COURSE CODE: **BCH 201**

COURSE TITLE: **General Biochemistry 1**

NUMBER OF UNITS: **3 Units**

COURSE DURATION: **Two hours per week; Three hours Practical**

COURSE DETAILS:

Course Coordinator: Mr. B. O. Onunkwor
Email: @yahoo.com
Office Location: Room B307, COLNAS
Other Lecturers:
1. Prof. (Mrs) E.A. Balogun
2. Prof. O. Ademuyiwa
3. Dr. O. A. Akinloye
4. Dr. (Mrs) R. N. Ugbaja
5. Dr. Sunmonu
6. Mrs. O. A. Dosumu

COURSE CONTENT:

Principles of the chemical basis of life. The molecular basis of cellular structure-polysaccharides, lipids, proteins, nucleic acids. The cellular basis of life. Buffers, acidity and alkalinity; pH and pKa values and their effects on cellular activities. Chemistry of carbohydrates, lipids, amino acids and proteins, nucleic acids and nucleoproteins. Enzymes and co-enzymes. Vitamins.

COURSE REQUIREMENTS:

Students are expected to participate in all the course activities and have minimum of 75% attendance to be able to write the final examination.

READING LIST:

1. Bohinski, R.C. 1983. Mordern concepts in Biochemistry. 4th Edition. Allyn and Bacon.
2. Murray, R.K., Granner, D.K., Mayes, P.A. and Rodwell, V.W. 2003. Harper's Illustrated Biochemistry. 26th Edition. McGraw-Hill.

LECTURE NOTES

Nucleotides and Nucleic acids- Mrs. Dosumu

Nucleotides and Nucleic acids are compounds containing Nitrogen bases (aromatic cyclic structures possessing nitrogen atoms) as part of their structure.N**ucleic acids** are linear, unbranchedpolymers of nucleotides.

Nucleotides consist of three parts:

1. A five-carbon sugar (hence a **pentose**). Two kinds are found:

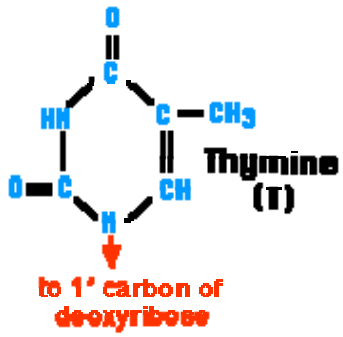
- **Deoxyribose**, which has a hydrogen atom attached to its #2 carbon atom (designated 2'), and
- **Ribose**, which has a hydroxyl group there.

Deoxyribose-containing nucleotides, the **deoxyribonucleotides**, are the monomers of deoxyribonucleic acids (**DNA**).

Ribose-containing nucleotides, the **ribonucleotides**, are the monomers of ribonucleic acids (**RNA**).

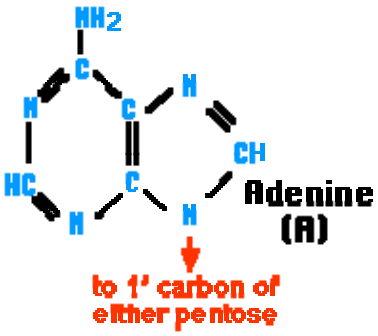
2. A nitrogen-containing ring structure called a **base**. The base is attached to the 1' carbon atom of the pentose. In **DNA**, four different bases are found:

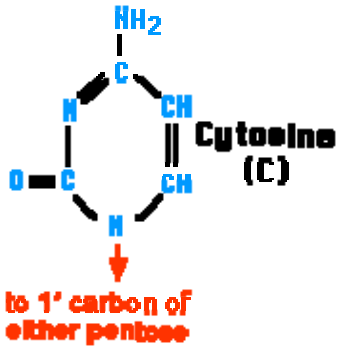
1. two **purines**, called **adenine** (**A**) and **guanine** (**G**)
2. two **pyrimidines**, called **thymine** (**T**) and **cytosine** (**C**)



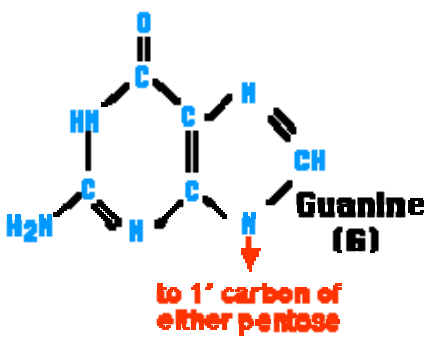
RNA contains:

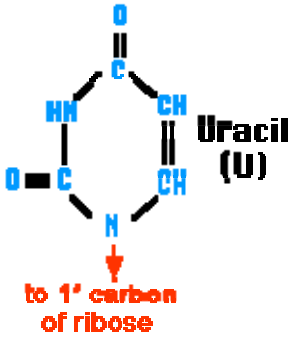
1. The same purines, **adenine** (**A**) and **guanine** (**G**).
2. RNA also uses the pyrimidine **cytosine** (**C**), but instead of thymine, it uses the pyrimidine **uracil** (**U**).





The Purines
The Pyrimidines





The combination of a base and a pentose is called a **nucleoside**.

The pentose sugar of the nucleotides is joined to the bases by **β-N-glycosidic bonds**. The β-N-glycosidic linkage is between $\mathbf{N^9\text{-}C^1}$ for purine bases and $\mathbf{N^1\text{-}C^1}$ for pyrimidine bases.

3. One (as shown in the first figure), two, or three **phosphate** groups. These are attached to the 5' carbon atom of the pentose. The product in each case is called a **nucleotide**.

Both DNA and RNA are assembled from **nucleoside triphosphates**.

For **DNA**, these are **dATP**, **dGTP**, **dCTP**, and **dTTP**.

For **RNA**, these are **ATP**, **GTP**, **CTP**, and **UTP**.

In both cases, as each nucleotide is attached, the second and third phosphates are removed.

The nucleosides and their mono-, di-, and triphosphates

	Base	Nucleoside	Nucleotides		
DNA	Adenine (A)	Deoxyadenosine	dAMP	dADP	dATP
	Guanine (G)	Deoxyguanosine	dGMP	dGDP	dGTP
	Cytosine (C)	Deoxycytidine	dCMP	dCDP	dCTP
	Thymine (T)	Deoxythymidine	dTMP	dTDP	dTTP
RNA	Adenine (A)	Adenosine	AMP	ADP	ATP
	Guanine (G)	Guanosine	GMP	GDP	GTP
	Cytosine (C)	Cytidine	CMP	CDP	CTP
	Uracil (U)	Uridine	UMP	UDP	UTP

Function of Nucleotides:

- Participate in the majority of biochemical reactions

- ATP - - energy currency (also ADP, AMP)

- UDP-glucose - - glycogen biosynthesis

- CoA, NAD+, NADP+, FAD are derivatives of nucleotides

- cAMP - - regulation of cellular processes (signaling)

- Nucleoside triphosphates (NTP) - - RNA

- Deoxynucleoside triphosphates (dNTP) - - DNA

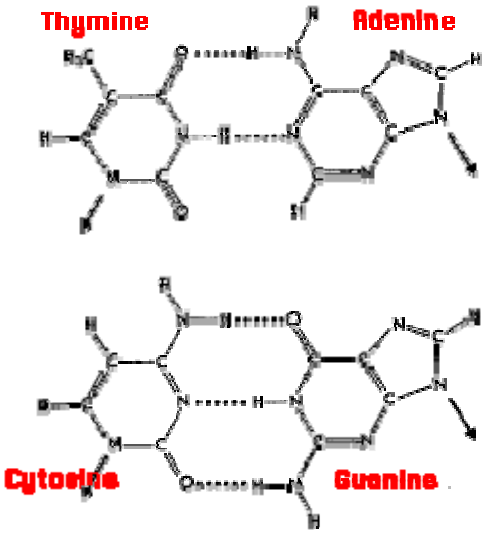
NUCLEIC ACIDS.

Nucleic acids are defined as biopolymers that are involved in the preservation/storage and transmission of genetic information from one generation to another. The nucleotides that make up the nucleic acids are linked by phosphodiester bonds between 3' and 5' positions of the sugars. The linkage is called **a 3'-5' phosphodiester bond.**

Base Pairing

The rules of base pairing (or nucleotide pairing) are:

- **A** with **T**: the purine **adenine** (A) always pairs with the pyrimidine **thymine** (T)
- **C** with **G**: the pyrimidine **cytosine** (C) always pairs with the purine **guanine** (G)



This is consistent with there not being enough space (20 Å) for two purines to fit within the helix and too much space for two pyrimidines to get close enough to each other to form hydrogen bonds between them.

But why not A with C and G with T?

The answer: only with A & T and with C & G are there opportunities to establish **hydrogen bonds** (shown here as dotted lines) between them (two between A & T; three between C & G). These relationships are often called the rules of **Watson-Crick** base pairing, named after the two scientists who discovered their structural basis.

The rules of base pairing tell us that if we can "read" the sequence of nucleotides on one strand of DNA, we can immediately deduce the complementary sequence on the other strand.

The rules of base pairing explain the phenomenon that whatever the amount of adenine (A) in the DNA of an organism, the amount of thymine (T) is the same (called Chargaff's rule). Similarly, whatever the amount of guanine (G), the amount of cytosine (C) is the same.

RIBONUCLEIC ACID (RNA)

Several types of RNA are synthesized in the nucleus of eukaryotic cells. Of particular interest are:

- **messenger RNA** (**mRNA**). This will later be **translated** into a polypeptide.
- **ribosomal RNA** (**rRNA**). This will be used in the building of ribosomes: machinery for synthesizing proteins by translating mRNA.
- **transfer RNA** (**tRNA**). RNA molecules that carry amino acids to the growing polypeptide.
- **small nuclear RNA** (**snRNA**). DNA transcription of the genes for mRNA, rRNA, and tRNA produces large precursor molecules (**"primary transcripts"**) that must be processed within the nucleus to produce the functional molecules for export to the cytosol. Some of these processing steps are mediated by snRNAs.
- **small nucleolar RNA** (**snoRNA**). These RNAs within the nucleolus have several functions (described below).
- **microRNA** (**miRNA**). These are tiny (~22 nucleotides) RNA molecules that regulate the expression of messenger RNA (mRNA) molecules. [Discussion]
- **XIST** RNA. This inactivates one of the two X chromosomes in female vertebrates. [Discussion]

Messenger RNA (mRNA)

Messenger RNA comes in a wide range of sizes reflecting the size of the polypeptide it encodes. Most cells produce small amounts of thousands of different mRNA molecules, each to be translated into a peptide needed by the cell.

Many mRNAs are common to most cells, encoding "housekeeping" proteins needed by all cells (e.g., the enzymes of glycolysis). Other mRNAs are specific for only certain types of cells. These encode proteins needed for the function of that particular cell (e.g., the mRNA for hemoglobin in the precursors of red blood cells).

Ribosomal RNA (rRNA)

There are 4 kinds. In eukaryotes, these are

- **18S rRNA**. One of these molecules, along with some 30 different protein molecules, is used to make the **small subunit** of the ribosome.
- **28S, 5.8S, and 5S rRNA**. One each of these molecules, along with some 45 different proteins, are used to make the **large subunit** of the ribosome.

The S number given each type of rRNA reflects the rate at which the molecules sediment in the ultracentrifuge. The larger the number, the larger the molecule (but not proportionally).

The 28S, 18S, and 5.8S molecules are produced by the processing of a single primary transcript from a cluster of identical copies of a single gene. The 5S molecules are produced from a different cluster of identical genes.

Transfer RNA (tRNA)

There are some 32 different kinds of tRNA in a typical eukaryotic cell.

- Each is the product of a separate gene.
- They are small (~4S), containing 73-93 nucleotides.
- Many of the bases in the chain pair with each other forming sections of double helix.
- The unpaired regions form 3 loops.
- Each kind of tRNA carries (at its 3′ end) one of the 20 **amino acids** (thus most amino acids have more than one tRNA responsible for them).
- At one loop, 3 unpaired bases form an **anticodon**.
- Base pairing between the anticodon and the complementary codon on a mRNA molecule brings the correct amino acid into the growing polypeptide chain.

Small Nuclear RNA (snRNA)

Approximately a dozen different genes for snRNAs, each present in multiple copies, have been identified. The snRNAs have various roles in the processing of the other classes of RNA. For example, several snRNAs are part of the **spliceosomes** that participate in converting pre-mRNA into mRNA by excising the introns and splicing the exons.

Small Nucleolar RNA (snoRNA)

As the name suggests, these small (60–300 nucleotides) RNAs are found in the nucleolus where they are responsible for several functions:

- Some participate in making ribosomes by helping to cut up the large RNA precursor of the 28S, 18S, and 5.8S molecules.
- Others chemically modify many of the nucleotides in rRNA, tRNA, and snRNA molecules, e.g., by adding methyl groups to ribose.
- Some have been implicated in the alternative splicing of pre-mRNA to different forms of mature mRNA.
- One snoRNA serves as the template for the synthesis of telomeres.

In vertebrates, the snoRNAs are made from **introns** removed during RNA processing.

Noncoding RNA

Only messenger RNA encodes polypeptides. All the other classes of RNA, including types not mentioned here, are thus called noncoding RNA. Much remains to be learned about the function(s) of some of them. But, taken together, noncoding RNAs probably account for two-thirds of the transcription going on in the nucleus.

HYDROLYSIS OF NUCLEIC ACIDS.

Hydrolysis of Nucleic acids by selective methods can be achieved chemically or enzymatically.

Chemical method of hydrolysis:

ACID HYDROLYSIS:

RNA is relatively resistant to the effects of dilute acid, but gentle treatment of DNA with 1Mm Hcl leads to hydrolysis of purine glycosidic bonds and the loss of purine bases from the DNA without affecting the pyrimidine deoxyribose bonds or the phosphodiester bonds of the backbone. At other chemical conditions, selective removal of pyrimidine bases occurs. In most cases, both Nucleic acids can be hydrolysed to their constituent bases by the treatment with 72% perchloric acid ($HClo_4^-$) for 1hour. The resulting nucleic acid derivative which is devoid of purine bases is called **APURINIC ACID**; while that devoid of pyrimidine bases is called **APYRIMIDINIC ACID.**

ALKALI HYDROLYSIS:

DNA is not susceptible to alkaline hydrolysis. On the other hand, RNA is alkali labile and is readily hydrolyzed by dilute sodium hydroxide.

Enzymatic hydrolysis of Nucleic acids:

Enzymes that hydrolyse nucleic acids are called **NUCLEASES.**

Some nucleases can hydrolyse linkages between 2 adjacent nucleotides at internal positions in the DNA or RNA strand and proceed stepwise from that end. Such nucleases are called **ENDONUCLEASES. Another class of nucleases can hydrolyse only the terminal nucleotide linkage, some at the 5' and others at the 3' end; these are called EXONUCLEASES.**

DNases (deoxyribonucleases) acts only on DNA

RNases(ribonucleases) are specific for RNA.

AMINO ACIDS AND PROTEINS- Mr. Onunkwor

`*The Amino Acids*

Proteins are formed by polymerizing monomers that are known as **amino acids** because they contain an amine ($-NH_2$) and a carboxylic acid ($-CO_2H$) functional group. With the exception of the amino acid proline, which is a secondary amine, the amino acids used to synthesize proteins are primary amines with the following generic formula.

An amino acid

These compounds are known as □-amino acids because the $-NH_2$ group is on the carbon atom next to the $-CO_2H$ group, the so-called carbon atom of the carboxylic acid.

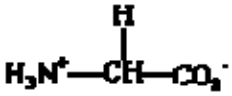
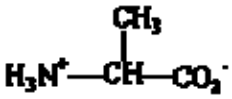
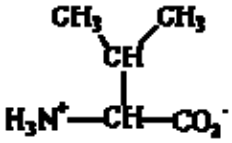
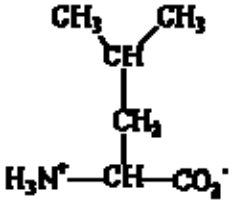
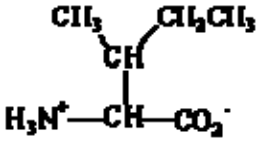
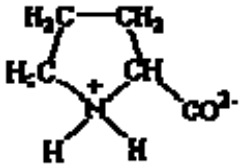
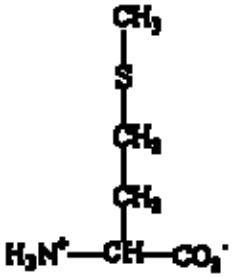
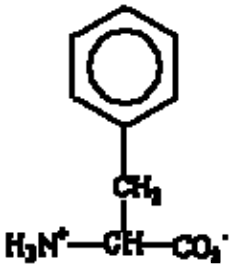
Zwitterions

The chemistry of amino acids is complicated by the fact that the $-NH_2$ group is a base and the $-CO_2H$ group is an acid. In aqueous solution, an H^+ ion is therefore transferred from one end of the molecule to the other to form a **zwitterion** (from the German meaning mongrel ion, or hybrid ion).

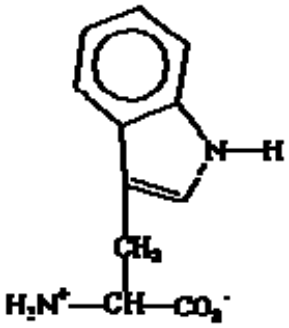
Zwitterions are simultaneously electrically charged and electrically neutral. They contain positive and negative charges, but the net charge on the molecule is zero.

The Amino Acids Used to Synthesize Proteins

More than 300 amino acids are known, but only the twenty amino acids in the table below are used to synthesize proteins. Most of these amino acids differ only in the nature of the *R* substituent. The standard amino acids are therefore classified on the basis of these *R* groups. Amino acids with nonpolar substituents are said to be *hydrophobic* (water-hating). Amino acids with polar *R* groups that form hydrogen bonds to water are classified as *hydrophilic* (water-loving). The remaining amino acids have substituents that carry either negative or positive charges in aqueous solution at neutral pH and are therefore strongly hydrophilic. *The 20 Standard Amino Acids are:*

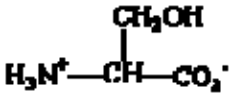
NAME	*STRUCTURE (AT NEUTRAL pH)*
Nonpolar (Hydrophobic) R Groups	
Glycine (Gly)	
Alanine (Ala)	
Valine (Val)	
Leucine (Leu)	
Isoleucine (Ile)	
Proline (Pro)	
Methionine (Met)	
Phenylalanine (Phe)	

Tryptophan (Trp)

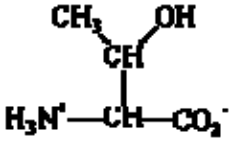


Polar (Hydrophilic) R Groups

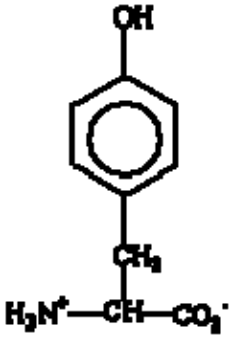
Serine
(ser)



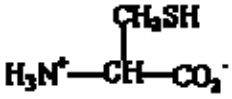
Threonine
(Thr)



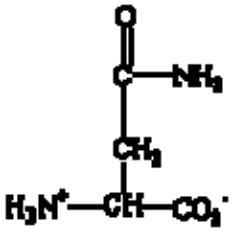
Tyrosine
(Tyr)



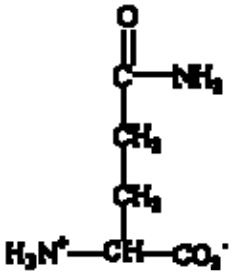
Cysteine
(Cys)



Asparagine
(Asn)

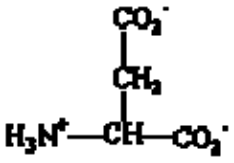


Glutamine
(Gln)

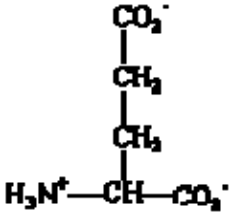


Negatively Charged R Groups

Aspartic acid
(Asp)

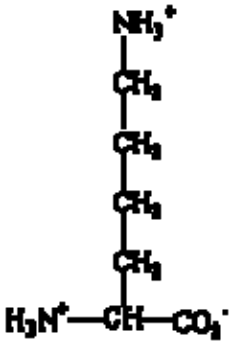


Glutamic acid
(Glu)

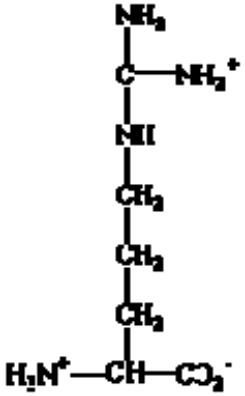


Positively Charged R Groups

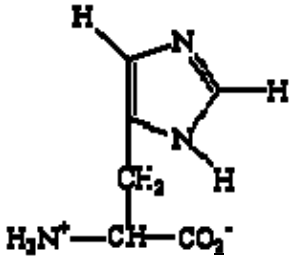
Lysine
(Lys)



Arginine
(Arg)



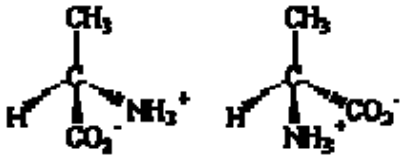
Histidine
(His)



Amino Acids as Stereoisomers

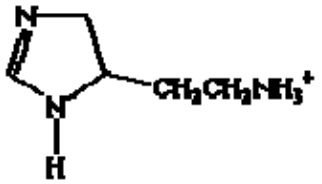
With the exception of glycine, the common amino acids all contain at least one chiral carbon atom. These amino acids therefore exist as pairs of stereoisomers. The structures of the D and L isomers of alanine are shown in the figure below. Although D amino acids can be found in nature, only the L isomers are used to form proteins. The D isomers are most often found attached to the cell walls of bacteria and in antibiotics

that attack bacteria. The presence of these D isomers protects the bacteria from enzymes the host organism uses to protect itself from bacterial infection by hydrolyzing the proteins in the bacterial cell wall.

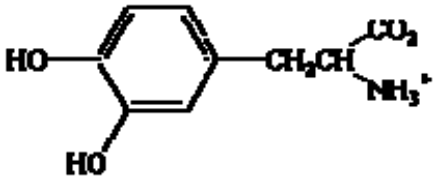


D-Alanine *L-Alanine*

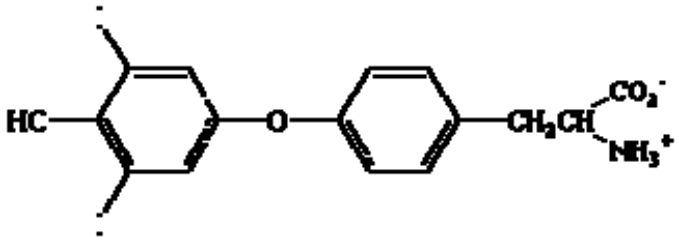
A few biologically important derivatives of the standard amino acids are shown in the figure below. Anyone who has used an "anti-histamine" to alleviate the symptoms of exposure to an allergen can appreciate the role that histamine — a decarboxylated derivative of histidine — plays in mediating the body's response to allergic reactions. L-DOPA, which is a derivative of tyrosine, has been used to treat Parkinson's disease. This compound received notoriety a few years ago in the film *Awakening*, which documented it's use as a treatment for other neurological disorders. Thyroxine, which is an iodinated ether of tyrosine, is a hormone that acts on the thyroid gland to stimulate the rate of metabolism.



Histamine



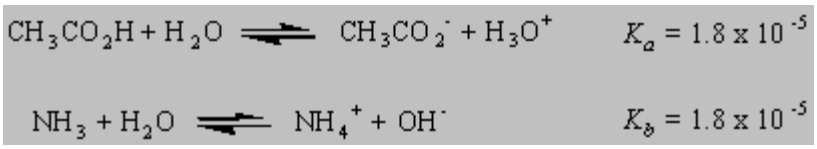
L-DOPA



Thyroxine

The Acid-Base Chemistry of the Amino Acids

Acetic acid and ammonia often play an important role in the discussion of the chemistry of acids and bases. One of these compounds is a weak acid; the other is a weak base.



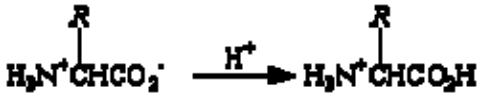
$$CH_3CO_2H + H_2O \rightleftharpoons CH_3CO_2^- + H_3O^+ \qquad K_a = 1.8 \times 10^{-5}$$

$$NH_3 + H_2O \rightleftharpoons NH_4^+ + OH^- \qquad K_b = 1.8 \times 10^{-5}$$

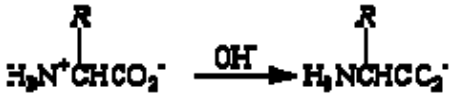
Thus, it is not surprising that an H^+ ion is transferred from one end of the molecule to the other when an amino acid dissolves in water.

The zwitterion is the dominant species in aqueous solutions at physiological pH (pH 7). The zwitterion can undergo acid-base reactions, howeer, if we add either a strong acid or a strong base to the solution.

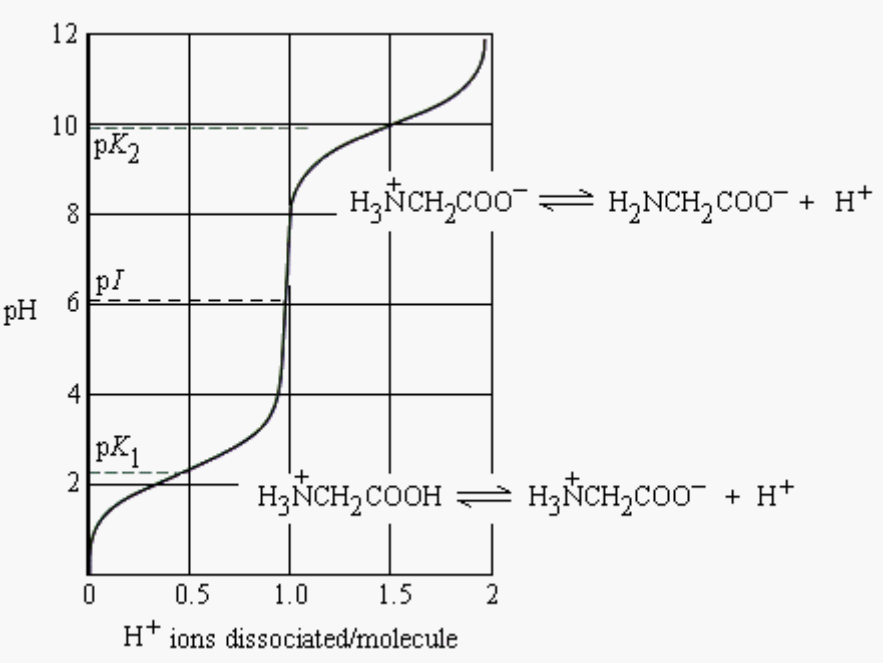
Imagine what would happen if we add a strong acid to a neutral solution of an amino acid in water. In the presence of a strong acid, the $-CO_2^-$ end of this molecule picks up an H^+ ion to form a molecule with a net positive charge.



In the presence of a strong base, the $-NH_3^+$ end of the molecule loses an H^+ ion to form a molecule with a net negative charge.



The figure below shows what happens to the pH of an acidic solution of glycine when this amino acid is titrated with a strong base, such as NaOH.



In order to understand this titration curve, let's start with the equation that describes the acid-dissocitation equilibrium constant expression for an acid, HA.

$$K_c = \frac{[H_3O^+][A^-]}{[HA]}$$

Let's now rearrange the K_a expression,

$$[H_3O^+] = K_a \times \frac{[HA]}{[A^-]}$$

take the log to the base 10 of both sides of this equation,

$$\log [H_3O^+] = \log K_a + \log \frac{[HA]}{[A^-]}$$

and then multiply both sides of the equation by -1.

$$- \log [H_3O^+] = - \log K_a - \log \frac{[HA]}{[A^-]}$$

By definition, the term on the left side of this equation is the pH of the solution and the first term on the right side is the pK_a of the acid.

$$pH = pK_a - \log \frac{[HA]}{[A^-]}$$

The negative sign on this right side of this equation is often viewed as "inconvenient." The derivation therefore continues by taking advantage of the following feature of logarithmic mathematics

$$- \log \frac{[HA]}{[A^-]} = \log \frac{[A^-]}{[HA]}$$

to give the following form of this equation.

$$pH = pK_a + \log \frac{[A^-]}{[HA]}$$

This equation is known as the **Henderson-Hasselbach equation**, and it can be used to calculate the pH of the solution at any point in the titration curve.

The following occurs as we go from left to right across this titration curve.

- The pH initially increases as we add base to the solution because the base deprotonates some of the positively charged $H_3N^+CH_2CO_2H$ ions that were present in the strongly acidic solution.
- The pH then levels off because we form a buffer solution in which we have reasonable concentrations of both an acid, $H_3N^+CH_2CO_2H$, and its conjugate base, $H_3N^+CH_2CO_2^-$.
- When virtually all of the $H_3N^+CH_2CO_2H$ molecules have been deprotonated, we no longer have a buffer solution and the pH rises rapidly when more NaOH is added to the solution.
- The pH then levels off as some of the neutral $H_3N^+CH_2CO_2^-$ molecules lose protons to form negatively charged $H_2NCH_2CO_2^-$ ions. When these ions are formed, we once again get a buffer solution in which the pH remains relatively constant until essentially all of the $H_3N^+CH_2CO_2H$ molecules have been converted into $H_2NCH_2CO_2^-$ ions.
- At this point, the pH rises rapidly until it reaches the value observed for a strong base.

The pH titration curve tells us the volume of base required to titrate the positively charged $H_3N^+CH_2CO_2H$ molecule to the $H_3N^+CH_2CO_2^-$ zwitterion. If we only add half as much base, only half of the positive ions would be titrated to zwitterions. In other words, the concentration of the $H_3N^+CH_2CO_2H$ and $H_3N^+CH_2CO_2^-$ ions would be the same. Or, using the symbolism in the Henderson-Hasselbach equation:

$$[HA] = [A^-]$$

Because the concentrations of these ions is the same, the logarithm of the ratio of their concentrations is zero.

$$\log \frac{[A^-]}{[HA]} = 0$$

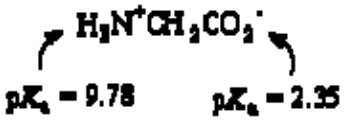
Thus, at this particular point in the titration curve, the Henderson-Hasselbach equation gives the following equality.

$$pH = pK_a$$

We can therefore determine the pK_a of an acid by measuring the pH of a solution in which the acid has been half-titrated.

Because there are two titratable groups in glycine, we get two points at which the amino acid is half-titrated. The first occurs when half of the positive $H_3N^+CH_2CO_2H$ molecules have been converted to neutral $H_3N^+CH_2CO_2^-$ ions. The second occurs when half of the $H_3N^+CH_2CO_2^-$ zwitterions have been converted to negatively charged $H_2NCH_2CO_2^-$ ions.

The following results are obtained when this technique is applied to glycine.

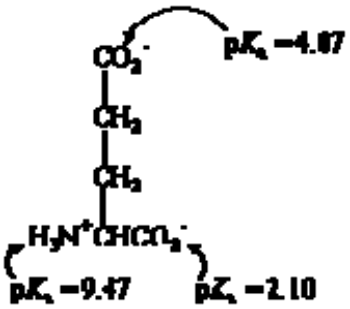


Let's compare these values with the pK_a's of acetic acid and the ammonium ion.

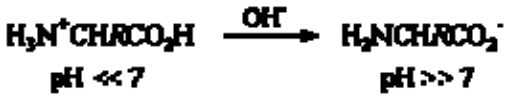
CH_3CO_2H	pK_a = 4.74
NH_4^+	pK_a = 9.24

The acid/base properties of the α-amino group in an amino acid are very similar to the properties of ammonia and the ammonium ion. The α-amine, however, has a significant effect on the acidity of the carboxylic acid. The -amine increases the value of K_a for the carboxylic acid by a factor of about 100.

The inductive effect of the α-amine can only be felt at the α-CO_2H group. If we look at the chemistry of glutamic acid, for example, the α-CO_2H group on the *R* substituent has an acidity that is close to that of acetic acid.



When we titrate an amino acid from the low end of the pH scale (pH 1) to the high end (pH 13), we start with an ion that has a net positive charge and end up with an ion that has a net negative charge.



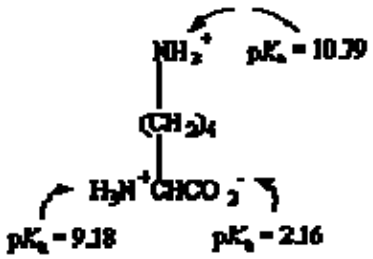
Somewhere between these extremes, we have to find a situation in which the vast majority of the amino acids are present as the zwitterion — with no net electric charge. This point is called the ***isoelectric point (pI)*** of the amino acid.

For simple amino acids, in which the *R* group doesn't contain any titratable groups, the isoelectric point can be calculated by averaging the pK_a values for the α-carboxylic acid and α-amino groups. Glycine, for example, has a p*I* of about 6.

$$pI = \frac{2.35 + 9.78}{2} = 6.1$$

At pH 6, more than 99.98% of the glycine molecules in this solution are present as the neutral $H_3N^+CH_2CO_2H$ zwitterion.

When calculating the p*I* of an amino acid that has a titratable group on the *R* side chain, it is useful to start by writing the structure of the amino acid at physiological pH (pH 7). Lysine, for example, could be represented by the following diagram.



At physiological pH, lysine has a net positive charge. Thus, we have to increase the pH of the solution to remove positive charge in order to reach the isoelectric point. The p*I* for lysine is simply the average of the pK_a's of the two $-NH_3^+$ groups.

$$pI = \frac{9.18 + 10.79}{2} = 10.0$$

At this pH, all of the carboxylic acid groups are present as $-CO_2^-$ ions and the total population of the $-NH_3^+$ groups is equal to one. Thus, the net charge on the molecule at this pH is zero.

If we apply the same technique to the pK_a data for glutamic acid, given above, we get a p*I* of about 3.1. The three amino acids in this section therefore have very different p*I* values.

Glutamic acid	(*R* = $-CH_2CH_2CO_2^-$):	p*I* = 3.1
Glycine	(*R* = -H):	p*I* = 6.1
Lysine	(*R* = $-CH_2CH_2CH_2CH_2NH_3^+$):	p*I* = 10.0

Thus, it isn't surprising that a common technique for separating amino acids (or the proteins they form) involves placing a mixture in the center of a gel and then applying a strong voltage across this gel. This technique, which is known as **gel electrophoresis**, is based on the fact that amino acids or proteins that carry a net positive charge at the pH at which the separation is done will move toward the negative electrode, whereas those with a net negative charge will move toward the positive electrode.

There are 20 **amino acids** and 10 of them can be produced by human beings. The rest need to be acquired through eating food. If only one of the essential **amino acids** that the body cannot produce is not taken into the body in an appropriate amount, then a deficiency occurs. When a deficiency occurs, proteins that rely on the **amino acids** that are not present in sufficient quantity will degrade. The human body dos not store excess **amino acids** so the ones it doesn't produce need to be regularly obtained through the diet.

Cyclic standard amino acids

,L-proline, (is often wrongfully referred to as an imino acid)

Glucogenic amino acids

,Isoleucine, Valine, Threonine, Aspartic acid, Alanine, Alanine, Methionine, Glutamine, Phenylalanine, Glycine, Cysteine,, Serine, Asparagine, Arginine, Tyrosine, Tryptophan, Glutamate, L-proline

Ketogenic amino acids

Lysine, Branched Chain Amino Acids, Isoleucine, Leucine, Phenylalanine, Phenylalanine, Threonine, Tryptophan, Tyrosine

Essential amino acids

The following 10 standard amino acids are referred to as essential because they are required from diet-Histidine, Arginine, Isoleucine, Leucine, Lysine, Methionine, Phenylalanine, Threonine, Tryptophan, Valine. The remaining 10 standard amino acids are **Non essential amino** acids since they can be biologically synthesized in humans

Sulfur amino acids

Methionine, Cysteine

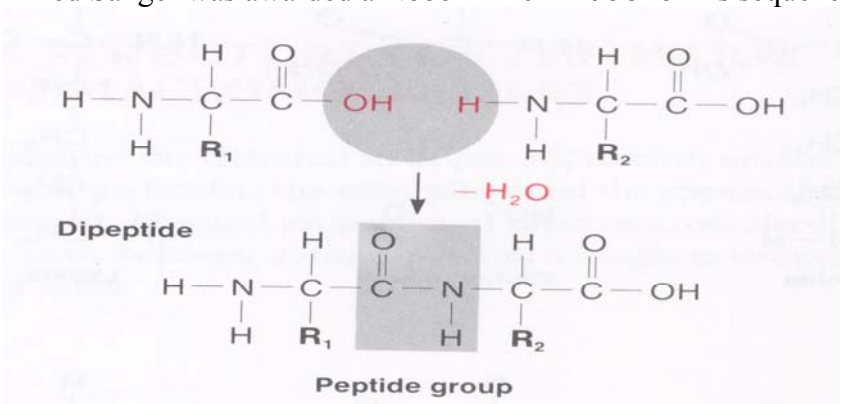
Cystine is not an amino acid but the product of the oxidation of two cysteins and is joined by a disulfide bond and is richly found in hair, tortoise shell,feathers, finger nails etc

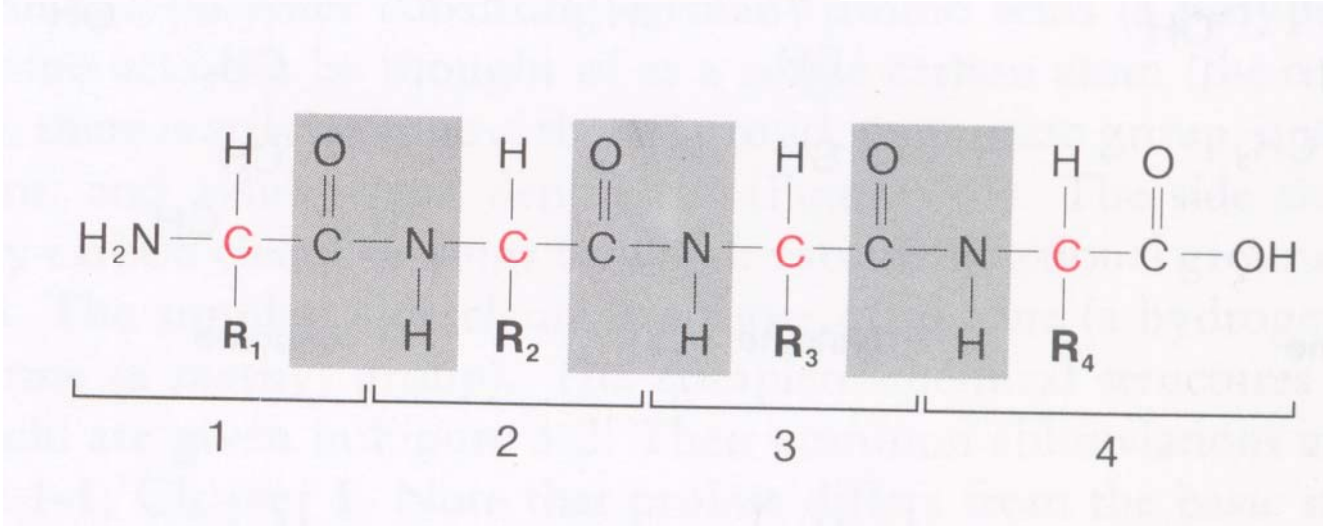
Proteins

Proteins are a polymer of amino acids (a polypeptide). Each amino acid in polypeptide is connected with peptide bond (a result of the chemical reaction between amino group of one amino acid with carboxyl group of another with the release of a water molecule)

-Amino acids are joined together by peptide bond to form a polypeptide chain
-Two ends of every protein are Amino group (NH2) & Carboxyl group (COOH) Known as N terminal (amino end terminal) & C terminal (carboxylic end terminal) respectively.
-The central chain, without the R group (side chain) is called: The Polypeptide backbone
-When 20 different amino acids are arranged in a polypeptide this is referred to as 20 amino acid sequence

-Determining of these sequences is referred to as protein sequencing (a very important technique in protein chemistry)
* Fred Sanger was awarded a Nobel Prize in 1958 for his sequencing work on insulin polypeptide





Functions of Proteins
Enzymes, Signalling, Transport, & storage, Structure & Movement, Nutrition (Casien & Ovalbumin) Defence, Immunity.

Classes of protein
1 Structural protein : Keratin, Collagen (give support to cells)
2 Dynamic protein : Hormone, enzyme (for catalytic purpose)
Based on the structure, protein can be divided to :

* Fibrin : Blood clotting
* Fibrous : Myosin (from muscle), keratin (from hair, finger nail, tortoise shell, birds feathers)
* Globular : Half sphere form/structure – eg. Enzyme

Size : Varied depending on functions
1amino acid = 110 Daltons
Most protein are highly folded

ENZYMES AND COENZYMES - Dr. Akinloye

Introduction

The use of enzymes in the diagnosis of disease is one of the important benefits derived from the intensive research in biochemistry since the 1940's. Enzymes have provided the basis for the field of clinical chemistry.

It is, however, only within the recent past few decades that interest in diagnostic enzymology has multiplied. Many methods currently on record in the literature are not in wide use, and there are still large areas of medical research in which the diagnostic potential of enzyme reactions has not been explored at all.

Early Enzyme Discoveries

The existence of enzymes has been known for well over a century. Some of the earliest studies were performed in 1835 by the Swedish chemist Jon Jakob Berzelius who termed their chemical action catalytic. It was not until 1926, however, that the first enzyme was obtained in pure form, a feat accomplished by James B. Sumner of Cornell University. Sumner was able to isolate and crystallize the enzyme urease from the jack bean. His work was to earn him the 1947 Nobel Prize.

John H. Northrop and Wendell M. Stanley of the Rockefeller Institute for Medical Research shared the 1947 Nobel Prize with Sumner. They discovered a complex procedure for isolating pepsin. This precipitation technique devised by Northrop and Stanley has been used to crystallize several enzymes.

Definition.

Enzymes are often referred to as biological catalyst that speeds up the rate of chemical reactions by converting substrate(s) to product(s).

N.B Not all enzymes are protein because we have ribozyme that is nucleic acid in nature.

Almost all processes in biological cells needs enzyme action(s) in order to occur at significant of appreciable rate.

Naming and Classification

Except for some of the originally studied enzymes such as pepsin, rennin, and trypsin, most enzyme names end in "ase". The International Union of Biochemistry (I.U.B.) initiated standards of enzyme nomenclature which recommend that enzyme names indicate both the substrate acted upon and the type of reaction catalyzed. Under this system, the enzyme uricase is called urate: O_2

oxidoreductase, while the enzyme glutamic oxaloacetic transaminase (GOT) is called L-aspartate: 2-oxoglutarate aminotransferase.

Enzymes can be classified by the kind of chemical reaction catalyzed.

1. Addition or removal of water
    A. Hydrolases - these include esterases, carbohydrases, nucleases, deaminases, amidases, and proteases
    B. Hydrases such as fumarase, enolase, aconitase and carbonic anhydrase
2. Transfer of electrons
    A. Oxidases
    B. Dehydrogenases
3. Transfer of a radical
    A. Transglycosidases - of monosaccharides
    B. Transphosphorylases and phosphomutases - of a phosphate group
    C. Transaminases - of amino group
    D. Transmethylases - of a methyl group
    E. Transacetylases - of an acetyl group
4. Splitting or forming a C-C bond
    A. Desmolases
5. Changing geometry or structure of a molecule
    A. Isomerases
6. Joining two molecules through hydrolysis of pyrophosphate bond in ATP or other tri-phosphate
    A. Ligases

Many enzymes are named by adding the suffix '-ase' to the name of their substrate e.g urease catalysis the hydrolysis of urea. However, this is not always true for all enzymes e.g pepsin, trypsin acts on protein. The classification based on the International Union of Biochemistry is broadly into six (6) classes thus:

CLASS1- OXIDOREDUCTASE- This group of enzyme catalyze the oxidation of one substrate with simultaneous reduction of another substrate .e.g alcohol dehydrogenase

CLASS 2- TRANSFERASE- They catalyze the transfer of functional group(s) other than hydrogen from one substrate to another; e.g hexose-6-phosphate transferase

CLASS 3- HYDROLASE- This class of enzyme hydrolyse ester, ether, peptide or glycosidic bonds by adding water and the breaks the bond, e.g acetylcholine hydrolase

CLASS 4- LYASE: This group of enzyme removes group from a particular substrate or breaks bonds by mechanism other than hydrolysis, e.g aldolase

CLASS 5- ISOMERASE-They catalyse the conversion of one isomer to the other. They produce optical geometric or positional isomer of substrate, e.g triose phosphate isomerase.

CLASS 6- LIGASE- This group catalyses the linkage of two substrates together, usually with the simultaneous hydrolysis of ATP, e.g acetyl CoA carboxylase.

GENERAL PROPERTIES OF ENZYME.

(i) Enzymes have enormous catalytic power i.e they can accelerate reaction rate by at least a million
(ii) Enzymes are highly specific i.e highly specific both in the choice of substrate and in reaction catalysed
(iii) Activities of some enzymes are regulated i.e different kind of regulatory mechanisms affect enzyme catalysed reaction.
(iv) Enzymes do not alter the reaction equilibria i.e enzymes do not alter the equilibrium position, meaning that they accelerates the forward and back ward reactions by precisely the same factor.
(v) Enzymes decrease the activation energy e.g the lowers the activation energy by reducing the transition state / activation complex
(vi) Enzymes transform different kinds of energy i.e energy of reactant could be converted into different form with high efficiency.

Chemical Nature of Enzymes

All known enzymes are proteins. They are high molecular weight compounds made up principally of chains of amino acids linked together by peptide bonds. See Figure 1.

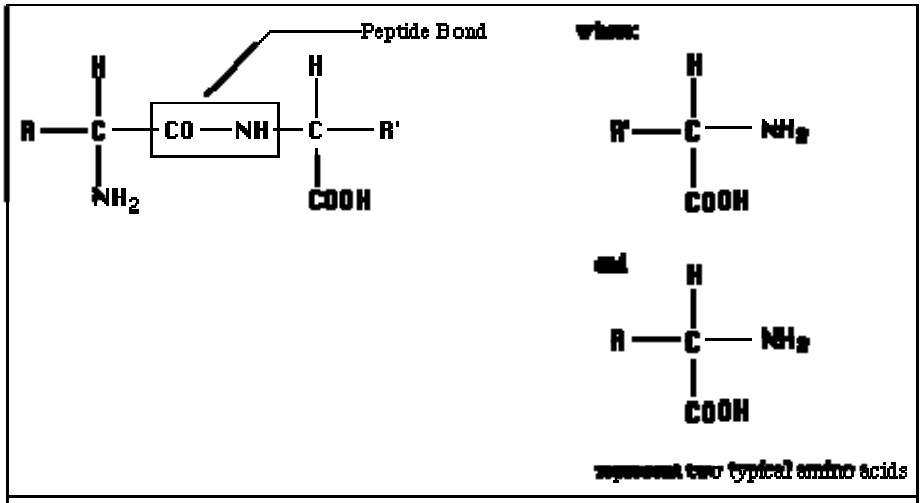
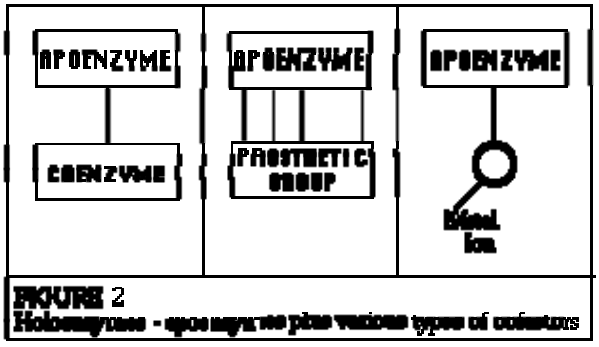


FIGURE 1
Typical protein structure - two amino acids joined by a peptide bond.

Enzymes can be denatured and precipitated with salts, solvents and other reagents. They have molecular weights ranging from 10,000 to 2,000,000.

Many enzymes require the presence of other compounds - cofactors - before their catalytic activity can be exerted. This entire active complex is referred to as the holoenzyme; i.e., apoenzyme (protein portion) plus the cofactor (coenzyme, prosthetic group or metal-ion-activator) is called the holoenzyme.



Apoenzyme + Cofactor = Holoenzyme

According to Holum, the cofactor may be:

1. A coenzyme - a non-protein organic substance which is dialyzable, thermostable and loosely attached to the protein part.

2. A prosthetic group - an organic substance which is dialyzable and thermostable which is firmly attached to the protein or apoenzyme portion.

3. A metal-ion-activator - these include K^{+}, Fe^{++}, Fe^{+++}, Cu^{++}, Co^{++}, Zn^{++}, Mn^{++}, Mg^{++}, Ca^{++}, and Mo^{+++}.

FACTORS AFFECTING THE RATE OF ENZYME CATALYSIS

Factors affecting the rate of enzyme catalyzed reactions include among others:

(i) temperature
(ii) pH
(iii) Substrate concentration
(iv) Presence or absence of activator(s) and/or inhibitor(s)

Temperature: The rate of an enzyme reaction varies with temperature according to the Arrhenius equation i.e rate=Ae (-E/RT). The equation explains the sensitivity of enzyme to temperature because of the relationship between the rate and temperature is exponential. Each enzyme has optimum temperature after which is starts to denature

pH: The state of ionization of amino residues in the active site of an enzyme is pH dependent. A typical enzyme has an optimum pH of activity.

Effect of substrate concentration: At constant enzyme concentration, when the sudstrate concentration is low, the rate of reaction is very low. However, this increases with an increase in substrate concentration. Later, a point will be reached beyond which further increase in substrate concentration will not produce significant increase in reaction velocity.

Influence of inhibitor /activator: Enzyme inhibitors combine specifically with an enzyme to reduce its ability to convert substrate to products while activator enhances the rate of an enzyme catalyzed reaction. There are two types of inhibitors namely:

(i) reversible inhibitor-which binds with non-covalent bonds
(ii) irreversible inhibitor-which bind with covalent bonds.

Reversible inhibitors are further divided into:

(i) competitive inhibitor i.e the one that competes with the substrate for binding at the active site
(ii) non-competitive inhibitor i.e the one that binds at some other site apart from the active site of the enzyme.
(iii) Uncompetitive inhibitor i.e the one that did not bind to the enzyme but only bind to the enzyme –substrate (ES) complex..

Active site of an enzyme.

The active sits of an enzyme is that region of the enzyme where catalysis takes place. It is also the region that binds the substrate and contributes the residues that directly participates in the making and breaking of bonds.

Some features of active site are:

(i) it is a relatively small portion of the total enzyme volume
(ii) it is a three dimensional entity
(iii) substrate binds with relatively weak forces
(iv) it is a cleft or crevice
(v) the specificity of binding depends on the precisely defined arrangement of atom in an active site

Note that: the interaction of substrate and enzyme could be expressed in term of two models namel:

(i) lock and key model
(ii) induced fit model.

Enzyme Kinetics

Michaelis and Menten derived equation for enzyme catalyzed reaction involing a single substrate and single product thus:

$S \rightarrow P$

as

$v = Vmax \times [S]/Km + [S]$

where v = initial velocity
Vmax = maximum velocity
[S] = substrate concentration
Km = Michealis-Menten constant.
Note that: any enzyme that obeys M-M equation will give an hyperbolic curve when the plot of v vs [S] is made.

Significance of Km and Vmax.
Km is the substrate concentration at half the maximum velocity. It is a measure of affinity of an enzyme for substrate i.e the higher the Km the lower the affinity and vice versa.
Vmax is used to express the efficiency of an enzyme operation i.e often used to compare the catalytic efficiency of different enzyme.

ALLOSTERIC ENZYMES
These are regulatory enzymes that functions through reversible non-covalent binding of a modulatory molecule. They usually determine the rate of overall sequence of reaction because they catalyze the committed/slowest step. Such enzyme is usually the first in the sequence of a multienzyme reaction system. They are known to have the following properties:

(i) They have both catalytic and regulatory sites for binding of substrate
(ii) Generally larger and more complex than the simple enzyme
(iii) Shows deviation from classical M-M behaviour in that thet give sigmodial curve for the plot of v vs [S].
(iv) They undergo conformational changes in binding of modulatory molecule
(v) They may be inhibited by their modulator (-ve modulator) or stimulated by modulator (+ve modulator)

CO-ENZYMES.

These are additional non-protein part of an enzyme that is required for enzymatic activities. Inorganic forms of coenzyme are called cofactors. Tightly forms of coenzyme are called prosthetic group.

The role of a cofactor is either:

(i) to alter the three-dimensional structure of the protein and/or the bound substrate in order to activate the interaction of the enzyme with its substrate
(ii) to actually participate in overall reaction as another substrate.

Different types of coenzymes, type of reaction and group transfer are given below

COENZYMES	**TYPE OF REACTION**	**GROUP TRANSFER**
NAD+/NADP+	oxidation-reduction	hydrogen (electron)
FAD, FMN	oxidation-reduction	hydrogen (electron)
Coenzyme A	activation and transfer of acyl group	acyl group
Lipoic acid	acyl group transfer	acyl group
Thiamine pyrophosphate	acyl group transfer	acyl group

Biotin	carbon (iv) oxide fixation	carbon (iv) oxide
Pyridoxal phosphate	transamination	amide ($-NH_2$)
Tetrahydrofolic acid	metabolism of one carbon fragment	$-CH_3$, $-CH_2$

LIPIDS- Dr. R. N. Ugbaja

BIOMEDICAL IMPORTANCE

The lipids are a heterogeneous group of compounds, including fats, oils, steroids, waxes, and related compounds, which are related more by their physical than by their chemical properties. They have the common property of being (1) relatively **insoluble in water** and (2) **soluble in nonpolar solvents** such as ether and chloroform. They are important dietary constituents not only because of their high energy value but also because of the fat-soluble vitamins and the essential fatty acids contained in the fat of natural foods. Fat is stored in **adipose tissue,** where it also serves as a thermal insulator in the subcutaneous tissues and around certain organs.

Nonpolar lipids act as **electrical insulators,** allowing rapid propagation of depolarization waves along **myelinated nerves.** Combinations of lipid and protein (lipoproteins) are important cellular constituents, occurring both in the cell **membrane** and in the mitochondria, and serving also as the means of **transporting lipids** in the blood. Knowledge of lipid biochemistry is necessary in understanding many important biomedical areas, eg, **obesity, diabetes mellitus, atherosclerosis,** and the role of various **polyunsaturated fatty acids** in nutrition and health.

As molecules that are largely hydrocarbon in nature, lipids represent highly reduced forms of carbon and, upon oxidation in metabolism, yield large amounts of energy. Lipids are thus the molecules of choice for metabolic energy storage.

The lipids found in biological systems are either **hydrophobic** (containing only nonpolar groups) or **amphipathic,** which means they possess both polar and nonpolar groups. The hydrophobic nature of lipid molecules allows membranes to act as effective barriers to more polar molecules.

LIPIDS ARE CLASSIFIED AS SIMPLE OR COMPLEX

1. Simple lipids: Esters of fatty acids with various alcohols.

a. **Fats:** Esters of fatty acids with trihydric alcohol, glycerol. **Oils** are fats in the liquid state.

b. **Waxes:** Esters of fatty acids with higher molecular weight monohydric alcohols.

2. Complex lipids: Esters of fatty acids containing groups in addition to an alcohol and a fatty acid.

a. **Phospholipids:** Lipids containing, in addition to fatty acids and an alcohol, a phosphoric acid residue. They frequently have nitrogen containing bases and other substituents, e.g, in **glycerophospholipids e.g. the phosphatidyl compounds,** the alcohol is glycerol and in **sphingophospholipids e.g sphingomyelins,** the alcohol is sphingosine.

b. **Glycolipids (glycosphingolipids):** Lipids containing a fatty acid, sphingosine, and carbohydrate e.g Cerebrosides and gangliosides.

c. **Other complex lipids:** Lipids such as sulfolipids and aminolipids. Lipoproteins may also be placed in this category.

3. Precursor and derived lipids: These include fatty acids, glycerol, steroids, other alcohols, fatty aldehydes, and ketone bodies, hydrocarbons, lipid-soluble vitamins, and hormones.

Because they are uncharged, acylglycerols (glycerides), cholesterol, and cholesteryl esters are termed **neutral lipids.**

Lipids in 1 and 2 can de hydrolysed into their constituent parts on complete hydrolysis while the lipids in 3 cannot be hydrolysed.

FUNCTIONS OF LIPIDS

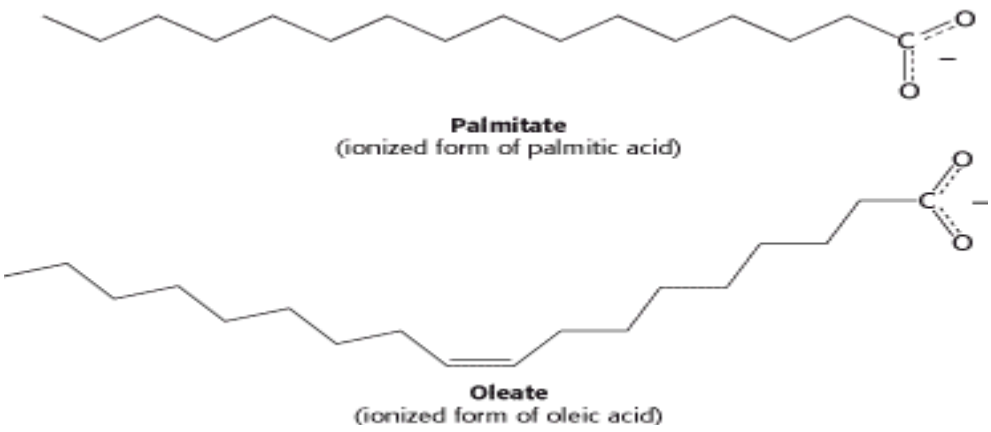
In the living organism, the functions of lipids are as follows:

1. **Provision of energy**
2. **As components of biological membranes**
3. **Homonal activity**
4. **Vitamin activity**
5. **Regulatory and Signal transducing effects**
6. **Immunological actions**
7. **Providing insulation against changes in external temp.**
8. **Protecting internal organs through cushioning effect**
9. **Giving shape and contour to body**

FATTY ACIDS

A **fatty acid** is composed of a long hydrocarbon chain ("tail") and a terminal carboxyl group (or "head"). The carboxyl group is normally ionized under physiological conditions. Fatty acids occur in large amounts in biological systems,but rarely in the free, uncomplexed state. They typically are esterified to glycerol or other backbone structures. Most of the fatty acids found in nature have an even number of carbon atoms (usually 14 to 24). Certain marine organisms, however, contain substantial amounts of fatty acids with odd numbers of carbon atoms. Fatty acids are either **saturated** (all carbon–carbon bonds are single bonds) or **unsaturated** (with one or more double bonds in the hydrocarbon chain). If a fatty acid has a single double bond, it is said to be **monounsaturated,** and if it has more than one, **polyunsaturated.** Fatty acids can be named or described in at least three ways, as listed in the Table below.

The systematic name for a fatty acid is derived from the name of its parent hydrocarbon by the substitution of *oic* for the final *e*. For example, the C18 saturated fatty acid is called *octadecanoic acid* because the parent hydrocarbon is octadecane. A C18 fatty acid with one double bond is called octadec*enoic* acid; with two double bonds, octadeca*dienoic* acid; and with three double bonds, octadeca*trienoic* acid. The notation 18:0 denotes a C18 fatty acid with no double bonds, whereas 18:2 signifies that there are two double bonds. The structures of the ionized forms of two common fatty acids palmitic acid (C16, saturated) and oleic acid (C18, monounsaturated) are shown in Figure below.



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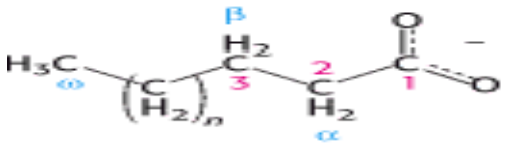
They are numbered starting at the carboxyl terminus, as shown in the margin. Carbon atoms 2 and 3 are often referred to as α and β, respectively. The methyl carbon atom at the distal end of the chain is called the ω-*carbon atom.*



Alternatively, the position of a double bond can be denoted by counting from the distal end, with the ω -carbon atom (the methyl carbon) as number 1. An ω -3 fatty acid, for example, has the structure shown in the margin.

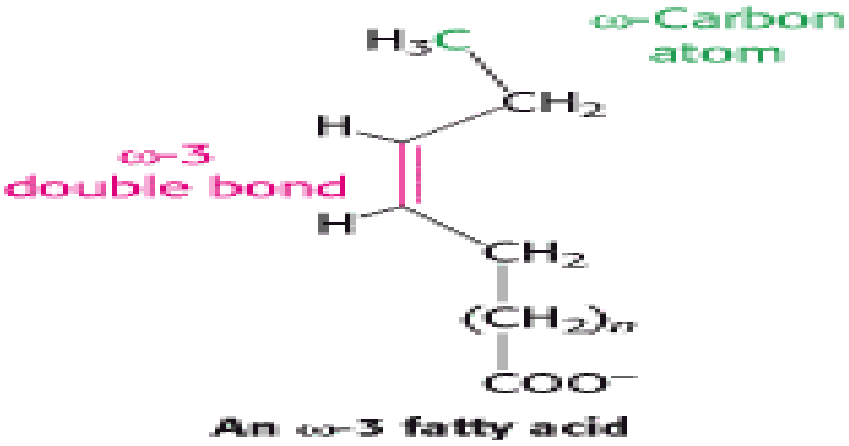


Table showing some Common Biological Fatty Acids

Number of Carbons	Common Name	Systematic Name	Symbol	Structure
Saturated fatty acids				
12	Lauric acid	Dodecanoic acid	12:0	CH3(CH2)10COOH
14	Myristic acid	Tetradecanoic acid	14:0	CH3(CH2)12COOH
16	Palmitic acid	Hexadecanoic acid	16:0	CH3(CH2)14COOH
18	Stearic acid	Octadecanoic acid	18:0	CH3(CH2)16COOH
20	Arachidic acid	Eicosanoic acid	20:0	CH3(CH2)18COOH
22	Behenic acid	Docosanoic acid	22:0	CH3(CH2)20COOH
24	Lignoceric acid	Tetracosanoic acid	24:0	CH3(CH2)22COOH
Unsaturated fatty acids (all double bonds are *cis*)				
16	Palmitoleic acid	9-Hexadecenoic acid	16:1	CH3(CH2)5CH=CH(CH2)7COOH
18	Oleic acid	9-Octadecenoic acid	18:1	CH3(CH2)7CH=CH(CH2)7COOH
18	Linoleic acid	9,12-Octadecadienoic acid	18:2	CH3(CH2)4(CH=CHCH2)2(CH2)6COOH
18	α-Linolenic acid	9,12,15-Octadecatrienoic acid	18:3	CH3CH2(CH=CHCH2)3(CH2)6COOH
18	γ-Linolenic acid	6,9,12-Octadecatrienoic acid	18:3	CH3(CH2)4(CH=CHCH2)3(CH2)3COOH
20	Arachidonic acid	5,8,11,14-Eicosatetraenoic acid	20:4	CH3(CH2)4(CH=CHCH2)4(CH2)2COOH
24	Nervonic acid	15-Tetracosenoic acid	24:1	CH3(CH2)7CH=CH(CH2)13COOH

Unsaturated fatty acids are slightly more abundant in nature than saturated fatty acids, especially in higher plants. The most common unsaturated fatty acid is **oleic acid,** or 18:1(9), with the number in parentheses indicating that the double bond is between carbons 9 and 10. The number of double bonds in an unsaturated fatty acid varies typically from one to four, but, in the fatty acids found in most bacteria, this number rarely exceeds one.

The double bonds found in fatty acids are nearly always in the *cis* configuration. As shown in Figure below, this causes a bend or "kink" in the fatty acid chain. This bend has very important consequences for the structure of biological membranes. Saturated fatty acid chains can pack closely together to form ordered, rigid arrays under certain conditions, but unsaturated fatty acids prevent such close packing and produce flexible, fluid aggregates.

Some fatty acids are not synthesized by mammals and yet are necessary for normal growth and life. These *essential fatty acids* include **linoleic** and **γ-linolenic acids.** These must be obtained by mammals in their diet (specifically from plant sources). **Arachidonic acid,** which is not found in plants, can only be synthesized by mammals from linoleic acid. At least one function of the essential fattyacids is to serve as a precursor for the synthesis of **eicosanoids,** such as *prostaglandins,* a class of compounds that exert hormone-like effects in many physiological processes.

In addition to unsaturated fatty acids, several other modified fatty acids are found in nature. Microorganisms, for example, often contain branched-chain fatty acids, such as **tuberculostearic acid.** When these fatty acids are incorporated in membranes, the methyl group constitutes a local structural perturbation in a manner similar to the double bonds in unsaturated fatty acids. Some bacteria also synthesize fatty acids containing cyclic structures such as cyclopropane, cyclopropene, and even cyclopentane rings.

The properties of fatty acids and of lipids derived from them are markedly dependent on chain length and degree of saturation. Unsaturated fatty acids have lower melting points than saturated fatty acids of the same length. For example, the melting point of stearic acid is 69.6°C, whereas that of oleic acid (which contains one cis double bond) is 13.4°C. The melting points of polyunsaturated fatty acids of the C18 series are even lower. Chain length also affects the

melting point, as illustrated by the fact that the melting temperature of palmitic acid (C16) is 6.5 degrees lower than that of stearic acid (C18). Thus, *short chain length and unsaturation enhance the fluidity of fatty acids and of their derivatives*

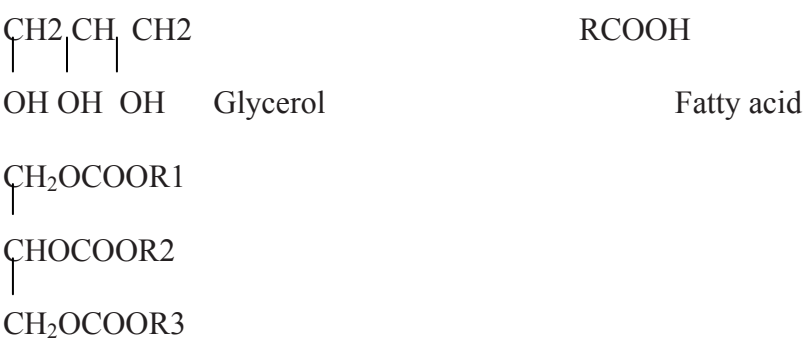
Fatty Acids in Food: Saturated Versus Unsaturated

Fats consumed in the modern human diet vary widely in their fatty acid compositions. The incidence of cardiovascular disease is correlated with diets high in saturated fatty acids. By contrast, a diet that is relatively higher in unsaturated fatty acids (especially polyunsaturated fatty acids) may reduce the risk of heart attacks and strokes. Corn oil, abundant in the United States and high in (polyunsaturated) linoleic acid, is an attractive dietary choice. *Margarine* made from corn, safflower, or sunflower oils is much lower in saturated fatty acids than is butter, which is made from milk fat. However, margarine may present its own health risks. Its fatty acids contain *trans*-double bonds (introduced by the hydrogenation process), which may also contribute to cardiovascular disease. Although vegetable oils usually contain a higher proportion of unsaturated fatty acids than do animal oils and fats, several plant oils are actually high in saturated fats. Palm oil is low in polyunsaturated fatty acids and particularly high in (saturated) palmitic acid (whence the name *palmitic*). Coconut oil is particularly high in lauric and myristic acids (both saturated) and contains very few unsaturated fatty acids.

Some of the fatty acids found in the diets of developed nations (often 1 to 10 g of daily fatty acid intake) are *trans* fatty acids— fatty acids with one or more double bonds in the *trans* configuration. Some of these derive from dairy fat and ruminant meats, but the bulk is provided by partially hydrogenated vegetable or fish oils. Substantial evidence now exists to indicate that *trans* fatty acids may have deleterious health consequences. Numerous studies have shown that *trans* fatty acids raise plasma LDL cholesterol levels when exchanged for *cis*-unsaturated fatty acids in the diet and may also lower HDL cholesterol levels and raise triglyceride levels. The effects of *trans* fatty acids on LDL, HDL, and cholesterol levels are similar to those of saturated fatty acids, and diets aimed at reducing the risk of coronary heart disease should be low in both *trans* and saturated fatty acids.

Triacylglycerols

A significant number of the fatty acids in plants and animals exist in the form of **triacylglycerols** (also called **triglycerides**). Triacylglycerols are a major energy reserve and the principal neutral derivatives of glycerol found in animals. These molecules consist of a glycerol esterified with three fatty acids.



TRIACYLGLYCEROL

If all three fatty acid groups are the same, the molecule is called a simple triacylglycerol. Examples include **tristearoylglycerol** (common name *tristearin*) and **trioleoylglycerol** (*triolein*). Mixed triacylglycerols contain two or three different fatty acids. Triacylglycerols in animals are found primarily in the adipose tissue (body fat), which serves as a depot or storage site for lipids. Monoacylglycerols and diacylglycerols also exist, but are far less common than the triacylglycerols. Most natural plant and animal fat is composed of mixtures of simple and mixed triacylglycerols.

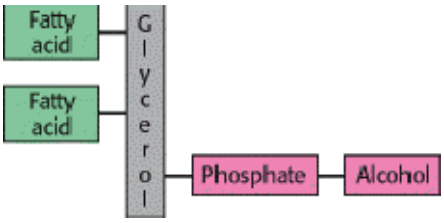
Acylglycerols can be hydrolyzed by heating with acid or base or by treatment with lipases. Hydrolysis with alkali is called **saponification** and yields salts of free fatty acids and glycerol. This is how **soap** (a metal salt of an acid derived from fat) was made by our ancestors. One method used potassium hydroxide (*potash*) leached from wood ashes to hydrolyze animal fat (mostly triacylglycerols).

(The tendency of such soaps to be precipitated by Mg and Ca ions in hard water makes them less useful than modern detergents.) When the fatty acids esterified at the first and third carbons of glycerol are different, the second carbon is asymmetric.

The various acylglycerols are normally soluble in benzene, chloroform, ether, and hot ethanol. Although triacylglycerols are insoluble in water, mono- and diacylglycerols readily form organized structures in water, owing to the polarity of their free hydroxyl groups. Triacylglycerols are rich in highly reduced carbons and thus yield large amounts of energy in the oxidative reactions of metabolism. Complete oxidation of 1 g of triacylglycerols yields about 38 kJ of energy, whereas proteins and carbohydrates yield only about 17 kJ/g. Also, their hydrophobic nature allows them to aggregate in highly anhydrous forms, whereas polysaccharides and proteins are highly hydrated. For these reasons, triacylglycerols are the molecules of choice for energy storage in animals. Body fat (mainly triacylglycerols) also provides good insulation.

GLYCEROPHOSPHOLIPIDS

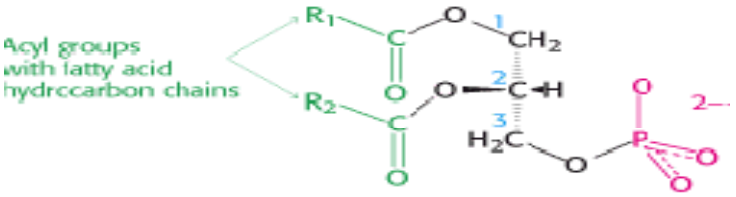
A 1,2-diacylglycerol that has a phosphate group esterified at carbon atom 3 of the glycerol backbone is a **glycerophospholipid,** also known as a *phosphoglyceride* or a *glycerol phosphatide.* These lipids form one of the largest classes of natural lipids and one of the most important. They are essential components of cell membranes and are found in small concentrations in otherparts of the cell. It should be noted that all glycerophospholipids are members of the broader class of lipids known as **phospholipids.** The numbering and nomenclature of glycerophospholipids present a dilemma in that the number 2 carbon of the glycerol backbone of a phospholipid is asymmetric. It is possible to name these molecules either as D- or L-isomers. Thus, glycerol phosphate itself can be referred to either as D-glycerol-1-phosphate or as L-glycerol-3-phosphate. Instead of naming the glycerol phosphatides in this way, biochemists have adopted the ***stereospecific numbering* or *sn*- system**. In this system, the *pro-S* position of a prochiral atom is denoted as the *1-position,* the prochiral atom as the *2-position,* and so on. When this scheme is used, the prefix *sn*- precedes the molecule name (glycerol phosphate in this case) and distinguishes this nomenclature from other approaches. In this way, the glycerol phosphate in natural phosphoglycerides is named *sn*-glycerol-3-phosphate.



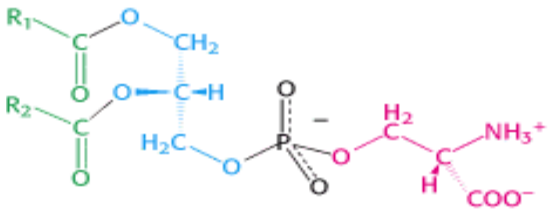
Schematic Structure of a Phospholipid.

The Most Common Phospholipids

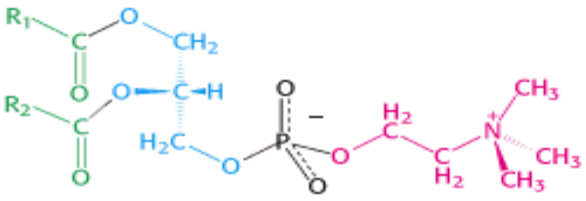
Phosphatidic acid, the parent compound for the glycerol-based phospholipids consists of *sn*-glycerol-3-phosphate, with fatty acids esterified at the 1- and 2-positions. Phosphatidic acid is found in small amounts in most natural systems and is an important intermediate in the biosynthesis of the more common glycerophospholipids In these compounds, a variety of polar groups are esterified to the phosphoric acid moiety of the molecule. The phosphate, together with such esterified entities, is referred to as a "head" group. Phosphatides with choline or ethanolamine are referred to as **phosphatidylcholine** (known commonly as **lecithin**) or **phosphatidylethanolamine,** respectively. These phosphatides are two of the most common constituents of biological membranes. Other common *head groups* found in phosphatides include glycerol, serine, and inositol. Another kind of glycerol phosphatide found in many tissues is **diphosphatidylglycerol.** First observed in heart tissue, it is also called **cardiolipin.** In cardiolipin, a phosphatidylglycerol is esterified through the C-1 hydroxyl group of the glycerol moiety of the head group to the phosphoryl group of another phosphatidicacid molecule.



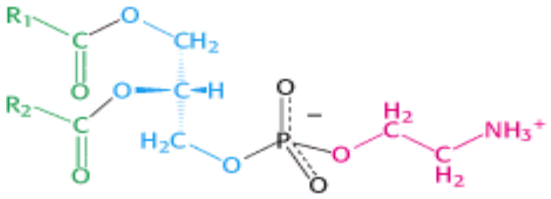
Phosphatidate
(Diacylglycerol 3-phosphate)



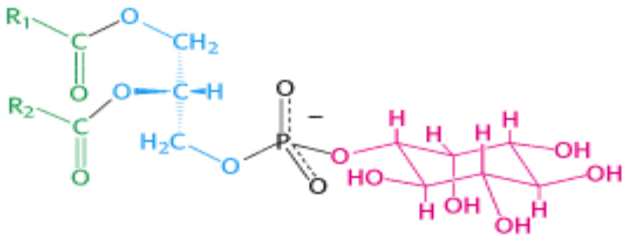
Phosphatidyl serine



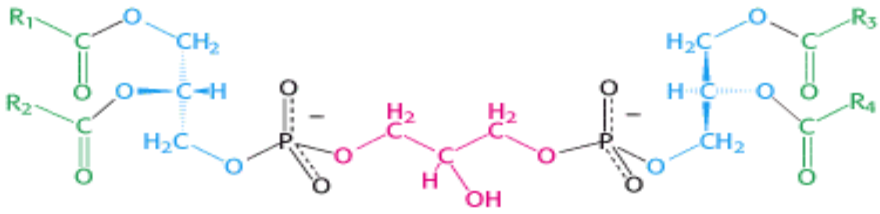
Phosphatidyl choline



Phosphatidyl ethanolamine



Phosphatidyl inositol

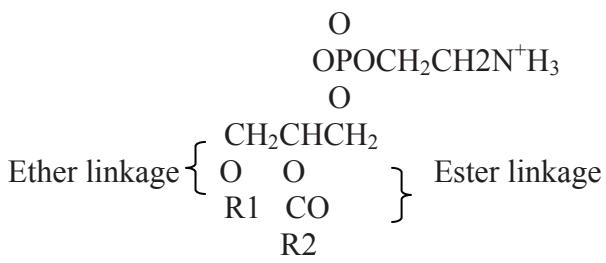


Diphosphatidyl glycerol (cardiolipin)

Phosphatides exist in many different varieties, depending on the fatty acids esterified to the glycerol group. As we shall see, the nature of the fatty acids can greatly affect the chemical and physical properties of the phosphatides and the membranes that contain them. In most cases, glycerol phosphatides have a saturated fatty acid at position 1 and an unsaturated fatty acid at position 2 of the glycerol. Thus, **1-stearoyl-2-oleoyl-phosphatidylcholine** is a common constituent in natural membranes, but **1-linoleoyl-2 palmitoylphosphatidylcholine** is not.

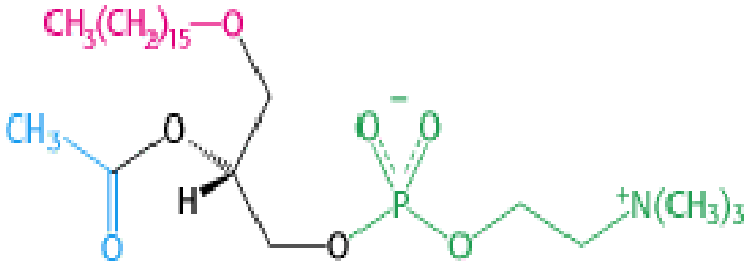
ETHER GLYCEROPHOSPHOLIPIDS

Ether glycerophospholipids possess an ether linkage instead of an acyl group at the C-1 position of glycerol.



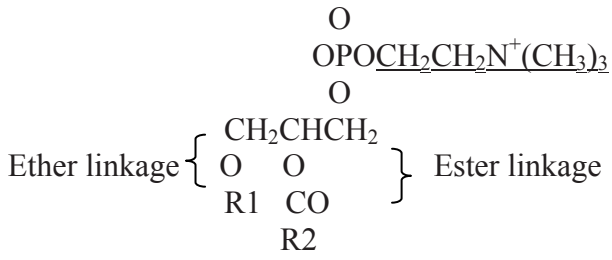
1-alkyl-2-acyl phosphatidylethanolamine, an ether glycerophospholipid.

One of the most versatile biochemical signal molecules found in mammals is **platelet activating factor,** or **PAF,** a unique ether glycerophospholipid called 1-alkyl 2-acetyl-phosphatidylcholine. The alkyl group at C-1 of PAF is typically a 16-carbon chain, but the acyl group at C-2 is a 2-carbon acetate unit. By virtue of this acetate group, PAF is much more water-soluble than other lipids, allowing PAF to function as a soluble messenger in signal transduction.



Platelet activating factor (PAF)

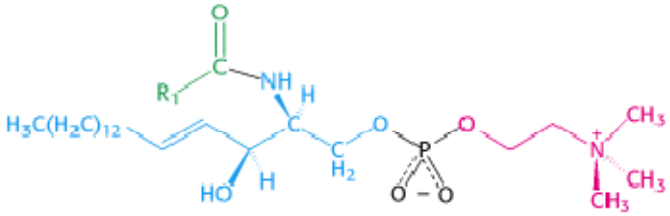
Plasmalogens are ether glycerophospholipids in which the alkyl moiety is *cis*-α,β-unsaturated. Common plasmalogen head groups include choline, ethanolamine, and serine. These lipids are referred to as phosphatidal choline, phosphatidal ethanolamine, and phosphatidal serine.



For phosphatidal choline,
R1 = -CH=CH(CH2)13CH3
R2 = -(CH2)16CH3
For phosphatidal ethanolamine, ethanolamine is in place of choline above.

SPHINGOLIPIDS
These represent another class of lipids found frequently in biological membranes. An 18-carbon amino alcohol, **sphingosine**, forms the backbone of these lipids rather than glycerol. Typically, a fatty acid is joined to a sphingosine via an amide linkage to form a **ceramide. Sphingomyelins** represent a phosphorus-containing subclass of sphingolipids and are especially important in the nervous tissue of higher animals. It is therefore the only set of sphingolipids also known as a phospholipid. A **sphingomyelin** is formed by the esterification of a phosphorylcholine or a phosphorylethanolamine to the 1-hydroxy group of a ceramide.

Sphingosine



Sphingomyelin

GLYCOSPHINGOLIPIDSLIPIDS

There is another class of ceramide-based lipids which, like the sphingomyelins, are important components of muscle and nerve membranes in animals. These are the **glycosphingolipids,** and they consist of a ceramide with one or more sugar residues in aα-glycosidic linkage at the 1-hydroxyl moiety.
The neutral glycosphingolipids contain only neutral (uncharged) sugar residues. When a single glucose or galactose is bound in this manner, the molecule is a **cerebroside**. Another class of lipids is formed when a sulfate is esterified at the 3-position of the galactose to make a **sulfatide.**

Gangliosides are more complex glycosphingolipids that consist of a ceramide backbone with three or more sugars esterified, one of these being a **sialic acid** such as ***N*-acetylneuraminic acid.** These latter compounds are referred to as *acidic glycosphingolipids,* and they have a net negative charge at neutral pH.
The glycosphingolipids have a number of important cellular functions, despite the fact that they are present only in small amounts in most membranes.
Glycosphingolipids at cell surfaces appear to determine, at least in part, certain elements of tissue and organ specificity. Cell–cell recognition and tissue immunity appear to depend upon specific glycosphingolipids.
Gangliosides are present in nerve endings and appear to be important in nerve impulse transmission. A number of genetically transmitted diseases involve the accumulation of specific glycosphingolipids due to an absence of the enzymes needed for their degradation. Such is the case for ganglioside GM2 in the brains of *Tay-Sachs disease* victims, a rare but fatal disease characterized by a red spot on the retina, gradual blindness, and loss of weight, especially in infants and children.

Waxes

Waxes are esters of long-chain alcohols with long-chain fatty acids. The resulting molecule can be viewed (in analogy to the glycerolipids) as having a weakly polar head group (the ester moiety itself) and a long, nonpolar tail (the hydrocarbon chains). Fatty acids found in waxes are

usually saturated and very long. The alcohols found in waxes are also long chain and may be saturated or unsaturated and may include sterols, such as cholesterol. Waxes are water-insoluble due to the weakly polar nature of the ester group. As a result, this class of molecules confers water-repellant character to animal skin, to the leaves of certain plants, and to bird feathers. The glossy surface of a polished apple results from a wax coating.
Lanolin, a component of wool wax, is used as a base for pharmaceutical and cosmetic products because it is rapidly assimilated by human skin.

DERIVED LIPIDS
TERPENES
The **terpenes** are a class of lipids formed from combinations of two or more molecules of 2-methyl-1,3-butadiene, better known as **isoprene** (a five-carbon unit that is abbreviated C5). A **monoterpene** (C10) consists of two isoprene units, a **sesquiterpene** (C15) consists of three isoprene units, a **diterpene** (C20) has four isoprene units, and so on. Isoprene units can be linked in terpenes to form straight chain or cyclic molecules, and the usual method of linking isoprene units is head to tail (Figure 8.16). Monoterpenes occur in all higher plants, while sesquiterpenes and diterpenes are less widely known. The **triterpenes** are C30 terpenes and include **squalene** and **lanosterol,** two of the precursors of cholesterol and other steroids (discussed later). **Tetraterpenes** (C40) are less common but include the carotenoids, a class of colorful photosynthetic pigments.
β-Carotene is the precursor of vitamin A, while lycopene, similar to β- carotene but lacking the cyclopentene rings, is a pigment found in tomatoes.
Long-chain polyisoprenoid molecules with a terminal alcohol moiety are called **polyprenols.** The **dolichols,** one class of polyprenols (Figure 8.18), consist of 16 to 22 isoprene units and, in the form of dolichyl phosphates, function to carry carbohydrate units in the biosynthesis of glycoproteins in animals. Polyprenyl groups serve to *anchor* certain proteins to biological membranes.

STEROIDS
CHOLESTEROL
A large and important class of terpene-based lipids is the **steroids.** This molecular family, whose members effect an amazing array of cellular functions, is based on a common structural motif of three six-membered rings and one five membered ring all fused together. They are derivatives of saturated tetracyclic hydrocarbons known as **PERHYDROCYCLOPENTANOPHENANTHRENE**, a 17C constituent. to form . **Cholesterol** is the most common steroid in animals and the precursor for all other animal steroids. The numbering system for cholesterol applies to all such molecules. Many steroids contain methyl groups at positions 10 and 13 and an 8- to 10-carbon alkyl side chain at position 17. The polyprenyl nature of this compound is particularly evident in the side chain. Many steroids contain an oxygen at C-3, either a hydroxyl group in sterols or a carbonyl group in other steroids. Note also that the carbons at positions 10 and 13 and the alkyl group at position 17 are nearly always oriented on the same side of the steroid nucleus, the α-orientation. Alkyl groups that extend from the other side of the steroid backbone are in an α-orientation. Cholesterol is a principal component of animal cell plasma membranes, and much smaller amounts of cholesterol are found in the membranes of intracellular organelles. The relatively rigid fused ring system of cholesterol and the weakly polar alcohol group at the C-3 position have important consequences

for the properties of plasma membranes. Cholesterol is also a component of *lipoprotein complexes* in the blood, and it is one of the constituents of *plaques* that form on arterial walls in *atherosclerosis.*

Steroid Hormones

Steroids derived from cholesterol in animals include five families of hormones (the androgens, estrogens, progestins, glucocorticoids and mineralocorticoids) and bile acids. **Androgens** such as **testosterone** and **estrogens** such as **estradiol** mediate the development of sexual characteristics and sexual function in animals. The **progestins** such as **progesterone** participate in control of the menstrual cycle and pregnancy. **Glucocorticoids (cortisol,** for example) participate in the control of carbohydrate, protein, and lipid metabolism, whereas the **mineralocorticoids** regulate salt (Na+, K+, and Cl-) balances in tissues. The **bile acids** (including **cholic** and **deoxycholic acid**) are detergent molecules secreted in bile from the gallbladder that assist in the absorption of dietary lipids in the intestine.

PROPERTIES OF LIPIDS

-Paper transparency

-Solubility

Chemical Properties

-Saponification value

-Acid value

-Iodine value

PROBLEMS

1. Draw the structures of all the possible triacylglycerols that can be formed from glycerol with stearic and arachidonic acid.

2. Describe in your own words the structural features of

a. a ceramide, and how it differs from a cerebroside.

b. a phosphatidylethanolamine, and how it differs from a phosphatidylcholine.

c. an ether glycerophospholipid, and how it differs from a plasmalogen.

d. a ganglioside, and how it differs from a cerebroside.

e. testosterone, and how it differs from estradiol.

3. From your memory of the structures, name

a. the glycerophospholipids that carry a net positive charge.

b. the glycerophospholipids that carry a net negative charge.

c. the glycerophospholipids that have zero net charge.

4. Compare and contrast two individuals, one of whose diet consistslargely of meats containing high levels of cholesterol, and the other of whose diet is rich in plant sterols. Are their risks of cardiovascular disease likely to be similar or different? Explain your reasoning.