the system.

> How is equilibrium best explained? Alright, as an example set yourself on the living room carpet with your most gullible younger relative (a little nephew, niece or cousin will work fine). Take out a set of Monopoly, take one ten dollar bill for yourself and give your little relative the rest. Now both of you give the other 5% of all that you have. Do this again, and again, and again-again-again until eventually... you both have the same amount of money. This is **precisely** what the equilibrium of a reaction means, though equilibrium only very rarely results in an even, 50-50% split of products and reactants.

ΔG naturally varies with the concentration of reactants and products. When ΔG reaches 0, the reaction rate in the forward direction and the reaction rate in the reverse direction are the same, and the concentration of reactants and products no longer appears to change; this state is called the *point of chemical equilibrium*. You and your gullible little relative have stopped gaining and losing Monopoly money, respectively; you both keep exchanging the same amount each turn. Note again that equilibrium is *dynamic*. Chemical reaction does not cease at equilibrium, but products are converted to reactants and reactants are converted to products at exactly the same rate. A small ΔG (that is, a value of ΔG close to 0) indicates that a reaction is somewhat reversible; the reaction can actually run backwards, converting products back to reactants. A very large ΔG (that is, $\Delta G >> 0$ or $\Delta G << 0$) is precisely the opposite, because it indicates that a given reaction is irreversible, i.e., once the reactants become products there are very few molecules that go back to reactants.

2.3 Metabolic pathways

The food we consume is processed to become a part of our cells; DNA, proteins, etc. If the biochemical reactions involved in this process were reversible, we would convert our own DNA back to food molecules if we stop eating even for a short period of time. To prevent

this from happening, our *metabolism* is organized in *metabolic pathways.* These pathways are a series of biochemical reactions which are, as a whole, irreversible. The reactions of a pathway occur in a row, with the products of the first reaction being the reactants of the second, and so on:

A $\rightleftharpoons$ B $\rightleftharpoons$ C $\rightleftharpoons$ D $\rightleftharpoons$ E

At least one of these reactions has to be irreversible, e.g.:

A $\rightharpoonup$ B $\rightleftharpoons$ C $\rightleftharpoons$ D $\rightharpoonup$ E
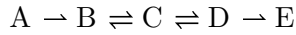
The control of the irreversible steps (e.g., A → B) enables the cell to control the whole pathway and, thus, the amount of reactants used, as well as the amount of products generated. Some metabolic pathways do have a "way back", but it is not the same pathway backwards. Instead, while using the reversible steps of the existing pathway, at least one of the irreversible reactions is bypassed by another (irreversible) one on the way back from E to A:

E $\rightharpoonup$ **X** $\rightleftharpoons$ C $\rightleftharpoons$ B $\rightharpoonup$ A

This reaction is itself controlled, letting the cell choose the direction in which the pathway is running.

2.4 Free energy and equilibrium

For ΔG, the free energy of a reaction, standard conditions were defined:

- concentration of reactants and products at 1M
- temperature at 25°C
- acidity at pH 7.0

Under these standard conditions, $\mathbf{\Delta G^{0'}}$ is defined as the **standard free energy change**. For a reaction

A + B $\rightleftharpoons$ C + D
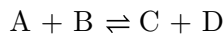
the ratio of products to reactants is given by $\mathbf{k_{eq}'}$ (=k_{eq} at pH 7.0):

$k'_{eq} = \frac{products}{reactants} = \frac{[C][D]}{[A][B]}$

The relationship of $\Delta G^{0'}$ and k_{eq}' is

$\Delta G^{0'}$ = - R T ln k_{eq}' = - R T 2.030 $\log_{10}$ k_{eq}'

with

R = 8.315 [J mol^{-1} K^{-1}] (molar gas constant[2])

T = temperature [K]

---

2   `http://en.wikibooks.org//en.wikipedia.org/wiki/molar_gas_constant`

In theory, we can now decide if a reaction is favorable ($\Delta G^{0'} < 0$). However, the reaction might need a *catalyst* to occur within a reasonable amount of time. In biochemistry, such a catalyst is often called an *enzyme*.

The purpose of DNA melting or DNA denaturation is emphasizing and demonstrating the life cycles of all organisms and the origin of replication. The origin of replication specific structure varies from species to species. Furthermore, the particular sequence of the origin of replication is in a genome which is the human genes. Nevertheless, DNA replication is also part of origin of replication which examen in the living organism such as prokaryotes and eukaryotes. Thermodynamically, there are two important contributions on the DNA denaturation. One of them is the breaking all of the hydrogen bonds between the bases in the double helix; the other one is to overcome the stacking stability/energy of bases on top of each other. There are several methods to denature DNA; heat is known as the most common one use in laboratory. We just have to heat the sample to reach above its melting point, the unstack ability of DNA can be then monitored. Melting point and denaturation of DNA depend on several factors: the length of DNA, base-composition of DNA, the condition of the DNA and also the composition of buffer. For instance, the longer DNA will contain more H-bonds and more intermolecular forces compared to the shorter one; therefore, denaturations of longer DNA requires more time and more heat. Base-composition of DNA can also play as a key factor because A:T requires two hydrogen bonds and G:C interaction requires three hydrogen bonds. The region of DNA which contains more A:T will melt/denature more rapidly compared to G:C. We can also see how the condition of DNA is important because condition of DNA is related to whether the DNA is relax, supercoiled, linear or heavily nicked. It is important because it allow us to examine how much intermolecular forces existing in the double helix. Finally, condition of buffer is also playing an essential role to study DNA denaturation because it allow us to control the amount of ions present in the solution during the entire process. Biologically, DNA denaturation can happen inside the cell during DNA replication or translation. In both cases, DNA denaturation is an essential step and a beginning to start each of the process. Most of the time, denaturation happened because of binding of protein or enzymes to a specific region of DNA, the binding will likely lead to open or denature of the helix. However, the actual meaning of the DNA melting is the denaturation of DNA which changes the structure of DNA from double stranded into single stranded. The processes of DNA denaturation is unwinding the double stranded deoxyribonucleic acid and breaks it into two single stranded by breaking the hydrogen bonding between the bases. DNA denaturation is also known of DNA annealing because it is reservable . The main steps DNA annealing are double helical will go through the denaturation to become partially denatured DNA then it will separated the strands into two single strand of DNA in random coils.

2.5 Catalysis

Catalysis refers to the acceleration of the rate of a chemical reaction by a substance, called a catalyst, that is itself unchanged by the overall reaction. Catalysis is crucial for any known form of life, as it makes chemical reactions happen much faster than they would "by themselves", sometimes by a factor of several million times. A common misunderstanding is that catalysis "makes the reaction happen", that the reaction would not otherwise proceed without the presence of the catalyst. However, a catalyst cannot make a thermodynamically

unfavorable reaction proceed. Rather, it can only speed up a reaction that is already thermodynamically favorable. Such a reaction in the absence of a catalyst would proceed, even without the catalyst, although perhaps too slowly to be observed or to be useful in a given context. Catalysts accelerate the chemical reaction by providing a lower energy pathway between the reactants and the products. This usually involves the formation of an intermediate, which cannot be formed without the catalyst. The formation of this intermediate and subsequent reaction generally has a much lower activation energy barrier than is required for the direct reaction of reactants to products. Catalysis is a very important process from an industrial point of view since the production of most industrially important chemicals involve catalysis. Research into catalysis is a major field in applied science, and involves many fields of chemistry and physics. Two types of catalysis are generally distinguished. In homogeneous catalysis the reactants and catalyst are in the same phase. For example acids (H+ ion donors) are common catalysts in many aqueous reactions. In this case both the reactants and the catalysts are in the aqueous phase. In heterogeneous catalysis the catalyst is in a different phase than the reactants and products. Usually, the catalyst is a solid and the reactants and products are gases or liquids. In order for the reaction to occur one or more of the reactants must diffuse to the catalyst surface and adsorb onto it. After reaction, the products must desorb from the surface and diffuse away from the solid surface. Frequently, this transport of reactants and products from one phase to another plays a dominant role in limiting the rate of reaction. Understanding these transport phenomena is an important area of heterogeneous catalyst research.

2.6 Enzymes

Enzyme (from Greek, *in ferment*) are special protein molecules whose function is to facilitate or otherwise accelerate most chemical reactions in cells. Many chemical reactions occur within biological cells, but without catalysts most of them happen too slowly in the test tube to be biologically relevant. Enzymes can also serve to couple two or more reactions together, so that a thermodynamically favorable reaction can be used to "drive" a thermodynamically unfavorable one. One of the most common examples is enzymes which use the dephosphorylation of ATP to drive some otherwise unrelated chemical reaction. Chemical reactions need a certain amount of activation energy to take place. Enzymes can increase the reaction speed by favoring or enabling a different reaction path with a lower activation energy (Fig. 1), making it easier for the reaction to occur. Enzymes are large globular proteins that catalyze (accelerate) chemical reactions. They are essential for the function of cells. Enzymes are very specific as to the reactions they catalyze and the chemicals (substrates) that are involved in the reactions. Substrates fit their enzymes like a key fits its lock (Fig. 2). Many enzymes are composed of several proteins that act together as a unit. Most parts of an enzyme have regulatory or structural purposes. The catalyzed reaction takes place in only a small part of the enzyme called the active site, which is made up of approximately 2 - 20 amino acids.
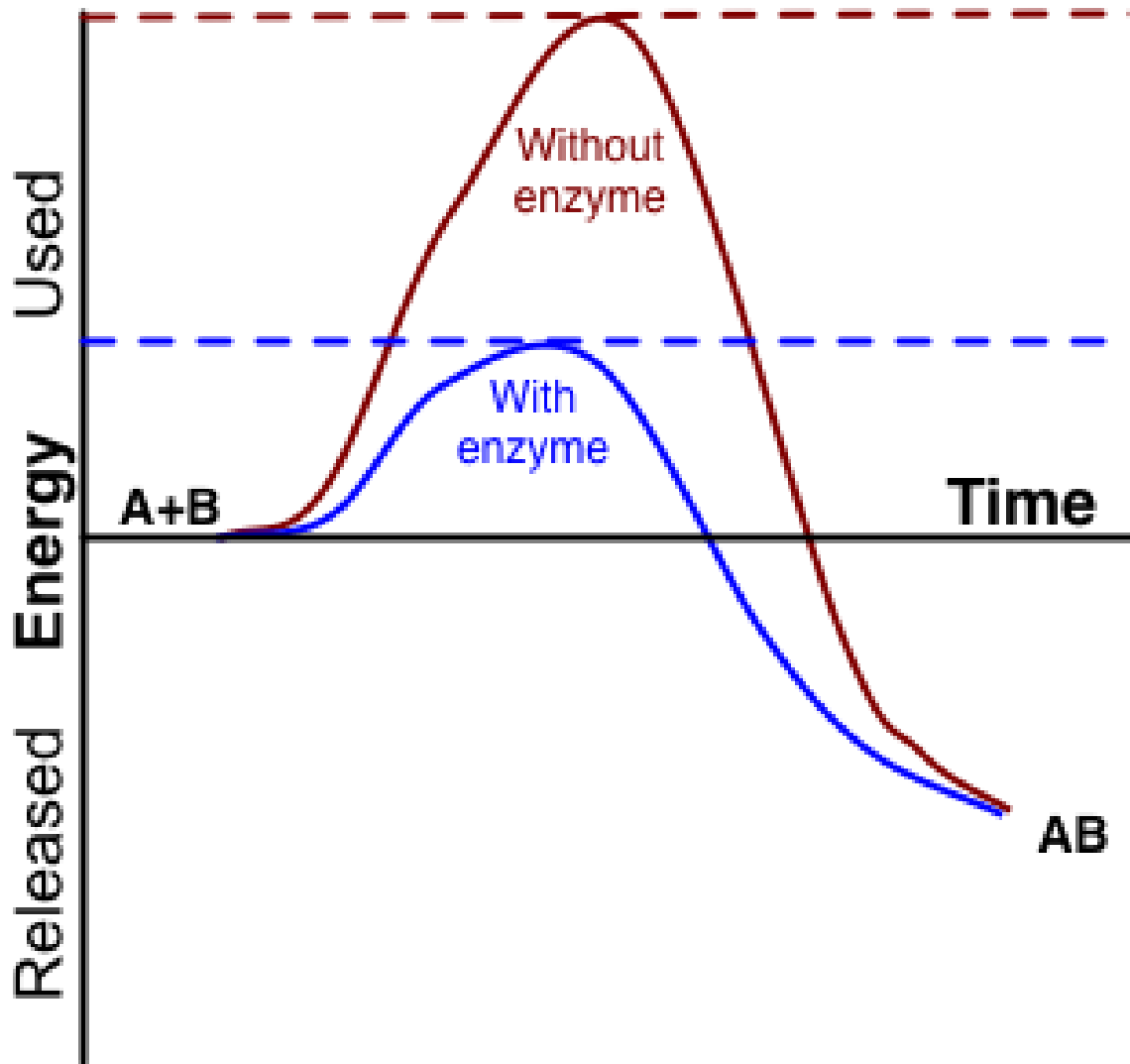
Figure 2   **Figure 1**: Diagram of a catalytic reaction, showing the energy needed (E) against time (t).

The substrates (A and B) need a large amount of energy (E_1) to reach the intermediate state A···B, which then reacts to form the end product (AB). The enzyme (E) creates a microenvironment in which A and B can reach the intermediate state (A···E···B) more easily, reducing the amount of energy needed (E_2). As a result, the reaction is more likely to take place, thus improving the reaction speed.
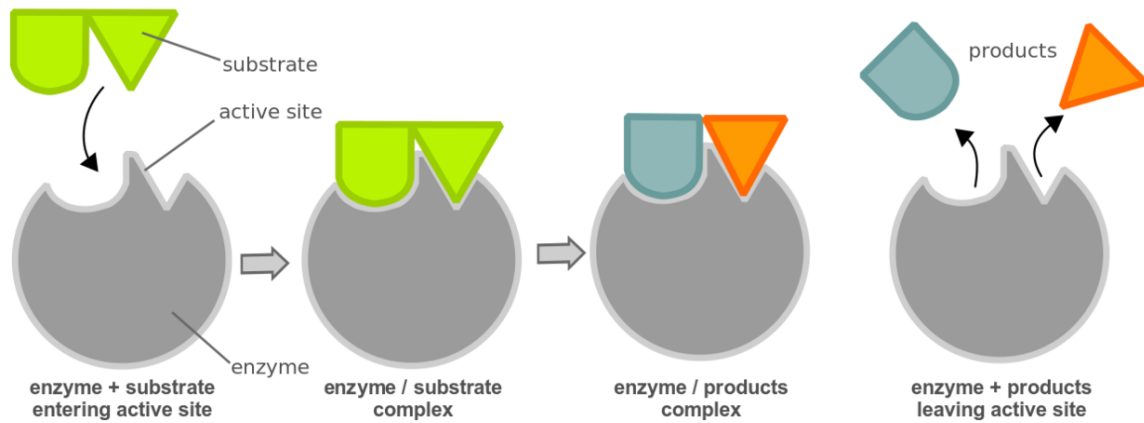
Figure 3 **Figure 2:** An enzyme catalyzes the reaction of two substrates and to form one product.

Enzymes can perform up to several million catalytic reactions per second. To determine the maximum speed of an enzymatic reaction, the substrate concentration is increased until a constant rate of product formation is achieved (Fig. 3). This is the maximum velocity ($V_{\max}$) of the enzyme. In this state, all enzyme active sites are saturated with substrate. This was proposed in 1913 by Leonor Michaelis and Maud Menten. Since the substrate concentration at $\mathbf{V}_{\max}$ cannot be measured exactly, enzymes are characterized by the substrate concentration at which the rate of reaction is half its maximum. This substrate concentration is called the Michaelis-Menten constant (K_{M}). Many enzymes obey Michaelis-Menten kinetics.
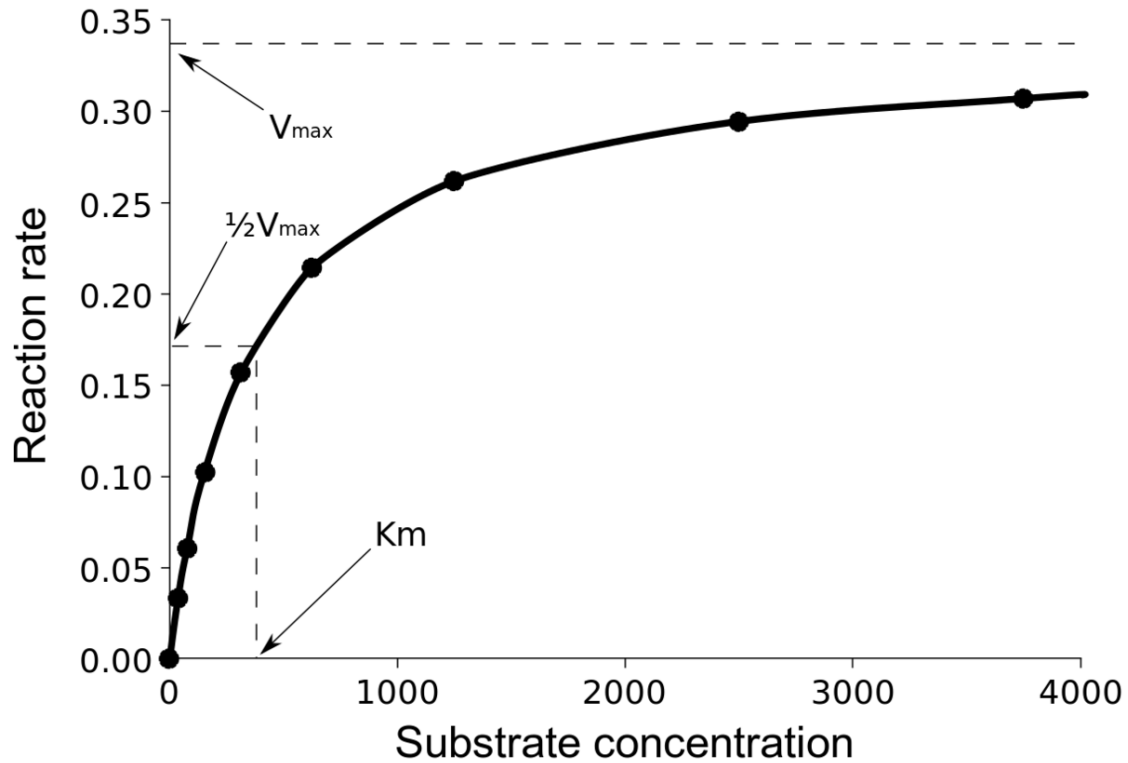
Figure 4   **Figure 3**: Diagram of reaction speed and Michaelis-Menten constant.

The speed V means the number of reactions per second that are catalyzed by an enzyme. With increasing substrate concentration [S], the enzyme is asymptotically approaching its maximum speed V_{max}, but never actually reaching it. Because of that, no [S] for V_{max} can be given. Instead, the characteristic value for the enzyme is defined by the substrate concentration at its half-maximum speed ($V_{max}/2$). This K_M value is also called Michaelis-Menten constant. Several factors can influence the reaction speed, catalytic activity, and specificity of an enzyme. Besides de novo synthesis (the production of more enzyme molecules to increase catalysis rates), properties such as pH or temperature can denature an enzyme (alter its shape) so that it can no longer function. More specific regulation is possible by posttranslational modification (e.g., phosphorylation) of the enzyme or by adding cofactors like metal ions or organic molecules (e.g., NAD^+, FAD, CoA, or vitamins) that interact with the enzyme. Allosteric enzymes are composed of several subunits (proteins) that interact with each other and thus influence each other's catalytic activity. Enzymes can also be regulated by competitive inhibitors (Fig. 4) and non-competitive inhibitors and activators (Fig. 5). Inhibitors and activators are often used as medicines, but they can also be poisonous.
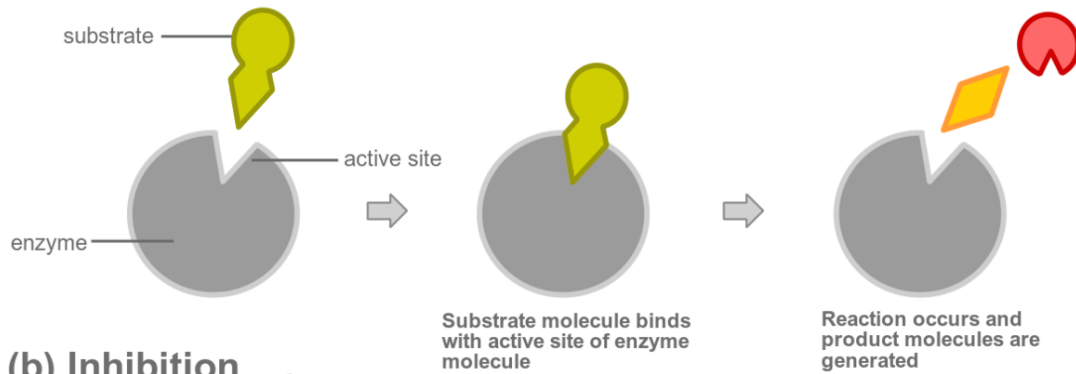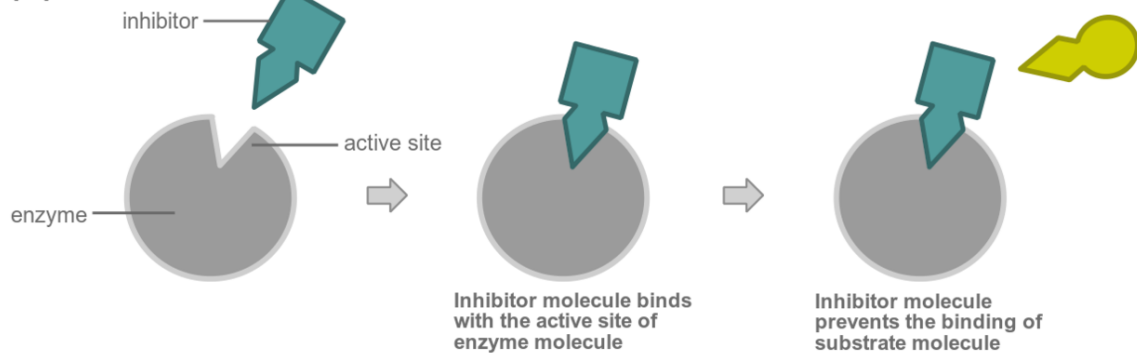
Figure 5 **Figure** 4: Competitive inhibition.

A competitive inhibitor fits the enzyme as well as its real substrate, sometimes even better. The inhibitor takes the place of the substrate in the active center, but cannot undergo the catalytic reaction, thus inhibiting the enzyme from binding with a substrate molecule. Some inhibitors form covalent bonds with the enzyme, deactivating it permanently (suicide inhibitors). In terms of the kinetics of a competitive inhibitor, it will increase K_m but leave V_{max} unchanged.
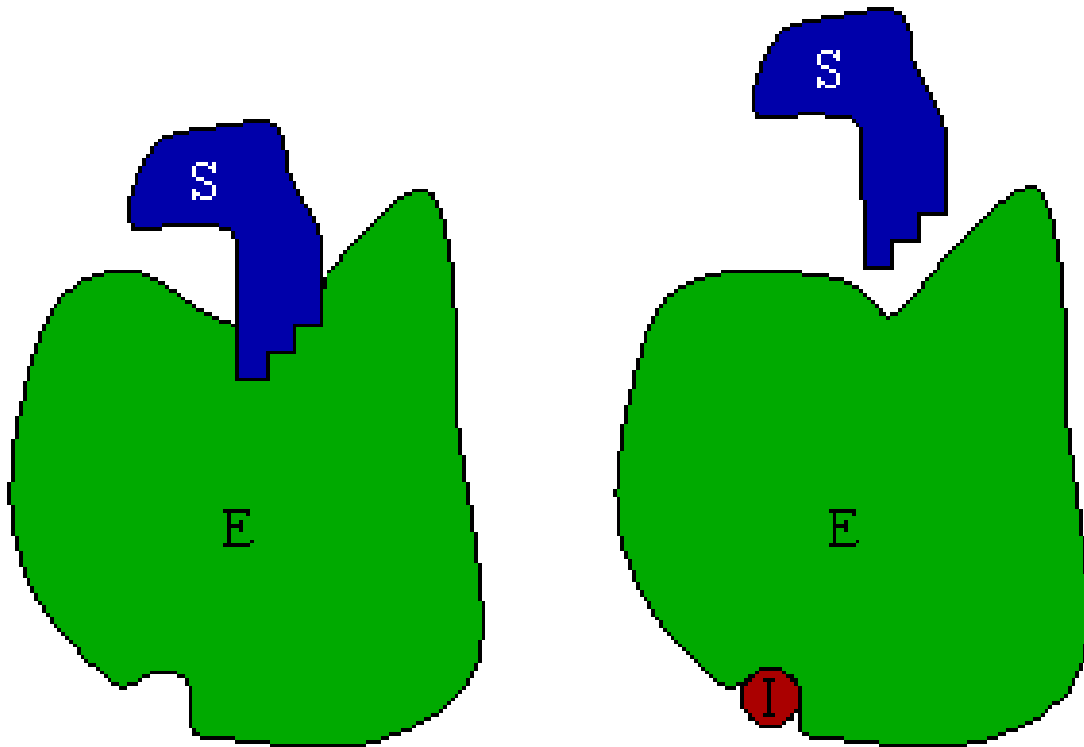
Figure 6   **Figure 5**: Non-competitive inhibition.

Non-competitive inhibitors/activators (I) do not bind to the active center, but to other parts of the enzyme (E) that can be far away from the substrate (S) binding site. By changing the conformation (the three-dimensional structure) of the enzyme (E), they disable or enable the ability of the enzyme (E) to bind its substrate (S) and catalyze the desired reaction. The noncompetitive inhibitor will lower V_{max} but leave K_m unchanged. An *uncompetitive* inhibitor will only bind to the enzyme-substrate complex forming an enzyme-substrate-inhibitor (ESI) complex and cannot be overcome by additional substrate. Since the ESI is nonreactive, V_{max} is effectively lowered. The uncompetitive inhibitor will in turn lower the K_m due to a lower concentration of substrate needed to achieve half the maximum concentration of ES. Several enzymes can work together in a specific order, creating metabolic pathways (e.g., the citric acid cycle, a series of enzymatic reactions in the cells of aerobic organisms, important in cellular respiration). In a metabolic pathway, one enzyme takes the product of another enzyme as a substrate. After the catalytic reaction, the product is then passed on to another enzyme. The end product(s) of such a pathway are often non-competitive inhibitors (Fig. 5) for one of the first enzymes of the pathway (usually the first irreversible step, called *committed step*), thus regulating the amount of end product made by the pathway (Fig. 6).
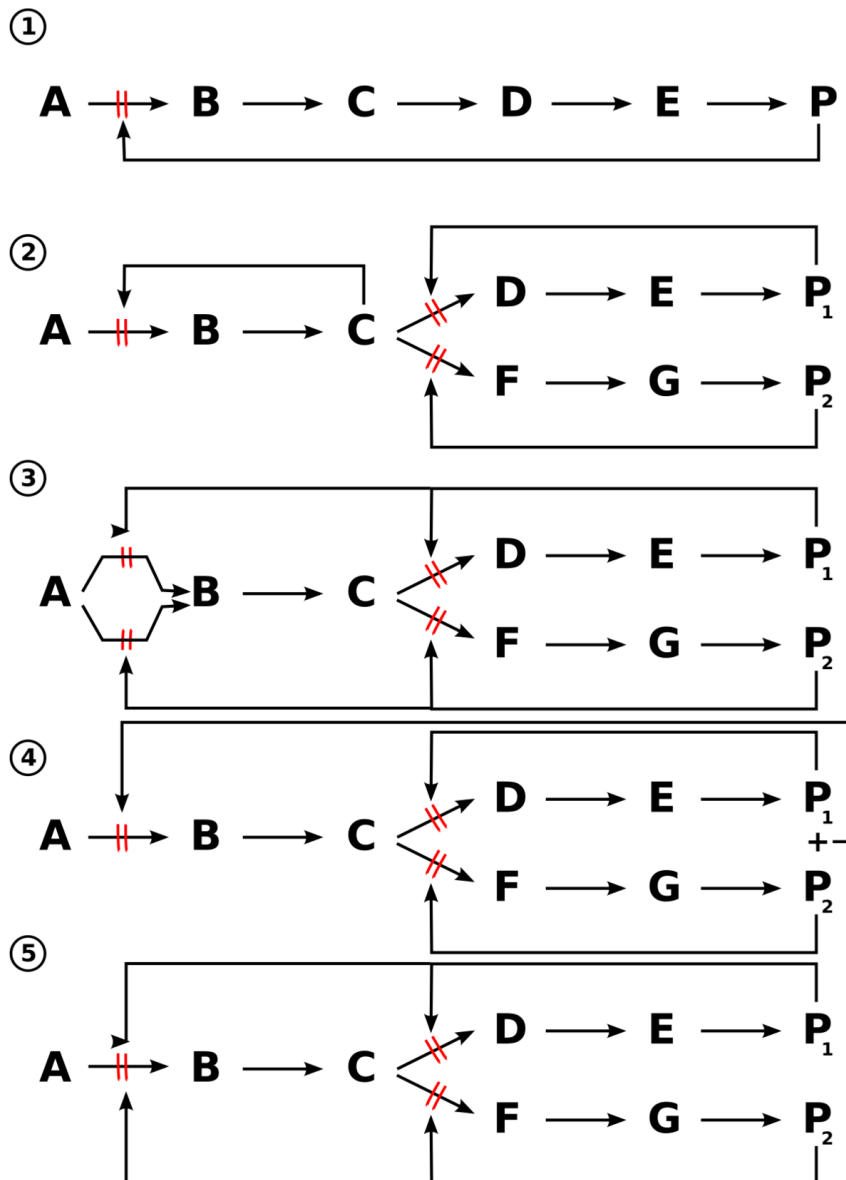
Figure 7   Figure 6: Common feedback inhibition mechanisms.

1. The basic feedback inhibition mechanism, where the product (P) inhibits the committed step (A⇢B).

2. *Sequential feedback inhibition.* The end products P_1 and P_2 inhibit the first committed step of their individual pathway (C⇢D or C⇢F). If both products are present in abundance, all pathways from C are blocked. This leads to a buildup of C, which in turn inhibits the first common committed step A⇢B.

3. *Enzyme multiplicity.* Each end product inhibits both the first individual committed step and one of the enzymes performing the first common committed step.

4. *Concerted feedback inhibition.* Each end product inhibits the first individual committed step. *Together*, they inhibit the first common committed step.
5. *Cumulative feedback inhibition.* Each end product inhibits the first individual committed step. Also, each end product *partially* inhibits the first common committed step.

Enzymes are essential to living organisms, and a malfunction *of even a single enzyme* out of approximately 2,000 present in our bodies can lead to severe or lethal illness. An example of a disease caused by an enzyme malfunction in humans is phenylketonuria (PKU). The enzyme phenylalanine hydroxylase, which usually converts the essential amino acid phenylalanine into tyrosine does not work, resulting in a buildup of phenylalanine that leads to mental retardation. Enzymes in the human body can also be influenced by inhibitors in good or bad ways. Aspirin, for example, inhibits an enzyme that produces prostaglandins (inflammation messengers), thus suppressing pain. But not all enzymes are in living things. Enzymes are also used in everyday products such as biological washing detergents where they speed up chemical reactions, (to get your clothes clean).

2.7 Digestive and Metabolic Enzymes

In the previous section we have been talking about the digestive enzymes, both the ones produced by the body, such as salivary amylase, and the food enzymes. Their primary role is for the digestion of food. Another class of enzymes is called metabolic enzymes. Their role is to catalyze chemical reactions involving every process in the body, including the absorption of oxygen. Our cells would literally starve for oxygen even with an abundance of oxygen without the action of the enzyme, cytochrome oxidase. Enzymes are also necessary for muscle contraction and relaxation. The fact is, without both of these classes of enzymes, (digestive and metabolic,) life could not exist. Digestive enzymes function as biological catalysts in which it helps to breakdown carbohydrates, proteins, and fats. On the other hand, metabolic enzymes function as a remodel of cells. Digestion of food has a high priority and demand for enzymes; digestive enzymes get priority over metabolic enzymes. Any deficiency in metabolic enzyme can lead to over work, which could lead to enlarge organs in order to perform the increased workload. The result is unhealthy and could cause enlarged heart or pancreas. The deficiencies of metabolic enzymes can have a tremendous impact on health. As we grow older enzyme level decline and the efficiency in the body decline.

2.8 Enzyme naming conventions

By common convention, an enzyme's name consists of a description of what it does, with the word ending in "-ase". Examples are alcohol dehydrogenase and DNA polymerase. Kinases are enzymes that transfer phosphate groups. The International Union of Biochemistry and Molecular Biology has developed a nomenclature for enzymes, the EC numbers; each enzyme is described by a sequence of four numbers, preceded by "EC". The first number broadly classifies the enzyme based on its mechanism:

- EC 1 *Oxidoreductases*: catalyze oxidation/reduction reactions
- EC 2 *Transferases*: transfer a functional group (e.g., a methyl or phosphate group)

- EC 3 *Hydrolases*: catalyze the hydrolysis of various bonds
- EC 4 *Lyases*: cleave various bonds by means other than hydrolysis and oxidation
- EC 5 *Isomerases*: catalyze isomerization changes within a single molecule
- EC 6 *Ligases*: join two molecules with covalent bonds

Some other important enzymes are: Protease: breaks the protein into amino acids in high acidity environments such as stomach,pancreatic and intestinal juices. Act on bacteria, viruses and some cancerous cells. Amylase: Break complex carbohydrates such as starch into simpler sugars (dextrin and maltose). It found in the intestines, pancreas and also in salivary glands. Lipase: breaks down fats and some fat soluble vitamins (A,E,K, and D). helpful in treating cardiovascular diseases. Cellulase: break down cellulose that found in fruits, grains, and vegetables. It increases the nutritional values of vegetables, and fruits. Pectinase: break down pectin that found in citrus fruits, carrots, beets, tomatoes, and apple. Antioxidants: protect from free radical negative effect that can damage cell in the body. Cathepsin: break animal protein down. Lactase: break down lactose that found in milk products. the production of lactase decrease with age. Invertase: assimilate sucrose that can contribute to digestive stress if not digested properly. Papain: break down protein and help the body in digestion. Bromelain: Break proteins that found in plants and animals. it could help the body to fight cancer and treat inflammation. Glucoamylase: break down maltose that found in all grains in to two glucose molecules.

2.9 Metabolism and energy

Biochemistry[3] << Catalysis[4] | pKa values >>[5]

2.10 Metabolism

2.10.1 Anabolism and catabolism

Metabolism (Fig. 1) is, broadly speaking, the conversion of food into energy, cell components, and waste products.

3   http://en.wikibooks.org/wiki/Biochemistry
4   http://en.wikibooks.org/wiki/Biochemistry/Catalysis
5   http://en.wikibooks.org/wiki/Biochemistry/pKa_values
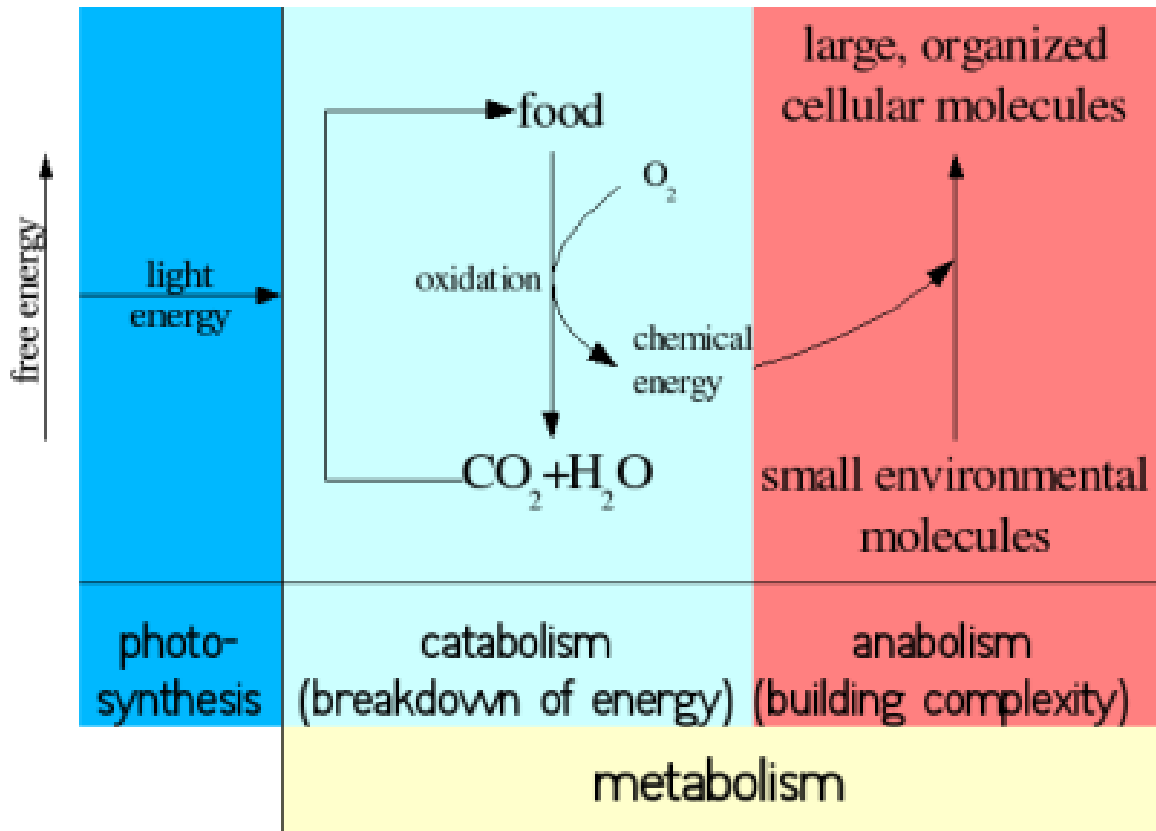
Figure 8

Figure 1: Overview of metabolism The above diagram shows the different parts of metabolism:

- *energy source*, which is, after all, the sun, whose energy is harvested through photosynthesis
- *catabolism*, the breakdown of food into chemical energy, which is needed in
- *anabolism*, the construction of complex cell molecules from small environmental molecules, utilizing chemical energy

Catabolic reactions release energy and are therefore `exergonic`, while anabolic reactions use up energy and are therefore `endergonic`.

2.10.2 High-energy phosphates

Due to the large variety of food compounds, and the large number of biochemical reactions which need energy in anabolism, it would be quite inefficient to couple a specific anabolic reaction to a specific energy source in catabolism. Instead, the cell uses an intermediate compound, a kind of universal energy currency. This intermediate is called *high-energy phosphate*. But when is a phosphate group called "high-energy", and how does it differ from a "low-energy" phosphate? A giveaway is the $\Delta G^{0'}$ of hydrolysis. Hydrolysis separates a phosphate from a compound by adding water:

O   O

```
R-OP-OH + H₂O  Ì  R-OH + HO-P-OH
   O                        O
```
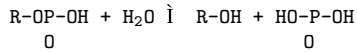
The $\Delta G^{0'}$ of a low-energy (or "inorganic") phosphate group (called P_i) is 9-20 kJ mol^{-1}, while the $\Delta G^{0'}$ of a high-energy phosphate (denoted Å) is ~30 kJ mol^{-1}.

2.10.3 pKa value

Now what makes this Å so special? To explain this, we must take a little excourse into pH and pKa values. A phosphate group has between zero and three `OH` groups. This allows Å to exist in up to four different forms (0, 1, 2, and 3 `OH` groups, Fig. 2), depending on the pH value of the surrounding solution. A pKa value gives us the pH value at which 50% of the molecules are in one form (e.g., 1 `OH` group) and another (e.g., 2 `OH` groups). This is expressed by the *Henderson-Hasselbalch equation* :

$$pH = pK_a + log\frac{[base]}{[acid]}$$
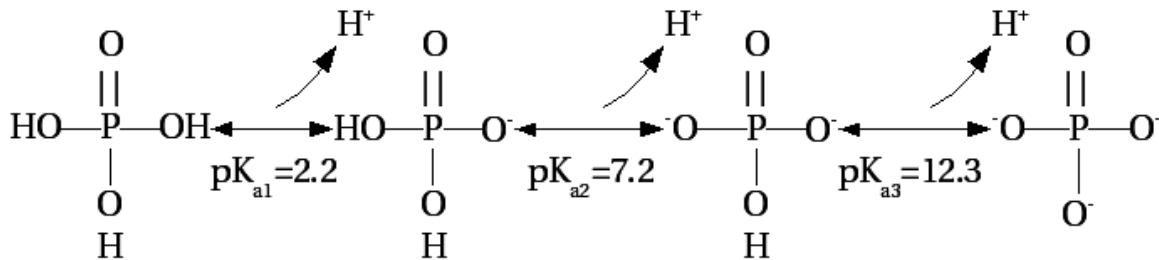
Figure 9

Figure 2: The four possible forms of a phosphate group. pK_{a2} represents the conditions in the cell. Now to the promised difference between Å and PP_i. The breaking of the ester bond of an ROÅ releases more energy than the breaking of a PP_i bond (Fig. 3), because of

1. *electrostatic repulsion* between the two phosphate groups in PP_i
2. *resonance stabilization* of two P_i groups, compared to PP_i (Fig. 4)
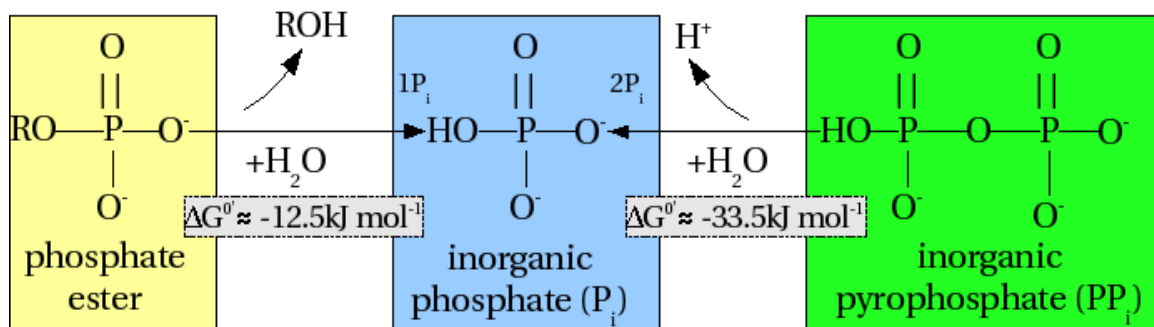
Figure 10

Figure 3: Hydrolysis of Å and PP_i.
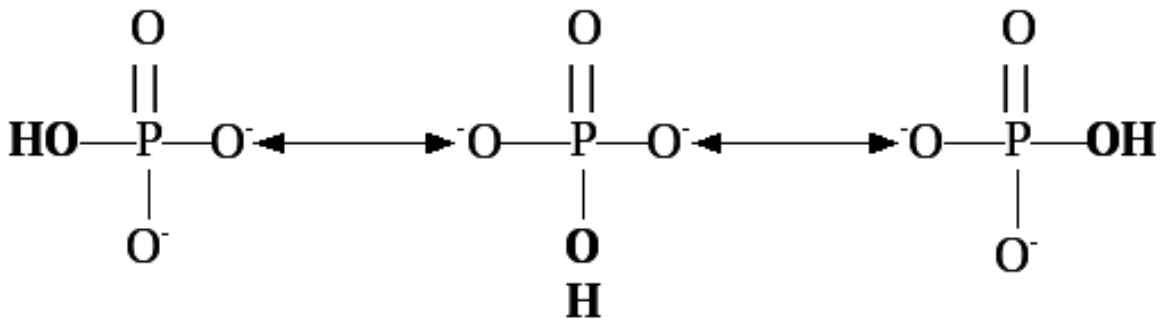
Figure 11

Figure 4: Resonance stabilization of P_i. Resonance stabilization means that both `OH` and `=O` can "travel" around the phosphate. Of course, this is a crude analogy; they do not really move, the electrons are just "smeared" around the phosphate atom. This is also indicated by the use of the $\leftrightarrow$ arrow, instead of $\rightleftharpoons$; the three forms do not exist, they are just a way of writing down the chemical reality. As you can see in Fig. 3, the $\Delta G^{0'}$ value for $PP_i \rightleftharpoons 2P_i$ is $\ll 0$, shifting the reaction strongly in favor of the $2P_i$.

2.11 Molecules using high-energy phosphates

2.11.1 Anhydride between phosphoric acid and carboxyl group

Hydrolysis : $\Delta G^{0'} = $ -49.3 kJ mol^{-1}
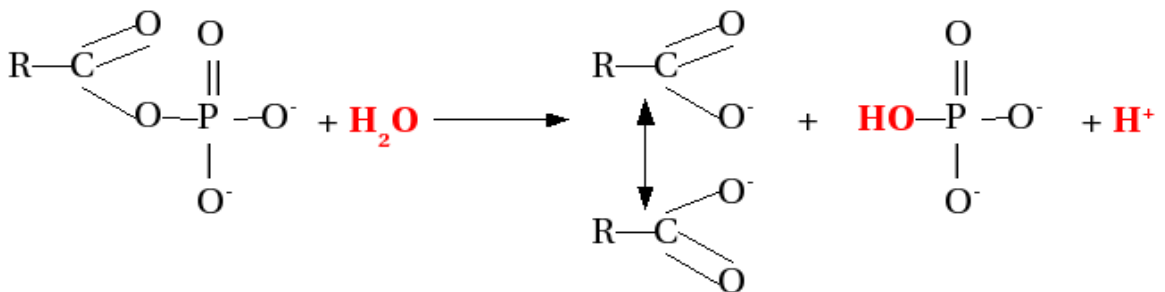
Figure 12

2.11.2 Guanidine phosphate

Hydrolysis : $\Delta G^{0'} = $ -43.0 kJ mol^{-1}
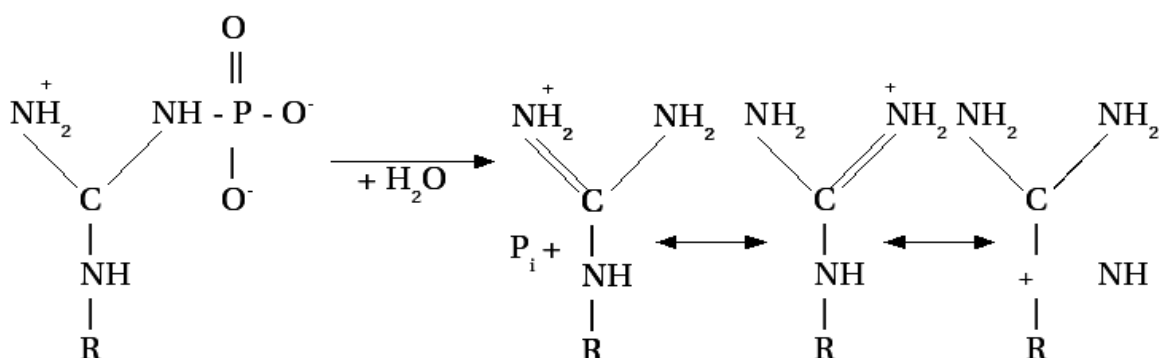
Figure 13

2.11.3 Enol phosphate

For example, this picture is wrong, the carbon has 5 bonds to it after it tautomerizes, which is impossible and a rookie organic chemistry error. please fix phosphoenolpyruvate

Hydrolysis : $\Delta G^{0'}$ = -61.9 kJ mol^{-1}
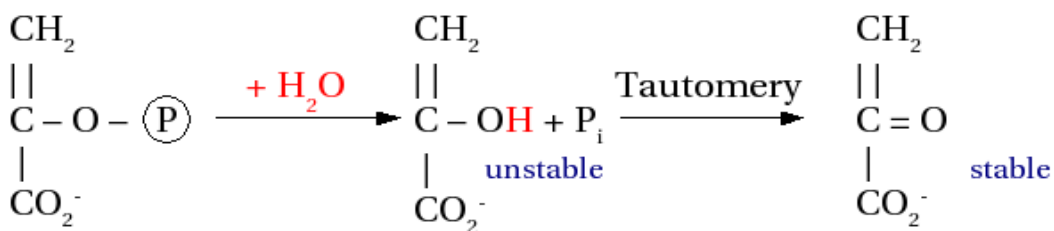
Figure 14

2.11.4 ATP

Adenosine triphosphate contains one low-energy and two high-energy phosphate bonds:
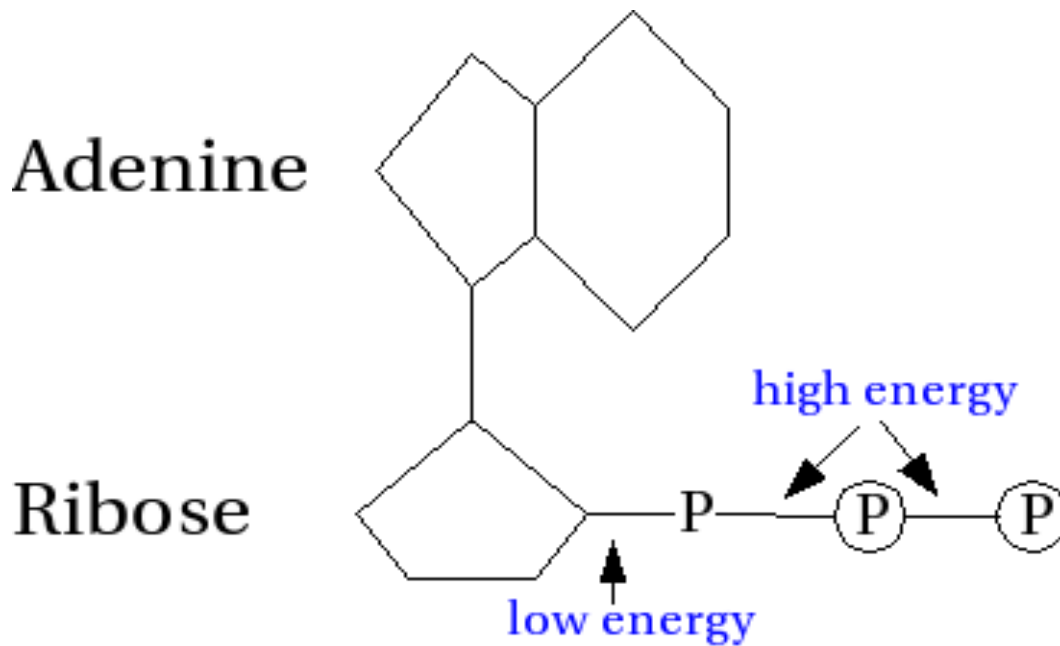
Figure 15

Low energy : $\Delta G^{0'}$ = -14,2 kJ mol^{-1}

High energy : $\Delta G^{0'}$ = -30.5 kJ mol^{-1}

- ATP is regenerated from ADP (adenosine diphosphate), P_i and energy (from food); H_2O is released in the process.
- ATP is the short-term energy "currency" of the cell.
- ATP concentration in the cell is low (ATP: 2-8mM; ADP:0,2-0,8mM). ATP is generated in high "turn over".
- ATP performs its chemical work through *coupled reactions*.
- Coupled reactions are always Å *transfers*, never direct hydrolysis

Basically, any ATP-driven reaction is reversible, building ATP from ADP and P_i in the process. However, some ATP-driven reactions should never be reversed; these include nucleotide and protein synthesis. If these were reversed, the organism would disassemble its own DNA and proteins for energy, a rather unfortunate strategy. For reactions that should never be reversed, ATP can be broken down into AMP (adenosine monophosphate) and PP_i, which in turn becomes $2\times P_i$. This reaction has a $\Delta G^{0'}$ of -65,7 kJ mol^{-1}, which is totally irreversible under *in vivo* conditions. It should be noted that AMP can not directly be converted to ATP again. Instead, the enzyme *AMP kinase* forms two ADP molecules from one ATP and one AMP. The resulting ADPs are then treated as described above.

2.12 Non-covalent bonds

The destruction of covalent bonds takes up huge amounts of energy. The breakdown of an O_2 molecule into two oxygen atoms needs ~460 kJ mol^{-1}. Thus, nowhere in "living" biochemistry are covalent bonds actually destroyed; if one is broken, another one is created. This is where non-covalent bonds come in, they are weak enough to be broken down easily, and to form "bonds" again. For this reason, many biochemical functions are using so-called weak/secondary/non-covalent bonds. Weak bonds are created and destroyed much more easily than covalent ones. The typical range of energy needed to destroy such a weak bond is 4-30 kJ mol^{-1}. Thus, the formation of weak bonds is energetically favorable, but these bonds are also easily broken by kinetic (thermal) energy (the normal movement of molecules). Biochemical interactions are often temporary (e.g., a substrate has to leave an enzyme quickly after being processed), for which the weakness of these bonds is essential. Also, biochemical specificity (e.g., enzyme-substrate-recognition) is achieved through weak bonds, utilizing two of their major properties:

- Since individual weak bonds are, well, weak, several of them have to occur in a specific pattern at the same time in roughly the same place.
- The short range of weak bonds.

The link that follows demonstrates the type of non-covalent forces: [1][6] There are three basic types of weak bonds, and a fourth "pseudo-bond":

2.12.1 Ionic bonds

Ionic bonds are electrostatic attractions between permanently charged groups. Ionic bonds are not directed. Example:

 X-CO$_2$$^-$ ⋯⋯ H$_3$$^+$N-Y
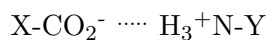
~ 20 kJ mol^{-1}

2.12.2 Hydrogen bonds

Hydrogen bonds are also established by electrostatic attraction. These attractions do not occur between permanently charged groups, but rather between atoms temporarily charged by a *dipole moment*, resulting from the different electronegativity[7] of atoms within a group. Hydrogen bonds are even weaker than ionic bonds, and they are highly directional, usually along a straight line. Besides being weaker than ionic bonds, hydrogen bonds are also weaker, and longer than similar covalent bonds. Hydrogen bonds are unique because they only exist when the Hydrogen is bonded to an oxygen (O), Nitrogen (N), or Fluorine (F), but the most common hydrogen bonds in biochemistry are:

---

6   `http://www.pearsonhighered.com/mathews/ch02/fi2p2.htm`
7   `http://en.wikibooks.org/w/index.php?title=Electronegativity&action=edit&`
     `redlink=1`

X-OH ⋯⋯ O-Y

X-OH ⋯⋯ N-Y

X-NH ⋯⋯ O-Y

X-NH ⋯⋯ N-Y
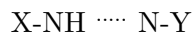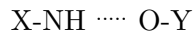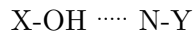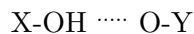
Hydrogen bonds equal an energy between 12-29 kJ mol, whereas covalent bonds are much higher. For example, the covalent bond between oxygen and hydrogen is about 492 KJ mol-1^{-1}.

2.13 *Hydrogen Bonds and Water*

Water has unique properties, after all, it is chosen to be the universal solvent. The unique properties of water are due to hydrogen bonding between all the oxygens and hydrogens of the content. The hydrogen bonds occurring in water are about 2 angstroms apart from each other. Although hydrogen bonding is only about 5% as strong as covalent bond, they still cause water to have a high boiling point, and a high surface tension. The following link will take you to the structure of water and its Hydrogen Bonding: [[2][8]]

2.13.1 Van der Waals attractions

Van der Waals attractions are established between electron density-induced dipoles. They form when the outer electron shells of two atoms *almost* (but not quite) touch. The distance of the atoms is very important for these weak interactions. If the atoms are too far apart, the interactions are too weak to establish; if the atoms are too close to each other, their electron shells will repel each other. Van der Waals attractions are highly unspecific; they can occur between virtually any two atoms. Their energy is between 4-8 kJ mol^{-1}. [[File:Example.jpg]]

2.13.2 Hydrophobic forces

Hydrophobic forces are not actually bonds, so this list has four items, but still just three bond types. In a way hydrophobic forces are the negation of the hydrogen bonds of a polar solute, usually water, enclosing a nonpolar molecule. For a polar solute like water, it is energetically unfavorable to "waste" a possible hydrogen bond by exposing it towards a nonpolar molecule. Thus, water will arrange itself around any nonpolar molecule in such a way that no hydrogen bonds point towards that molecule. This results in a higher order, compared to "freely" moving water, which leads to a lower entropy level and is thus energetically unfavorable. If there is more than one nonpolar molecule in the solute, it is favorable for the nonpolar molecules to aggregate in one place, reducing their surrounding, ordered "shell" of water to a minimal surface. Also, in large molecules, such as proteins, the hydrophobic (nonpolar)

---

8   `http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/H/Hbonds_water.gif`

parts of the molecule will tend to turn towards the inside, while the polar parts will tend to turn towards the surface of the molecule.

2.13.3 References

Cooke, Rosa-lee. *Properties of Water.* Lecture 10. Mountain Empire Community College. n.d. Web. [9] Kimball, John W.. *Hydrogen Bonds.* Kimball's Biology Pages. Feb. 12, 2011. Web. [10] Lower, Stephen. *States of matter: Water and hydrogen bonding.* General Chemistry Virtual Textbooks. 2009. Aug. 26, 2010. Web. [11] n.p. *Covalent vs. Non-Covalent Bonds.* n.d. [12] W. W. Norton & Company. *Hydrogen Bonding in Water.* Web. 2012. [13] WyzAnt Tutoring. *WyzAnt Tutoring.* Bonds. 2012. Web. [14]

<< Catalysis[15] | pKa values >>[16] Biochemistry[17]

2.14 pKa values

Biochemistry[18] << Metabolism and energy[19]

It is difficult to discuss the subject of biochemistry without a firm foundation in general chemistry and organic chemistry, but if one doesn't remember the concept of the Acid Dissociation Constant (pK_a) from Organic Chemistry[20], one can read up on the topic below.

Buffers are essential to biochemical reactions, as they provide a (more or less) stable pH value for reactions to take place under constantly changing circumstances. The pH value in living cells tends to fall between 7.2 and 7.4, and this pH level is generally maintained by weak acids. (The pH values in lysosomes and peroxisomes differ from this value, as do the pH measurements of the stomach and other organs found in various types of plants and animals.)

An *acid* is here defined simply as any molecule that can release a proton (H^+) into a solution. Stronger acids are more likely to release a proton, due to their atomic and molecular properties. The tendency of an acid to release a proton is called the *dissociation constant* (K_a) of that substance, with

9   http://water.me.vccs.edu/courses/env211/lesson10_print.htm
10   http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/H/Hbonds_water.gif
11   http://www.chem1.com/acad/webtext/states/water.html
12   http://www.pearsonhighered.com/mathews/ch02/c02cv.htm
13   http://www.wwnorton.com/college/chemistry/gilbert2/tutorials/chapter_10/water_h_bond/
14   http://www.wyzant.com/Help/Science/Chemistry/Bonds/
15   http://en.wikibooks.org/wiki/Biochemistry/Catalysis
16   http://en.wikibooks.org/wiki/Biochemistry/pKa_values
17   http://en.wikibooks.org/wiki/Biochemistry
18   http://en.wikibooks.org/wiki/Biochemistry
19   http://en.wikibooks.org/wiki/Biochemistry/Metabolism_and_energy
20   http://en.wikibooks.org/w/index.php?title=Organic_ChemistryFoundational_concepts_of_organic_chemistry/Acids_and_bases&action=edit&redlink=1

$$K_a = \frac{[H^+][A^-]}{[HA]}$$

for HA <-> H$^+$ + A$^-$.

A larger K_a value means a greater tendency to dissociate a proton, and thus it means the substance is a stronger acid. The pH at which 50% of the protons are dissociated can therefore be calculated as:

pK_a = -log (K_a)

This equation is known as the Henderson-Hasselbalch equation.

$$pH = pK_a + log\frac{[base]}{[acid]}$$

The Henderson-Hasselbalch equation is derived from the adjacent K_a expression. By taking the logarithm of base ten to both sides, the next part of the equation is obtained. Using the logarithmic property of multiplication, the [H$^+$] breaks from the expression. Since log K_a is equal to -pK_a and log [H$^+$] is equal to -pH, they are then substituted. To obtain what is known as the Henderson-Hasselbalch equation, -pK_a and -pH are subtracted from their respective sides to yield a positive equation.

The Henderson-Hasselbalch equation interestingly enough predicts the behavior of buffer solutions. A solution of 1 M ethanoic (acetic) acid [HA] and 1 M sodium ethanoate [A$^-$] will have a pH equal to the pK_a of ethanoic acid: 4.76.

If we added acid to pure water up to a concentration of 0.1 M, the pH would become 1. If we add the same concentration of acid to the buffer solution, it will react with the ethanoate to form ethanoic acid. The ethanoate concentration would drop to 0.9 M and the ethanoic acid concentration should rise to 1.1 M. The pH becomes 4.67 - very different from the pH=1 *without* a buffer.

Similarly, adding 0.1 M alkali changes the pH of the buffer to 4.85, instead of pH=13 without buffer. Due to the amphipathic nature of amino acids - which are the monomer building blocks of all proteins, physiological conditions are always considered to be buffered, which plays a major role in the conformations and reactivities of substrates in the cell's liquid interior, its cytosol.

A very small (which would include a large negative value) pK_a indicates a very strong acid. A pK_a value between 4 and 5 is the most common range for organic acid compounds.

<< Metabolism and energy[21] Biochemistry[22]

---

21   http://en.wikibooks.org/wiki/Biochemistry/Metabolism_and_energy
22   http://en.wikibooks.org/wiki/Biochemistry

3 Proteins

Proteins are a primary constituent of living things and one of the chief classes of molecules studied in biochemistry. Proteins provide most of the molecular machinery of cells. Many are enzymes or subunits of enzymes. Other proteins play structural or mechanical roles, such as those that form the struts and joints of the cytoskeleton. Each protein is linear polymers built of amino acid. Proteins are also nutrient sources for organisms that do not produce their own energy from sunlight and/or are unable to fix nitrogen. Proteins can interact with one another and with others molecules to form complexes.

Index of chapters and main sections

1. Introduction[1]

Protein role in biology[2]

Proteins as polymers of amino acids[3]

Protein function depends on structure[4]

Protein synthesis[5]

The chemistry of proteins[6]
   Amino acid structure and chemistry[7] Disulfide bonds[8] Post-translational modifications[9]

2. Protein structure and folding[10]

1   http://en.wikibooks.org/wiki/Biochemistry/Proteins/Introduction
2   http://en.wikibooks.org/wiki/Biochemistry/Proteins/Introduction#Protein_role_in_
     biology
3   http://en.wikibooks.org/wiki/Biochemistry/Proteins/Introduction#Proteins_as_polymers_
     of_aminoacids
4   http://en.wikibooks.org/wiki/Biochemistry/Proteins/Introduction#Protein_function_
     depends_on_structure
5   http://en.wikibooks.org/wiki/Biochemistry/Proteins/Introduction#Protein_synthesis
6   http://en.wikibooks.org/wiki/Biochemistry/Proteins/The_chemistry_of_proteins
7   http://en.wikibooks.org/wiki/Biochemistry/Proteins/The_chemistry_of_proteins#
     Aminoacid_structure_and_chemistry
8   http://en.wikibooks.org/wiki/Biochemistry/Proteins/The_chemistry_of_proteins#
     Disulfide_bonds
9   http://en.wikibooks.org/wiki/Biochemistry/Proteins/The_chemistry_of_proteins#
     Post-translational_modifications
10   http://en.wikibooks.org/wiki/Biochemistry/Proteins/Protein_structure_and_folding

What is protein folding?[11] Protein structural levels[12] Secondary structure elements[13]
Tertiary structure elements[14] Common folds and modules[15]

3. Types of Protein[16]
Oxygen Binding Proteins[17]

3.1 Introduction

3.2 Protein role and importance

Proteins are among the fundamental molecules of biology. They are common to all life present on Earth today, and are responsible for most of the complex functions that make life possible. They are also the major structural constituent of living beings. According to the Central Dogma[18] of Molecular Biology (proposed by Francis Crick in 1958), information is transferred from DNA to RNA to proteins. DNA functions as a storage medium for the information necessary to synthesize proteins, and RNA is responsible for (among other things) the translation of this information into protein molecules, as part of the ribosome. Virtually all the complex chemical functions of the living cell are performed by protein-based catalysts called enzymes. Specifically, enzymes either make or break chemical bonds. Protein enzymes should not be confused with RNA-based enzymes (also called ribozymes), a group of macromolecules that perform functions similar to protein enzymes. Further, most of the scaffolding that holds cells and organelles together is made of proteins. In addition to their catalytic functions, proteins can transmit and commute signals from the extracellular environment, duplicate genetic information, assist in transforming the energy in light and chemicals with astonishing efficiency, convert chemical energy into mechanical work, and carry molecules between cell compartments.

3.3 Functions not performed by proteins

Proteins do so much that it's important to note what proteins **don't** do. Currently there are no known proteins that can directly replicate themselves. Prions are no exception to this rule. It is theorized that prions may be able to act as a structural template for other chemically (but not structurally) identical proteins, but they can't function as a native

---

11 http://en.wikibooks.org/wiki/Biochemistry/Proteins/Protein_structure_and_folding#What_is_protein_folding.3F
12 http://en.wikibooks.org/wiki/Biochemistry/Proteins/Protein_structure_and_folding#Protein_structural_levels
13 http://en.wikibooks.org/wiki/Biochemistry/Proteins/Protein_structure_and_folding#Secondary_structure_elements
14 http://en.wikibooks.org/wiki/Biochemistry/Proteins/Protein_structure_and_folding#Tertiary_structure_elements
15 http://en.wikibooks.org/wiki/Biochemistry/Proteins/Protein_structure_and_folding#Common_folds_and_modules
16 http://en.wikibooks.org/wiki/Biochemistry/Proteins/Types_of_Protein
17 http://en.wikibooks.org/wiki/Biochemistry/Proteins/Types_of_Protein#Oxygen_Binding_Proteins
18 http://en.wikibooks.org/w/index.php?title=Biochemistry/Print_version/Central_Dogma&action=edit&redlink=1

template for protein synthesis *de novo*. Proteins don't act as fundamental energy reserves in most organisms, as their metabolism is slower and inefficient compared to sugars or lipids. They are, on the other hand, a fundamental nitrogen and amino acid reserve for many organisms. Proteins do not directly function as a membrane in most organisms, except viruses; however, they are often important components of these structures, lending both stability and structural support.

4 Proteins as polymers of aminoacids

4.1 Composition and Features

Proteins are composed of a linear (not branched and not forming rings) polymer of amino acids. The twenty genetically encoded amino acids are molecules that share a central core: The α-carbon is bonded to a primary amino (-NH$_2$) terminus, a carboxylic acid (-COOH) terminus, a hydrogen atom, and the amino acid side chain, also called the "R-group". The R-group determines the identity of the amino acid. In an aqueous solution, at physiological pH (~6.8), the amino group will be in the protonated -NH$_3^+$ form, and the carboxylic acid will be in the deprotonated -COO$^-$ form, forming a zwitterion. Most amino acids that make up proteins are L-isomers, although a few exotic creatures use D-isomers in their proteins. It is important to note that levorotatory (L) and dextrorotatory (D) are not specific to rectus (R) and sinister (S) configurations. A levorotatory form of a protein can be either R or S configuration. Levorotatory and dextrorotatory refer to how the proteins bend light in a polarimeter.
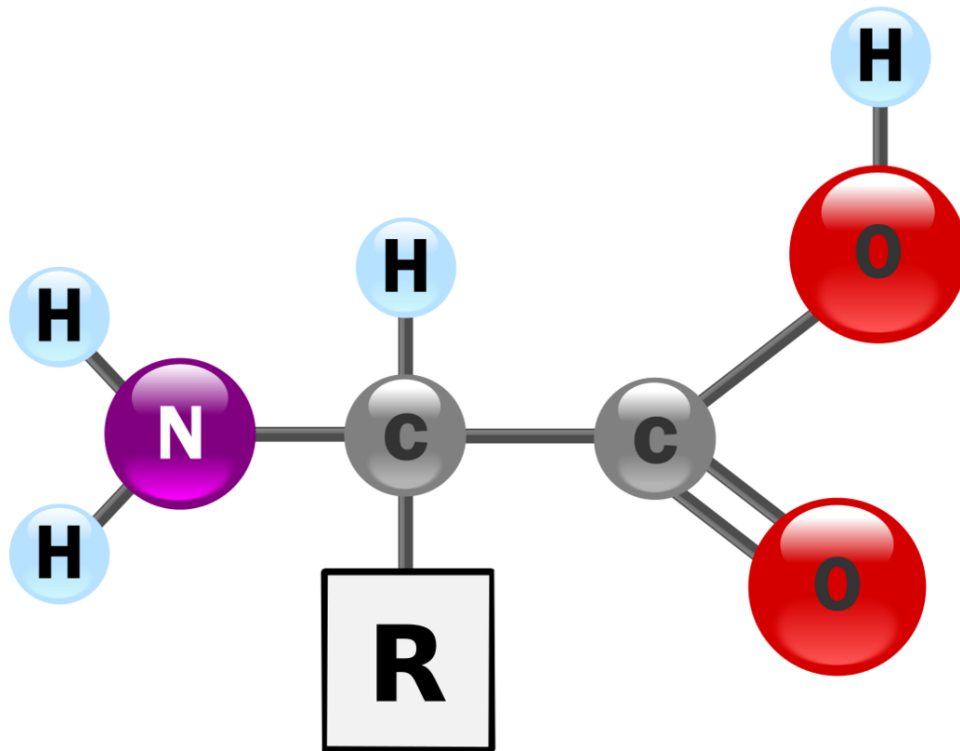
Figure 16   The structure of an amino acid.

Amino acids polymerize via peptide bonds, which is a type of amide bond. A peptide bond is formed upon the dehydration of the carboxy-terminus of one amino acid with the amine terminus of a second amino acid. The resulting carbonyl group's carbon atom is directly bound to the nitrogen atom of a secondary amine. A peptide chain will have an unbound amino group free at one end (called the N-terminus) and a single free carboxylate group at the other end (called the C-terminus). The written list of the amino acids linked together in a protein, in order, is called its primary structure. By convention, peptide sequences are written from N-terminus to C-terminus. This convention mimics the way polypeptides are synthesized by the ribosome in the cell. Small polymers of less than 20 amino acids long are more often called peptides or polypeptides. Proteins can have sequences as short as 20-30 amino acids to gigantic molecules of more than 3,000 aminoacids (like Titin,a human muscle protein).

4.2 Genetically-Encoded Amino Acids

While there are theoretically billions of possible amino acids, most proteins are formed of only 20 amino acids: the genetically-encoded (or more precisely, *proteogenic*) amino acids. Note that all amino acids except glycine have a chiral center at their α-carbons. (Glycine has two hydrogens on its α-carbon, and therefore it is achiral.) Besides glycine, all proteogenic amino acids are L-amino acids, meaning they have the same absolute configuration as L-glyceraldehyde. This is the same as the S-configuration, with the exception of cysteine, which contains a sulfur atom in its side chain, and so the naming priority changes. D-amino acids are sometimes found in nature, as in the cell walls of certain bacteria, but they are rarely incorporated into protein chains. The side-chains of proteogenic amino acids are quite varied: they range from a single hydrogen atom (as for glycine, the simplest amino acid) to the indole heterocycle, as found in tryptophan. There are polar, charged and hydrophobic amino acids. The chemical richness of amino acids is at the base of the complexity and versatility of proteins.

4.3 Post-Translational Modification

There are two major types of post-translational modifications to protein: those that cleave the bonds of the peptide backbone and those that add or remove functional groups to the sidechains of individual amino acids. In the first type of post-translational modification, specialized enzymes called proteases recognize specific amino acids of a protein and break the associated peptide bond, thereby irreversibly modifying the primary structure. In the second major type of modification, the amino acid side chains of a given protein are chemically modified by enzymatic reactions or are spontaneously formed (non-enzymatic). Examples of sidechain modifications are quite numerous, but common ones include oxidation, acylation, glycosylation (addition of a glycan, or sugar), methylation, and phosphorylation. Both types of post-translational modifications are capable of exerting positive and negative control over a given protein or enzyme.

4.4 The importance of protein structure

Generally speaking, the function of a protein is completely determined by its structure. Molecules like DNA, which perform a fairly small set of functions, have an almost fixed structure that's fairly independent from sequence. By contrast, protein molecules perform functions as different as digesting sugars or moving muscles. To perform so many different functions, proteins come in many different structures. The protein function is almost completely dependent on protein structure. Enzymes must recognize and react with their substrates with precise positioning of critical chemical groups in the three-dimensional space. Scaffold proteins must be able to precisely dock other proteins or components and position them in space in the correct fashion. Structural proteins like Collagen[1] must face mechanical stresses and be able to build a regular matrix where cells can adhere and proliferate. Motor proteins must reversibly convert chemical energy in movement, in a precise fashion.

4.5 Protein folding depends on sequence

As Anfinsen demonstrated in the 1960's, proteins acquire their structure by spontaneous folding of the polypeptide chain into the minimal energy configuration. Most proteins require no external factors in order to fold (although specialized protein exist in cells, called chaperones, that help other, misfolded, proteins acquire their correct structure) — the protein sequence itself uniquely determines the structure. Often the whole process takes place in milliseconds. Despite the apparent chemical simplicity of proteins, the vast number of permutations of twenty amino acids in a linear sequence leads to an amazing number of different protein folds. Nevertheless, protein structures share several characteristics in common: they are almost all built of a few secondary structure elements (short-range structural patterns that are recurrent in protein structures) and even the way these elements combine is often repeated in common motifs. Nonetheless, it is still impossible to know what structure a given protein sequence will yield in solution. This is known as the protein folding problem, and it is one of the most important open problems in modern molecular biology.

4.6 Protein denaturation

Proteins can lose their structure if put in unsuitable chemical (e.g. high or low pH; high salt concentrations; hydrophobic environment) or physical (e.g. high temperature; high pressure) conditions. This process is call denaturation. Denatured proteins have no defined structure and, especially if concentrated, tend to aggregate into insoluble masses. Protein denaturation is by no means an exotic event: a boiled egg becomes solid just because of denaturation and subsequent aggregation of its proteins. Denatured proteins can sometimes refold when put again in the correct environment, but sometimes the process is irreversible (especially after aggregation: the boiled egg is again an example). It is finally the proteins which are responsible for susceptibility or resistance to a pathogen or parasite.

---

1   http://en.wikibooks.org//en.wikipedia.org/wiki/Collagen

4.7 Proteins can fold into domains

A significant number of proteins, especially large proteins, have a structure divided into several independent domains. These domains can often perform specific functions in a protein. For example, a cell membrane receptor might have an extracellular domain to bind a target molecule and an intracellular domain that binds other proteins inside the cell, thereby transducing a signal across the cell membrane. The domain of a protein is determined by the secondary structure of a protein there are four main types of domain structures: alpha-helix, beta-sheet, beta-turns, and random coil. The alpha-helix is when the poly-peptide chain forms a helix shape with the amino acids side chains sticking out, usually about 10 amino acids long. The alpha-helix gets its strength by forming internal hydrogen bonds, that occur between amino acid 1 and 4 along the length of the helix. A high concentration of Glycine's in a row tend towards the alpha-helix conformation. The beta-sheet structure is composed of poly-peptide chains stacking forming hydrogen bonds between the sheets. You can from parallel sheets by stacking in the same direction N-C terminal on top of N-C terminal or form anti-parallel by stacking in opposite directions N-C terminal on top of C-N terminal. Beta-turns link two anti-parallel beta strands by a 4 amino acid loop in a defined conformation. A random coil is a portion of the protein that has no defined secondary structure. Domains of a protein then come from unique portions of the peptide that are made up of these types of secondary structure.

4.8 The chemistry of proteins

4.9 Amino acid structure and chemistry

4.9.1 General features

Amino acids consist of a primary amine bound to an aliphatic carbon atom (the so-called α-carbon), which in turn is bound to a carboxylic acid group. At least one hydrogen atom is bound to the α-carbon; in addition, the α-carbon bears a side chain, which is different for different amino acids. In a neutral aqueous solution, amino acids exist in two forms. A very small fraction of amino acid molecules will be neutral, with a deprotonated amino group and a protonated carboxylic acid group. However, the overwhelming majority of molecules will be in a *Zwitterion[2]* tautomer[3], with a positive charge on the (protonated) amino group and a negative charge on the (deprotonated) carboxylate group. Amino acids are linked via a Peptide bond[4]. (From an organic chemistry perspective, a peptide bond is a type of amide[5] group.) A peptide bond consists of a carbonyl group's carbon atom directly bound to the nitrogen atom of a secondary amine. A peptide chain will have an unbound amino group free at one end (called the N-terminus) and a single free carboxylate group at the other end (called the C-terminus). The peptide bond is planar, because resonance between the carbonyl group and the amino nitrogen lends the C-N bond a partial double-bond character.

---

2   `http://en.wikibooks.org//en.wikipedia.org/wiki/Zwitterion`

3   `http://en.wikibooks.org//en.wikipedia.org/wiki/tautomer`

4   `http://en.wikibooks.org//en.wikipedia.org/wiki/Peptide_bond`

5   `http://en.wikibooks.org//en.wikipedia.org/wiki/amide`

(It is possible to draw a resonance structure with a double bond between the carbon atom and the nitrogen atom, with a formal negative charge on the oxygen atom and a formal positive charge on the nitrogen.) This prevents rotation around the C-N bond, locking the peptide bond in the *trans* conformation, and holding six atoms in a plane: the α-carbon of one amino acid, the carbonyl carbon and oxygen atoms, the amino nitrogen and hydrogen atoms, and the &alpha-carbon of the second amino acid are all co-planar. The structure of a peptide chain's "backbone" can be described uniquely by the torsion angles between adjacent peptide units. The φ-angle[6] is the torsion angle between the α-carbon and the carbonyl carbon of one amino acid; the ψ-angle[7] is the torsion angle between the amino nitrogen and the α-carbon.
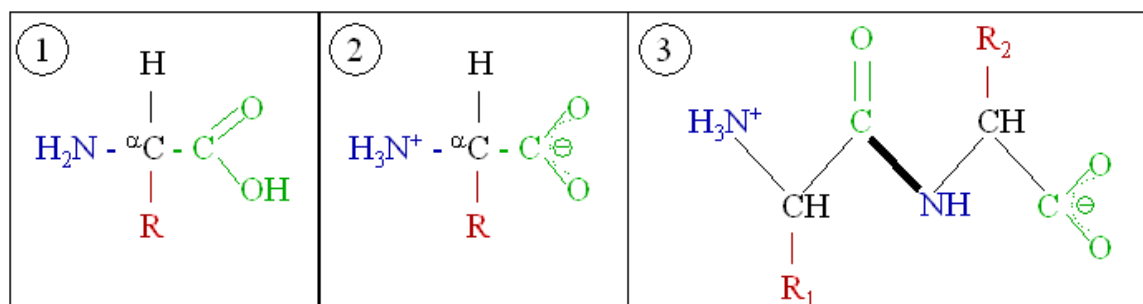
Figure 17   Peptide bond formation

Peptide bond formation

Two amino acids with different side chains react when the amino-terminus (red) of one joins the carboxy-terminus (blue) of another are linked by an amide bond (green). See also the mechanism of peptide bond formation[8].)

It is important to note that the formation of this peptide bond is highly unfavorable under standard conditions. However, in the human body, there are enzymes that assist in facilitating this reaction, making peptide bond formation and proteins possible.

4.9.2 Chemical classification of aminoacids

The 20 amino acids encoded by the genetic code[9] are:

---

6   http://en.wikibooks.org//en.wikipedia.org/wiki/%CF%86-angle
7   http://en.wikibooks.org//en.wikipedia.org/wiki/%CF%88-angle
8   http://en.wikibooks.org/wiki/Biochemistry/Peptide_Bond_Formation
9   http://en.wikibooks.org//en.wikipedia.org/wiki/Wikipedia:genetic_code
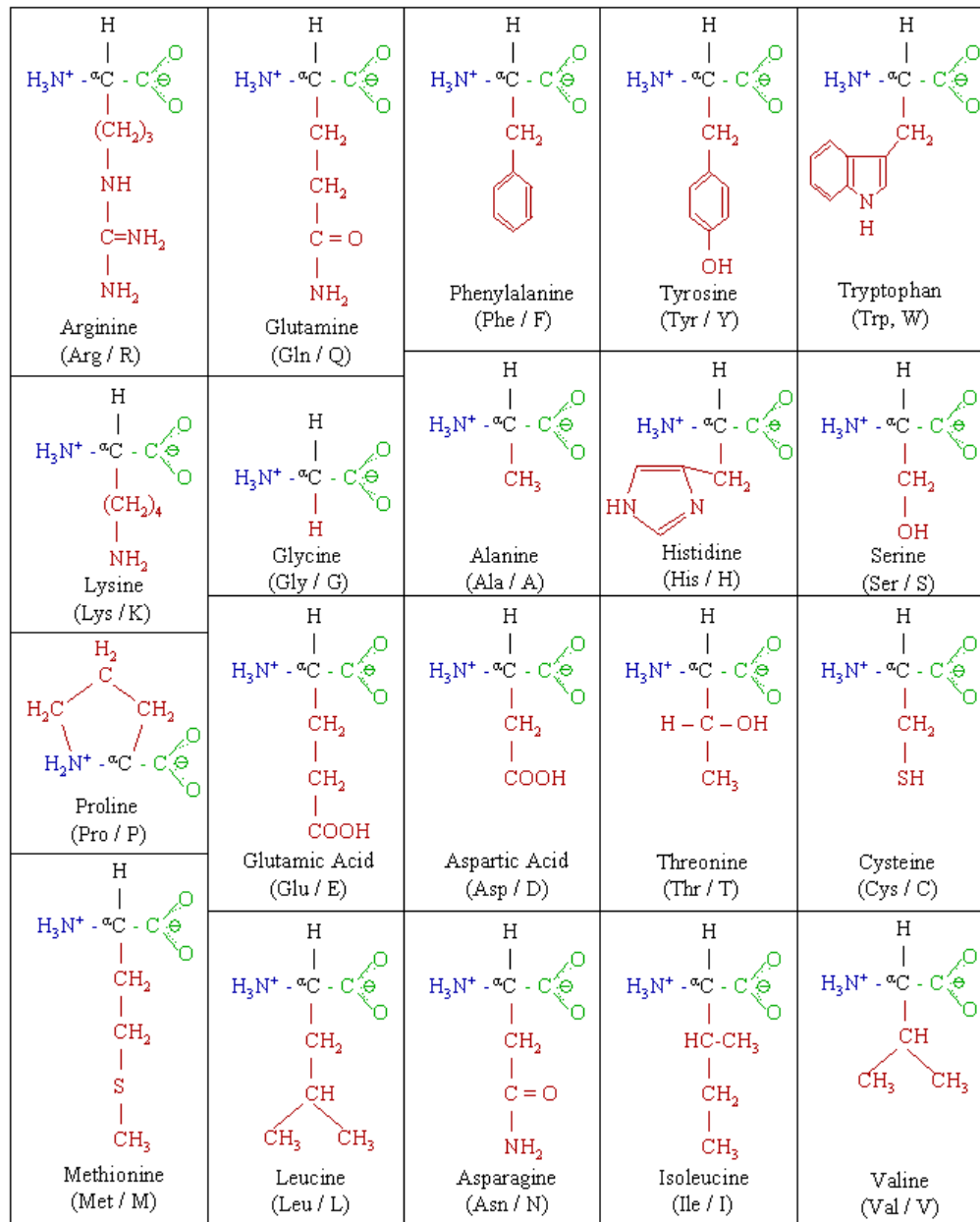
Figure 18 The 20 Genetically Encoded Amino Acids

They are not shown in their zwitterionic state for clarity. At physiological pH (~pH 6.8), all would be so, except proline, which is a five-membered ring. Charged side-chains are shown ionic when they exist as such at physiological pH.

4.9.3 Protein Electrophoresis

Protein Electrophoresis is a method in which a mixture of proteins can be separated and analyzed. Electrophoresis is based on the mobility of ions in an electric field. The charge distribution of the molecules is critical in the separation of all electrophoresis. In an electric

field, electrophoresis is a passage of charged molecules in solution. Positively charged ions have tendency toward a negative electrode and inversely, negatively charged ions have tendency toward a positive electrode. The molecular weight results to a molecular friction which is directly proportional to the molecular charge and its voltage and inversely proportional to a molecule's mobility in an electric field. Gel electrophoresis is performed to analyze the molecular weights and the charge of the protein and is mostly used in electrophoresis of the protein. The gel electrophoresis is carried out in a thin piece of polyacrylamide. The crosslinked of acrylamide and N,N'-methylene-bis-acrylamide forms the polyacrylamide by polymerization. The size has an important effect in the movement of the protein molecule. The smaller molecules of protein would result in a faster passage of the molecules through the gel pores. The separation of the protein molecules in the gel affects the protein activity. In this process, first the protein reduces the disruption of disulfide bonding by heating which results in purification and denaturalization. Next, the sodium dodecyl sulfate, abbreviated as SDS, (and ionic detergent) is added. SDS is an anionic detergent which dissolve hydrophobic molecules and denatures protein molecules without breaking peptide bonds. This result in the dislocation of the structure of the protein changes the secondary, tertiary and quaternary to the primary structure with negative charge. Then the protein is passed through the gel. For denatured proteins, SDS can form a steady charge mass ratio in binding with proteins. The polyacrylamide gel electrophoresis is a very sensitive method capable of a bearing a high resolution and it is analytically used in the studies in the separation techniques.

4.9.4 Isoelectric point

The isoelectric point[10] (pI) is the pH-value in which a protein is neutral, that is, has zero net charge. To be clear, it is not the pH value where a protein has all bases deprotonated and all acids protonated, but rather the value where positive and negative charges cancel out to zero. **Calculating pI:** An aminoacid with n ionizable groups with their respective pKa values pK1, pK2, ... pkn will have the pI equal to the average of the group pkas: pI=(pK1+pK2+...+pkn)/n Most proteins have many ionizable sidechains in addition to their amino- and carboxy- terminal groups. The pI is different for each protein and it can be theoretically calculated according Henderson-Hasselbalch equation[11] if we know amino acids composition of protein. In order to experimentally determine a protein's pI 2-Dimensional Electrophoresis (2-DE) can be used. The proteins of a cell lysate are applied to a pH immobilized gradient strip, upon electrophoresis the proteins migrate to their pI within the strip. The second dimension of 2-DE is the separation of proteins by MW using a SDS-gel. To clearly understand isoelectric point, you have to keep in mind that the positively charged groups are balanced by the negatively charged groups. For simple amino acid " alanine", the isoelectric point is an average of the PKa of the carboxyl with PK1 of (2.34) and ammonium with PK2 of (9.69) groups. So, the PI for the simple amino acid " alanine" is calculated as:(2.34+9.69)/2 which is equal to 6.02. When additional basic or acidic groups are added as a side-chain functions, the isoelectric point pI will be the average of the pKa's of the most similar acids. Example for this concept could be aspartic acid in which the similar acids are alpha-carboxyl function with pKa of 2.1 and the side chain carboxyl function with a pKa

---

10   http://en.wikibooks.org//en.wikipedia.org/wiki/Isoelectric_point
11   http://en.wikibooks.org//en.wikipedia.org/wiki/Henderson-Hasselbalch_equation

of 3.9. Thus, the pI for aspartic acid is (2.1+3.9)/2=3.0. Another example is arginine, its similar acids are guanidinium on the side chain with pKa of 12.5 and the alpha-ammonium function with pKa of 9.0. Thus the calculated pI for arginine= (12.5+9.0)/2=10.75. pI does not has a unite.

4.9.5 The peptidic bond

Two amino acid molecules can be covalently joined through a substituted amide linkage, termed a **peptide bond,** to yield a dipeptide. this link is formed by dehydration (removal of the water molecules - one hydrogen atom from one amino acid and an OH group from the other). This process can then continue to join other amino acids and yield in an amino acid chain. When there are few amino acids in a chain, it is called an **oligopeptide**, when there are many it is called a **polypeptide**. although the terms "protein" and "polypeptide" are sometimes used to describe the same thing, the term polypeptide is generally used when the molecular weight of the chain is below 10,000. An amino acid unit in a peptide is often called a residue.

4.10 Disulfide bonds

Disulfide bonds form between the sulfur atoms of two cysteine side chains in a protein. The side chains undergo a reversible oxidation of the sulfhydryl groups of the cysteine, wich results in covalent bonding of the sulfur atoms (S-S). This bonding is called a "disulfide bridge". Typically, disulfides don't form on the surface of proteins because of the presence of reducing agents in the cytoplasm. These bonds are of great importance concerning the shaping of protein structure; their formation guides the folding of peptide chains as the proteins are produced. Structural proteins that have to be rigorously stable (for example, Keratin, which is found in nail, horn and crustacean shell) often contain a large number of disulfide bonds.
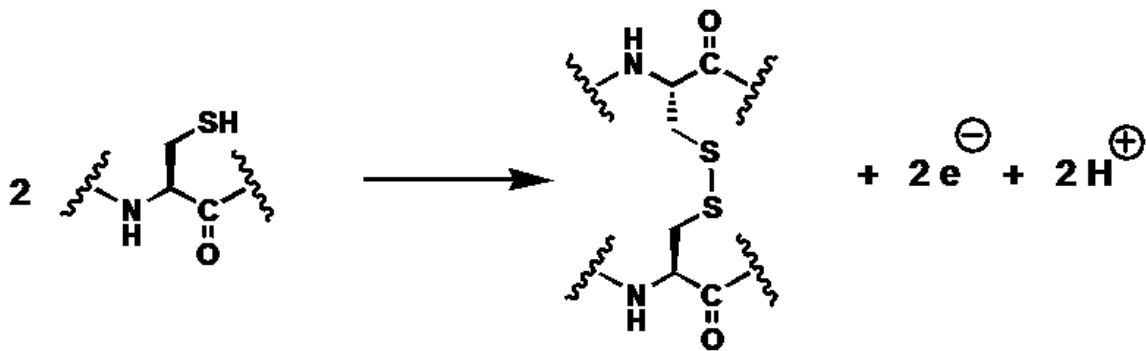
Figure 19   The reaction scheme for disulfide bond formation. Two cysteine residues undergo a two electron oxidation, relaeasing two electrons and two protons. Such reactions are typically catalyzed by flavin-enzymes.

4.11 Post-translational modifications

After a protein is synthesized inside the cell is usually modified by addition of extra functional groups the polypeptide chain. These can be sugar or phosphate groups and may confer to the protein special functions such as: the ability to recognize other molecules, to integrate in the plasma membrane, to catalyze biochemical reactions, and various other processes. It is in the interest of the biochemist to understand what proteins are modified, what the modification is, and where it is located. An easy way to do this is by using mass spectrometry. In a sample of protein submitted to mass spectrometry you will see both modified and unmodified protein signals, the change in mass between these signals will correspond to the change in mass of your protein due to your post-translation modification.

4.12 Protein structure and folding

4.13 What is protein folding?

Protein folding is commonly a fast or very fast process, often but not always reversible, taking no more than a few milliseconds to occur. It can be viewed as a complex compromise between the different chemical interactions that can happen between the amino acid sidechains, the amidic backbone and the solvent. There are literally millions of possible three-dimensional configurations, often with minimal energetic differences between them. That's why we're still almost unable to predict the folding of a given polypeptide chain *ab initio*.Protein folding problem is that scientist still has failed to crack the code that governs folding. Moreover, the ability of biological polymers such as protein fold into well-defined structures is remarkable thermodynamically. An unfolded polymer exists are random coils, each copy of an unfolded polymers will have different conformation, yielding a mixture of many possible conformation.

4.14 Folding depends only on primary structure

In 1954 Christian Anfinsen demonstrated that the folding of a protein in a given environment depends only on its primary structure - the amino acid sequence. This conclusion was by no means obvious, given the complexity of the folding process and the paucity of biochemical knowledge at the time. The process of folding often begins co-translationally, so that the N-terminus of the protein begins to fold while the C-terminal portion of the protein is still being synthesized by the ribosome. Specialized proteins called chaperones assist in the folding of other proteins. Meanwhile, protein folding is a thermodynamically driven process: that is, proteins fold by reaching their thermodynamically most stable structure. The path followed by the protein in the potential energy landscape is far from obvious, however. Many local and non-local interactions take part in the process, and the space of possible structures is enormous. As of today molecular dynamics simulations are giving invaluable hints on the first stages of the folding process. It is known now that the unfolded state still retains key long-range interactions and that the local propensity of the sequence to fold in a given secondary structure element narrow the "search" in the so-called conformational space. This seems to mean that biological proteins somehow evolved to properly fold. In fact, many

random amino acid sequences only acquire ill-defined structures (molten globules) or no structure at all. There are some general rules, however. Hydrophobic amino acids will tend to be kept inside the structure, with little or no contact with the surrounding water; conversely, polar or charged amino acids will be often exposed to solvent. Very long proteins will often fold in various distinct modules, instead of in a single large structure.

4.15 The Ramachandran plot

The Ramachandran plot was invented by professor G.N.Ramachandran, a very eminent scientist from India. He discovered the triple helix structure of collagen in 1954 a year after the double helix structure was put forth. In a polypeptide the main chain N-Calpha and Calpha-C bonds relatively are free to rotate. These rotations are represented by the torsion angles phi and psi, respectively. G N Ramachandran used computer models of small polypeptides to systematically vary phi and psi with the objective of finding stable conformations. For each conformation, the structure was examined for close contacts between atoms. Atoms were treated as hard spheres with dimensions corresponding to their van der Waals radii. Therefore, phi and psi angles which cause spheres to collide correspond to sterically disallowed conformations of the polypeptide backbone

4.16 Intramolecular forces in protein folding

The tertiary structure is held together by hydrogen bonds, hydrophobic interactions, ionic interactions, and/or disulfide bonds.

4.17 The protein folding problem

The protein folding problem relates to what is known as the Levinthal paradox. Levinthal calculated that if a fairly small protein is composed of 100 amino acids and each amino acid residue has only 3 possible conformations (an underestimate) then the entire protein can fold into 3^{100}-1 or $5\text{x}10^{47}$ possible conformations. Even if it takes only 10^{-13} of a second to try each conformation it would take 10^{27} years to try them all. Obviously a protein doesn't take that long to fold, so randomly trying out all possible conformations is not the way proteins fold. Since most proteins fold on a timescale of the order of milliseconds it is clear that the process is directed in some manner dependent on the constituents of the chain. The protein folding problem which has perplexed scientists for over thirty years is that of understanding how the tertiary structure of a protein is related to its primary structure, because it has been proven that the primary structure of a protein holds the only information necessary for the protein to fold. Ultimately the aim is also to be able to predict what pathway the protein will take.

4.18 Folding in extreme environments

Most proteins are not capable of maintaining their three-dimensional shape when they are exposed to environmental extremes such as a low or high pH, or a highly variable temperature. Changes in the pH of the proteins environment may alter the charges on the amino acid side chains that form the whole protein, so that repulsive or attractive forces may form, altering the secondary and tertiary structure of the protein as a whole, as a result the shape of the enzyme is warped, and the now non-functional protein is said to be denatured. A high or exceptionally low temperature can cause the constitutive bonds on the protein to be broken, again resulting in the protein being rendered non-functional. It is important to note that certain proteins, mainly digestive enzymes such as trypsin, are capable of with-standing a pH as low as 1. If the pH of such an enzymes environment were to increase to approximately pH5, it would be inactivated.

4.19 Protein misfolding

The way that a protein folds is one of the most important factors influencing its properties, as this is what determines which active groups are exposed for interaction. If the protein misfolds, its properties can be markedly changed. One example of this is in Transmissible Spongiform Encephalopathies, such as BSE, and Scrapie. In these, the prion protein, which is involved in the brain's copper metabolism, misfolds, and starts forming plaques, which destroy brain tissue.

5 Protein structural levels

Biochemists refer to four distinct aspects of a protein's structure:

5.1 Primary structure

Primary structure is practically a synonym of the amino acid sequence. It can also contain informations on amino acids linked by peptide bonds. Primary structure is typically written as a string of three letter sequences, each representing an amino acid. Peptides and proteins must have the correct sequence of amino acids.

5.2 Secondary structure

Secondary structure elements are elementary structural patterns that are present in most,if not all,known proteins. These are highly patterned sub-structures --alpha helix and beta sheet-- consisting of loops between elements or segments of polypeptide chain that assume no stable shape. Secondary structure elements, when mapped on the sequence and depicted in the relative position they have in respect to each other, define the *topology* of the protein. It is also relevant to note that hydrogen bonding between residues is the cause for secondary structure features; secondary structure is usually described to beginning biochemists as (almost) entirely independent of residue side-chain interactions.

5.3 Tertiary structure

Tertiary structure is the name given to refer to the overall shape of a single protein molecule. Although tertiary structure is sometimes described (especially to beginning biology and biochemistry students) as being a result of interactions between amino acid residue side chains, a more correct understanding of tertiary structure is the interactions between elements of secondary protein structure, i.e. alpha-helices and beta-pleated sheets. Tertiary structure is often referred to as the "fold structure" of a protein, since it is the result of the complex three-dimensional interplay of other structural and environmental elements.

5.4 Super-tertiary structure (protein modules)

Some literature refers to elements of super-tertiary structure, which often refers to elements of folding that, for whatever reason, do not neatly fit into the category of tertiary structure.

Often this level of distinction is saved for graduate level coursework. Protein denaturation can be a reversible or an irreversible process, i.e., it may be possible or impossible to make the protein regain its original spatial conformation.

5.5 Quaternary structure

Quaternary structure is the shape or structure that results from the union of more than one protein molecule, usually called *subunit proteins subunits* in this context, which function as part of the larger assembly or protein complex.

```
And it refers to the regular association of two or more polypeptide
 chains to form a complex. A multi-subunit protein may be composed of
 two or more identical polypeptides, or it may include different
 polypeptides.
```

Quaternary structure tends to be stabilized mainly by weak interactions between residues exposed on surfaces polypeptides within a complex.

6 Secondary structure elements

6.1 The alpha helix
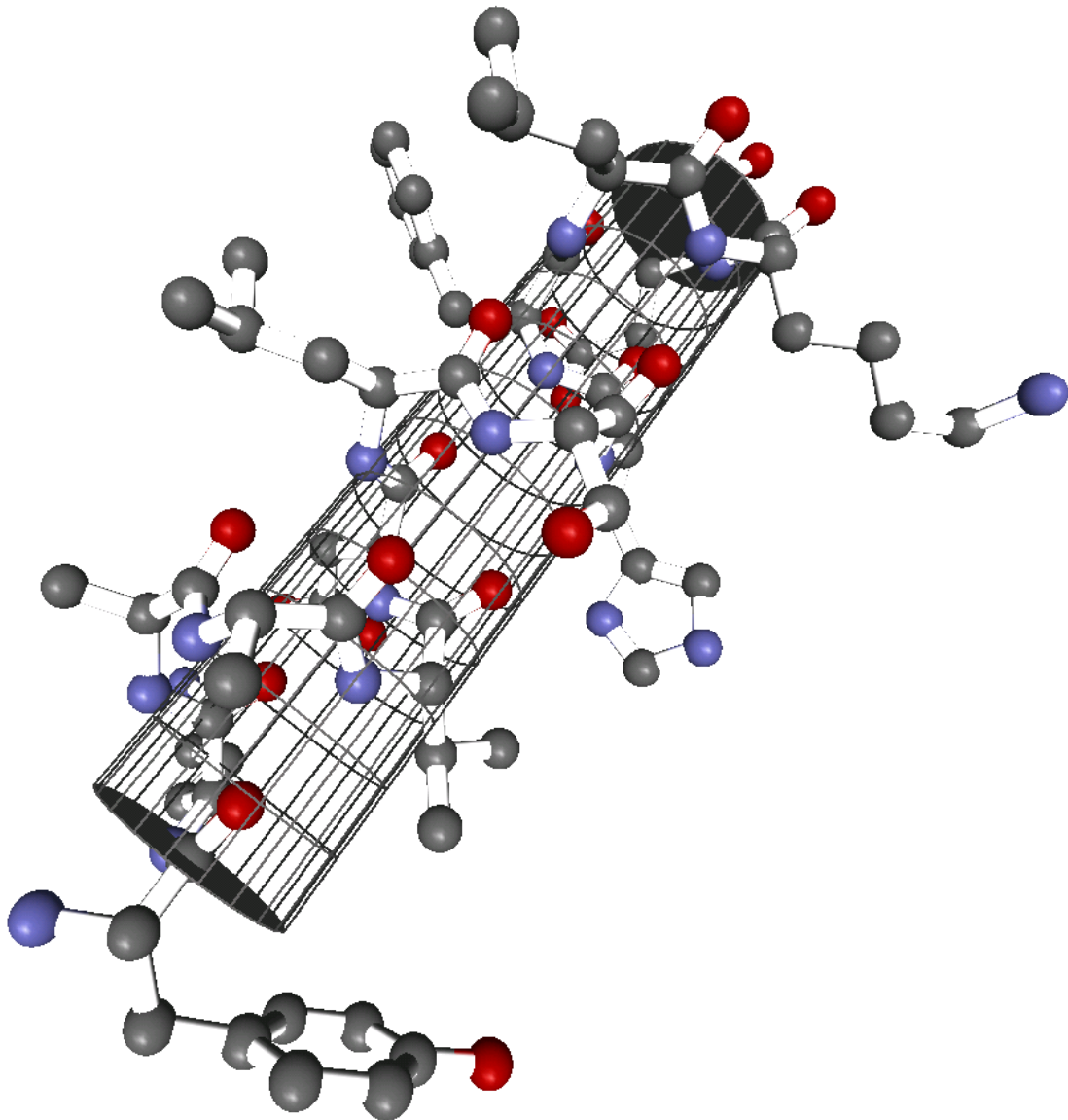
Figure 20   Alpha Helix

The alpha helix is a periodic structure formed when main-chain atoms from residues spaced four residues apart hydrogen bond with one another. This gives rise to a helical structure, which in natural proteins is always right-handed. Each turn of the helix comprises 3.6 amino acids. Alpha helices are stiff, rod-like structures which are found in many unrelated proteins. One feature of these structures is that they tend to show a bias in the distribution of hydrophobic residues such that they tend to occur primarily on one face of the helix. The amino acids in an α helix are arranged in a helical structure, about 5 Å wide. Each amino acid results in a 100° turn in the helix, and corresponds to a translation of 1.5Å along the helical axis. The helix is tightly packed; there is almost no free space within the helix. All amino acid side-chains are arranged at the outside of the helix. The N-H group of amino acid (n) can establish a hydrogen bond with the C=O group of amino acid (n+4). Short polypeptides usually are not able to adopt the alpha helical structure, since the entropic cost associated with the folding of the polypeptide chain is too high. Some amino acids (called *helix breakers*) like proline will disrupt the helical structure. Ordinarily, a helix has a buildup of positive charge at the N-terminal end and negative charge at the C-terminal end which is a destabilizing influence. As a result, α helices are often capped at the N-terminal end by a negatively charged amino acid (like glutamic acid) in order to stabilize the helix dipole. Less common (and less effective) is C-terminal capping with a positively charged protein like lysine. α helices have particular significance in DNA binding motifs, including helix-turn-helix motifs, leucine zipper motifs and zinc finger motifs. This is because of the structural coincidence of the α helix diameter of 12Å being the same as the width of the major groove in B-form DNA. α helices are one of the basic structural elements in proteins, together with beta sheets. The peptide backbone of an α helix has 3.6 amino acids per turn.

6.2 The beta sheet
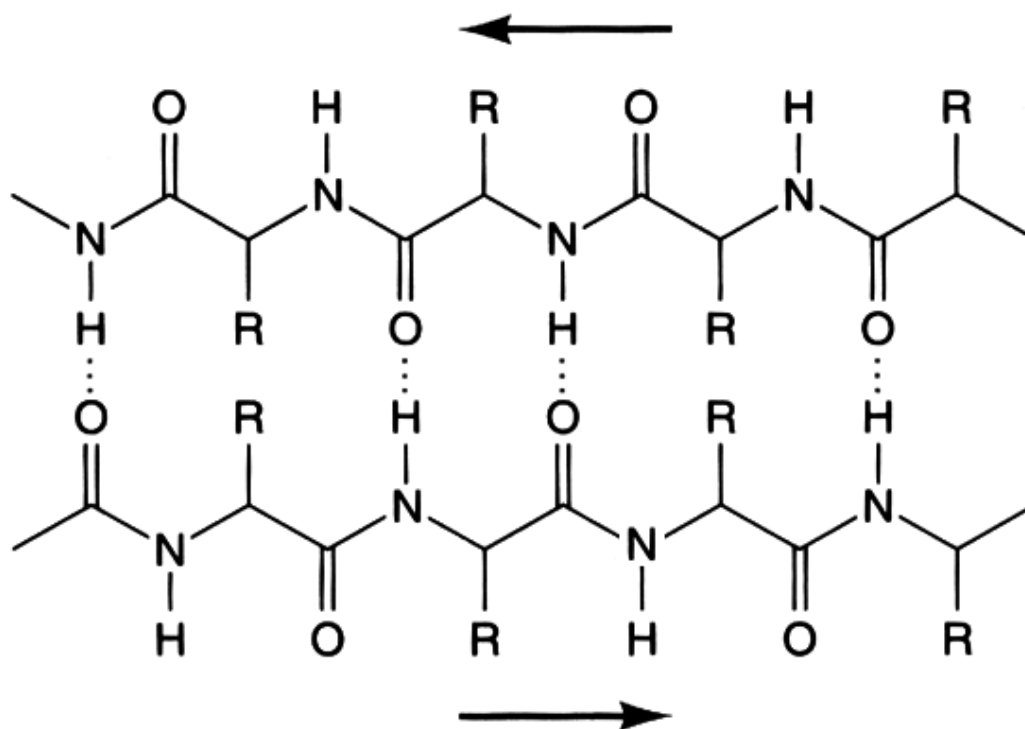
Figure 21   Diagram of B-Pleated sheet and bond structure of protein

The **β sheet** (also **β-pleated sheet**) is a commonly occurring form of regular secondary structure in proteins, first proposed by Linus Pauling and Robert Corey in 1951. It consists of two or more amino acid sequences within the same protein that are arranged adjacently and in parallel, but with alternating orientation such that hydrogen bonds can form between the two strands. The amino acid chain is almost fully extended throughout a β strand, lessening the probability of bulky steric clashes. The Ramachandran plot shows optimal conformation with angle phi = -120 to -60 degrees and angle psi = 120 to 160 degrees. The N-H groups in the backbone of one strand establish hydrogen bonds with the C=O groups in the backbone of the adjacent, parallel strand(s). The cumulative effect of multiple such hydrogen bonds arranged in this way contributes to the sheet's stability and structural rigidity and integrity: for example, cellulose's beta-1,4 glucose structure. The side chains from the amino acid residues found in a β sheet structure may also be arranged such that many of the adjacent side chains on one side of the sheet are hydrophobic, while many of those adjacent to each other on the alternate side of the sheet are polar or charged (hydrophilic). Some sequences involved in a β sheet, when traced along the backbone, take a *hairpin turn* in orientation (direction), sometimes through one or more prolines. The α-C atoms of adjacent strands stand 3.5Å apart. In addition to the parallel beta sheet, there is also the anti-parallel beta sheet. The hydrogen-bond still exists in this conformation but the main difference lies in the directionality of the protein. In this conformation the proteins run in opposite directions, but the resulting hydrogen bonds connect directly across from one

another instead of the diagonal. In short, beta sheets can be purely parallel, anti-parallel, or even mixed.

6.3 The random coil

A protein that completely lacks secondary structure is a **random coil**. In random coil, the only fixed relationship between amino acids is that between adjacent residues through the peptide bond. As a result, random coil can be detected from the absence of the signals in a multidimensional nuclear magnetic resonance experiment that depend on particular peptide-peptide interactions. Likewise in the images produced in crystallography experiments, pieces of random coil appear simply as an absence of "electron density" or contrast. Random coil is also easily distinguished by circular dichroism. Denaturing reduces a protein entirely to random coil.

6.4 Less common secondary structure elements

Certain other periodic structures rarely appear in proteins, some of which are similar to the more common types. For example, two variants of the alpha-helix also occur, the 3-10 helix and the pi helix. These have helical pitches of 3 and 4.4 residues per turn respectively, corresponding to hydrogen bonds forming between residue i and i+3 for the 3-10 helix and between i and i+5 for the pi helix. Both are usually very short (1 turn or so) and have been seen only at the ends of alpha helices.

7 Tertiary structure elements

7.1 Alpha-only structures

Structures that contain only Alpha Helices

7.2 Beta-only structures

8 Common folds and modules

8.1 The Rossman fold

The Rossmann fold is a protein structural motif found in proteins that bind nucleotides, especially the cofactor NAD. The structure is composed of three or more parallel beta strands linked by two alpha helices in the topological order beta-alpha-beta-alpha-beta. Because each Rossmann fold can bind one nucleotide, binding domains for dinucleotides such as NAD consist of two paired Rossmann folds that each bind one nucleotide moiety of the cofactor molecule. Single Rossmann folds can bind mononucleotides such as the cofactor FMN. The motif is named for Michael Rossmann who first pointed out that this was a frequently occurring motif in nucleotide binding proteins.

8.2 The hemoglobin fold

8.3 The immunoglobulin fold

8.4 Keratin

Found in hair and in the palms of human hands.

8.5 Collagen

8.6 Types of protein

Biochemistry/Proteins/Types of protein[1]

---

1   `http://en.wikibooks.org/w/index.php?title=Biochemistry/Proteins/Types_of_protein&` `action=edit&redlink=1`

9 Carbohydrates

9.1 Introduction

Carbohydrates are one of the fundamental classes of macromolecules found in biology[1]. Carbohydrates are commonly found in most organisms, and play important roles in organism structure, and are a primary energy source for animals and plants. Most carbohydrates are sugars or composed mainly of sugars. By far, the most common carbohydrate found in nature is glucose, which plays a major role in cellular respiration and photosynthesis. Some carbohydrates are for structural purposes, such as cellulose (which composes plants' cell walls) and chitin (a major component of insect exoskeletons). However, the majority of carbohydrates are used for energy purposes, especially in animals. Carbohydrates are made up of a 1:2:1 ratio of Carbon, Hydrogen, and Oxygen $(CH_2O)_n$

9.2 Simple Carbohydrates (Monosaccharides)

These are used only for energy in living organisms. Simple carbohydrates are also known as "Monosaccharides".The chemical formula for all the monosaccharides is CnH2nOn. They are all structural isomers of each other. There are two main types of monosaccharides. The first type are aldoses, containing an aldehyde on the first carbon, and the second type are ketoses, which have a ketone on the second carbon (This carbonyl group is always located on the second carbon).

```
Name            Formula         Aldoses             Ketoses
Trioses         C3 H6 O3        Glycerose
Dhydroxyacetone
Tetroses        C4 H8 O3        Erythrose
Erythrulose
Pentoses        C5 H10 O5       Ribose              Ribulose
Hexoses         C6 H12 O6       Glucose             Fructose
Heptose         C7 H14 O7       Glucoheptose
Sodoheptulose
```

The suffix -oses is kept for the aldoses & the suffix -uloses is kept for the ketoses. Except fructose ketoses are as common as aldoses.The most abundant monosaccharide in nature is the 6-Carbon sugar -i.e.D-Glucose. All monosachharides are non reducing sugar.

---

1   http://en.wikibooks.org/wiki/Biology

9.2.1 Monomers

There are two main forms of monomer, aldoses and ketoses, of which, all have the same backbone. The simplest for of monomers are trioses. The trioses consist of three carbons in the backbone.

9.2.2 Fructose

Fructose is a ketohexose with the molecular formula of C6H12O6 ; which is same as the molecular formula of Glucose but different structure. The structure of fructose differs from glucose at carbon 1 and 2 by the location of the carbonyl group. Fructose is the sweetest naturally occurring sugar. Fructose is also called levulose and fruit sugar; fructose is also found in fruits, root vegetables(such as sweet potato and onion) and honey. It is a isomer of glucose and a ketose simple sugar. Fructose has the highest solubility among all sugars. Fructose can be converted to its isomer glucose, after it enters the blood stream.

```
        CH2OH

         C=O

     OH-C-H


      H-C-OH


      H-C-OH


       CH2OH
```
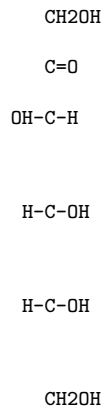
9.2.3 Glucose

The hexoses glucose,galactose, and fructose are important monosaccharides.Glucose is the most prevalent monosaccharide in diet. The most common hexose, D-glucose, C6H12O6 also known as dextrose and blood sugar, is found in fruits,vegetables, corn syrup, and honey. Glucose is a building block of the disaccharides(sucrose,maltose,lactose) and polysaccharides (glycogen,cellulose,starch). In the body, excess glucose is converted to glycogen and then stored in the muscle and liver.

```
         CHO


      H-C-OH


      OH-C-H
```
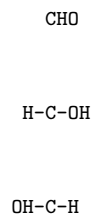
```
H-C-OH


H-C-OH


CH2OH
```

9.2.4 Galactose

Galactose is a sugar component of the disaccharide lactose, as found in milk. It is not as sweet as glucose and is separated from the glucose in lactose via hydrolysis.In addition, galactose is an aldohexose that does not occur in the free form in nature; galactose has an important role in the cellular membrane of the brain and nervous system. D-galactose has a structure similar to D-glucose and the only difference between them is in the arrangement of the –OH group on carbon number 4. Galactose can be found in human breast milk and is incorporated into the structure of Human Milk Oligosaccharides. The backbone of Human Milk Oligosaccharides is the disaccharide lactose, which is formed by the linkage between galactose and glucose sugars.

`Oligosachharides`

Tha oligosachharides yields 2 to 10 monosachharides on hydrolysis. Disachharides are most common oligosachharide found in nature.

9.2.5 Disaccharides

Monosaccharides are simple forms of sugars consisting of one sugar, disacharides on the other hand, consists of two. The two sugars are bonded together via a glycosidic bond. Most linkages between sugars occur between the first and fourth carbon on the sugars.

9.2.6 Polysaccharides

Sugars do not stop at disaccharides, they are much more complex than that. Multiple sugars are linked together forming many polysaccharides. Polysaccharides are made up of monosaccharides linked together.

9.3 Compound Carbohydrates (Disaccharides)

These are used by living organisms for energy. They are composed of two monosaccharides joined together by the process known as dehydration synthesis. In this process, one molecule loses one hydrogen atom, while the other loses one hydrogen atom and one oxygen atom. The reverse of this is hydrolysis, where water is added to break down a molecule into two or more simpler molecules. Disaccharides have the chemical equation $C_{12}H_{22}O_{11}$. The reason

it does not follow the 1:2:1 ratio is, obviously, due to the H_2O taken way from it. Some examples of disaccharides are:

```
                           Disachharides



              Non reducing                    reducing



   C1-C1 glycosidic    C1-C2 glycosidic    C1-C4 glycosidic
 C1-C6 glycosidic
       linkage            linkage             linkage
    linkage
    Trehalose           Sucrose             Lactose
  Gentiobiose
                                            Maltose
   Melibiose
                                            Cellobiose
   Isomaltose
```

9.3.1 Maltose

1. Chemistry-

Maltose is a white crystal sugar, also known as malt sugar and a reducing disaccharide made from two glucose units. The bonding of two glucose units is called 1-4 glycosidic linkage which joins the carbon number 1 of one glucose to carbon number 4 of the second glucose.In the presence of enzyme maltase,1-4 linkage of two glucose is broken down and maltose is hydrolized into glucose

1. occurrence-

Maltose is present in sprouting barely. It is the major product of hydrolysis of starch.
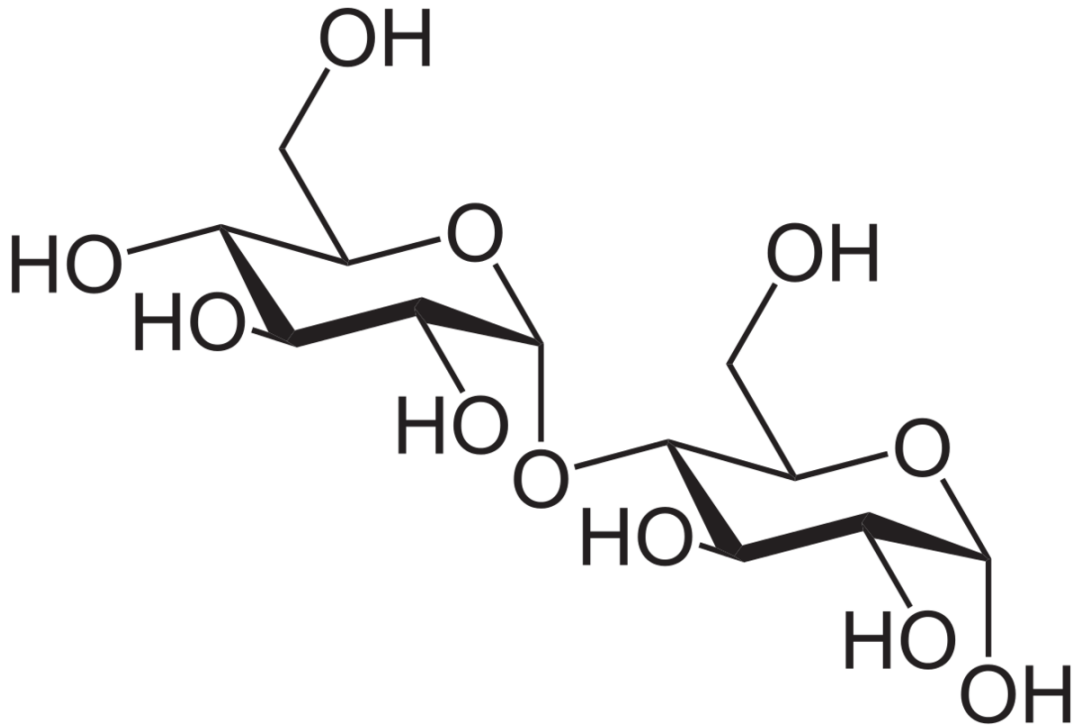
Figure 22   alpha - maltose
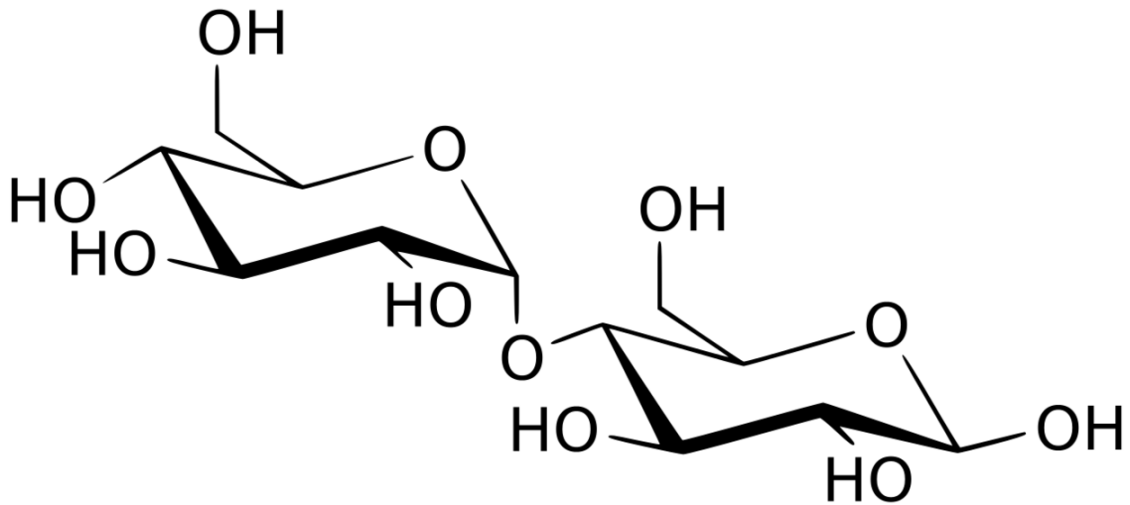
Figure 23   beta - maltose

9.3.2 Sucrose

3Occurrence- It is the costituent of sugarcane. it is also found in Beet,carrot,etc. Nector of flower is particularly rich in sucrose. 3Chemistry- Sucrose is white crystal sugar.When heat above its melting point it forms a brown substance known as "Caramel". it is non reducing sugar. Sucrose is formed by the C1-C2 glucosidic bond between glucose & fructose. Sucrose

on hydrolysis yield equimolar mixture of glucose & fructose which is offen called as "invert sugar" by the "enzyme".
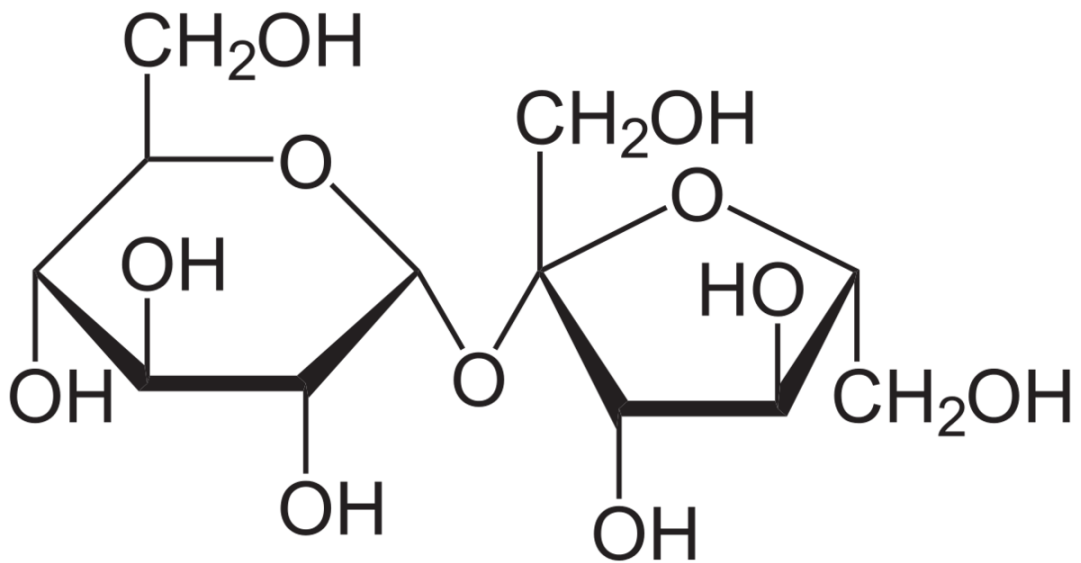
Figure 24
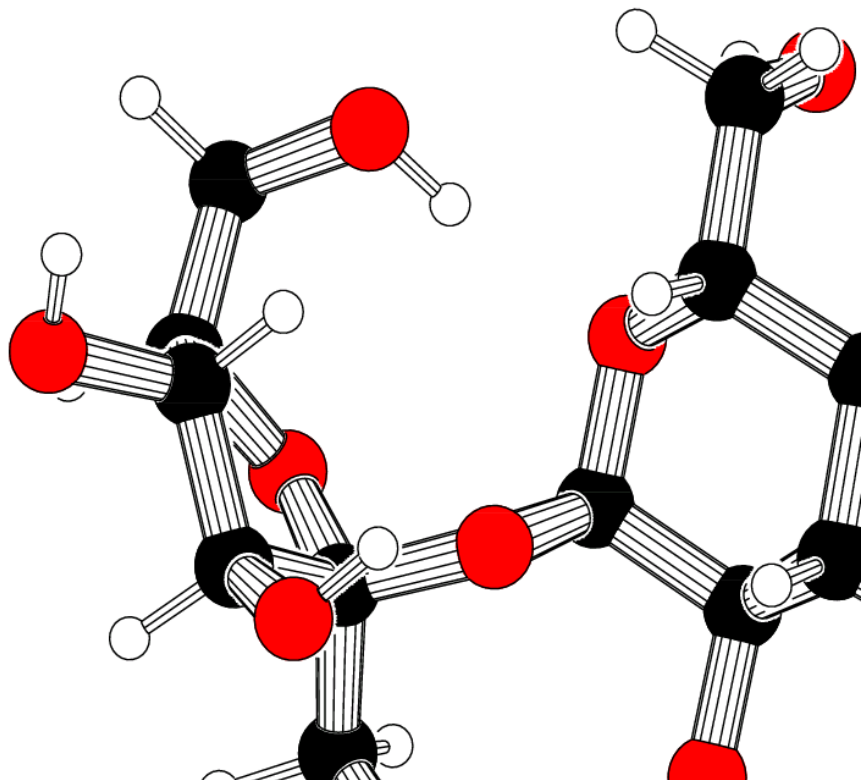
Figure 25

9.3.3 Lactose

3 Occurrence- It is found in milk of animal. It is also Milk sugar. *The percentage of latose in human milk is 6.8 % & in cow milk is 4.8 %* 3 Chemistry- Lactose is made by the C1-C4 glycosidic bond between galactose & glucose. The hydrolysis of Lactose gives the monosacchride: glucose + glactose by the enzyme lactase. Lactose can be a reducing sugar.
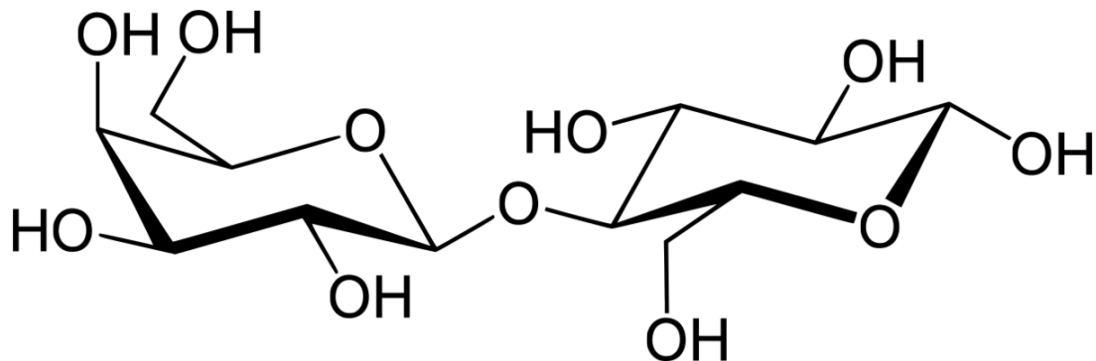
Figure 26   β-D-galactopyranosyl-(1→4)-D-glucose
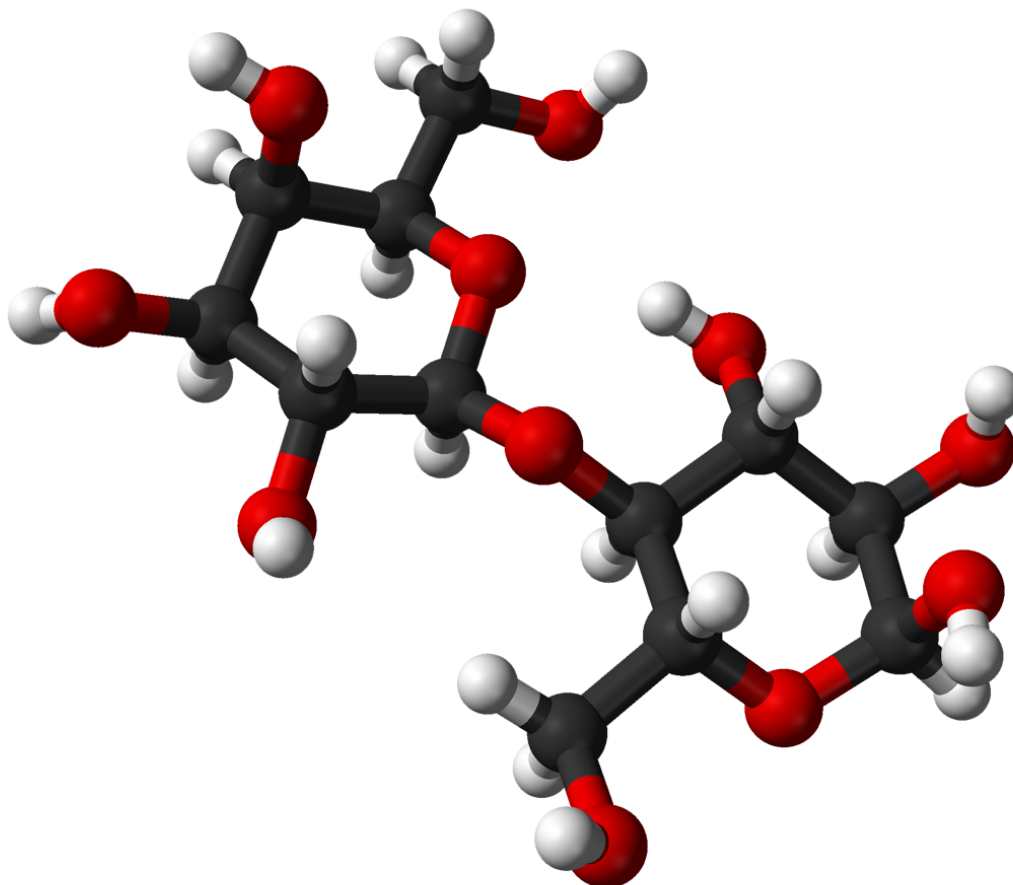
Figure 27 The molecular structure of α-lactose, as determined by X-ray crystallography.

9.4 Complex Carbohydrates (Polysaccharides)

Polysachharide are also called as glycanes are high molecular weight which on hydrolysis yield monosachharides.Some source of complex carbohydrates are pasta, brown bread, brown rice, corn, beans, potatoes, and peas. Digestion of complex carbohydrates could take more time because digestive enzyme have to work harder to break down the chain into individual sugars. Both monosaccharides and disaccharides are used only for energy. Another difference is that while monosaccharides can be used for energy immediately, and disaccharides can be used for energy relatively rapidly because they can be broken down quickly, polysaccharides release their energy slowly. chemicaly , the polysachharide may be distinguished into homopolysachharide & heteropolysachharide. Homopolysachharides which on hydrolysis yield only single monosachharide. Eg. starch, cellulose, etc. Hateropolysachharide which on hydrolysis yield two or more monosachharide. Eg. heparin, chondroitin, etc.

```
POLYSACHHARIDE
```

```
            Homopolysachharide
Heteropolysachharide
            Starch                      Mucopolysachharids
            Glucogen                    Chondroitin
            Inulin                      Hyaluronic acid
            Cellulose                   Keratosulfate
            Pectin                      Heparin
            Chitin                      Vegetable gum
            Xylan                       Agar agar
```

polysachharide may be classified on the basis of functional aspect as Nutritive & Structural. Nutritive- starch,glycogen,inulin etc. Structural- cellulose,chitin,pectin,etc.

9.4.1 Cellulose (Fiber)

Cellulose is a special kind of carbohydrate. It is insoluble and most organisms can not produce enzymes to break it down. It is made by β-D-glucose. It is found only in plants, and it's found in the cell wall. It is composed of β glucose molecules, which create a more rigid structure. Cotton used to make clothes etc. is cellulose from around the seeds of the cotton plant. Fibre helps the plant keep a strong structure. Humans can't digest fibre.it gives no color with iodine.It is completely absent in meat, egg, fish & milk.

9.4.2 Starch

Starch is the energy storage molecule of plants.It is formed by long chains of α glucose molecules linked together. It is mostly found in potato. Starch is actually a mixture of 2 types of molecules, amylose and amylopectin. These large polysaccharides are very good for energy storage because they have lots of glucose molecules crammed into a small space which can then be easily broken off, one at a time, by hydrolysis and then used for energy. It is hydrolysed by enzyme Amylase.It is non reducinh sugar. It gives blue,purple colour with iodine

9.4.3 Glycogen

Glycogen is the energy storage molecule of animals. It is formed by branched chains of alpha glucose molecules with 1-4 glycosidic bonds on the main chains and 1-6 glycosidic bonds to form the branches. Humans store small amounts of glycogen in the liver and muscles. It is created when there are high blood sugar levels. The pancreas secretes insulin, which stimulates the creation of glycogen from glucose and signals the body to use glucose as its main form of energy. It is non reducing sugar. It gives red colour with iodine.

9.4.4 References

n.p. *Carbohydrates Notes.* n.d. www.chem.ucla.edu/harding/notes/notes__14C__carbos.pdf

10 Membranes and Lipids

Biochemistry[1]
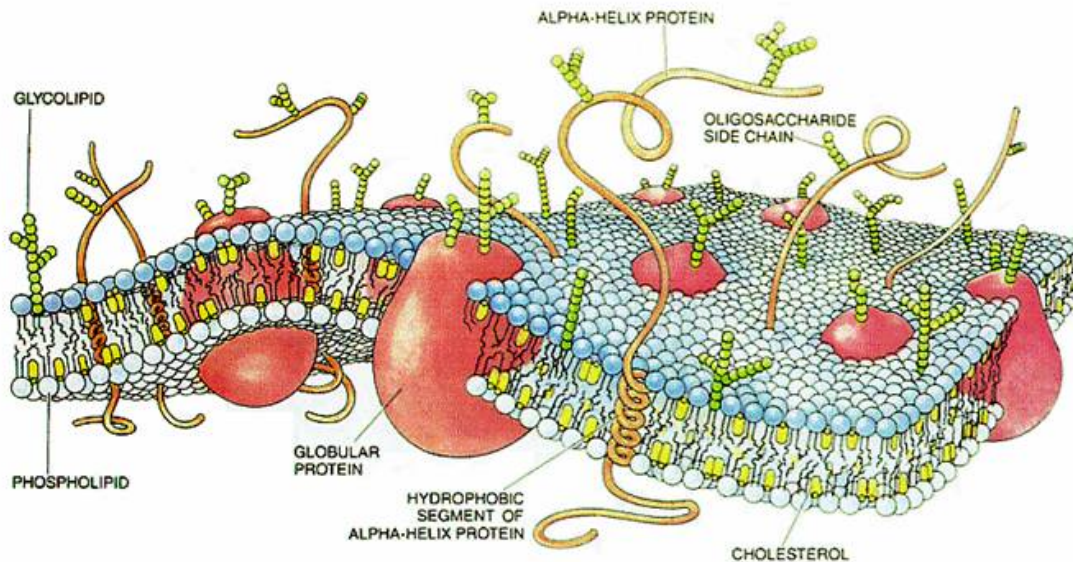
Figure 28

10.1 Membranes And Lipids

All cells, from simple prokaryotic bacteria to complex multicellular organisms are surrounded by a **membrane**. The membrane acts as a selective barrier, letting some substances into the cell and preventing other substances from entering. The membrane also actively transports substances between the inside and outside of the cell, using cellular energy to do so. This is important in regulating the concentration of many substances which must be maintained within strict limits. The cell also contains membrane bound compartments, where the membrane acts as a barrier for the separation of different environments, like lysosomes which have a high pH that would be toxic to the rest of the cell. This chapter is about the chemical composition of the membrane and how this creates the functionality of the membrane.

---

1   http://en.wikibooks.org/wiki/Biochemistry

10.2 The Lipid Bilayer

The first person to link lipids with the cell membrane may have been Charles Ernest Overton, who was studying heredity in plants. Part of his studies involved studying which substances absorbed into plant cells most quickly. After characterizing a large number of substances, he came up with the idea that cell membranes were composed of something similar to the lipids found in vegetable oils, and that substances are absorbed into the cells by dissolving through the membrane. The next big step came when Gorter and Grendel extracted the lipids from red blood cells and compared the surface area of the lipids spread out on water compared to the surface area of the red blood cells. They found the lipid surface area was twice that of the red blood cells, and concluded that the lipids must be arranged in a layer two lipid molecules thick. This is what biologists now call the **lipid bilayer**, and to understand the construction of the bilayer, we need to understand the lipids themselves.

10.2.1 Fatty Acids

A fatty acid is simply a linear carbon chain with a carboxylic acid group at one end. Fatty acids usually have between 14 and 24 carbon atoms and may have one or more double bonds. These double bonds are almost always in the cis configuration.
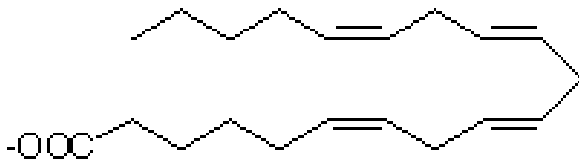
Figure 29

Fatty acids have both common names and systematic names. The systematic name is based on the alkane or alkene with the same number of carbon atoms, with the final *e* of the hydrocarbon replaced with *oic acid*. For example Laurate (Figure x)

Figure 30

is a fatty acid with 12 carbon atoms and no double bonds, so the systematic name is dodecanoic acid, and the ionized form is dodecanoate. (saturated fatty acid table) If there are double bonds, the location is marked with the symbol Δ with a superscript number indicating the location of the double bond and preceded by *cis* or *trans* to indicate configuration (but almost invariably *cis* in natural fatty acids). Carbon atoms are counted from the carboxyl end, as in Figure x, so a dodecenoate acid with a cis double bond between carbon atoms 9 and 10 would be: *cis*-Δ^9-dodecenoate. The number 2, 3 and last carbon

atom are called the α, β and ω atoms, respectively.

One common shorthand for describing fatty acids is based on the fact that multiple double bonds are placed three bonds apart. This allows the acid to be described by the length of its carbon chain, number of double bonds, and location of the final double bond, relative to the ω carbon. Under this system, for instance, Docosahexaenoic Acid (DHA),a very important constituent of membranes in the brain and eyes, can by listed as 22:6 ω-3. This corresponds to: 22 carbons and 6 double bonds,of which the final double bond is at ω-3. This is equivalent to 22:6 $\Delta^{4, 7, 10, 13, 16, 19}$, or, in its full IUPAC form, cis-docosa - 4, 7, 10, 13, 16, 19 - hexaenoic acid.

10.2.2 Phosphoglycerides

Phosphoglycerides are composed of a glycerol back bone with substituents in the following arrangements:

- Hydroxyl #1 of glycerol is usually esterified to a saturated fatty acid
- Hydroxyl #2 of glycerol is usually esterified to an unsaturated fatty acid
- Hydroxyl #3 of glycerol is esterified to a phosphate group
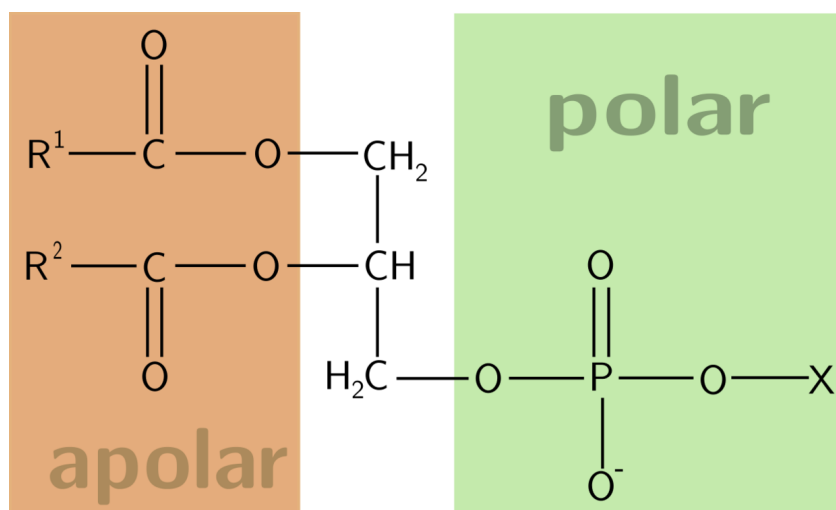
Figure 31

The simplest phosphoglyceride is phosphatidate (picture). Other phosphoglycerides can be made when a group with a hydroxyl is esterified to the phosphate group of phosphatidate. There are four common substituents for phosphatidate. Serine, ethanolamine and choline are structurally similar, while inositol is different:

Figure 32

Figure 33

When the fatty acid esterified to hydroxyl #2 is a cis- configured polyunsaturated fatty acid (i.e. one with more than one double bond), it tends to curl, and thus prevents the molecules from packing so closely together in a membrane. This makes the membrane more flexible.

10.2.3 Sphingolipids

Sphingolipids', have the same overall shape as phosphoglycerides but have different chemistry, using sphingosine in place of glycerol. Sphingosine has a long hydrocarbon tail similar to fatty acids attached to a structure that is similar to the amino acid serine. A fatty acid can attach to the amine group, and a "head" group can attach to a hydroxyl (see Figure x). Sphingolipids are named according to this head group:

- If there is no head group it is called a ceramide
- If the head group is phosphate and choline, it is called sphingomyelin
- If the head group is a sugar, it is called a glycosphingolipid (or a glycolipid)

The majority of sphingolipids are of the third type, glycosphingolipids. It is thought they have functions in cell recognition and protection in addition to their structural role in the membrane.

Sphingolipids are known to regulate activity in cells, such as immune responses, production of cells, and development of specialized cells. Although these are under the spatial and temporal control, it was recently discovered that sphingosine kinases will be focused on therapeutic effects on enzymes for people with cancer and other conditions.
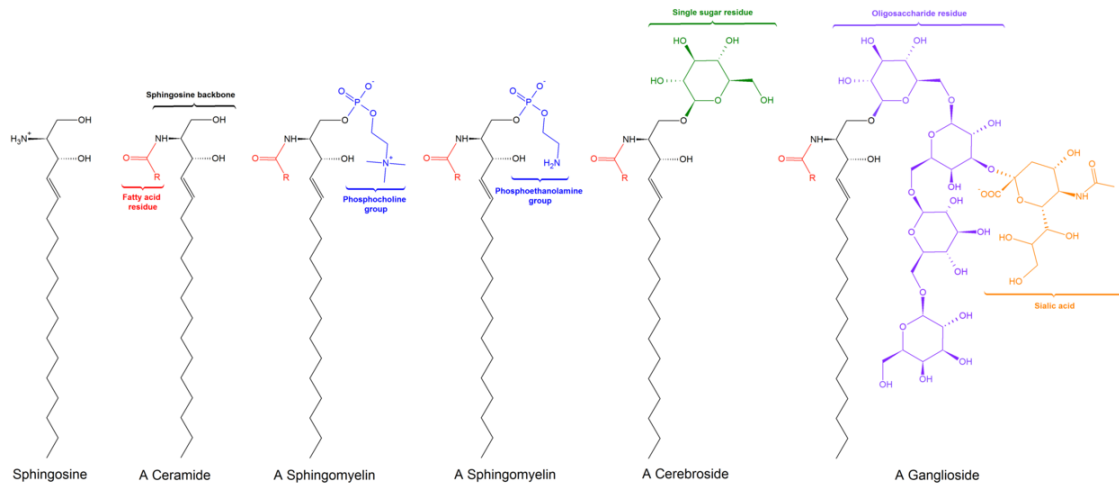
Figure 34   General structures of sphingolipids

10.2.4 Formation Of The Bilayer

If we compare the structures of phosphoglycerides and sphingolipids, we see that they are very similar compounds. Each lipid has two long hydrophobic hydrocarbon "tails" and a single polar "head". Since the molecule has both polar and nonpolar moieties, it is said to be **amphipathic**. It is the amphipathic nature of these molecules that causes them to form bilayers, mediated by four forces:

- The hydrophobic effect- this causes the hydrophobic tails to come together. This is the strongest force driving the formation of the bilayer. It is a consequence of the increased entropy in water molecules when non polar substances are aggregated in water.
- Van der Walls forces between the hydrophobic tails.
- Electrostatic forces of the head groups.
- Hydrogen bonds between the head groups.

One possible structure that satisfies the above forces is called a **micelle** (pictures). This is common with free fatty acids, but not with most phosphoglycerides and sphingolipids because these groups have twice as many acyl chains per head than the fatty acid (picture), and it is difficult to pack them all into the center of the micelle. Phospholipids and sphingolipids more often form a bilayer in a sheet or a sphere (picture). This is the so called lipid bilayer.

10.2.5 Lipid Motion

Studying Motion

NMR, ESR, X-Ray, Differential scanning calorimetry FRAP is a good way to measure diffusion of receptors through a lipid membrane after tagging the protein of interest with a GFP construct

Intramolecular Motion

There are three basic kinds of motion within the lipid molecules: stretching between bonds, rotating between bonds, and wagging between bonds (?). [picture] 99% of motion within liquid crystal is due to rotation about carbon-carbon bonds. Unsaturated fatty acids of membrane lipids rotate more often. This is because of the packed arrangement of the lipid bilayer. When there is rotation about one bond, an adjacent bond rotates to compensate for steric clashes [pictures]. Since double bonds in fatty acids are nearly always cis, they introduce kinks in the fatty acid. When a bond adjacent to a double bond rotates, the other bond adjacent to the double bond also rotates, and the whole thing moves like an old fashioned bit and brace [pictures]. It takes more energy to rotate the double bonds closer to the head groups due to [angle thing picture]. Double bonds react with O2 readily and create poisons [more] bacteria have [cyclopropane picture].

Types of Membrane Protein Diffusion

Rotational Lateral Liquid crystal Flip Flop

10.3 Plasma Membrane Function

10.3.1 Barrier

Interior of cell is separated from surrounding environment. to keep undesirable substances out keep desirable substances in intracellulat membranes:act to compartmentalize functions within eukaryotic cells(ex:mitochondria,chloroplast)

10.3.2 Transport Regulation

The lipid membrane is impermeable to many essential nutrients to the cell, such as glucose. In order to get the proper nutrients into the cell protein transporters are constantly moving through the membrane. For the example of glucose, there are several isoforms of the glucose transporter, some of which are specific for only certain types of cells. One of the more interesting transporter is the water transporter: aquaporins. Even though water can get through the membrane, at times the cells needs more water then can be provided through diffusion.

10.3.3 Cell Communication

Gap junctions are channels found between cells that allow for the cytoplasm to exchange between the cells. This is how very fast signalling can occur by cells close together.

10.3.4 Cell Adhesion

Several classes of proteins are responsible for cells sticking together: Cadherins

10.4 Summary

11 Lipids and the Plasma Membrane
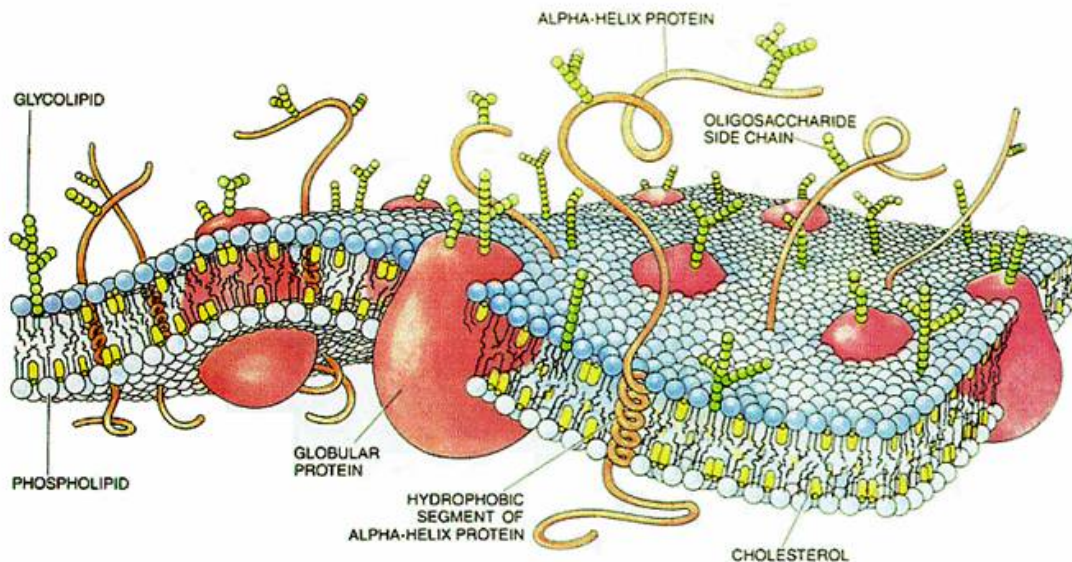
Figure 35

11.1 Membranes And Lipids

All cells, from simple prokaryotic bacteria to complex multicellular organisms are surrounded by a **membrane**. The membrane acts as a selective barrier, letting some substances into the cell and preventing other substances from entering,and it also actively transports substances between the inside and outside of the cell, using cellular energy to do so. This is important in regulating the concentration of many substances which must be maintained within strict limits. The cell also contains membrane bound compartments, where the membrane acts as a barrier for the separation of different environments, like lysosomes which have a high pH that would be toxic to the rest of the cell. Furthermore, lipids are non-polar compound and soluble in organic solvent. Their ability to form membranes are as a result of their hydrophobic properties, which is contributed by their fatty acids. Membranes are amphipatic.

11.2 The Lipid Bilayer

The first person to link lipids with the cell membrane may have been Charles Ernest Overton, who was studying heredity in plants. Part of his studies involved studying which substances

absorbed into plant cells most quickly. After characterizing a large number of substances, he came up with the idea that cell membranes were composed of something similar to the lipids found in vegetable oils, and that substances are absorbed into the cells by dissolving through the membrane. The next big step came when Gorter and Grendel extracted the lipids from red blood cells and compared the surface area of the lipids spread out on water compared to the surface area of the red blood cells. They found the lipid surface area was twice that of the red blood cells, and concluded that the lipids must be arranged in a layer two lipid molecules thick. Lipid bilayer is a double layer membrane formed from phospholipids. Phospholipids are composed of a polar head group and non-polar fatty acid tails. The arrangement of the phospholipids makes the cell membrane permeable.

11.2.1 Fatty Acids

A fatty acid is a linear of carboxylic acid with a long hydrocarbon chain. In fatty acids, the non-polar hydrocarbon chain gives the molecule a non- polar character Fatty acids usually have between 14 and 24 carbon atoms and their carbon chain may have one or more double bonds. In naturally occurring fatty acids, these double bonds are mostly in cis configuration. Fatty acids have both common names and systematic names. The systematic name is based on the alkane or alkene with the same number of carbon atoms, with the final *e* of the hydrocarbon replaced with *oic acid*, if the carbon chain of the fatty acid is saturated (without double bond in its carbon chain), and "enoic acid" if there is double in its carbon chain. For example Laurate (Figure x)

Figure 36

is a fatty acid with 12 carbon atoms and no double bonds, so the systematic name is dodecanoic acid, and the ionized form is dodecanoate. (saturated fatty acid table) If there are double bonds, the location is marked with the symbol Δ with a superscript number indicating the location of the double bond and preceded by *cis* or *trans* to indicate configuration (but almost invariably *cis*). Carbon atoms are counted from the carboxyl end, as in Figure x, so a dodecenoate acid with a cis double bond between carbon atoms 9 and 10 would be: *cis*-Δ^9-dodecenoate. The number 2, 3 and last carbon atom are called the α, β and ω atoms, respectively.

Biological fatty acids usually contain an even number of carbon atoms, with numbers of 16 and 18 being most common. The length of the fatty acid chain and the degree of saturation contributes largely to their distinct properties. The shorter the chain length and the more unsaturation in the fatty acid enhances their fluidity, thus lowering their melting point.

11.2.2 Phosphoglycerides

Phosphoglycerides are esters of two fatty acids, phosphoric acid and a trifunctional alcohol-glycerol. The fatty acids are attached to the glycerol at the 1 and 2 positions on glyc-

erol through ester bonds. Phosphoglycerides are composed of a glycerol back bone with substituents in the following arrangements:

- Hydroxyl #1 of glycerol is usually esterified to a saturated fatty acid
- Hydroxyl #2 of glycerol is usually esterified to an unsaturated fatty acid
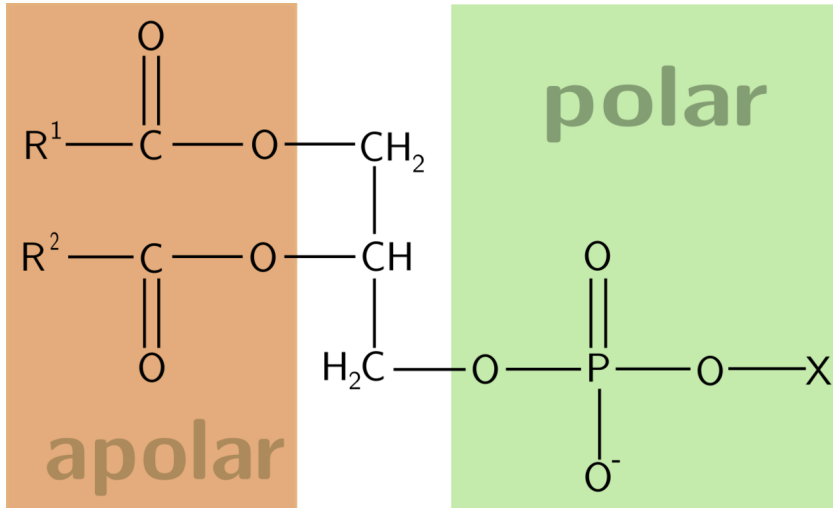- Hydroxyl #3 of glycerol is esterified to a phosphate group

Figure 37

The simplest phosphoglyceride is phosphatidate (picture). Other phosphoglycerides can be made when a group with a hydroxyl is esterified to the phosphate group of phosphatidate. There are four common substituents for phosphatidate. Serine, ethanolamine and choline are structurally similar, while inositol is different:
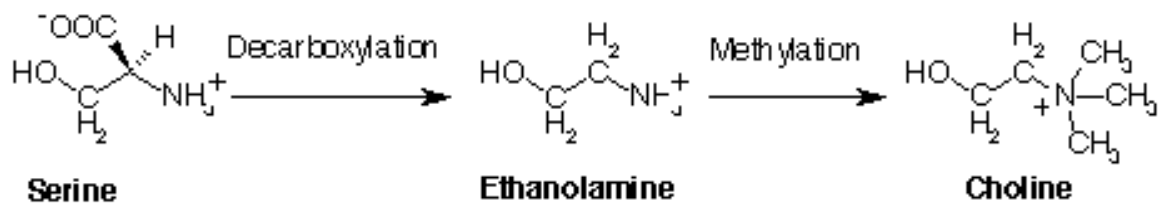
Figure 38
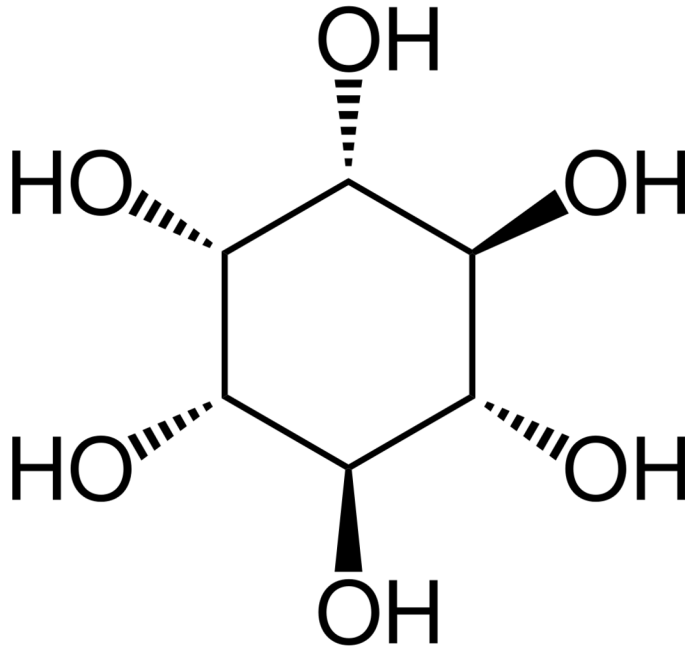
Figure 39

11.2.3 Sphingolipids

Sphingolipids are a second type of lipid found in cell membranes, particularly nerve cells and brain tissues. They do not contain glycerol, but retain the two alcohols with the middle position occupied by an amine and have the same overall shape as phosphoglycerides but have different chemistry, using sphingosine in place of glycerol. Sphingosine has a long hydrocarbon tail similar to fatty acids attached to a structure that is similar to the amino acid serine. A fatty acid can attach to the amine group, and a "head" group can attach to a hydroxyl (see Figure x). Sphingolipids are named according to this head group:

- If there is no head group it is called a ceramide
- If the head group is phosphate and choline, it is called sphingomyelin
- If the head group is a sugar, it is called a glycosphingolipid (or a glycolipid)

The majority of sphingolipids are of the third type, glycosphingolipids. It is thought they have functions in cell recognition and protection in addition to their structural role in the membrane.

11.2.4 Formation Of The Bilayer

If we compare the structures of phosphoglycerides and sphingolipids, we see that they are very similar compounds. Each lipid has two long hydrophobic hydrocarbon "tails" and a single polar "head". Since the molecule has both polar and nonpolar moieties, it is said to

be **amphipathic**. It is the amphipathic nature of these molecules that causes them to form bilayers, mediated by four forces:

- The hydrophobic effect- this causes the hydrophobic tails to come together. This is the strongest force driving the formation of the bilayer. It is a consequence of the increased entropy in water molecules when non polar substances are aggregated in water.
- Van der Walls forces between the hydrophobic tails.
- Electrostatic forces of the head groups.
- Hydrogen bonds between the head groups.

One possible structure that satisfies the above forces is called a **micelle** (pictures). This is common with free fatty acids, but not with most phosphoglycerides and sphingolipids because these groups have twice as many acyl chains per head than the fatty acid (picture), and it is difficult to pack them all into the center of the micelle. Phospholipids and sphingolipids more often form a bilayer in a sheet or a sphere (picture). This is the so called lipid bilayer. Lipid bilayer is permeable to water molecules and other small, uncharged molecules like oxygen, carbon dioxide. A bimolecular sheet formed by amphipathic molecules in which the hydrophobic moieties are on the inside of the sheet and the hydrophilic ones are on the aqueous outside.

11.2.5 Lipid Motion

Studying Motion

NMR, ESR, X-Ray, Differential scanning calorimetry

Intramolecular Motion

There are three basic kinds of motion within the lipid molecules: stretching between bonds, rotating between bonds, and wagging between bonds (?). [picture] 99% of motion within liquid crystal is due to rotation about carbon-carbon bonds. Unsaturated fatty acids of membrane lipids rotate more often. This is because of the packed arrangement of the lipid bilayer. When there is rotation about one bond, an adjacent bond rotates to compensate for steric clashes [pictures]. Since double bonds in fatty acids are nearly always cis, they introduce kinks in the fatty acid. When a bond adjacent to a double bond rotates, the other bond adjacent to the double bond also rotates, and the whole thing moves like an old fashioned bit and brace [pictures]. It takes more energy to rotate the double bonds closer to the head groups due to [angle thing picture]. Double bonds react with O2 readily and create poisons [more] bacteria have [cyclopropane picture]. Furthermore, NMR rotating frame relaxation studies of the intramolecular motion in methionine-enkephalin.

Diffusion

One of the most notable properties of the lipid membrane is the ability of a single lipid to diffuse. This property was discovered using the "fluorescence recovery after photobleaching," also known as FRAP. In this experiment, the cell surface is first labeled with a fluorescent chromophore. Afterwards, a specific region is then bleached with intense light, leaving a

prominent mark. In the bleached area, the experimenters noticed that as time progressed, bleached molecules moved out from the bleached area and unbleached molecules moved towards the bleached area. This shows that the lipid bilayer allows for molecule movement within the membrane. There are two kinds of diffusion: lateral and transverse (flip-flop). The lateral diffusion is exactly as it sounds like. In a lipid bilayer, one lipid molecule can move pass the adjacent one. The transverse molecule works a bit differently. It still displaces one over its adjacent counterpart, and crosses over to the other side of the lipid bilayer. It is important to note that the lateral diffusion is substantially faster than the transverse. Studies show that the transverse diffusion only happens once every few hours. Diffusion is the movement of a particle from area of high concentration to low concentration, and it is not to be confused with osmosis, which is the movement of water through a semi-permeable membrane. Diffusion depends on CO2 and O2 concentrate inside and outside of alveolar sack.

11.3 Plasma Membrane Function

11.3.1 Transportation Methods

The membrane regulates molecule transport through a variety of means. There are two general categories that each transportation regulation falls into: passive transport and active transport. Passive transport is the movement of molecules down its concentration gradient: in other words, higher concentration to lower concentration. The diffusion of water, a special case, is called "osmosis." Not all molecules can diffuse through the phospholipid bilayer. The ones that can are usually hydrophobic molecules (i.e. oxygen), nonpolar molecules (i.e. benzene), or small uncharged polar molecules (i.e. water). The hydrophobic and nonpolar molecules can dissolve through the bilayer because of the similar polarites of the bilayer and the molecule itself. The small uncharged polar molecules usually require a membrane channel to diffuse i.e. aquaporins for water. The molecules that do not pass through the layer with ease are large & uncharged, polar, or ions. Examples of each are glucose, sucrose, and hydrogen ion, respectively. To sum up the defining characteristics of what passes or does not pass through the membrane, they are size, charge, and polarity. Normally, hydrophilic molecules would not be able to cross the phospholipid bilayer due to the difference in polarities. However, it is observed that hydrophilic molecules do cross the membrane protein through a different mechanism than the hydrophobic molecules. While hydrophobic molecules diffuse through the membrane due to their similar polarities, hydrophilic molecules require a molecule called a membrane protein (integral or peripheral), which transfers the molecule in both active and passive transport. A few examples of passive transport are simple diffusion and facilitated diffusion. Simple diffusion is when a solute moves from an area of high concentration to an area of low concentration to establish equilibrium via a channel protein. Facilitated diffusion still moves from an area of higher concentration to lower concentration but instead of a channel protein, it is a carrier protein. This carrier protein, also called permeases, accelerates the rate of transport greatly. Active transport moves the molecules from an area of low concentration to an area of high concentration (the opposite of passive transport). This transport moving molecules up its concentration gradient requires energy, usually from ATP (adenosine triphosphate). A common example used to explain this concept is the sodium-potassium pump. In the human cell, there is usually a high

concentration of sodium on the outside the cell and a low concentration of sodium inside the cell. The opposite applies to the concentrations of potassium: high on the inside and low on the outside. The mechanism begins by sodium ion binding to the sodium-potassium pump protein with its gap close off from the cell's external environment. The binding of sodium ion signals to the cell to use convert ATP to ADP (adenosine triphosphate). This reaction will release energy and add a phosphate group to the protein, enabling the protein to change its conformation and release the bound sodium ions to the external cell environment. After the release of sodium ions, potassium ions from the same area will bind to the protein. The potassium ion binding will signal to the protein to convert the bound phosphate group to unbound inorganic phosphate and change the conformation of the protein to release the potassium ions into the cell. Afterwards, the sodium ions bind again and the cycle repeats. A few other energy-driven processes are endocytosis and exocytosis. Endocytosis are when large molecules are transported into the cell. Exocytosis are when molecules are expelled from the cell. An example of receptor-mediated endocytosis occurs in eukaryotes. It is important for the different membranes in the cell to either join or separate in order to perform certain functions such as engulf, transport, or even release molecules essential for the body. This process is facilitated by LDL, a low-density lipoprotein. External cellular LDL float first binds to its corresponding receptor, called the LDL receptor. The cell membrane then engulfs the LDL and the LDL receptor into the cell, forming a vesicle to keep it unharmed from the cell hydrophilic environment. The complex then separates to protein and receptor counterparts. The LDL fuses with a lysosome, breaking down the LDL with the release of cholesterol as a by-product. A short example of exocytosis would be either the insulin secretion in the blood, or the transport of neurotransmitters at the gap junction during an action potential. A more specific example lies in Golgi's vesicular transport. In eukaryotes, the endoplasmic reticulum (ER) sends transport vesicles to the Golgi apparatus created from proteins. Some of the vesicles fuse with the lysosomes resulting in digestion. Some of the other transport vesicles with essential proteins for the plasma membrane will diffuse in to the membrane due to their similar polarities, releasing the protein content out into the cell's environment. Different concentrations with regards to internal and external cell environment have different effects on animal cells and plant cells. In a hypertonic solution (concentration of solute is greater on the outside than on the inside), the cell would attempt to create an equilibrium by releasing its water content to the external cell environment. By doing so, the concentrations may be equal if there is enough water in the cell. Also, the cell would end up being shriveled. In the plant cell, the same things occurs. However, because plant cells have cell walls, instead of being shriveled, it is plasmolyzed. On the other extreme, hypotonic solution, when the concentration is solute is lower on the outside than the inside, the cell will become lysed. The attempt to establish equilibrium lies in the water's entrance into the cell to lower the concentration in the cell. In the plant cell, this is called turgid instead of lysed. On a sidenote, this is the normal preference of the plant cell. In an isotonic solution (concentrations are equal on both sides of the cell), the net content of water in and out of the cell remains the cell. This is the eukaryote's preferred state. In the plant cell, it is termed flaccid. Membrance proteins called transporters facilitate the passage of molecules across membrance, there is involved at least three step: dinding, conformational change fo the protein, and release. There are two types of transporter: passive transporters which use no energy, and active transporters that employ ATP to drive transport.

11.3.2 Cell Communication

cell-to- cell communication is essential for multicellular organisms such as human or an oak creatures in which they must communicate in order for them to develop from fertilized egg and then survive and reproduce in turn. Communication between cells is very important for many unicellular organisms that must find matters and food in order for them to develop and to sexually reproduce. An early example of cell communication is Saccharomyces cerevisiae in which the cells of that yeast use chemical signaling to identify cells of opposite mating and to start the mating process. The study of cell communication could help to answer some of the most important questions and medicine in areas ranging between embryological development to hormone action to the development of cancer and other related diseases. On a side note, not only is cell-to-cell communication imperative, but also with other molecules. Different kinds of molecules communicate differently with the internal programming of the cell. For example, synaptic transmitters, such as acetylcholine, bind to a certain G-protein and activates intracellular 2nd messengers. Another way for synaptic transmitters to bind is via a protein complex (integral/membrane protein) including an ion channel. The net result of both pathways change the state of ion channel conformation in the membrane. Another example, steroid hormones, diffuse through the cell membrane and eventually changes the protein programming in certain cells. As for the peptide hormone, it binds to a G-protein, activates intracellular 2nd messengers, and changes the transcription paradigm. Due to the peptide hormone's hydrophobic character and the cell membrane nonpolarity, it requires secondary messenger interaction whereas the steroid hormone (also nonpolar) can directly communicate and diffuse into the cell. Cell-cell communication is essential for multicellular organisms. Cells usually communicate by releasing chemical messenger targeted for cell. Some messengers travel short distances such as molecules are alled local regulators. Animal growth factors which are compounds that stimulate nearby target cells to grow and multiply. Numerous cells can simulaneously receive and respond to the molecules of growth factors produced by single cell in their vicinity.

11.3.3 Cell Adhesion

Cell adhesion is any glandular cells in ctenophores, turbellarian used for adhesion to a substrate and for capture. Cell adhesion occurs when one cell binds to another surface such as another cell or some other inanimate surface. Cell adhesion molecules serve as intermediates that hold the cell to another surface.

11.4 Summary

12 Complex Lipid Metabolism and Cholesterol

12.1 Introduction

Dietary fats serve a variety of function in the body. One of the most important function is as a metabolic fuel, per gram, fats yield twice as much energy as carbohydrate. All steroid hormones are derived initially from lipid, and these in turn are responsible for the regulation of body functions such as growth, development and the appearance of sexual characteristics as a result of sex hormones. Essential fatty acids, such as linoleic acid and α-linolenic acid are prostaglandin and ecosanoid precursors.

12.2 Lipid Droplets (LDs)

The LDs takes part of a hydrophobic phase in the cytosol and are composed of an organic phase of neutral lipids. This hydrophobic core stores metabolic energy and membrane components making them the central region for lipid metabolism. They also take part in protein storage, degradation, and viral replication. The formal definition for Lipid Droplets is "the cytoplasmic organelle composed of a hydrophobic core of neutral lipids bounded by a phospholipid monolayer and specific proteins" [1]

12.2.1 How were they Discovered

These droplets were found in the nineteenth century by the method of light microscopy of cellular organelles. Lipid Droplets were often called liposomes until the in vitro vesicles were generated in 1960's and this new vesicles were named liposomes. Since this moment the Lipid Droplets were referred as lipid bodies, fat bodies, spherosomes, or simply LDs. LDs were mainly ignored on research until a protein that specifically localized to LD surface was identified in 1991 [1]

12.2.2 Where are they found?

LDs are found in most cells. LDs can be found among bacterias and eukaryote, although their number and size vary greatly in different cell types.

12.2.3 What is their functions?

The LDs provide building blocks for membranes for energy metabolism. LDs have the ability to package hydrophobic lipids, such as TGs, without water, providing a very efficient form of energy storage. It has also been found that there are links between lipid synthesis pathways and LDs. LDs also serve as organizing center for specific lipids and have been linked to protein storage with the possibility of also storing unfolded membrane proteins. LDs have also been found to be involved in the hepatitis C virus assembly.

12.3 Digestion and Absorption of Lipids

Lingual Lipase

Short Chain Fatty Acids Long Chain Fatty Acids

12.4 Bile

Primary Bile Acids Bile Salts Secondary Bile Acids

12.5 Chylomicron Formation

Micelles

12.6 Very Low Density Lipoprotein (vLDL)

12.7 Low Density Lipoprotein (LDL)

Low density lipoprotein (LDL) is known as bad cholesterol. When the level of LDL is high in the blood, it could slowly build up inside the arteries that pump the blood to the brain and the heart. LDL and other substances can cause atherosclerosis. Atherosclerosis is a disease that happened when the arteries became less flexible and narrow due to the plaque that LDL and other substances form.

12.8 High Density Lipoprotein (HDL)

Cholesterol can not dissolved in the blood there for it has to be transported in and out of the cells by carrier called lipoprotein. There are two type of lipoprotein: low-density lipoprotein (LDL) and high-density lipoprotein HDL. high density lipoproteins (HDL) are known as good cholesterol because high level of HDL could prevent from heat attacks. Some

scientists believe that high-density lipoprotein (HDL) slow the build up of plaque in which it removes cholesterol from arteries.

13 Reference

[1] Walther, Tobias C., and Robert V. Farese, Jr. "Lipid Droplets and Cellular Lipid Metabolism." National Center for Biotechnology Information. U.S. National Library of Medicine, n.d. Web. 06 Dec. 2012. $<$ [1] $>$.

---

1   http://www.ncbi.nlm.nih.gov/pubmed/22524315

14 Enzymes

14.1 Introduction

Enzymes are proteins that act as catalysts in most of the body's biochemical reactions that have catalytic functions indispensable to maintenance and activity of life. A small subset of enzymes are composed of RNA and are known as **ribozymes**. The general mechanism by which enzymes work is by reducing the energy required to start the reaction, otherwise known as the activation energy. Enzymes have extremely varied uses: ethanol production, forensic science, and polymerase chain reaction amongst them. And there are all chemical reactions occurring in a living organism which are dependent on the catalytic actions of enzymes. Biologists aren't the only people interested in enzymes, however. These diverse molecules form a huge part of industries such as brewing and pharmaceuticals; the use of enzymes alone in the brewing process of Guinness is worth over a billion euro to the Irish economy.

14.2 Structure of Enzymes

Enzymes are composed of many amino acids, joined in long chains, forming proteins, which are manufactured in the cell(s) of every organism. Furthermore, enzymes are globular proteins, folding conformation creates the active site. These long amino chains fold in specific ways, so as to make the reactions the enzyme catalyses specific to that particular enzyme.The nature and arrangement of amino acids in the active site make it specific for only one type of substrate.

14.3 Mechanism of Enzymes

The mechanism of enzymes work is by lowering the activation energy of the reaction the specific enzyme is to catalyse. Each enzyme is entirely specific to the reactants; it will only work for one specific reaction, this is due to the unique active site of the enzyme. The enzyme's active site is in a unique 3D shape, with a unique pattern of electrostatic charge and hydrophobicity. Here the specific substrates, the substances upon which the enzyme works, are temporarily combined with the enzyme, forming an enzyme substrate complex, or ESC. The reaction then occurs, and the product of the reaction separates from the enzyme, at which point it may be used. The enzyme is then capable of repeating the reaction many thousands of times, although they may have to be replaced every so often. **Activity of Enzymes** Control of Enzyme activity in Cell The production of enzyme can be enhanced or diminished by changing the internal environment of the cell. This is called

enzyme induction and inhabitation. The enzyme inhibitor or the enzyme inductor will be bound to the enzyme and either decrease or increase the activity of the enzyme. The inhibitor stops the entre of substrate to the active site. It can be reversible or irreversible. Enzyme activities are compartmentalized that means reduce the activity of one enzyme can lower the productivity of other since they are linked. Some enzymes will be activated by the environmental factor in side your body, for example the lowering of pH changes the activity of hemagglutinin in the influenza. Thus,the lower the potential energy barrier to reaction, the more reactants have sufficient energy and, hence, the faster the reaction will occur. All catalysts, including enzymes, function by forming a transition state, with the reactants, of lower free energy than would be found in the uncatalysed reaction.

Enzyme Graph Enzymes do not modify the equilibrium of a reaction. Enzymes only catalyze or speeds up the rate of reaction by stabilizing the transition state and do not participate in the chemical reaction. The transition state is the highest activation energy (Ea) on a reaction coordinate in which the molecule is only partially reacted. The activation energy is lowered when a catalyst (enzyme) is used. When an enzyme is not used, the activation energy of a reaction is much higher, which will take the reaction much longer to proceed. The enzyme is complementary to the transition state and not the substrate, in order to facilitate the reaction efficiently. A catalyst is present in the reactants and also appears in the end products.
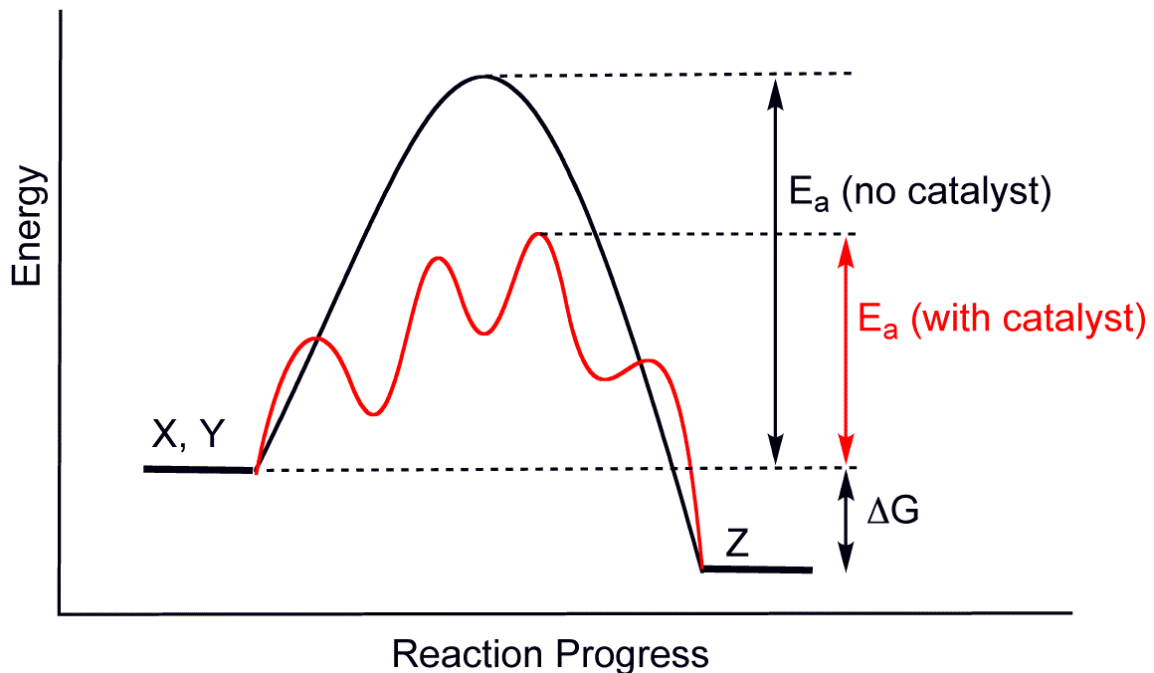

Figure 40

14.4 Properties of Enzymes

1. Enzymes are sensitive to heat, and are denatured by excess heat or (inactivated by excess cold), i.e. their active site becomes permanently warped, thus the enzyme is unable to

form an enzyme substrate complex. This is what happens when you fry an egg, the egg white (augmenten, a type of protein, not an enzyme), is denatured. 2. Enzymes are created in cells but are capable of functioning outside of the cell. This allows the enzymes to be immobilised, without *killing* them. 3. Enzymes are sensitive to pH, the rate at which they can conduct reaction is dependent upon the pH of where the reaction is taking place, e.g. pepsin in the stomach has an optimum pH of about pH2. Whereas salivary amylase has an optimum pH of about 7. 4. Enzymes are reusable, and some enzymes are capable of catalysing many hundreds of thousands of reactions every second, e.g. catalase working on hydrogen peroxide. 5. Enzymes will only catalyse one reaction, e.g. invertase will only produce glucose and fructose, when a glucose solution is passed over beads of enzyme (see Immobilisation). 6. The reaction rate for an enzyme is limited by its saturation point. Saturation is essentially the point at which an enzyme can no longer speed up the reaction to compensate for the increase in substrate concentration (push the equilibrium towards the products). Graphically this is portrayed as a horizontal asymptote and is also called the maximum velocity. Each enzyme has its own maximum velocity for a given substrate. Measuring the rate of product formation with varying concentrations of substrate is one way of determining the maximum velocity of an enzyme. 7. Enzymes are capable of working in reverse, this acts as a cut off point for the amount of product being produced. If there are excess reactants the reaction will keep going and be reversed, so that there is no overload or build up of product. Enzymes are catalysts in the breaking down of substances by bonding with them briefly. Hydrogen peroxide they break it down into water and oxygen much faster than it would naturally occur. Inside the body they are used to break down food and to start off the digestion process. They are proteins they are affected by the pH level of the substance and also the temperature of the substance. The last important note is that obviously the concentration of the enzyme will affect how well it works.They are substrate specific which is illustrated by the lock and key hypothesis. Enzymes may be immobilised in two ways, firstly by bonding them chemically to some sort of rigid structure, and secondly by physically placing the enzymes on an insolube support structure, e.g. a gel or bead. The second method of enzyme immobilisation is much more common, is cheaper, but it does reduce the enzyme's ability to catalyse reactions.

Steps in Enzyme immobilization 1. Make up a 5% w/v solution of sodium alginate, and leave to stand overnight, to mix properly. 2. Add the enzyme, or in this case, a cell e.g. yeast, to the alginate solution, and mix in well. At this point the mixture will be very thick, and should be left for twenty minutes to eliminate the air bubbles, from the solution; such air bubbles would increase the surface area of the beads, making them less efficient. 3. Take a syringe, and fill it with the alginate/cell matrix. 4. Make a 2% w/v solution of Calcium Chloride. 5. Hold the syringe about 20 cm perpendicularly above the calcium chloride solution, and drip in the alginate solution, trying to create the smallest possible beads, approx 3-4mm in diameter. 6. Remove and discard any alginate strings or floating beads from the chloride solution. 7. Rinse the beads in deionised water. 8. Using a retort stand set up two tap funnels above each other, placing the beads in the bottom funnel, and strong glucose in the top. 9. Allow the glucose to run through the beads, and draw off the product. The product is a type of alcohol, which should not be ingested as it is impossible to tell whether it is ethanol or methanol, the later being toxic.

14.5 Naming Enzymes

Until quite recently enzymes have been given arbitrary names, for example, ptylin (amylase), this tells us nothing of what the enzyme acts upon, instead biologists have devised a means of naming enzymes, whereby enzymes end -ase; e.g. DNA Helicase, they have also included a clue as to what the enzyme acts upon; DNA Helicase working to separate nucleotides in a chain of DNA. Other common enzymes include protease, lipases, pepsin, amylase, and lysomase.

14.6 References

Berg, Jeremy M. Biochemistry 6th Edition. New York, W. H. Freeman and Company, 2007.

15 Metabolism

Biochemistry[1] << Enzymes[2] | DNA and RNA >>[3]

What is life? What makes life work? Life is not static; it is defined by activity. A cell has to perform certain tasks to stay alive. This activity has to be powered by fuel; the fuel is converted into energy, to keep the cell running, building blocks for new biomolecules, and waste products. This process is called **metabolism**.

15.1 ATP as an intermediate energy carrier

We have already seen[4] how ATP is a universal "energy currency" in the cell, and how its high-energy phosphate can store and release this energy efficiently. But ATP is only a short-term energy storage; a hard-working human can use up to one pound of ATP per minute. How can that be? The answer lies in the medium- and long-term energy storage molecules, notably *phosphoenol pyruvate* (PEP), *acetyl phosphate*, and *creatine phosphate*. Each of these molecules carries a phosphate group with an ever higher transfer potential than that of ATP. ATP is continuously regenerated by transferring phosphate groups from these molecules to ADP. For example, the reaction of creatine phosphate is

$$\text{Creatine phosphate} + \text{ADP} + \text{H}^+ \leftrightharpoons \text{ATP} + \text{creatine}$$

a reaction which is catalyzed by the creatine kinase, and highly in favor of the "ATP side" of the equation.

15.2 Electron carriers

Most of the energy of eucaryotic lifeforms derives from oxidating fuel molecules, that is, the transfer of electrons from fuel molecules to oxygen. The fuel molecules are oxidized, while the oxygen is reduced. A rather radical form of oxidation is fire, which oxidizes its fuel directly with oxygen. As spontaneous combustion one ones cells is rather unpleasant, the electrons are carried through several steps from the fuel molecules to the final electron acceptor, oxygen. The flow of electrons causes a proton gradient in the mitochondrial membranes, which in turn is used by the enzyme *ATPase* (or *proton pump*) to generate ATP. This process, utilizing an *electron transport chain*[5], is called *oxidative phosphorylation*. The

---

1   http://en.wikibooks.org/wiki/Biochemistry
2   http://en.wikibooks.org/wiki/Biochemistry/Enzymes
3   http://en.wikibooks.org/wiki/Biochemistry/DNA_and_RNA
4   http://en.wikibooks.org/wiki/Biochemistry/Metabolism_and_energy
5   http://en.wikibooks.org/wiki/Biochemistry/Electron_Transport_Chain

electron carrier molecules of choice are *NADH* and *FADH₂*. They are *pyridine nucleotides* (also called *flavions*).

	Reduced form	**Oxidized form**
NADH	NADH	NAD$^+$
FADH₂	FADH$_2$	FAD

Both molecules are also for biosyntheses that require reducing power. Especially a variant of NADH, *NADPH*, is used for this purpose. With all these high-energy bonds and electron-carrying molecules around, one would expect to see wild, uncontrolled chemical reactions occurring in the cell. However, the opposite is the case: *Without a catalyst*, ATP is quite slow to hydrolize and give its phosphate group to water. Also, all three electron-carrying molecules, NADH, NADPH, and FADH$_2$, are slow to react with O$_2$ *without a catalyst*.

15.3 Cancer Cell Metabolism

Glutamine addiction: a new therapeutic target in cancer The Warburg observation that cancer cells use up an unusually high level of glucose and produce more lactic acid has been around since the 1920's. It wasn't noted till 1955 by Eagle, that cancer cells also use up a high level of the amino acid glutamine. In the article, Glutamine addiction: a new therapeutic target in cancer, Wise and Thompson highlight the role glutamine plays in cancer cell growth, protein translation, anaplerosis, and macromolecular synthesis. Glutamine is the chief donor of nitrogen to cancer cells for the cells to multiply. When glutamine gives up its amide, it is converted into glutamic acid, which is the main nitrogen donor for nonessential amino acid synthesis. This amino acid biosynthesis makes glutamine a main ingredient for the cancer cells' need for protein translation. Stemming off of biosynthesis is Myc, a basic protein that binds 11 of the genes involved in nucleotide biosynthesis. Myc activation is a commonly observed in cancer and is identified as the driving force behind the spread of lymphomas and small cell lung cancer. Myc facilitates glutamine consumption and supports the change of glutamine into glutamic acid, and finally into lactic acid. This satisfies the Warburg observation of the unusual lactic acid production of cancer cells. The target of rapamycine (TOR) controls many cell functions such as cell growth, reproduction, motility, and protein synthesis. The "master regulator of protein translation, the mammalian target of rapamycin complex (TOR), is responsive to glutamine levels" (Wise and Thompson, 2010). This was initially discovered in yeast, where TOR activation was found to be dependent on glutamine levels. The article also describes how glutamine adds to macromolecular (DNA, RNA, and proteins) synthesis by producing NADPH (nicotinamide adenide dinucleotide phosphate, a reducing agent in anabolic reactions) and providing the cancer cell with a substrate source which refills the mitochondrial carbon pool (anaplerosis). This refilling of the carbon pool is necessary for the synthesis of proteins, lipids, and nucleotides. All of these facts relating glutamine importance to cancer cells may be a potential target in altering cancer cell metabolism. However, it is still unknown if this is a viable target for the treatment of cancer. Ideas include suppressing the glutamine uptake, TOR activation, or the anaplerosis of the cancer cells. Using an enzyme to lower blood glutamine levels is another possible method.

References Wise & Thompson, Glutamine addiction: a new therapeutic target in cancer, 2010, Department of Cancer Biology, University of Pennsylvania, Elsevier Ltd.

15.4 Glycolysis

Biochemistry/Glycholysis[6]

15.5 Pyruvate Dehydrogenase

15.6 Pyruvate Dehydrogenase

Metabolism, as all other functions in the human body, is not a simple mechanism. Although performed extremely fast, metabolism has many complex steps. For the multiple steps, there are multiple enzymes catalyzing the process. One of the steps of metabolism is taking pyruvate (from glycolysis) and converting it into CO2 and acetyl-CoA (used in the krebs cycle). In order to perform this action, so it will not be a lengthy process, an enzyme is necessary. The enzyme that catalyzes this action is pyruvate dehydrogenase. Aside from being an enzyme catalyzing the complex steps of metabolism, pyruvate dehydrogenase too is very complex. The pyruvate dehydrogenase is an extremely complex structure made of several enzymes: E1, E2, and E3. Since it does contain a complex structure, these enzymes are present within the structure multiple times. Overall, pyruvate dyhydrogenase is a dehydrogenase. As most dehydrogenase do, pyruvate dehydrogenase performs it job with the cofactor of NAD+. NAD+ performs its function by converting NAD+ to NADH+H^+.

15.7 Pyruvate Dehydrogenase Deficiency

As previously stated, pyruvate dehydrogenase is the enzyme in charge of changing pyruvate into acetyl-CoA (acetyl coenzyme A), a necessity to start the Krebs's cycle. The question at hang is can pyruvate dehydrogenase go wrong? The answer is yes, and although uncommon, pyruvate dehydrogenase deficiency is an example of what occurs when pyruvate dehydrogenase cannot perform its job. The question left unanswered is what pyruvate dehydrogenase deficiency is? Pyruvate dehydrogenase deficiency is a buildup of lactic acid, or lactic acidosis, which can be life-threatening for many. The most common forms of this lactic acid buildup can cause symptoms such as nausea, abnormal heartbeat, and vomiting. Pyruvate dehydrogenase is also characterized by varies neurological problems. The most common issues occurring with neurological abnormalities are slow mental abilities and motor skills such as sitting. Other neurological problems range from having poor coordination to difficulty walking to being intellectually disabled.

---

[6] `http://en.wikibooks.org/w/index.php?title=Biochemistry/Glycholysis&action=edit&redlink=1`

15.8 References

Diwan, Joyce J. 2007. *Pyruvate Dehydrogenous and The Kreb's Cycle.* Web. [7] *Pyruvate Dehydrogenous and The Kreb's Cycle.* College of Science / Microbiology. Aug. 3, 1999. Web. [8] Genetics Home Reference. *Pyruvate dehydrogenase deficiency.* Nov. 19. 2012. Web. [9]

15.9 Tricarboxylic Acid Cycle

Biochemistry/Tricarboxylic Acid Cycle[10]

15.10 Electron Transport Chain

The **electron transport chain** is a system of molecules through which electrons are transferred to generate ATP. It has an important role in both photosynthesis and cellular respiration. **ETS in photosynthesis** In photosynthesis, when sunlight is absorbed in photosystem 2, electrons are energized. They are transferred to the reaction center. From the reaction center, the electrons enter the electron transport chain and pass the etransport chain molecules. Then the 2 deenergized electrons are reenergized in Photosystem 1 (is second because PSII was discovered first)and they go to the NAPD+ reductase which transfers ther electrons to a coenzyme, converting it to NADPH+ The two electrons used must be replaced, so water is broken down producing 2 protons (H+) which concentrate in the thylakoid membrane, 2 electrons replaced in PSII and oxygen released as O2. Protons go down proton pumps and a concentration gradient forms as protons move from the stroma into the thylakoid space. Protons move down gradient through ATP synthase which forms ATP. Energy is stored. **ETS in respiration** In respiration, an electron transport chain is used to break the fall of electrons to oxygen into several energy-releasing steps. It slows down the releasing process of energy. There are a number of molecules that consists an electron transport chain. Most of these molecules are proteins, built into the inner membrane of mitochondria of eukaryotic cells and the plasma membrane of aerobically respiring prokaryotes. On the "top" end of the chain, where the energy is highest, electrons removed from oxidation of glucose are shuttled by NADH. On the "bottom" end, where the energy is lowest, Oxygen molecules pick up the electrons, and together with protons existing in the cellular environment, oxygen molecules form water. The total energy released from the oxidation of glucose by oxygen is huge, about 2880kJ/mol. (The overall oxidation of glucose is shown by reaction C6H12O6 (aq) + 6 O2 (g) $\rightarrow$ 6 CO2 (g) + 6 H2O). This energy can't be picked up by cells in a short amount of time. Therefore, electron transport chain applies a cascade of reactions to slowly release the energy, maximizing the usage of glucose. In the cascade, each "downhill" carrier is more electronegative than their "uphill" neighbors, so that the electrons can be passed "downhill" by oxidation. Oxygen is the last electron acceptor.

---

7   http://www.rpi.edu/dept/bcbp/molbiochem/MBWeb/mb1/part2/krebs.htm

8   http://www.science.siu.edu/microbiology/micr425/425Notes/05-PyrKrebs.html

9   http://ghr.nlm.nih.gov/condition/pyruvate-dehydrogenase-deficiency

10   http://en.wikibooks.org/w/index.php?title=Biochemistry/Tricarboxylic_Acid_Cycle&action=edit&redlink=1

There are four unique electron carrier complexes in the respiratory chain:

Complex I: NADH dehydrogenase

- Composed of 42 different polypeptide chains.
- Has several Fe-S centers.
- NADH + H + FMN <---> NAD + FMNH2
- Transfers 2 electrons from NADH to Q.
- Pumps 4 protons from matrix to IMS.

Complex II: Succinate Dehydrogense

- Transfers 2 electrons from succinate to coenzyme Q.
- Contains 4 different protein subunits:

Subunit A: FAD, succinate binding

Subunit B: 3 Fe-S centers

Subunit C: integral membrane protein, Q binding

Subunit D: does not participate in electron transfer to Q, instead reduces the frequency of some leaking electrons form reactive oxygen Species: ROS.

Complex III: Ubiquinone Cytochrome C Oxidoreductase

- Composed of 22 subunits: heme groups and Fe-S centers
- Transfers of 2 electrons QH2 to Cytochrome C.
- Pumps 4 protons into IMS.

Complex IV: Cytochrome C Oxidase

- Contains 13 subunits, prosthetic groups such as cytochromes and Fe-S centers.
- Transfers 2 electrons from cytochrome C to oxygen, reducing it to H2O.
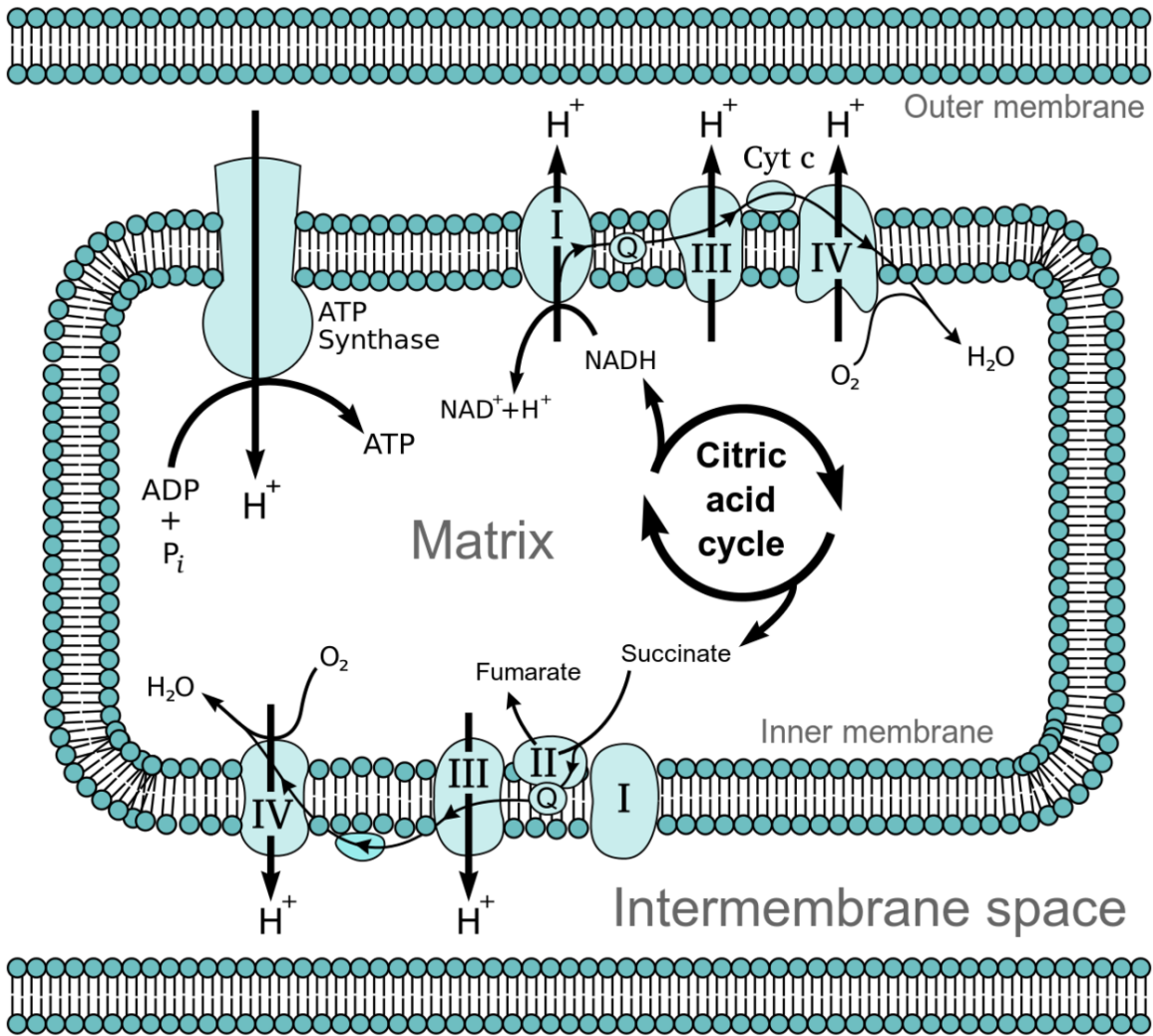- Pumps 2 protons into IMS

Figure 41   the detailed graph of the mechanism of electron transfer chain

image citation: [11]

11   http://en.wikipedia.org/wiki/File:Mitochondrial_electron_transport_chain%E2%80%94Etc4.svg

16 DNA and RNA

Deoxyribonucleic acid (DNA) and **ribonucleic acid** (RNA) are the information storage molecules and working templates for the construction of proteins. Every living cell and virus encodes its genetic information using either DNA or RNA. It is a true marvel of evolution that the vast amount of information needed to produce a human being can fit inside cells. Friedrich Miescher first isolated DNA and RNA from used surgical bandages in 1869. A series of experiments done by Oswald Avery, Colin MacLeod, and Maclyn McCarty in 1944 defined DNA as the genetic information. They injected virulent encapsulated bacteria into a mouse. As it a result, it died. On the contrary, nonvirulent nonencapsulated bacteria did not kill the mouse. Also, heating the virulent encapsulated bacteria and injecting did not kill the mouse either. The surprise lied in mixing the heat-killed virulent encapsulated bacteria with nonvirulent nonencapsulated bacteria did kill the mouse. Avery's group kept isolating factors until they discovered what killed the mouse, which was the DNA. In conclusion, the nonpathogenic bacteria was made pathogenic by DNA transfer from a pathogenic stain. A.D. Hershey and Martha Chase's work in 1952 reconfirmed that DNA is the carrier of genetic information. Hershey and Chase's group conducted an experiment involving two phages. One of the phages was radioactively labeled with heavy phosphorus to highlight the DNA and heavy sulfur to highlight the protein. The two phages inserted their "genetic material" into the bacterial cell. Afterwards, these cells were "blended" to remove the phage heads. The resulting radioactively-labeled heavy phosphorus was found in the cell. This experiment showed that DNA alone was sufficient to make new viruses, and therefore, defined as the genetic information. Rosalind Franklin, Chargaff, and other chemists provided information that led to the discovery of DNA as well. Franklin provided crystallographic data that implied a helical structure. Chargaff's rule said that the adenine content was equal to the thymine content and cytosine was equal to guanine. Also the aromaticity of the electron-rich purine and pyrimidine rings allowed for tautomer shifts between the keto and enol form. The keto tautomer (lactam) dominated at physiological pH. Garnering all this information, James Watson and Francis Crick discovered the structure of DNA (double-helix) in 1953.

16.1 Structure of DNA and RNA

DNA is a polymer of nucleotides. Nucleotides consist of a pentose sugar (2-deoxyribose for DNA and ribose for RNA), a phosphate group (H_3PO_4), and a nitrogen-containing base. There are five nitrogen bases - adenine, guanine, thymine, cytosine and uracil. Individual nucleotides are connected by phosphodiester bonds to form polynucleotides.
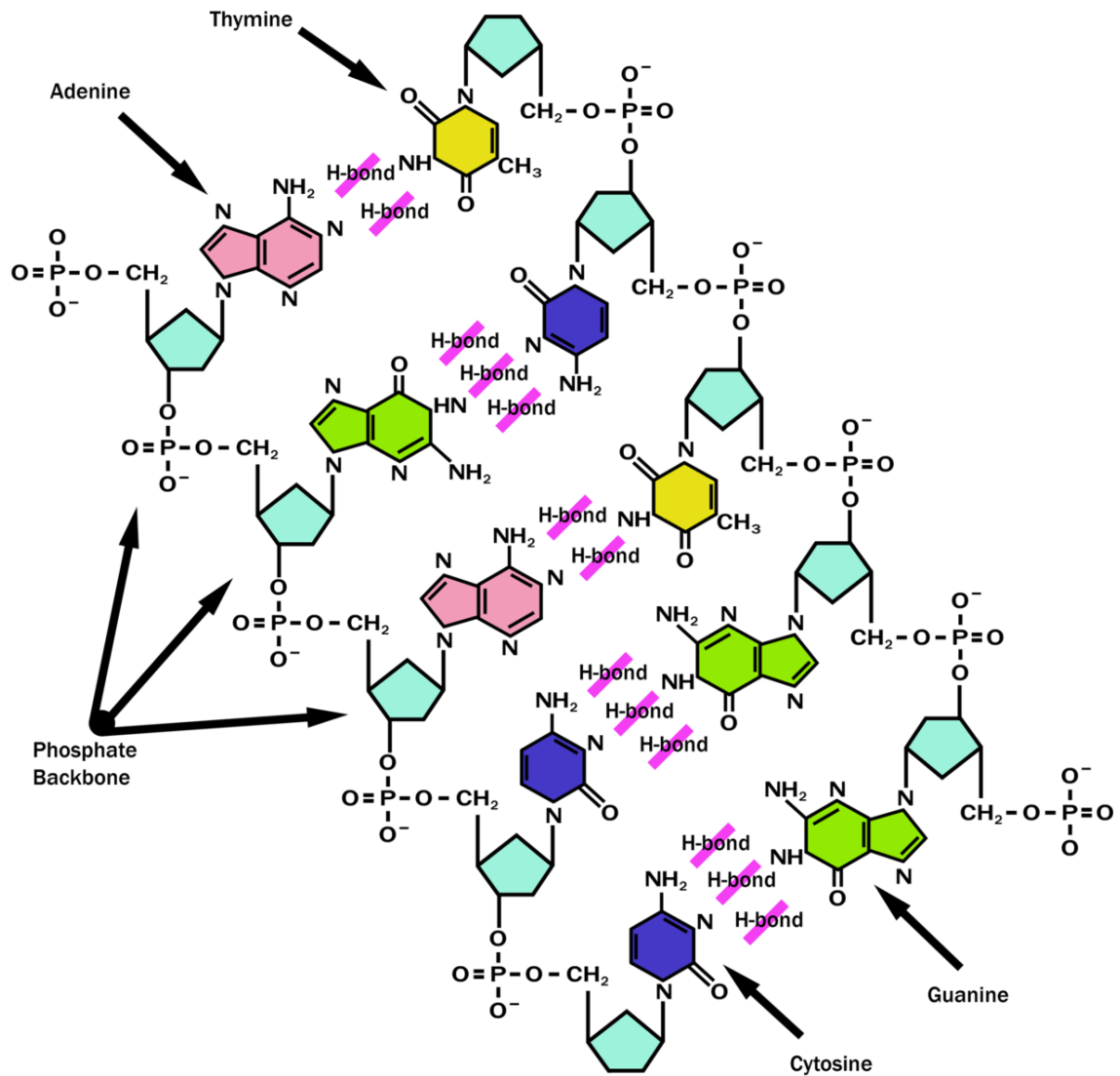
Figure 42   DNA

DNA exists as a double helix of two (2) strands of polynucleotides. According to the principle of complementarity nucleotides A (adenine) bases are bound with a hydrogen bridge to the T (thymine) or U (uracil in case of RNA) on the other thread and similarly C (cytosine) bind themselves to G (guanine). This principle allows for DNA and RNA replication and for limited possibilities of repairing the genetic information when one of the threads gets damaged.

16.2 Difference between DNA and RNA

DNA is the permanent genetic information storage medium, found in the nucleus of most cells of most living organisms. RNA is, in the case of eukaryotes, the medium that transfers the genetic information from the nucleus to the cytoplasm where proteins are synthesized. The most basic structural difference between a DNA molecule and an RNA molecule is that

DNA lacks an hydroxyl group at its 2' carbon while RNA has an hydroxyl group at its 2' carbon. There are three major types of RNA:

- messenger RNA (mRNA) - temporarily created RNA used for transferring the information from the DNA in the nucleus to the ribosome in the cytoplasm where it is "read," or translated, and a protein is synthesized.Each gene produce a separate mRNA molecule when a certain protein is needed in the cell. In addition, the size of an mRNA depends on the number of nucletides in that particular gene.

- transfer RNA (tRNA) - Is the smallest of the RNA molecules and it is associated with the ribosome and delivers an amino acid to be attached to the growing protein. Only the tRna can translate the genetic information into amino acid for proteins.

Each tRNA contains an "anticodon" which is a series of 3 bases that complements 3 bases on mRNA.

- ribosomal RNA (rRNA) - a major constituent of the ribosome with both structural and catalytic properties.

Additionally, by 2005, other types of RNA, such as siRNA (small, interfering RNA) and miRNA have been characterized. DNA structure is typically a double stranded molecule with long chain of nucleotides, while RNA is a single stranded molecule " in most biological roles" and it has shorter chain of nucleotide. Deoxyribose sugar in DNA is less reactive because of the carbon- hydrogen bonds. DNA has small grooves to prevent the enzymes to attack DNA. on the other hand, RNA contain ribose sugar which is more reactive because of the carbon-hydroxyl bonds. RNA not stable in its alkaline conditions.RNA has larger grooves than that of DNA, which makes RNA easier to be attacked by enzymes. DNA has a unique features because of its helix geometry which is in B-form. DNA is protected completely by the body in which the body destroys any enzymes that cleave DNA. In contrast the helix geometry of RNA is in the A- form; RNA can be broken down and reused. RNA is more resistance to be damage by Ultra- violet rays, while DNA can be damaged if exposed to Ultra-violet rays. DNA can be found in nucleus, while RNA can be found in nucleus and cytoplasm. The bases of DNA are A,T,C,and G, DNA is a long polymer with phosphate and deoxyribose backbone. In contrast, RNA bases are A,U, C,and G, RNA backbone contain ribose and phosphate. The job of DNA on the cell is to store and transmit genetic information. RNA job is to transfer the genetic codes that are needed for the creation of proteins from the nucleus to the ribosome, this process prevents the DNA from leaving the nucleus, so DNA stays safe.

16.3 Plasmid

Plasmids are small, circular DNA molecules that replicate separately from the much larger bacterial chromosome can be use in laboratory to manipulate genes. Plasmids can carry almost any gene and they can passed it on from one generation of bacteria to the next generation, therefore plasmids are key tools for "gene cloning" which is the production of multiple identical copies of a gene carrying piece of DNA

17 DNA Replication ;Semi-conservative replication

In the process of DNA replication, as cells divide, copies of DNA must be produced in order to transfer the genetic information. In DNA replication, the strands in the original DNA separate, and then each of the parent strands make copies by synthesizing complementary strands. Enzyme called helicase will start The process of replication by catalyzing the unwinding of protein of the double helix. This enzyme can break the H bonds between the complementary bases. These single strands now act as templates for the synthesis of new complementary strands. The energy for the reaction can be provided when a nucleoside triphosphate bonds to a sugar (at the end of a growing strand), and 2 phosphate group are cleaved. Then a T in the template forms H bond with an A in ATP, and a G on the template forms H bond with CTP. After the entire double helix of the parent DNA is copied, the new DNA molecule will form . The new DNA molecule is consist of one strand from the parent (original) DNA and one strand from the daughter strand(newly synthesized strand). This process is called "semi-conservative replication".
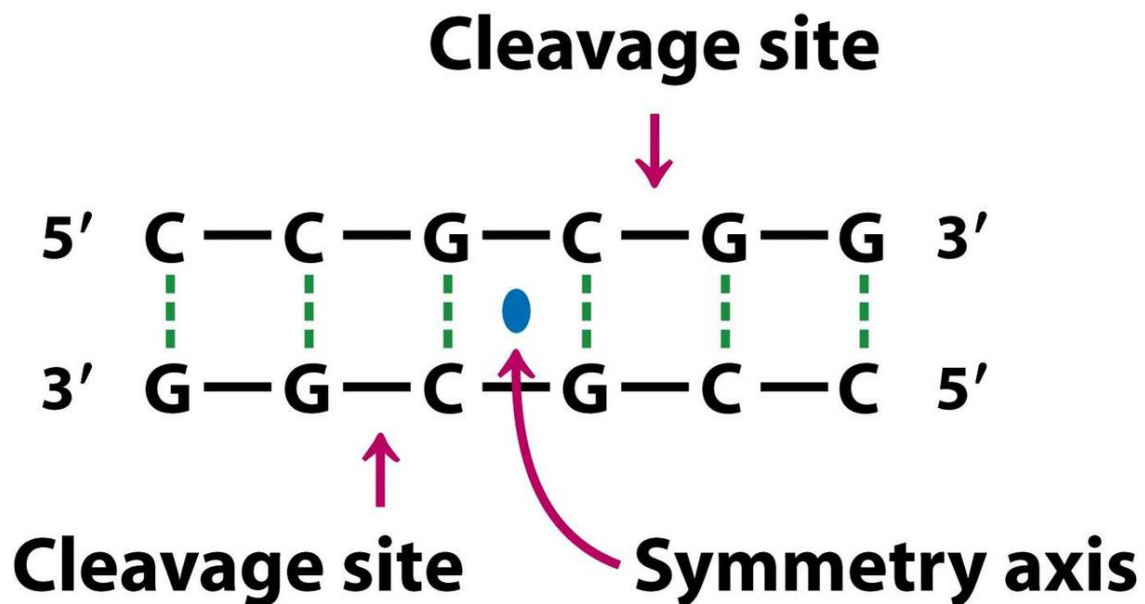
17.1 Recombination

Recombination refers to ways in which DNA modification leads to distinct gene expressions and functions. Deletions, insertions and substitutions are the most useful changes implemented for the synthesis of new genes. Recombination techniques and recombinant DNA technologies has made possible the modification and creation of new genes for specific proposes. These techniques helps clone, amplified and introduce new DNA into suitable cells for replication. Restriction enzymes and DNA ligase play a vital role in the in the production of recombinant DNA. Vectors are useful for cloning. Plasmids or viral DNA (lambda Phage) used to introduced a DNA sequence into a cell is refer as vector. The semi-conservative hypothesis was confirmed by both Matthew Meselson and Franklin Stahl in 1958. The parent DNA was labeled with "heavy nitrogen," the radioisotope of fifteen. This radioactive nitrogen was made through growing E. coli with ammonium chloride (also with "heavy nitrogen") environment. Directly after the labeling of "heavy nitrogen" to the parent DNA strand, it was transferred to a medium with only regular nitrogen (isotope of fourteen). The proposed question was "What is the distribution between the two nitrogens in the DNA molecules after successive rounds of replication?" Using density-gradient equilibrium sedimentation, this question was answered. In this centrifugation, similar densities were grouped together in the tube. Afterwards, an absorbance was applied with ultra-violet light to determine the density bands of all the different kinds of DNA in the solution. After one generation, a unique single density band was shown. It was not exactly the band of heavy nitrogen nor the regular band of regular nitrogen, but rather halfway between the two. This proved that DNA was not

conservative but semi-conservative due to this nitrogen hybrid. One generation after this one, there were two nitrogen-14 molecules and two hybrid nitrogen molecules. The probability and statistics of this second generation reassures the semi-conservative hypothesis. As DNA strands can combine and form molecules, it can also be melted, resulting in separation or denaturation. It is important to note that the heat from the melting process does not disrupt the DNA structure, but rather DNA helicase takes energy from the heat, separating the DNA strands. The denaturation process occurs at about ninety-four degrees Fahrenheit where the double strands are converted to single stands. Absorbance readings were taken to reconfirm the denaturation process. When the DNA molecule is in its double-stranded form, it absorbs less UV light. In its single-stranded form, it absorbs more UV light because the single strand is more exposed than its double-strand counterpart. This is known as the hypochromic effect. In a PCR, polymerase chain reaction, it utilizes this denaturation capability in its primary step. Afterwards, it is then annealed at fifty to sixty degrees Fahrenheit. In this step, this is where the primers bind to their corresponding flanking DNA base sequence. In the last step, DNA polymerization, it requires a few ingredients: heat-resistant DNA polymerase, magnesium ion, buffer, and corresponding nucleotides. The DNA polymerase requires heat resistance because throughout the entire process, it undergoes a constant temperatures changes. The magnesium stabilizes the phosphate backbone. The buffer prevents drastic changes in pH, because if the pH lies near the extremes on the scale, it may denature the DNA so drastically that it will not recombine. This final step of DNA polymerization happens at the "melting temperature." The melting temperature is the temperature at which half of the DNA is single-stranded and the other half is double-stranded. At this optimal temperature of seventy-two degrees Fahrenheit, the DNA is amplified with its respective primers, replicating at an exponential rate.

18 DNA manipulation: human's effort to desire DNA and "read" DNA

18.1 Restriction endonucleases

What's endonuclease?--Endonucleases are enzymes that can recognize specific base sequences in double-helical DNA and cleave, at specific places, both strands of that duplex. Many prokaryotes contain restriction enzymes. Biologically, they cleave foreign DNA molecules. Because the sites recognized by its own restriction enzymes are metylated, the cells' own DNA are not cleaved. One striking properties of endonucleases is that they recognize DNA cleavage sites by "two-fold symmetry". In another word, within the cleavage site domain (usually 4 to 8 base pair), there exists a center, around which the cleavage site rotate 180 degree, the whole cleavage site would be indistinguishable. The following graph shows an example of how endonuclease recognize cleavage site by symmetry.

Figure 43   The *Symmetry center* recognized by endonuclease.

.After cleavage from several different endonulcease, a DNA sample would be sliced into many different duplex fragments, which are then separated by electrophoresis. The restriction fragments are then denatured to form single stranded DNA, which are then transferred to a

nitrocellulose sheet, where they are exposed to a 32P-labeled single-stranded DNA probe. The single-stranded DNA are then hybridized by the radio active complementary strand. These duplex are then examined by autoradiography, which reveals the sequence of the DNA. This technique is named Southern plot, after Edwin South, who invented the technique. The following image shows the steps for creating a southern plot.
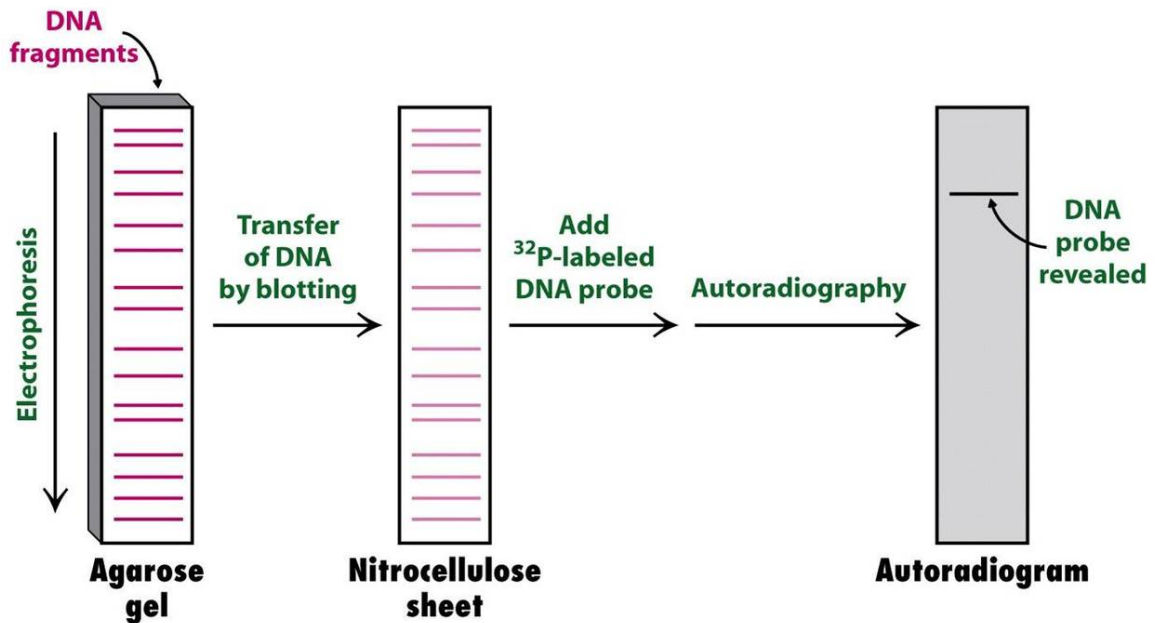

Figure 44   steps to create a southern plot.

18.2 DNA sequencing

DNA subunits are dGMP, dAMP, dCMP and dTMP. In a DNA molecule, these subunits are connected in a way that their phosphate on 5 prime carbon are bonded to another subunit's hydroxy group on 3 prime carbon. If there is no hydroxy group on 3 prime carbon, then the DNA polymeration will be terminated. Utilizing this idea, subunit analogies were invented, ddGMP, ddAMP, ddCMP and ddTMP. These analogies have the same structure as their parent molecule except that they have hydrogen on 3 prime carbon instead of hydroxyl group. In practice, target DNA template is put into four polymeration environments, dNTP+ddGMP or ddAMP or ddCMP or ddTMP, respectively.
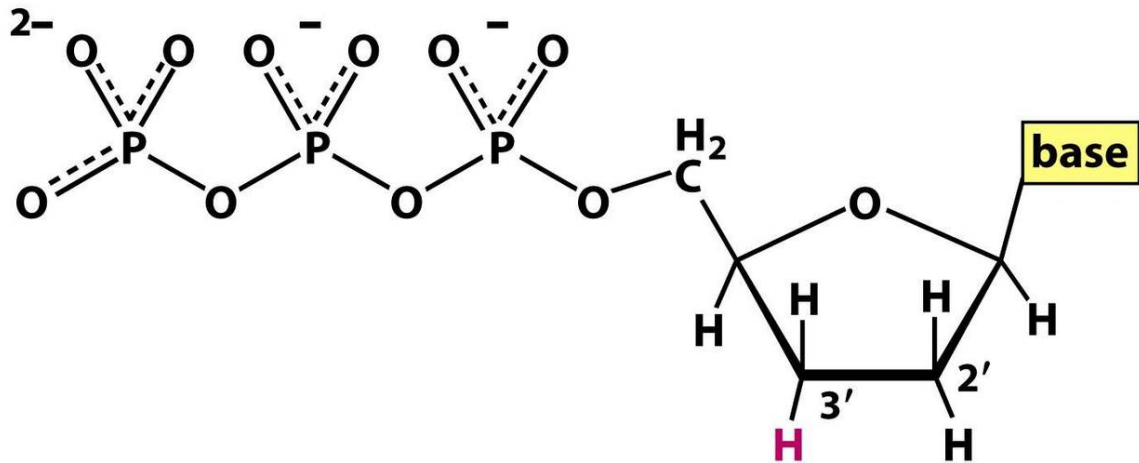
2', 3'-Dideoxy analog

Unnumbered figure pg 138
Biochemistry, Sixth Edition
© 2007 W. H. Freeman and Company

Figure 45   the analogy of dNTP

. Statically, fragments with different length terminated at base, G, A, C, T will be formed, respectively. Then these fragments will be separated by electrophoresis, which will tell the sequence of the target DNA.

18.3 DNA synthesis

Although nature synthesize DNA from 5 prime carbon to 3 prime carbon, human synthesize DNA from 3 prime carbon to 5 prime carbon. The following monomer is commercially available for each of the base and is used as basic building block for DNA synthesis.
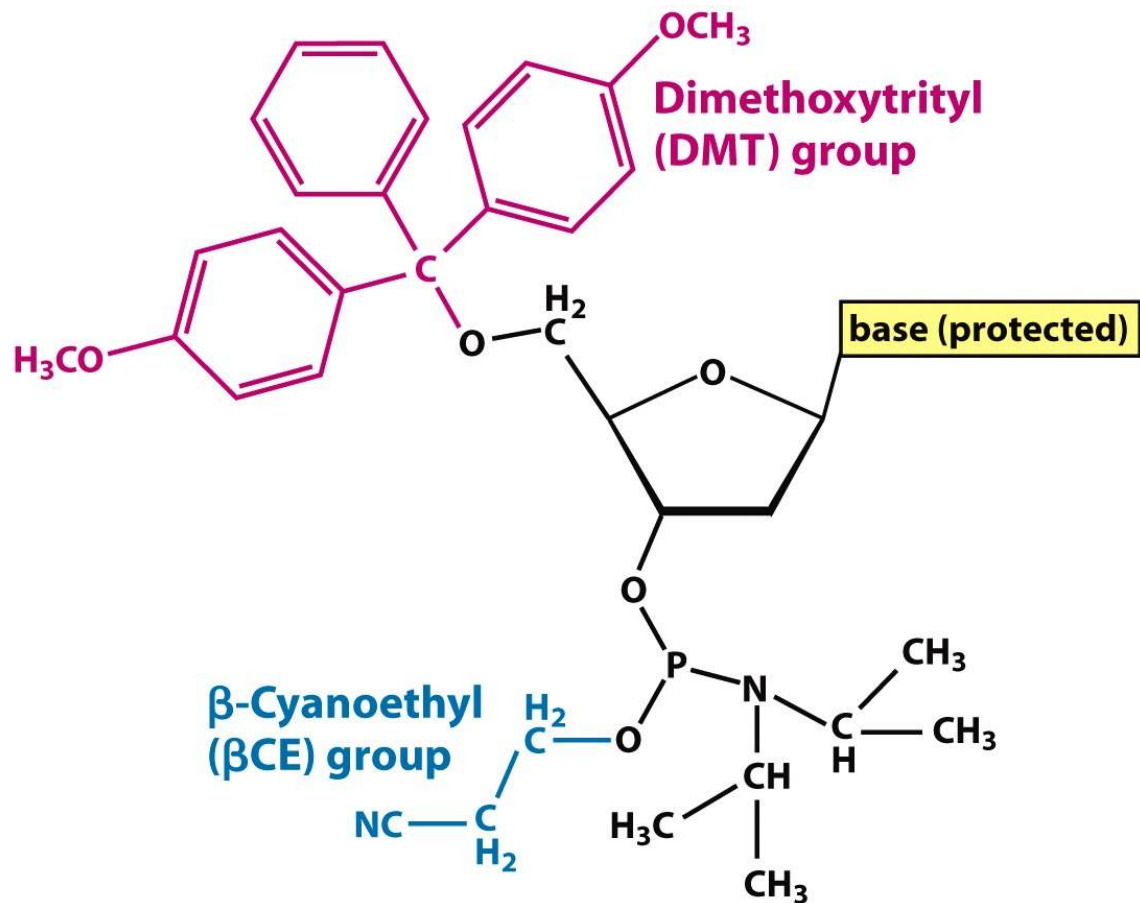
Figure 46   5 prime protected and 3 prime activated monomer.

It starts with the subunit with the first base bonded to resin by the 3 prime hydroxyl group. The second monomer with its own 5 hydroxyl protected by DMT (protecting group) was activated on its 3 prime phosphate, which is attached to the 5 prime hydroxyl group by nucleophilic substitution. Then the phosphite is oxidized by iodine to yield phosphate. Then DMT is removed to make the 5 prime hydroxyl available for the incoming of the third monomer. The graphical representation is shown below.
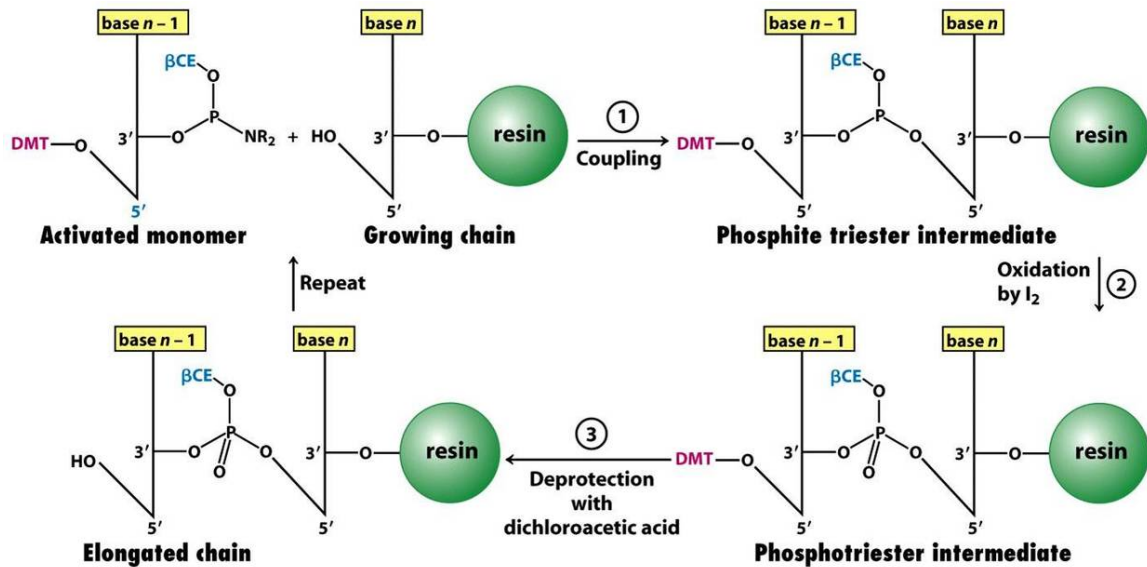
Figure 5-6
Biochemistry, Sixth Edition
© 2007 W. H. Freeman and Company

Figure 47   DNA synthesis.

18.4 Polymerase Chain Reaction (PCR)

It is used for amplification of DNA. It includes three stages: DNA denaturation, oligonucleotide annealing, and DNA polymerization. In each cycle, the DNA duplex is denatured at first, then the primers for both strands (forward and reverse) bind to the desired sites and at last both strands polymerize to form the desired sequence. What needed in PCR are the DNA containing the desired sequence, primers for both strands, heat resistant DNA polymerase, dNTP and constantly changing temperature. The following image shows the detailed steps in PCR.
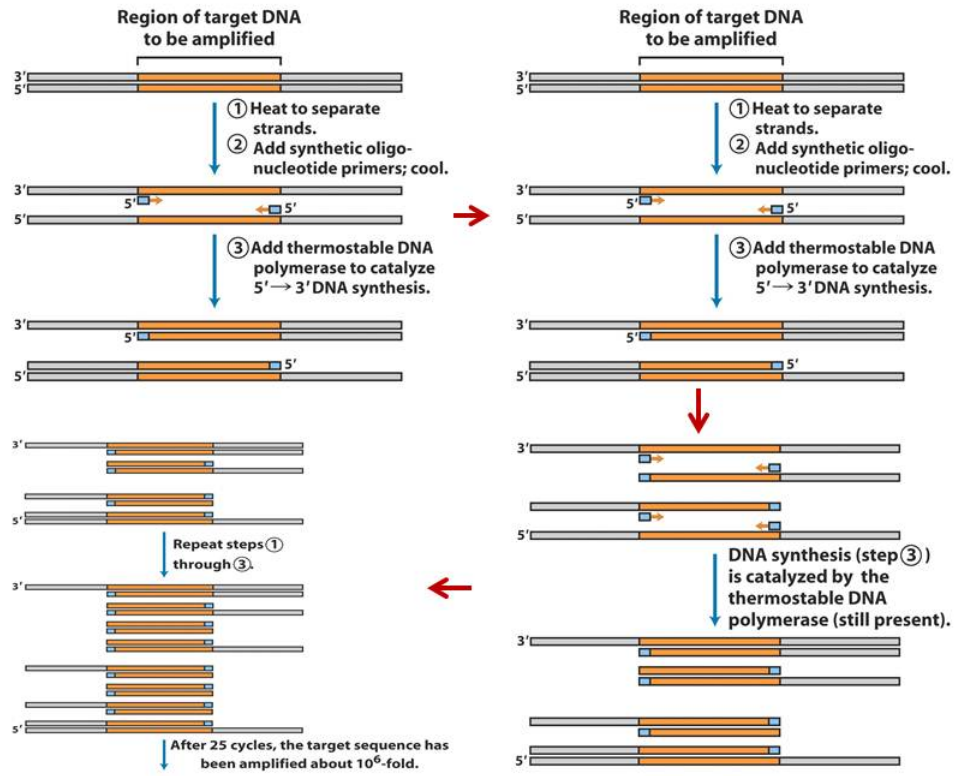
Figure 48   PCR steps in graphs.

19 RNA Editing

19.1 Introduction

Adenosine deaminases (ADAR) are mRNA editing enzymes that alternate a double stranded RNA sequence by tuning it. When tuning the mRNA, ADARs retype the nucleotides of the mRNA at certain, specific points they wish to change. Because ADARs can change the sequence of mRNA, they can create or remove a splice site, delete or alter the meaning of a codon, and finally change the sequence of the RNA altogether. ADAR plays a vital part in editing mRNA and modulating the mRNA translational activity and is mainly found in the nervous system and is important in regulating the nervous systems. Lacking the ADAR gene is very detrimental to health.

19.2 Function

RNA splicing and RNA editing are very similar to one another. RNA splicing is more of a cut, copy, and past process while RNA editing is an alteration of one or more nucleotides. There are mainly two types of ADAR that alternate the sequence of a single nucleotide. The first type of ADAR retypes cytidine nucleotides into uridine nucleotides while a second type of ADAR retypes adenosine into inosine.

mRNA consisting of many inosine nucleotides in its sequence is known to be very abundant in the nucleus and only mRNAs that leave the nucleus can be translated. In this case, ADAR is modulating the translational activities of mRNAs. In modulating the translational activities of the specified mRNA, it is also modulating the abundance of the certain proteins that are expressed in the edited mRNA. Also, increasing inosine can stabilize the structure of mRNA. Hence, ADAR can function as an mRNA stabilizer too.

ADARs target the coding sequence, introns, and 5' and 3' untranslated reigions (UTR). ADAR targets the coding sequence by changing the nucleotide which then changes the geometry of the protein. Change in the geometry of the protein may yield to higher protein-protein affinity interaction. This high protein-protein affinity can improve biological reactions by facilitating those reactions. Alternation of the non-coding sites such as introns, 5', and 3' UTR is still unknown. The only thing that is known is that alternating the sequence within these sites can create new splice sites.

19.3 Importance of ADAR

Adar is mainly found in the nervous system because it plays a very important role in regulating the nervous system. One of Adars' primary functions is its regulation of the nervous system. Adar regulates the nervous system by editing the glutamate receptors' mRNA. These glutamate receptors construct the glutamate-gated ion channels which are important in transferring electrical signals between neurons. When ADARs edit gluR mRNA, they change the calcium permeability of these glutamate channels. They can either lower the calcium permeability for certain glutamate channel or increase calcium permeability for other glutamate channels. Regulation of calcium permeability is important to proper neurotransmission between neurons.

19.4 Disease connecting to ADAR

Adenosine deaminase Deficiency (ADD) is caused by the lack of ADAR gene. Hence ADARs editing is important for survival especially for human. Without ADAR, the body can not break down the toxin deoxyadenosine. The accumulation of this toxin destroys the immune cells making its host vulnerable to infection from bacteria and viruses. Seizures can also be caused by ADAR. Unedited version of gluR mRNA strongly causes seizures. When a seizure happens, the body mechanism increases the level of ADAR activity to edit gluR mRNA to prevent future seizures.

19.5 DNA Replication:Initiation

There are multiple mechanisms that have been proposed for the melting of the DNA strand and the subsequent unwinding of the double helix during the initiation process of DNA replication. Two recently proposed models are E1 and LTag. These two models function differently to attain the same overall goal of melting the DNA double helix due to differences in their overall structure. E1 model: In the E1 hexamer, there are six ß-hairpins in the central channel of the protein. They are arranged in a staircase-like structure. Two adjacent E1 trimers assemble at the origin point to melt the double-stranded DNA. A ring shaped E1 hexamer is then formed around the melted single-stranded DNA and the DNA is then pumped through the ring hexamer to from a fork that allows for the replication of the DNA. Ltag: The channel diameters of these hexamers vary between 13-17 angstroms. The hexamer responsible for melting has an assortment of planar ß-hairpins in the narrow channel. Two of these hexamers surround the origin point of the double-stranded DNA and squeeze the

area together. This causes the melting of the dsDNA by forcing the breakage of base pairing. The two single-stranded DNA strands are then pumped into a larger channeland finally out through two separate Zn-domains. This allows for the replication of the DNA. The two mechanisms here are built solely on the structural knowledge of the proteins involved in the process and as such more experimental support is needed to confirm. There are not an excessive amount of helicases. Currently, work is being done in discerning the more simple helicases, and trying to find the different mechanisms that they may have. While there are some similarities, for example helicases all seem to have a hexameric structure, there are also differences as well in their structures that contribute greatly to the differences in their mechanisms. -This article refers to a recently published article in Curr Opin Struct Biol. published 2010, Sep. 24.: "Origin DNA melting and unwinding in DNA replication" by Gai *et al.*

20 Respiratory Pigments

In humans, these pigments include hemoglobin and myoglobin, the latter is present in red muscle fibers. Hemoglobin is a coordination compound of iron. It had attachment sites for gases, namely oxygen and carbon dioxide. Its reactivity depends upon partial pressure of these gases. The presence of heme makes it appear red colored. Myoglobin is present in muscles to make available extra oxygen.

21 Cell Signaling Pathways

Biochemistry/Cell Signaling Pathways[1]

---

[1] http://en.wikibooks.org/w/index.php?title=Biochemistry/Cell_Signaling_Pathways&
action=edit&redlink=1

22 Bioinformatics

22.1 Homology

When scientists study the relationship between proteins between different species they must determine whether or not these proteins are homologous to one another. Homologous proteins are proteins that have a common ancestor. There are two classes of homologs. They are ortholog and paralog. Orthologs are homologous proteins that are found among different species but have similar function. Orthologs occurs due to speciation. Paralog are homologous proteins that are found within the same species. They have very similar structure but serve different functions within the organism. Paralogs are due to gene duplication within the specie. Scientists finds out whether the proteins are homologous or not by studying the DNA sequence, amino acid sequence, the tertiary structure of the protein. In other word, they study the sequence alignment and structural alignment of the protein.

22.2 Sequence Alignment

When analyzing the DNA sequence or the amino acid sequence similarity and differences , scientists study the alignment of the DNA sequence or the alignment of the amino acid alignment of the protein. Alignment of DNA sequences is less sensitive to that of the alignment of amino acid. DNA sequence only consists of 4 bases. This means that the chance that both residues are the same is 1 out of 4. For the amino acid sequence, the chance that both residues are the same is 1 out of 21. The scoring of the degree of similarity of the alignment of each case is determined by Matrix.

22.3 Matrix

Matrix is a method used to determine the degree or similarity between the sequences (DNA or Amino Acid). Matrix takes account of two factors which are conservation and frequency. Conversation determines whether or not a certain residue can substitute for another residue by comparing the residue's physical factors such as their hydrophobicity, charge, and size. For example if two residues are both hydrophobic they may substitute for each other. If two residues have different charges, chance that they can substitute for one another is very slim. Frequency states how often a residue occurs. For example, if sequence A has a 20% of residue A and 30% of residue B and if sequence B has 21% of residue A and 35% of residue B, these residues A and B prevalence may indicate that these sequences are related to one another. The techniques used by matrix are the sliding between sequences, the gap

introduced within the sequence, and the deletion of certain residues within the sequence. This method is used in order to improve the alignment's similarity. In sliding, one sequence is sided in reference to the other. For example after the sliding the sequence by one residue, the similarity between the sequences may increase because more residues are identical or similar to one another. Gaps are introduces within the sequence in order to increase the residue similarity. For example if a sequence A have this particular range of residues that are not important or critical to its function or structure but have an important residue rang after the non-important residue range and the other sequence B lacks the sequence unimportant residue range but does have the same important residue range of sequence A. Then a gap is introduces in sequence B in order to make for the non important residue range. For deletion, certain residues are omitted in the sequence. Unimportant residues are usually deleted in order to increase the similarity between the sequences. There are two types of matrices. They are identity matrix and substitution matrix. Identity matrix only assign points to residues that are identical where as substitution may assign points to residues that are different yet alike in a conservative perspective. Substitution matrix are more accurate than that of identity matrix because in taking account for conservation substitution, substitution matrix give large positive score to the sequence where frequent substitution occur and large negative score where rare substitution occur . Hence substitution matrix is more sensitive to that of the identity matrix.

22.4 Structural Alignment

Structural alignment is the analysis of the degree of similarity between primary, secondary and tertiary structure. In protein, the tertiary structure is more conserved than that of primary structure because tertiary structure is more closely related to the protein's function. The purpose of structural alignment is to improve sequence alignment (method discussed before this) by creating a sequence template. Since some regions are more conserved than other, sequence template is template that maps out the conserved amino acid residues that are structurally and functionally important to a particular protein family member. Hence sequence template helps find protein that belongs to certain family member. SIRCh Link for Bioinformatics[1] (Selected Internet Resourcess)

Category[2]:

- Biochemistry[3]

---

1   http://en.wikibooks.org/wiki/Chemical_Information_Sources/SIRCh/Bioinformatics
2   http://en.wikibooks.org/wiki/Special:Categories
3   http://en.wikibooks.org/wiki/Category:Biochemistry

23 Contributors

Edits	User
2	A5song[1]
9	Adrignola[2]
10	Alsocal[3]
2	Amlau[4]
3	Annettep[5]
1	Arthurvogel[6]
10	Avicennasis[7]
1	Awdhesh pal[8]
1	Az1568[9]
8	Brandon (usurped)[10]
3	Brim[11]
1	Chazz[12]
1	Chemicadian[13]
1	Codec[14]
1	CommonsDelinker[15]
1	Cspurrier[16]
11	Cyclop[17]
3	Dekay[18]
3	Derbeth[19]
1	Dhs001[20]
1	Dintymusk[21]

1   http://en.wikibooks.org/w/index.php?title=User:A5song
2   http://en.wikibooks.org/w/index.php?title=User:Adrignola
3   http://en.wikibooks.org/w/index.php?title=User:Alsocal
4   http://en.wikibooks.org/w/index.php?title=User:Amlau
5   http://en.wikibooks.org/w/index.php?title=User:Annettep
6   http://en.wikibooks.org/w/index.php?title=User:Arthurvogel
7   http://en.wikibooks.org/w/index.php?title=User:Avicennasis
8   http://en.wikibooks.org/w/index.php?title=User:Awdhesh_pal
9   http://en.wikibooks.org/w/index.php?title=User:Az1568
10 http://en.wikibooks.org/w/index.php?title=User:Brandon_%28usurped%29
11 http://en.wikibooks.org/w/index.php?title=User:Brim
12 http://en.wikibooks.org/w/index.php?title=User:Chazz
13 http://en.wikibooks.org/w/index.php?title=User:Chemicadian
14 http://en.wikibooks.org/w/index.php?title=User:Codec
15 http://en.wikibooks.org/w/index.php?title=User:CommonsDelinker
16 http://en.wikibooks.org/w/index.php?title=User:Cspurrier
17 http://en.wikibooks.org/w/index.php?title=User:Cyclop
18 http://en.wikibooks.org/w/index.php?title=User:Dekay
19 http://en.wikibooks.org/w/index.php?title=User:Derbeth
20 http://en.wikibooks.org/w/index.php?title=User:Dhs001
21 http://en.wikibooks.org/w/index.php?title=User:Dintymusk

1	Dirk Hünniger[22]
1	Drewbie[23]
10	Du Chem114A[24]
3	Ealarcon[25]
1	Emmaueldwomoh[26]
3	Fishpi[27]
2	Fmarisca[28]
1	Frazzydee[29]
1	GJeffery[30]
1	Gary Dorman Wiggins[31]
4	Geocachernemesis[32]
3	Guanabot[33]
2	Hagindaz[34]
4	Hal009[35]
1	Hapa[36]
1	Herbythyme[37]
5	Hichris[38]
1	Iamunknown[39]
3	Innov8[40]
9	JSJohnson[41]
2	Jboisson[42]
1	Jdiaz78[43]
36	Jguk[44]
1	Jhn015[45]
11	Jomegat[46]

22 http://en.wikibooks.org/w/index.php?title=User:Dirk_H%C3%BCnniger
23 http://en.wikibooks.org/w/index.php?title=User:Drewbie
24 http://en.wikibooks.org/w/index.php?title=User:Du_Chem114A
25 http://en.wikibooks.org/w/index.php?title=User:Ealarcon
26 http://en.wikibooks.org/w/index.php?title=User:Emmaueldwomoh
27 http://en.wikibooks.org/w/index.php?title=User:Fishpi
28 http://en.wikibooks.org/w/index.php?title=User:Fmarisca
29 http://en.wikibooks.org/w/index.php?title=User:Frazzydee
30 http://en.wikibooks.org/w/index.php?title=User:GJeffery
31 http://en.wikibooks.org/w/index.php?title=User:Gary_Dorman_Wiggins
32 http://en.wikibooks.org/w/index.php?title=User:Geocachernemesis
33 http://en.wikibooks.org/w/index.php?title=User:Guanabot
34 http://en.wikibooks.org/w/index.php?title=User:Hagindaz
35 http://en.wikibooks.org/w/index.php?title=User:Hal009
36 http://en.wikibooks.org/w/index.php?title=User:Hapa
37 http://en.wikibooks.org/w/index.php?title=User:Herbythyme
38 http://en.wikibooks.org/w/index.php?title=User:Hichris
39 http://en.wikibooks.org/w/index.php?title=User:Iamunknown
40 http://en.wikibooks.org/w/index.php?title=User:Innov8
41 http://en.wikibooks.org/w/index.php?title=User:JSJohnson
42 http://en.wikibooks.org/w/index.php?title=User:Jboisson
43 http://en.wikibooks.org/w/index.php?title=User:Jdiaz78
44 http://en.wikibooks.org/w/index.php?title=User:Jguk
45 http://en.wikibooks.org/w/index.php?title=User:Jhn015
46 http://en.wikibooks.org/w/index.php?title=User:Jomegat

2	Jonh kaldun[47]
2	Jyl059[48]
6	Karl Wick[49]
16	Ketruong[50]
3	Kiubee[51]
2	Kjhong[52]
2	LTN019[53]
12	Lineweaver[54]
2	Lrandolp[55]
4	LyNguyen[56]
2	Lynx7725[57]
1	Madkiller12345[58]
46	Magnus Manske[59]
7	Malfarra[60]
1	Mattb112885[61]
1	Matzman[62]
1	Metric1000[63]
3	Mike.lifeguard[64]
2	Mlinht[65]
3	Mmwong[66]
4	Mockeldritch[67]
2	Moleculesoflife[68]
10	Msoto[69]
23	Mtvan[70]
1	Nelson.blaha[71]

47 http://en.wikibooks.org/w/index.php?title=User:Jonh_kaldun
48 http://en.wikibooks.org/w/index.php?title=User:Jyl059
49 http://en.wikibooks.org/w/index.php?title=User:Karl_Wick
50 http://en.wikibooks.org/w/index.php?title=User:Ketruong
51 http://en.wikibooks.org/w/index.php?title=User:Kiubee
52 http://en.wikibooks.org/w/index.php?title=User:Kjhong
53 http://en.wikibooks.org/w/index.php?title=User:LTN019
54 http://en.wikibooks.org/w/index.php?title=User:Lineweaver
55 http://en.wikibooks.org/w/index.php?title=User:Lrandolp
56 http://en.wikibooks.org/w/index.php?title=User:LyNguyen
57 http://en.wikibooks.org/w/index.php?title=User:Lynx7725
58 http://en.wikibooks.org/w/index.php?title=User:Madkiller12345
59 http://en.wikibooks.org/w/index.php?title=User:Magnus_Manske
60 http://en.wikibooks.org/w/index.php?title=User:Malfarra
61 http://en.wikibooks.org/w/index.php?title=User:Mattb112885
62 http://en.wikibooks.org/w/index.php?title=User:Matzman
63 http://en.wikibooks.org/w/index.php?title=User:Metric1000
64 http://en.wikibooks.org/w/index.php?title=User:Mike.lifeguard
65 http://en.wikibooks.org/w/index.php?title=User:Mlinht
66 http://en.wikibooks.org/w/index.php?title=User:Mmwong
67 http://en.wikibooks.org/w/index.php?title=User:Mockeldritch
68 http://en.wikibooks.org/w/index.php?title=User:Moleculesoflife
69 http://en.wikibooks.org/w/index.php?title=User:Msoto
70 http://en.wikibooks.org/w/index.php?title=User:Mtvan
71 http://en.wikibooks.org/w/index.php?title=User:Nelson.blaha

1	Neoptolemus[72]
1	Noname713[73]
1	Oatjay[74]
3	Panic2k4[75]
1	PatríciaR[76]
9	Pharamund[77]
7	Philculmer[78]
1	Phosphole[79]
2	Pnnguyen[80]
1	Pwijesek[81]
2	Quiark[82]
2	Quietnight8[83]
17	QuiteUnusual[84]
9	Recent Runes[85]
1	Reece[86]
1	Robert Horning[87]
2	RoseParks[88]
1	SBJohnny[89]
9	Salaviza[90]
1	Savh[91]
24	Sdoroudi[92]
10	Seldon[93]
1	Sidney[94]
1	Sigma 7[95]
2	Srhat[96]

72 http://en.wikibooks.org/w/index.php?title=User:Neoptolemus
73 http://en.wikibooks.org/w/index.php?title=User:Noname713
74 http://en.wikibooks.org/w/index.php?title=User:Oatjay
75 http://en.wikibooks.org/w/index.php?title=User:Panic2k4
76 http://en.wikibooks.org/w/index.php?title=User:Patr%C3%ADciaR
77 http://en.wikibooks.org/w/index.php?title=User:Pharamund
78 http://en.wikibooks.org/w/index.php?title=User:Philculmer
79 http://en.wikibooks.org/w/index.php?title=User:Phosphole
80 http://en.wikibooks.org/w/index.php?title=User:Pnnguyen
81 http://en.wikibooks.org/w/index.php?title=User:Pwijesek
82 http://en.wikibooks.org/w/index.php?title=User:Quiark
83 http://en.wikibooks.org/w/index.php?title=User:Quietnight8
84 http://en.wikibooks.org/w/index.php?title=User:QuiteUnusual
85 http://en.wikibooks.org/w/index.php?title=User:Recent_Runes
86 http://en.wikibooks.org/w/index.php?title=User:Reece
87 http://en.wikibooks.org/w/index.php?title=User:Robert_Horning
88 http://en.wikibooks.org/w/index.php?title=User:RoseParks
89 http://en.wikibooks.org/w/index.php?title=User:SBJohnny
90 http://en.wikibooks.org/w/index.php?title=User:Salaviza
91 http://en.wikibooks.org/w/index.php?title=User:Savh
92 http://en.wikibooks.org/w/index.php?title=User:Sdoroudi
93 http://en.wikibooks.org/w/index.php?title=User:Seldon
94 http://en.wikibooks.org/w/index.php?title=User:Sidney
95 http://en.wikibooks.org/w/index.php?title=User:Sigma_7
96 http://en.wikibooks.org/w/index.php?title=User:Srhat

2 Stephanmir[97]
6 Stewiki[98]
2 Tannersf[99]
2 Thenub314[100]
2 TreborA[101]
1 Ugen64[102]
2 Uncle G[103]
1 User7194[104]
2 Vivek.science[105]
3 Webaware[106]
1 Westhaking[107]
1 Whtndks[108]
8 Xania[109]
1 Yikrazuul[110]
4 Yntran[111]

97 http://en.wikibooks.org/w/index.php?title=User:Stephanmir
98 http://en.wikibooks.org/w/index.php?title=User:Stewiki
99 http://en.wikibooks.org/w/index.php?title=User:Tannersf
100 http://en.wikibooks.org/w/index.php?title=User:Thenub314
101 http://en.wikibooks.org/w/index.php?title=User:TreborA
102 http://en.wikibooks.org/w/index.php?title=User:Ugen64
103 http://en.wikibooks.org/w/index.php?title=User:Uncle_G
104 http://en.wikibooks.org/w/index.php?title=User:User7194
105 http://en.wikibooks.org/w/index.php?title=User:Vivek.science
106 http://en.wikibooks.org/w/index.php?title=User:Webaware
107 http://en.wikibooks.org/w/index.php?title=User:Westhaking
108 http://en.wikibooks.org/w/index.php?title=User:Whtndks
109 http://en.wikibooks.org/w/index.php?title=User:Xania
110 http://en.wikibooks.org/w/index.php?title=User:Yikrazuul
111 http://en.wikibooks.org/w/index.php?title=User:Yntran

List of Figures

- GFDL: Gnu Free Documentation License. `http://www.gnu.org/licenses/fdl.html`

- cc-by-sa-3.0: Creative Commons Attribution ShareAlike 3.0 License. `http://creativecommons.org/licenses/by-sa/3.0/`

- cc-by-sa-2.5: Creative Commons Attribution ShareAlike 2.5 License. `http://creativecommons.org/licenses/by-sa/2.5/`

- cc-by-sa-2.0: Creative Commons Attribution ShareAlike 2.0 License. `http://creativecommons.org/licenses/by-sa/2.0/`

- cc-by-sa-1.0: Creative Commons Attribution ShareAlike 1.0 License. `http://creativecommons.org/licenses/by-sa/1.0/`

- cc-by-2.0: Creative Commons Attribution 2.0 License. `http://creativecommons.org/licenses/by/2.0/`

- cc-by-2.0: Creative Commons Attribution 2.0 License. `http://creativecommons.org/licenses/by/2.0/deed.en`

- cc-by-2.5: Creative Commons Attribution 2.5 License. `http://creativecommons.org/licenses/by/2.5/deed.en`

- cc-by-3.0: Creative Commons Attribution 3.0 License. `http://creativecommons.org/licenses/by/3.0/deed.en`

- GPL: GNU General Public License. `http://www.gnu.org/licenses/gpl-2.0.txt`

- LGPL: GNU Lesser General Public License. `http://www.gnu.org/licenses/lgpl.html`

- PD: This image is in the public domain.

- ATTR: The copyright holder of this file allows anyone to use it for any purpose, provided that the copyright holder is properly attributed. Redistribution, derivative work, commercial use, and all other use is permitted.

- EURO: This is the common (reverse) face of a euro coin. The copyright on the design of the common face of the euro coins belongs to the European Commission. Authorised is reproduction in a format without relief (drawings, paintings, films) provided they are not detrimental to the image of the euro.

- LFK: Lizenz Freie Kunst. `http://artlibre.org/licence/lal/de`

- CFR: Copyright free use.

- EPL: Eclipse Public License. `http://www.eclipse.org/org/documents/epl-v10.php`

Copies of the GPL, the LGPL as well as a GFDL are included in chapter Licenses[112]. Please note that images in the public domain do not require attribution. You may click on the image numbers in the following table to open the webpage of the images in your webbrower.

---

112 Chapter 24 on page 129

1	The people from the Tango! project[113]	PD
2		GFDL
3	Vectorized version UNKNOWN TEMPLATE user Srhat	PD
4	fullofstars	PD
5	SVG version: UNKNOWN TEMPLATE User Srhat	GFDL
6	Magnus Manske[114]	GFDL
7	- SVG version: User:Srhat[115] - Source file: b:File:Feedback_inhibition.png[116] → w:User:Magnus Manske[117]	cc-by-2.5
8		GFDL
9		GFDL
10		GFDL
11		GFDL
12		GFDL
13		GFDL
14		GFDL
15		GFDL
16	{{User:YassineMrabet/mysign	PD
17		GFDL
18		GFDL
19	Lineweaver	cc-by-sa-3.0
20		GFDL
21	Original uploader was G3pro[118] at en.wikipedia[119] Later versions were uploaded by Chodges[120] at en.wikipedia[121].	GFDL
22	NEUROtiker[122]	PD
23	Zippanova[123]	PD
24	NEUROtiker[124]	PD
25		GFDL

---

113 `http://tango.freedesktop.org/The_People`

114 `http://en.wikibooks.org/wiki/User%3AMagnus%20Manske`

115 `http://en.wikibooks.org/wiki/User%3ASrhat`

116 `http://en.wikibooks.org/wiki/File%3AFeedback_inhibition.png`

117 `http://en.wikipedia.org/wiki/User%3AMagnus%20Manske`

118 `http://en.wikibooks.org/wiki/%3Aen%3AUser%3AG3pro`

119 `http://en.wikipedia.org`

120 `http://en.wikibooks.org/wiki/%3Aen%3AUser%3AChodges`

121 `http://en.wikipedia.org`

122 `http://en.wikibooks.org/wiki/User%3ANEUROtiker`

123 `http://en.wikibooks.org/wiki/%3Aw%3AUser%3AZippanova`

124 `http://en.wikibooks.org/wiki/User%3ANEUROtiker`

26	Yikrazuul[125] (talk[126])	PD
27	Ben Mills[127]	PD
28		
29		GFDL
30		GFDL
31		PD
32		GFDL
33	User:Bryan Derksen[128]	PD
34	LHcheM[129]	GFDL
35		
36		GFDL
37		PD
38		GFDL
39	User:Bryan Derksen[130]	PD
40		PD
41	Fvasconcellos[131] 22:35, 9 September 2007 (UTC)	PD
42		GFDL
43		
44		
45		
46		
47	Jeremy M. Berg; John L. Tymoczko; Lubert Stryer	
48	Tpirojsi[132]	PD

125 http://en.wikibooks.org/wiki/User%3AYikrazuul
126 http://en.wikibooks.org/wiki/User%20talk%3AYikrazuul
127 http://en.wikibooks.org/wiki/User%3ABenjah-bmm27
128 http://en.wikibooks.org/wiki/User%3ABryan%20Derksen
129 http://en.wikibooks.org/wiki/User%3ALHcheM
130 http://en.wikibooks.org/wiki/User%3ABryan%20Derksen
131 http://en.wikibooks.org/wiki/User%3AFvasconcellos
132 http://en.wikibooks.org/wiki/User%3ATpirojsi

24 Licenses

24.1 GNU GENERAL PUBLIC LICENSE

Version 3, 29 June 2007

Copyright © 2007 Free Software Foundation, Inc. <http://fsf.org/>

Everyone is permitted to copy and distribute verbatim copies of this license document, but changing it is not allowed. Preamble

The GNU General Public License is a free, copyleft license for software and other kinds of works.

The licenses for most software and other practical works are designed to take away your freedom to share and change the works. By contrast, the GNU General Public License is intended to guarantee your freedom to share and change all versions of a program—to make sure it remains free software for all its users. We, the Free Software Foundation, use the GNU General Public License for most of our software; it applies also to any other work released this way by its authors. You can apply it to your programs, too.

When we speak of free software, we are referring to freedom, not price. Our General Public Licenses are designed to make sure that you have the freedom to distribute copies of free software (and charge for them if you wish), that you receive source code or can get it if you want it, that you can change the software or use pieces of it in new free programs, and that you know you can do these things.

To protect your rights, we need to prevent others from denying you these rights or asking you to surrender the rights. Therefore, you have certain responsibilities if you distribute copies of the software, or if you modify it: responsibilities to respect the freedom of others.

For example, if you distribute copies of such a program, whether gratis or for a fee, you must pass on to the recipients the same freedoms that you received. You must make sure that they, too, receive or can get the source code. And you must show them these terms so they know their rights.

Developers that use the GNU GPL protect your rights with two steps: (1) assert copyright on the software, and (2) offer you this License giving you legal permission to copy, distribute and/or modify it.

For the developers' and authors' protection, the GPL clearly explains that there is no warranty for this free software. For both users' and authors' sake, the GPL requires that modified versions be marked as changed, so that their problems will not be attributed erroneously to authors of previous versions.

Some devices are designed to deny users access to install or run modified versions of the software inside them, although the manufacturer can do so. This is fundamentally incompatible with the aim of protecting users' freedom to change the software. The systematic pattern of such abuse occurs in the area of products for individuals to use, which is precisely where it is most unacceptable. Therefore, we have designed this version of the GPL to prohibit the practice for those products. If such problems arise substantially in other domains, we stand ready to extend this provision to those domains in future versions of the GPL, as needed to protect the freedom of users.

Finally, every program is threatened constantly by software patents. States should not allow patents to restrict development and use of software on general-purpose computers, but in those that do, we wish to avoid the special danger that patents applied to a free program could make it effectively proprietary. To prevent this, the GPL assures that patents cannot be used to render the program non-free.

The precise terms and conditions for copying, distribution and modification follow. TERMS AND CONDITIONS 0. Definitions.

"This License" refers to version 3 of the GNU General Public License.

"Copyright" also means copyright-like laws that apply to other kinds of works, such as semiconductor masks.

"The Program" refers to any copyrightable work licensed under this License. Each licensee is addressed as "you". "Licensees" and "recipients" may be individuals or organizations.

To "modify" a work means to copy from or adapt all or part of the work in a fashion requiring copyright permission, other than the making of an exact copy. The resulting work is called a "modified version" of the earlier work or a work "based on" the earlier work.

A "covered work" means either the unmodified Program or a work based on the Program.

To "propagate" a work means to do anything with it that, without permission, would make you directly or secondarily liable for infringement under applicable copyright law, except executing it on a computer or modifying a private copy. Propagation includes copying, distribution (with or without modification), making available to the public, and in some countries other activities as well.

To "convey" a work means any kind of propagation that enables other parties to make or receive copies. Mere interaction with a user through a computer network, with no transfer of a copy, is not conveying.

An interactive user interface displays "Appropriate Legal Notices" to the extent that it includes a convenient and prominently visible feature that (1) displays an appropriate copyright notice, and (2) tells the user that there is no warranty for the work (except to the extent that warranties are provided), that licensees may convey the work under this License, and how to view a copy of this License. If the interface presents a list of user commands or options, such as a menu, a prominent item in the list meets this criterion. 1. Source Code.

The "source code" for a work means the preferred form of the work for making modifications to it. "Object code" means any non-source form of a work.

A "Standard Interface" means an interface that either is an official standard defined by a recognized standards body, or, in the case of interfaces specified for a particular programming language, one that is widely used among developers working in that language.

The "System Libraries" of an executable work include anything, other than the work as a whole, that (a) is included in the normal form of packaging a Major Component, but which is not part of that Major Component, and (b) serves only to enable use of the work with that Major Component, or to implement a Standard Interface for which an implementation is available to the public in source code form. A "Major Component", in this context, means a major essential component (kernel, window system, and so on) of the specific operating system (if any) on which the executable work runs, or a compiler used to produce the work, or an object code interpreter used to run it.

The "Corresponding Source" for a work in object code form means all the source code needed to generate, install, and (for an executable work) run the object code and to modify the work, including scripts to control those activities. However, it does not include the work's System Libraries, or general-purpose tools or generally available free programs which are used unmodified in performing those activities but which are not part of the work. For example, Corresponding Source includes interface definition files associated with source files for the work, and the source code for shared libraries and dynamically linked subprograms that the work is specifically designed to require, such as by intimate data communication or control flow between those subprograms and other parts of the work.

The Corresponding Source need not include anything that users can regenerate automatically from other parts of the Corresponding Source.

The Corresponding Source for a work in source code form is that same work. 2. Basic Permissions.

All rights granted under this License are granted for the term of copyright on the Program, and are irrevocable provided the stated conditions are met. This License explicitly affirms your unlimited permission to run the unmodified Program. The output from running a covered work is covered by this License only if the output, given its content, constitutes a covered work. This License acknowledges your rights of fair use or other equivalent, as provided by copyright law.

You may make, run and propagate covered works that you do not convey, without conditions so long as your license otherwise remains in force. You may convey covered works to others for the sole purpose of having them make modifications exclusively for you, or provide you with facilities for running those works, provided that you comply with the terms of this License in conveying all material for which you do not control copyright. Those thus making or running the covered works for you must do so exclusively on your behalf, under your direction and control, on terms that prohibit them from making any copies of your copyrighted material outside their relationship with you.

Conveying under any other circumstances is permitted solely under the conditions stated below. Sublicensing is not allowed; section 10 makes it unnecessary. 3. Protecting Users' Legal Rights From Anti-Circumvention Law.

No covered work shall be deemed part of an effective technological measure under any applicable law fulfilling obligations under article 11 of the WIPO copyright treaty adopted on 20 December 1996, or similar laws prohibiting or restricting circumvention of such measures.

When you convey a covered work, you waive any legal power to forbid circumvention of technological measures to the extent such circumvention is effected by exercising rights under this License with respect to the covered work, and you disclaim any intention to limit operation or modification of the work as a means of enforcing, against the work's users, your or third parties' legal rights to forbid circumvention of technological measures. 4. Conveying Verbatim Copies.

You may convey verbatim copies of the Program's source code as you receive it, in any medium, provided that you conspicuously and appropriately publish on each copy an appropriate copyright notice; keep intact all notices stating that this License and any non-permissive terms added in accord with section 7 apply to the code; keep intact all notices of the absence of any warranty; and give all recipients a copy of this License along with the Program.

You may charge any price or no price for each copy that you convey, and you may offer support or warranty protection for a fee. 5. Conveying Modified Source Versions.

You may convey a work based on the Program, or the modifications to produce it from the Program, in the form of source code under the terms of section 4, provided that you also meet all of these conditions:

* a) The work must carry prominent notices stating that you modified it, and giving a relevant date. * b) The work must carry prominent notices stating that it is released under this License and any conditions added under section 7. This requirement modifies the requirement in section 4 to "keep intact all notices". * c) You must license the entire work, as a whole, under this License to anyone who comes into possession of a copy. This License will therefore apply, along with any applicable section 7 additional terms, to the whole of the work, and all its parts, regardless of how they are packaged. This License gives no permission to license the work in any other way, but it does not invalidate such permission if you have separately received it. * d) If the work has interactive user interfaces, each must display Appropriate Legal Notices; however, if the Program has interactive interfaces that do not display Appropriate Legal Notices, your work need not make them do so.

A compilation of a covered work with other separate and independent works, which are not by their nature extensions of the covered work, and which are not combined with it such as to form a larger program, in or on a volume of a storage or distribution medium, is called an "aggregate" if the compilation and its resulting copyright are not used to limit the access or legal rights of the compilation's users beyond what the individual works permit. Inclusion of a covered work in an aggregate does not cause this License to apply to the other parts of the aggregate. 6. Conveying Non-Source Forms.

You may convey a covered work in object code form under the terms of sections 4 and 5, provided that you also convey the machine-readable Corresponding Source under the terms of this License, in one of these ways:

* a) Convey the object code in, or embodied in, a physical product (including a physical distribution medium), accompanied by the Corresponding Source fixed on a durable physical medium customarily used for software interchange. * b) Convey the object code in, or embodied in, a physical product (including a physical distribution medium), accompanied by a written offer, valid for at least three years and valid for as long as you offer spare parts or customer support for that product model, to give anyone who possesses the object code either (1) a copy of the Corresponding Source for all the software in the product that is covered by this License, on a durable physical medium customarily used for software interchange, for a price no more than your reasonable cost of physically performing this conveying of source, or (2) access to copy the Corresponding Source from a network server at no charge. * c) Convey individual copies of the object code with a copy of the written offer to provide the Corresponding Source. This alternative is allowed only occasionally and noncommercially, and only if you received the object code with such an offer, in accord with subsection 6b. * d) Convey the object code by offering access from a designated place (gratis or for a charge), and offer equivalent access to the Corresponding Source in the same way through the same place at no further charge. You need not require recipients to copy the Corresponding Source along with the object code. If the place to copy the object code is a network server, the Corresponding Source may be on a different server (operated by you or a third party) that supports equivalent copying facilities, provided you maintain clear directions next to the object code saying where to find the Corresponding Source. Regardless of what server hosts the Corresponding Source, you remain obligated to ensure that it is available for as long as needed to satisfy these requirements. * e) Convey the object code using peer-to-peer transmission, provided you inform other peers where the object code and Corresponding Source of the work are being offered to the general public at no charge under subsection 6d.

A separable portion of the object code, whose source code is excluded from the Corresponding Source as a System Library, need not be included in conveying the object code work.

A "User Product" is either (1) a "consumer product", which means any tangible personal property which is normally used for personal, family, or household purposes, or (2) anything designed or sold for incorporation into a dwelling. In determining whether a product is a consumer product, doubtful cases shall be resolved in favor of coverage. For a particular product received by a particular user, "normally used" refers to a typical or common use of that class of product, regardless of the status of the particular user or of the way in which the particular user actually uses, or expects or is expected to use, the product. A product is a consumer product regardless of whether the product has substantial commercial, industrial or non-consumer uses, unless such uses represent the only significant mode of use of the product.

"Installation Information" for a User Product means any methods, procedures, authorization keys, or other information required to install and execute modified versions of a covered work in that User Product from a modified version of its Corresponding Source. The information must suffice to ensure that the continued functioning of the modified object code is in no case prevented or interfered with solely because modification has been made.

If you convey an object code work under this section in, or with, or specifically for use in, a User Product, and the conveying occurs as part of a transaction in which the right of possession and use of the User Product is transferred to the recipient in perpetuity or for a fixed term (regardless of how the transaction is characterized), the Corresponding Source conveyed under this section must be accompanied by the Installation Information. But this requirement does not apply if neither you nor any third party retains the ability to install modified object code on the User Product (for example, the work has been installed in ROM).

The requirement to provide Installation Information does not include a requirement to continue to provide support service, warranty, or updates for a work that has been modified or installed by the recipient, or for the User Product in which it has been modified or installed. Access to a network may be denied when the modification itself materially and adversely affects the operation of the network or violates the rules and protocols for communication across the network.

Corresponding Source conveyed, and Installation Information provided, in accord with this section must be in a format that is publicly documented (and with an implementation available to the public in source code form), and must require no special password or key for unpacking, reading or copying. 7. Additional Terms.

"Additional permissions" are terms that supplement the terms of this License by making exceptions from one or more of its conditions. Additional permissions that are applicable to the entire Program shall be treated as though they were included in this License, to the extent that they are valid under applicable law. If additional permissions apply only to part of the Program, that part may be used separately under those permissions, but the entire Program remains governed by this License without regard to the additional permissions.

When you convey a copy of a covered work, you may at your option remove any additional permissions from that copy, or from any part of it. (Additional permissions may be written to require their own removal in certain cases when you modify the work.) You may place additional permissions on material, added by you to a covered work, for which you have or can give appropriate copyright permission.

Notwithstanding any other provision of this License, for material you add to a covered work, you may (if authorized by the copyright holders of that material) supplement the terms of this License with terms:

* a) Disclaiming warranty or limiting liability differently from the terms of sections 15 and 16 of this License; or * b) Requiring preservation of specified reasonable legal notices or author attributions in that material or in the Appropriate Legal Notices displayed by works containing it; or * c) Prohibiting misrepresentation of the origin of that material, or requiring that modified versions of such material be marked in reasonable ways as different from the original version; or * d) Limiting the use for publicity purposes of names of licensors or authors of the material; or * e) Declining to grant rights under trademark law for use of some trade names, trademarks, or service marks; or * f) Requiring indemnification of licensors and authors of that material by anyone who conveys the material (or modified versions of it) with contractual assumptions of liability to the recipient, for any liability that these contractual assumptions directly impose on those licensors and authors.

All other non-permissive additional terms are considered "further restrictions" within the meaning of section 10. If the Program as you received it, or any part of it, contains a notice stating that it is governed by this License along with a term that is a further restriction, you may remove that term. If a license document contains a further restriction but permits relicensing or conveying under this License, you may add to a covered work material governed by the terms of that license document, provided that the further restriction does not survive such relicensing or conveying.

If you add terms to a covered work in accord with this section, you must place, in the relevant source files, a statement of the additional terms that apply to those files, or a notice indicating where to find the applicable terms.

Additional terms, permissive or non-permissive, may be stated in the form of a separately written license, or stated as exceptions; the above requirements apply either way. 8. Termination.

You may not propagate or modify a covered work except as expressly provided under this License. Any attempt otherwise to propagate or modify it is void, and will automatically terminate your rights under this License (including any patent licenses granted under the third paragraph of section 11).

However, if you cease all violation of this License, then your license from a particular copyright holder is reinstated (a) provisionally, unless and until the copyright holder explicitly and finally terminates your license, and (b) permanently, if the copyright holder fails to notify you of the violation by some reasonable means prior to 60 days after the cessation.

Moreover, your license from a particular copyright holder is reinstated permanently if the copyright holder notifies you of the violation by some reasonable means, this is the first time you have received notice of violation of this License (for any work)

from that copyright holder, and you cure the violation prior to 30 days after your receipt of the notice.

Termination of your rights under this section does not terminate the licenses of parties who have received copies or rights from you under this License. If your rights have been terminated and not permanently reinstated, you do not qualify to receive new licenses for the same material under section 10. 9. Acceptance Not Required for Having Copies.

You are not required to accept this License in order to receive or run a copy of the Program. Ancillary propagation of a covered work occurring solely as a consequence of using peer-to-peer transmission to receive a copy likewise does not require acceptance. However, nothing other than this License grants you permission to propagate or modify any covered work. These actions infringe copyright if you do not accept this License. Therefore, by modifying or propagating a covered work, you indicate your acceptance of this License to do so. 10. Automatic Licensing of Downstream Recipients.

Each time you convey a covered work, the recipient automatically receives a license from the original licensors, to run, modify and propagate that work, subject to this License. You are not responsible for enforcing compliance by third parties with this License.

An "entity transaction" is a transaction transferring control of an organization, or substantially all assets of one, or subdividing an organization, or merging organizations. If propagation of a covered work results from an entity transaction, each party to that transaction who receives a copy of the work also receives whatever licenses to the work the party's predecessor in interest had or could give under the previous paragraph, plus a right to possession of the Corresponding Source of the work from the predecessor in interest, if the predecessor has it or can get it with reasonable efforts.

You may not impose any further restrictions on the exercise of the rights granted or affirmed under this License. For example, you may not impose a license fee, royalty, or other charge for exercise of rights granted under this License, and you may not initiate litigation (including a cross-claim or counterclaim in a lawsuit) alleging that any patent claim is infringed by making, using, selling, offering for sale, or importing the Program or any portion of it. 11. Patents.

A "contributor" is a copyright holder who authorizes use under this License of the Program or a work on which the Program is based. The work thus licensed is called the contributor's "contributor version".

A contributor's "essential patent claims" are all patent claims owned or controlled by the contributor, whether already acquired or hereafter acquired, that would be infringed by some manner, permitted by this License, of making, using, or selling its contributor version, but do not include claims that would be infringed only as a consequence of further modification of the contributor version. For purposes of this definition, "control" includes the right to grant patent sublicenses in a manner consistent with the requirements of this License.

Each contributor grants you a non-exclusive, worldwide, royalty-free patent license under the contributor's essential patent claims, to make, use, sell, offer for sale, import and otherwise run, modify and propagate the contents of its contributor version.

In the following three paragraphs, a "patent license" is any express agreement or commitment, however denominated, not to enforce a patent (such as an express permission to practice a patent or covenant not to sue for patent infringement). To "grant" such a patent license to a party means to make such an agreement or commitment not to enforce a patent against the party.

If you convey a covered work, knowingly relying on a patent license, and the Corresponding Source of the work is not available for anyone to copy, free of charge and under the terms of this License, through a publicly available network server or other readily accessible means, then you must either (1) cause the Corresponding Source to be so available, or (2) arrange to deprive yourself of the benefit of the patent license for this particular work, or (3) arrange, in a manner consistent with the requirements of this License, to extend the patent license to downstream recipients. "Knowingly relying" means you have actual knowledge that, but for the patent license, your conveying the covered work in a country, or your recipient's use of the covered work in a country, would infringe one or more identifiable patents in that country that you have reason to believe are valid.

If, pursuant to or in connection with a single transaction or arrangement, you convey, or propagate by procuring conveyance of, a covered work, and grant a patent license to some of the parties receiving the covered work authorizing them to use, propagate, modify or convey a specific copy of the covered work, then the patent license you grant is automatically extended to all recipients of the covered work and works based on it.

A patent license is "discriminatory" if it does not include within the scope of its coverage, prohibits the exercise of, or is conditioned on the non-exercise of one or more of the rights that are specifically granted under this License. You may not convey a covered work if you are a party to an arrangement with a third party that is in the business of distributing software, under which you make payment to the third party based on the extent of your activity of conveying the work, and under which the third party grants, to any of the parties who would receive the covered work from you, a discriminatory patent license (a) in connection with copies of the covered work conveyed by you (or copies made from those copies), or (b) primarily for and in connection with specific products or compilations that contain the covered work, unless you entered into that arrangement, or that patent license was granted, prior to 28 March 2007.

Nothing in this License shall be construed as excluding or limiting any implied license or other defenses to infringement that may otherwise be available to you under applicable patent law. 12. No Surrender of Others' Freedom.

If conditions are imposed on you (whether by court order, agreement or otherwise) that contradict the conditions of this License, they do not excuse you from the conditions of this License. If you cannot convey a covered work so as to satisfy simultaneously your obligations under this License and any other pertinent obligations, then as a consequence you may not convey it at all. For example, if you agree to terms that obligate you to collect a royalty for further conveying from those to whom you convey the Program, the only way you could satisfy both those terms and this License would be to refrain entirely from conveying the Program. 13. Use with the GNU Affero General Public License.

Notwithstanding any other provision of this License, you have permission to link or combine any covered work with a work licensed under version 3 of the GNU Affero General Public License into a single combined work, and to convey the resulting work. The terms of this License will continue to apply to the part which is the covered work, but the special requirements of the GNU Affero General Public License, section 13, concerning interaction through a network will apply to the combination as such. 14. Revised Versions of this License.

The Free Software Foundation may publish revised and/or new versions of the GNU General Public License from time to time. Such new versions will be similar in spirit to the present version, but may differ in detail to address new problems or concerns.

Each version is given a distinguishing version number. If the Program specifies that a certain numbered version of the GNU General Public License "or any later version" applies to it, you have the option of following the terms and conditions either of that numbered version or of any later version published by the Free Software Foundation. If the Program does not specify a version number of the GNU General Public License, you may choose any version ever published by the Free Software Foundation.

If the Program specifies that a proxy can decide which future versions of the GNU General Public License can be used, that proxy's public statement of acceptance of a version permanently authorizes you to choose that version for the Program.

Later license versions may give you additional or different permissions. However, no additional obligations are imposed on any author or copyright holder as a result of your choosing to follow a later version. 15. Disclaimer of Warranty.

THERE IS NO WARRANTY FOR THE PROGRAM, TO THE EXTENT PERMITTED BY APPLICABLE LAW. EXCEPT WHEN OTHERWISE STATED IN WRITING THE COPYRIGHT HOLDERS AND/OR OTHER PARTIES PROVIDE THE PROGRAM "AS IS" WITHOUT WARRANTY OF ANY KIND, EITHER EXPRESSED OR IMPLIED, INCLUDING, BUT NOT LIMITED TO, THE IMPLIED WARRANTIES OF MERCHANTABILITY AND FITNESS FOR A PARTICULAR PURPOSE. THE ENTIRE RISK AS TO THE QUALITY AND PERFORMANCE OF THE PROGRAM IS WITH YOU. SHOULD THE PROGRAM PROVE DEFECTIVE, YOU ASSUME THE COST OF ALL NECESSARY SERVICING, REPAIR OR CORRECTION. 16. Limitation of Liability.

IN NO EVENT UNLESS REQUIRED BY APPLICABLE LAW OR AGREED TO IN WRITING WILL ANY COPYRIGHT HOLDER, OR ANY OTHER PARTY WHO MODIFIES AND/OR CONVEYS THE PROGRAM AS PERMITTED ABOVE, BE LIABLE TO YOU FOR DAMAGES, INCLUDING ANY GENERAL, SPECIAL, INCIDENTAL OR CONSEQUENTIAL DAMAGES ARISING OUT OF THE USE OR INABILITY TO USE THE PROGRAM (INCLUDING BUT NOT LIMITED TO LOSS OF DATA OR DATA BEING RENDERED INACCURATE OR LOSSES SUSTAINED BY YOU OR THIRD PARTIES OR A FAILURE OF THE PROGRAM TO OPERATE WITH ANY OTHER PROGRAMS), EVEN IF SUCH HOLDER OR OTHER PARTY HAS BEEN ADVISED OF THE POSSIBILITY OF SUCH DAMAGES. 17. Interpretation of Sections 15 and 16.

If the disclaimer of warranty and limitation of liability provided above cannot be given local legal ef-

fect according to their terms, reviewing courts shall apply local law that most closely approximates an absolute waiver of all civil liability in connection with the Program, unless a warranty or assumption of liability accompanies a copy of the Program in return for a fee.

END OF TERMS AND CONDITIONS How to Apply These Terms to Your New Programs

If you develop a new program, and you want it to be of the greatest possible use to the public, the best way to achieve this is to make it free software which everyone can redistribute and change under these terms.

To do so, attach the following notices to the program. It is safest to attach them to the start of each source file to most effectively state the exclusion of warranty; and each file should have at least the "copyright" line and a pointer to where the full notice is found.

<one line to give the program's name and a brief idea of what it does.> Copyright (C) <year> <name of author>

This program is free software: you can redistribute it and/or modify it under the terms of the GNU General Public License as published by the Free Software Foundation, either version 3 of the License, or (at your option) any later version.

This program is distributed in the hope that it will be useful, but WITHOUT ANY WARRANTY; without even the implied warranty of MERCHANTABILITY or FITNESS FOR A PARTICULAR PURPOSE. See the GNU General Public License for more details.

You should have received a copy of the GNU General Public License along with this program. If not, see <http://www.gnu.org/licenses/>.

Also add information on how to contact you by electronic and paper mail.

If the program does terminal interaction, make it output a short notice like this when it starts in an interactive mode:

<program> Copyright (C) <year> <name of author> This program comes with ABSOLUTELY NO WARRANTY; for details type 'show w'. This is free software, and you are welcome to redistribute it under certain conditions; type 'show c' for details.

The hypothetical commands 'show w' and 'show c' should show the appropriate parts of the General Public License. Of course, your program's commands might be different; for a GUI interface, you would use an "about box".

You should also get your employer (if you work as a programmer) or school, if any, to sign a "copyright disclaimer" for the program, if necessary. For more information on this, and how to apply and follow the GNU GPL, see <http://www.gnu.org/licenses/>.

The GNU General Public License does not permit incorporating your program into proprietary programs. If your program is a subroutine library, you may consider it more useful to permit linking proprietary applications with the library. If this is what you want to do, use the GNU Lesser General Public License instead of this License. But first, please read <http://www.gnu.org/philosophy/why-not-lgpl.html>.

24.2 GNU Free Documentation License

Version 1.3, 3 November 2008

Copyright © 2000, 2001, 2002, 2007, 2008 Free Software Foundation, Inc. <http://fsf.org/>

Everyone is permitted to copy and distribute verbatim copies of this license document, but changing it is not allowed. 0. PREAMBLE

The purpose of this License is to make a manual, textbook, or other functional and useful document "free" in the sense of freedom: to assure everyone the effective freedom to copy and redistribute it, with or without modifying it, either commercially or noncommercially. Secondarily, this License preserves for the author and publisher a way to get credit for their work, while not being considered responsible for modifications made by others.

This License is a kind of "copyleft", which means that derivative works of the document must themselves be free in the same sense. It complements the GNU General Public License, which is a copyleft license designed for free software.

We have designed this License in order to use it for manuals for free software, because free software needs free documentation: a free program should come with manuals providing the same freedoms that the software does. But this License is not limited to software manuals; it can be used for any textual work, regardless of subject matter or whether it is published as a printed book. We recommend this License principally for works whose purpose is instruction or reference. 1. APPLICABILITY AND DEFINITIONS

This License applies to any manual or other work, in any medium, that contains a notice placed by the copyright holder saying it can be distributed under the terms of this License. Such a notice grants a world-wide, royalty-free license, unlimited in duration, to use that work under the conditions stated herein. The "Document", below, refers to any such manual or work. Any member of the public is a licensee, and is addressed as "you". You accept the license if you copy, modify or distribute the work in a way requiring permission under copyright law.

A "Modified Version" of the Document means any work containing the Document or a portion of it, either copied verbatim, or with modifications and/or translated into another language.

A "Secondary Section" is a named appendix or a front-matter section of the Document that deals exclusively with the relationship of the publishers or

authors of the Document to the Document's overall subject (or to related matters) and contains nothing that could fall directly within that overall subject. (Thus, if the Document is in part a textbook of mathematics, a Secondary Section may not explain any mathematics.) The relationship could be a matter of historical connection with the subject or with related matters, or of legal, commercial, philosophical, ethical or political position regarding them.

The "Invariant Sections" are certain Secondary Sections whose titles are designated, as being those of Invariant Sections, in the notice that says that the Document is released under this License. If a section does not fit the above definition of Secondary then it is not allowed to be designated as Invariant. The Document may contain zero Invariant Sections. If the Document does not identify any Invariant Sections then there are none.

The "Cover Texts" are certain short passages of text that are listed, as Front-Cover Texts or Back-Cover Texts, in the notice that says that the Document is released under this License. A Front-Cover Text may be at most 5 words, and a Back-Cover Text may be at most 25 words.

A "Transparent" copy of the Document means a machine-readable copy, represented in a format whose specification is available to the general public, that is suitable for revising the document straightforwardly with generic text editors or (for images composed of pixels) generic paint programs or (for drawings) some widely available drawing editor, and that is suitable for input to text formatters or for automatic translation to a variety of formats suitable for input to text formatters. A copy made in an otherwise Transparent file format whose markup, or absence of markup, has been arranged to thwart or discourage subsequent modification by readers is not Transparent. An image format is not Transparent if used for any substantial amount of text. A copy that is not "Transparent" is called "Opaque".

Examples of suitable formats for Transparent copies include plain ASCII without markup, Texinfo input format, LaTeX input format, SGML or XML using a publicly available DTD, and standard-conforming simple HTML, PostScript or PDF designed for human modification. Examples of transparent image formats include PNG, XCF and JPG. Opaque formats include proprietary formats that can be read and edited only by proprietary word processors, SGML or XML for which the DTD and/or processing tools are not generally available, and the machine-generated HTML, PostScript or

PDF produced by some word processors for output purposes only.

The "Title Page" means, for a printed book, the title page itself, plus such following pages as are needed to hold, legibly, the material this License requires to appear in the title page. For works in formats which do not have any title page as such, "Title Page" means the text near the most prominent appearance of the work's title, preceding the beginning of the body of the text.

The "publisher" means any person or entity that distributes copies of the Document to the public.

A section "Entitled XYZ" means a named subunit of the Document whose title either is precisely XYZ or contains XYZ in parentheses following text that translates XYZ in another language. (Here XYZ stands for a specific section name mentioned below, such as "Acknowledgements", "Dedications", "Endorsements", or "History".) To "Preserve the Title" of such a section when you modify the Document means that it remains a section "Entitled XYZ" according to this definition.

The Document may include Warranty Disclaimers next to the notice which states that this License applies to the Document. These Warranty Disclaimers are considered to be included by reference in this License, but only as regards disclaiming warranties: any other implication that these Warranty Disclaimers may have is void and has no effect on the meaning of this License. 2. VERBATIM COPYING

You may copy and distribute the Document in any medium, either commercially or noncommercially, provided that this License, the copyright notices, and the license notice saying this License applies to the Document are reproduced in all copies, and that you add no other conditions whatsoever to those of this License. You may not use technical measures to obstruct or control the reading or further copying of the copies you make or distribute. However, you may accept compensation in exchange for copies. If you distribute a large enough number of copies you must also follow the conditions in section 3.

You may also lend copies, under the same conditions stated above, and you may publicly display copies. 3. COPYING IN QUANTITY

If you publish printed copies (or copies in media that commonly have printed covers) of the Document, numbering more than 100, and the Document's license notice requires Cover Texts, you

must enclose the copies in covers that carry, clearly and legibly, all these Cover Texts: Front-Cover Texts on the front cover, and Back-Cover Texts on the back cover. Both covers must also clearly and legibly identify you as the publisher of these copies. The front cover must present the full title with all words of the title equally prominent and visible. You may add other material on the covers in addition. Copying with changes limited to the covers, as long as they preserve the title of the Document and satisfy these conditions, can be treated as verbatim copying in other respects.

If the required texts for either cover are too voluminous to fit legibly, you should put the first ones listed (as many as fit reasonably) on the actual cover, and continue the rest onto adjacent pages.

If you publish or distribute Opaque copies of the Document numbering more than 100, you must either include a machine-readable Transparent copy along with each Opaque copy, or state in or with each Opaque copy a computer-network location from which the general network-using public has access to download using public-standard network protocols a complete Transparent copy of the Document, free of added material. If you use the latter option, you must take reasonably prudent steps, when you begin distribution of Opaque copies in quantity, to ensure that this Transparent copy will remain thus accessible at the stated location until at least one year after the last time you distribute an Opaque copy (directly or through your agents or retailers) of that edition to the public.

It is requested, but not required, that you contact the authors of the Document well before redistributing any large number of copies, to give them a chance to provide you with an updated version of the Document. 4. MODIFICATIONS

You may copy and distribute a Modified Version of the Document under the conditions of sections 2 and 3 above, provided that you release the Modified Version under precisely this License, with the Modified Version filling the role of the Document, thus licensing distribution and modification of the Modified Version to whoever possesses a copy of it. In addition, you must do these things in the Modified Version:

* A. Use in the Title Page (and on the covers, if any) a title distinct from that of the Document, and from those of previous versions (which should, if there were any, be listed in the History section of the Document). You may use the same title as a previous version if the original publisher of that version gives permission. * B. List on the Title

Page, as authors, one or more persons or entities responsible for authorship of the modifications in the Modified Version, together with at least five of the principal authors of the Document (all of its principal authors, if it has fewer than five), unless they release you from this requirement. * C. State on the Title page the name of the publisher of the Modified Version, as the publisher. * D. Preserve all the copyright notices of the Document. * E. Add an appropriate copyright notice for your modifications adjacent to the other copyright notices. * F. Include, immediately after the copyright notices, a license notice giving the public permission to use the Modified Version under the terms of this License, in the form shown in the Addendum below. * G. Preserve in that license notice the full lists of Invariant Sections and required Cover Texts given in the Document's license notice. * H. Include an unaltered copy of this License. * I. Preserve the section Entitled "History", Preserve its Title, and add to it an item stating at least the title, year, new authors, and publisher of the Modified Version as given on the Title Page. If there is no section Entitled "History" in the Document, create one stating the title, year, authors, and publisher of the Document as given on its Title Page, then add an item describing the Modified Version as stated in the previous sentence. * J. Preserve the network location, if any, given in the Document for public access to a Transparent copy of the Document, and likewise the network locations given in the Document for previous versions it was based on. These may be placed in the "History" section. You may omit a network location for a work that was published at least four years before the Document itself, or if the original publisher of the version it refers to gives permission. * K. For any section Entitled "Acknowledgements" or "Dedications", Preserve the Title of the section, and preserve in the section all the substance and tone of each of the contributor acknowledgements and/or dedications given therein. * L. Preserve all the Invariant Sections of the Document, unaltered in their text and in their titles. Section numbers or the equivalent are not considered part of the section titles. * M. Delete any section Entitled "Endorsements". Such a section may not be included in the Modified Version. * N. Do not retitle any existing section to be Entitled "Endorsements" or to conflict in title with any Invariant Section. * O. Preserve any Warranty Disclaimers.

If the Modified Version includes new front-matter sections or appendices that qualify as Secondary Sections and contain no material copied from the Document, you may at your option designate some or all of these sections as invariant. To do this, add their titles to the list of Invariant Sections in the Modified Version's license notice. These titles must be distinct from any other section titles.

You may add a section Entitled "Endorsements", provided it contains nothing but endorsements of your Modified Version by various parties—for example, statements of peer review or that the text has been approved by an organization as the authoritative definition of a standard.

You may add a passage of up to five words as a Front-Cover Text, and a passage of up to 25 words as a Back-Cover Text, to the end of the list of Cover Texts in the Modified Version. Only one passage of Front-Cover Text and one of Back-Cover Text may be added by (or through arrangements made by) any one entity. If the Document already includes a cover text for the same cover, previously added by you or by arrangement made by the same entity you are acting on behalf of, you may not add another; but you may replace the old one, on explicit permission from the previous publisher that added the old one.

The author(s) and publisher(s) of the Document do not by this License give permission to use their names for publicity or to assert or imply endorsement of any Modified Version. 5. COMBINING DOCUMENTS

You may combine the Document with other documents released under this License, under the terms defined in section 4 above for modified versions, provided that you include in the combination all of the Invariant Sections of all of the original documents, unmodified, and list them all as Invariant Sections of your combined work in its license notice, and that you preserve all their Warranty Disclaimers.

The combined work need only contain one copy of this License, and multiple identical Invariant Sections may be replaced with a single copy. If there are multiple Invariant Sections with the same name but different contents, make the title of each such section unique by adding at the end of it, in parentheses, the name of the original author or publisher of that section if known, or else a unique number. Make the same adjustment to the section titles in the list of Invariant Sections in the license notice of the combined work.

In the combination, you must combine any sections Entitled "History" in the various original documents, forming one section Entitled "History"; likewise combine any sections Entitled "Acknowledgements", and any sections Entitled "Dedications". You must delete all sections Entitled "Endorsements". 6. COLLECTIONS OF DOCUMENTS

You may make a collection consisting of the Document and other documents released under this License, and replace the individual copies of this License in the various documents with a single copy that is included in the collection, provided that you follow the rules of this License for verbatim copying of each of the documents in all other respects.

You may extract a single document from such a collection, and distribute it individually under this License, provided you insert a copy of this License into the extracted document, and follow this License in all other respects regarding verbatim copying of that document. 7. AGGREGATION WITH INDEPENDENT WORKS

A compilation of the Document or its derivatives with other separate and independent documents or works, in or on a volume of a storage or distribution medium, is called an "aggregate" if the copyright resulting from the compilation is not used to limit the legal rights of the compilation's users beyond what the individual works permit. When the Document is included in an aggregate, this License does not apply to the other works in the aggregate which are not themselves derivative works of the Document.

If the Cover Text requirement of section 3 is applicable to these copies of the Document, then if the Document is less than one half of the entire aggregate, the Document's Cover Texts may be placed on covers that bracket the Document within the aggregate, or the electronic equivalent of covers if the Document is in electronic form. Otherwise they must appear on printed covers that bracket the whole aggregate. 8. TRANSLATION

Translation is considered a kind of modification, so you may distribute translations of the Document under the terms of section 4. Replacing Invariant Sections with translations requires special permission from their copyright holders, but you may include translations of some or all Invariant Sections in addition to the original versions of these Invariant Sections. You may include a translation of this License, and all the license notices in the Document, and any Warranty Disclaimers, provided that you also include the original English version of this License and the original versions of those notices and disclaimers. In case of a disagreement between the translation and the original version of this License or a notice or disclaimer, the original version will prevail.

If a section in the Document is Entitled "Acknowledgements", "Dedications", or "History", the requirement (section 4) to Preserve its Title (section 1) will typically require changing the actual title. 9. TERMINATION

You may not copy, modify, sublicense, or distribute the Document except as expressly provided under this License. Any attempt otherwise to copy, modify, sublicense, or distribute it is void, and will automatically terminate your rights under this License.

However, if you cease all violation of this License, then your license from a particular copyright holder is reinstated (a) provisionally, unless and until the copyright holder explicitly and finally terminates your license, and (b) permanently, if the copyright holder fails to notify you of the violation by some reasonable means prior to 60 days after the cessation.

Moreover, your license from a particular copyright holder is reinstated permanently if the copyright holder notifies you of the violation by some reasonable means, this is the first time you have received notice of violation of this License (for any work) from that copyright holder, and you cure the violation prior to 30 days after your receipt of the notice.

Termination of your rights under this section does not terminate the licenses of parties who have received copies or rights from you under this License. If your rights have been terminated and not permanently reinstated, receipt of a copy of some or all of the same material does not give you any rights to use it. 10. FUTURE REVISIONS OF THIS LICENSE

The Free Software Foundation may publish new, revised versions of the GNU Free Documentation License from time to time. Such new versions will be similar in spirit to the present version, but may differ in detail to address new problems or concerns. See http://www.gnu.org/copyleft/.

Each version of the License is given a distinguishing version number. If the Document specifies that a particular numbered version of this License "or any later version" applies to it, you have the option of following the terms and conditions either of that specified version or of any later version that has been published (not as a draft) by the Free Software Foundation. If the Document does not specify a version number of this License, you may choose any version ever published (not as a draft) by the Free Software Foundation. If the Document specifies that a proxy can decide which future versions of this License can be used, that proxy's public statement of acceptance of a version permanently authorizes you to choose that version for the Document. 11. RELICENSING

"Massive Multiauthor Collaboration Site" (or "MMC Site") means any World Wide Web server that publishes copyrightable works and also provides prominent facilities for anybody to edit those works. A public wiki that anybody can edit is an example of such a server. A "Massive Multiauthor Collaboration" (or "MMC") contained in the site means any set of copyrightable works thus published on the MMC site.

"CC-BY-SA" means the Creative Commons Attribution-Share Alike 3.0 license published by Creative Commons Corporation, a not-for-profit corporation with a principal place of business in San Francisco, California, as well as future copyleft versions of that license published by that same organization.

"Incorporate" means to publish or republish a Document, in whole or in part, as part of another Document.

An MMC is "eligible for relicensing" if it is licensed under this License, and if all works that were first published under this License somewhere other than this MMC, and subsequently incorporated in whole or in part into the MMC, (1) had no cover texts or invariant sections, and (2) were thus incorporated prior to November 1, 2008.

The operator of an MMC Site may republish an MMC contained in the site under CC-BY-SA on the same site at any time before August 1, 2009, provided the MMC is eligible for relicensing. ADDENDUM: How to use this License for your documents

To use this License in a document you have written, include a copy of the License in the document and put the following copyright and license notices just after the title page:

Copyright (C) YEAR YOUR NAME. Permission is granted to copy, distribute and/or modify this document under the terms of the GNU Free Documentation License, Version 1.3 or any later version published by the Free Software Foundation; with no Invariant Sections, no Front-Cover Texts, and no Back-Cover Texts. A copy of the license is included in the section entitled "GNU Free Documentation License".

If you have Invariant Sections, Front-Cover Texts and Back-Cover Texts, replace the "with ... Texts." line with this:

with the Invariant Sections being LIST THEIR TITLES, with the Front-Cover Texts being LIST, and with the Back-Cover Texts being LIST.

If you have Invariant Sections without Cover Texts, or some other combination of the three, merge those two alternatives to suit the situation.

If your document contains nontrivial examples of program code, we recommend releasing these examples in parallel under your choice of free software license, such as the GNU General Public License, to permit their use in free software.

24.3 GNU Lesser General Public License