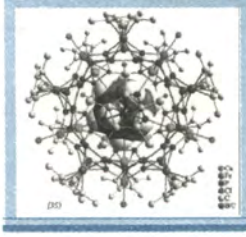
CHAPTER 2



Chemistry and Membrane Transport

CHEMISTRY

I. Carbohydrates: Structure and Function

Features

- Widely distributed
- Most abundant biologic molecule
- Have structural and metabolic role
- Polyhydroxy aldehydes or ketones
- General formula: $(CH_2)_n$, where $n \geq 3$

Importance of Glucose

- Universal fuel for fetus
- Major fuel for all tissues, even microbes in our intestines
- Form structural component of membrane
- Form carbohydrate with specific function:
  - Ribose – nucleotides
  - Galactose – lactose in milk
  - Glycolipid
  - Glycoprotein

Classification

Monosaccharide, disaccharide, oligosaccharide, polysaccharide

MONOSACCHARIDES

Formula: $(CH_2O)_n$

Based on number of carbon:

- with three carbons: triose
- with four carbons: tetrose
- with five carbons: pentose
- with six carbons: hexose

Based on functional groups:

Aldehyde: Aldose

Ketone: Ketose

Classification

Sr. No.	Class	Sub-class	Examples	
			Aldose	*Ketose*
1.	Monosaccharide	Triose	Glycerol	Dihydroxy acetone
		Tetrose	Erythrose	Erythrulose
		Pentose	Ribose	Ribulose
		Hexose	Glucose	Fructose
2.	Disaccharide (composed of two monosaccharide)	Maltose	2glucose units $\alpha 1 \rightarrow 4$ glycosidic bond	
		Sucrose	αD glucose +β D fructose $\alpha 1 \rightarrow \beta_2$ bond	
		Lactose	αD galactose + βD glucose$1 \rightarrow \beta_4$ bond	
3.	Oligosaccharide (composed of 2-10 monosaccharide units, O-linked) (attached to OH group) e.g. IgA	Malto-triose	Maltose and $\alpha 1 \rightarrow 6$ glucose	
	N-linked (attached to protein via N-glycosidic bond e.g. Aspartate side chain) e.g. dolichol phosphate			
4.	Polysaccharide (10 or more monosaccharide unit) and their derivatives	–	Amylose αDG: $\alpha 1 \rightarrow 4$ bond	
		–	Amylopectin 24–30 monomers (αD glucose) $1 \rightarrow 4$ linkage plus $\alpha 1 \rightarrow 6$ branching	
			Homopolysaccharide: Contain only one kind of monosacharides Starch: $\alpha 1 \rightarrow 4$ linkage and $\alpha 1 \rightarrow 6$ branching Glycogen: 12-14 monomers of D-glucose $\alpha 1 \rightarrow 4$ linkage and $\alpha 1 \rightarrow 6$ branching	
			Cellulose	βD glucose $\beta 1 \rightarrow 4$ bond
			Inulin	βD fructose
			Dextran*	αD glucose linked at $\alpha 1 \rightarrow 6$ bond, with few branches

* Dextran: breakdown product of starch

Limit dextrin: formed when hydrolysis of starch reaches a branch point.

Heteropolysaccharides

Composed of repeating units of monosaccharides, their derivative or proteoglycans.

Mucopolysaccharides (Glycosamainoglycans)	*Mucoprotein (Glycoprotein)*
Hyaluronic acid	Sialic acid
Chondroitin sulfate	Neuraminic acid
Dermatan sulfate	Gangliosides
Keratan sulfate	
Heparin sulfate	

GAG

GAG	*Repeating unit*	*Tissue distribution*	*Function*
Hyaluronic acid	GlcUA and GlcNA GlcUA —β1,3→ GlcNAc —β1→3→ GlcUA	Synovial fluid	Vitreous body, eye, cartilage, loose connective tissue
Chondroitin	GlcUA GalNAc	Cartilage	To maintain structure and function of tissues located at the site of calcification in endochondral bone
Dermatan sulfate	IdUA, GalNAc IdUA —β1,3→ GalNAc	Skin, valves, blood vessels lung, sclera	Sclera: maintain shape of eye ball
Keratin sulfate	GlcNAc, Gal GlcNAc —β1,3→ Gal —β1→4→ GlcNAc	Cornea bone, cartilage, horny structures, hair, nail	Corneal transparency
	Linked to protein via:		
	N-linkage (type1)	Cornea	Corneal transparency
	O-linkage (type 2)	Loose connective tissue	Ground substance
Heparan sulfate	GlcN, GlcUA, GlcN —α1→4→ GlcUA→ GlcN	Lung, muscle, liver, component of synapse, glomerulus, vesicles	Present on cell surface or bound to ECM, responsible for charge selectiveness
Heparin	GlcN, IdUA IdUA —α1,4→ GlcN	Granules of mast cells, liver, lung, skin	Function as intracellular binding site, anticoagulant, bind to lipoproteins lipase causing LP release

Key: GlcUA – glucuronic acid, GlcN – glucosamine, A – Acetyl

Organ and site specific functions of GAG

GAG	*Organ*	*Site*	*Function*
Heparin sulfate	Liver	Intracellular on cell surface	Anticoagulant
Heparan sulfate	Kidney	Renal basement membrane	Charge selectivity of glomerulus
Keratan sulfate Dermatan sulfate	Cornea	In between collagen fibers	Corneal transparency
Heparin	Mast cells	Secretory granules	Inflammatory response
Heparin sulfate	Vascular wall	-	Anticoagulant activation of lipoprotein lipase

Disease associated with GAG

GAG	*Disease*
Hyaluronic acid (HA)	Permit tumor cells to migrate through extracellular matrix Tumor cells induce fibroblasts to synthesize increased amounts of HA, facilitating their own spread
Heparan sulfate	Tumor cells lack this GAG, results: lack of adhesiveness of these cells
HA, chondroitin sulfate, heparin sulfate, dermatan sulfate	Arterial smooth muscle cell proliferation in atherosclerotic lesions and plaque
GAG	Arthritis, Autoantigens
Chondroitin sulfate	Diminishes in cartilage with age, contributing to development of osteo-arthritis
HA, keratin sulfate	Increase in cartilage with age, causes osteoarthritis
GAG in skin	Characteristic changes of age occur in skin

Carbohydrate derivatives

Sr. No.	*Derivatives*	*Example*	*Location/constituent of*
1.	Amino sugar	Glucosamine Galactosamine Neuraminic acid	Chitin Cartilage, chondroitin sulfate
2.	Sugar acids	Ascorbic acid, glucuronic acid	Vitamin C, proteoglycan
3.	Deoxy sugar	2-deoxyribose	DNA
4.	Sugar alcohol	D-sorbitol, D-mannitol	Minor pathway intermediates
5.	Phosphoric acid esters	D-glucose-1-phosphate	Minor pathway intermediates

OXIDATION OF CARBOHYDRATES

Oxidized forms

(a) Gluconic acid, 6 phosphogluconate
(b) Uronic acid, glucuronic acid

Structure: Monosaccharides

All monosaccharides are optically active due to presence of atleast one asymmetric carbon.

Asymmetric carbon

- Carbon atom bonded to four different atoms or groups of atoms is asymmetric carbon.

Isomers

Presence of asymmetric carbon allows formation of isomers and the number of isomers of a compound depends on the number of asymmetric carbon atoms (n) and is equal to 2^n, e.g. glucose has 4 asymmetric carbon atoms, thus $2^4 = 16$ isomers.

Anomer

New carbon formed during cyclization is anomeric carbon, e.g. during hemiacetal and hemiketal formation (e.g. C_1 in glucose).

Depending on the size of ring formed,

Blood group antigens

Antigen	*Structure*
O	Fucose – Galactose | N-Acetyl glucosamine | R
A	NAc Galactosamine | Fucose- Galactose | N-Acetyl glucosamine-R
B	Galactose | Fucose-Galactose | N-Acetyl | glucosamine | R

structure is designated pyranose, if it is 6-membered ring or furanose, if it is 5-membered ring. A six membered ring structure can adopt either a chair or boat configuration. D-glucose adopts a chair conformation.

Mutarotation

α and β-forms equilibrate via the straight chain aldehyde form. This occurs due to opening of hemiacetal ring.

Reducing sugar

Sugar possessing free anomeric carbon atom that is not involved in a glycosidic linkage is reducing sugar. The end containing free anomeric carbon is reducing end.

This free aldehyde or ketone group reduces alkaline copper sulfate, e.g. Lactose, maltose.

Sucrose is a non-reducing sugar, because glycosidic bond between anomeric carbon C1

Isomerism

Sr. No.	*Isomerism*	*Reasoning*	*Example*
1.	D-L isomerism (stereoisomer)	• Same chemical formula • Differ in position of –OH group on one or more asymmetric carbon (e.g. C5 in glucose) Mirror images of each other	D, L glucose (Fig. 2.1)
2.	Optical isomerism (Enantiomer)	Presence of asymmetric carbon rotate plane polarized light either to right [dextrorotatory, (+)] or to left [levorotatory (–)]	Enantiomer (+) isomer (–) isomer
3.	Epimerism	Differ as a result of variation in configuration of –OH and –H glucose on C-2, 3 and 4 of glucose, galactose at C_4 and mannose at C-2 or conformation that differ only at one carbon atom	Mannose at C-2 (Fig. 2.2) Galactose at C-4
4.	Anomerism	Differ in configuration at carbonyl or anomeric carbon	α anomer, β anomer (Fig. 2.3) α:- OH on anomeric is below plane of ring, β:- OH is above plane of ring
5.	Aldose-ketose isomerism	Same molecular formula, but due to position of carbonyl carbon	Glucose: C-1 is aldehyde fructose (Fig. 2.4). C-2 is keto (Fig. 2.5)

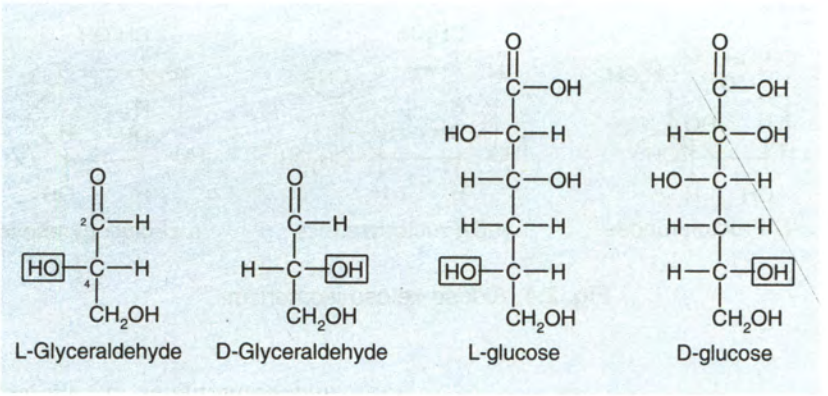


Fig. 2.1. D-L isomerism (Glyceraldehyde).

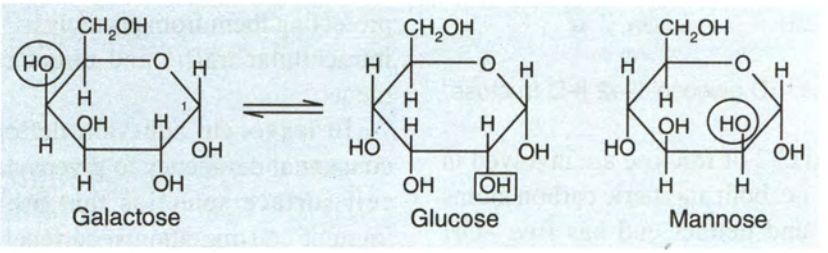


Fig. 2.2. Epimerism.

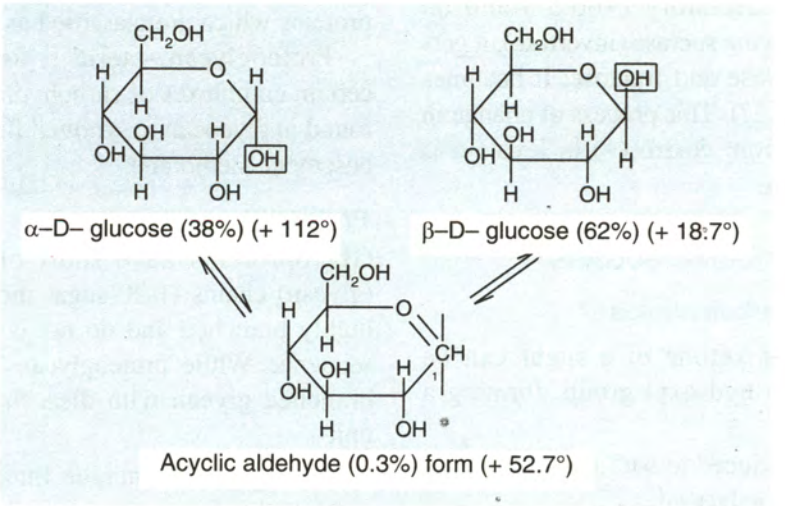


Fig. 2.3. Mutarotation (Anomerism).

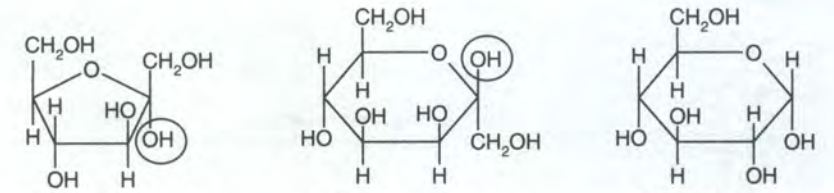


Fig. 2.4. Aldose-ketose isomerism.

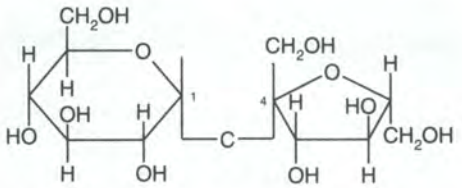


Fig. 2.5. Sucrose: αD glucose 1→2 β-D fructose.

of a glucose and C-2 of fructose are involved in bond formation i,e. both anomeric carbon atoms are substituted and neither end has free –OH group (Fig. 2.5).

Inversion of sugar

Sucrose is dextrorotatory (+66.5°) and on hydrolysis by enzyme sucrase (invertase), it gets converted to glucose and fructose. It becomes levorotatory (–28.2°). This process of change in optical rotation from dextro (+) to levo (–) is inversion of sugar.

TESTS FOR REDUCING SUGARS

Reduction of carbohydrates

- Aldehyde or ketone of a sugar can be reduced to a hydroxyl group, forming a polyol.
- Glucose is reduced to sorbitol
- Galactose to galactitol
- Sorbitol does not readily diffuse out of cells and accumulates in cells causing osmotic damage to neurons and cataract.

Glycosylation of proteins

This can alter protein and modify their function, protecting them from proteolysis, directing their intracellular traffic and direct cellular movements.

In leukocyte adhesion deficiency (LAD), congenital deficiency to glycosylate ligands for cell surface selectins that are required for immune cell migration; recurrent life threatening infections are common in such patients.

Glycoproteins (Mucoproteins)

Carbohydrate is found attached to many globular proteins which are classified as glycoproteins.

Proteoglycan – term is used to describe certain complexes of carbohydrate and proteins found in glycocalyx, synovial fluid of joints and basement membrane.

Proteoglycans

Glycoproteins have short oligosaccharide (glycan) chains (1-20 sugar moiety in length), highly branched and do not contain repeating sequence. While, proteoglycans are long, linear, branched glycan with disaccharide repeating units.

Glycoproteins include integral membrane proteins that function as receptors for hormones or other molecules.

Functions of glycoprotein

Function	*Example*
Structural	Component of plasma membrane
Lubricant	Component of mucus
Receptor	Hormone, other molecules
Hormone	hCG, thyrotropin, erythropoietin
Immune system	Immunoglobulin, complement, interferon
Transport	Recognition signal
Cell-cell interaction	Mediate cell to cell interaction
Stabilization	Stabilize protein against denaturation, proteolysis

Protein carbohydrate linkages

Protein carbohydrate linkages are of two types:

- O-linkage
- N-linkage

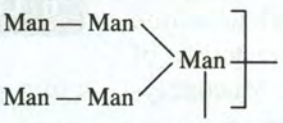
O-linkage: Sugar is attached via –OH group of a serine or threonine residue.

N-linkage: Sugar is attached via amide $-NH_2$ group of asparagine residue.

O-linked Oligosaccharides

Proteins in mucous secretions contain oligosaccharides linked by a glycosidic linkage

Glycoprotein structure and function

Collagen	Glycosyl-galactose disaccharide linked to hydroxylysine residue. **In tendon:** collagen is less glycosylated, form ordered fibrous structure **In basement membrane:** Collagen is heavily glycosylated, form meshwork structure
Mucin	O-linked oligosaccharides containing sialic acid, galactose and GalNAc mainly, sometimes GlcNAc and fucose **Salivary mucin:** contain unusually large number of serine or threonine residues glycosylated with a sialic acid-galactose GalNAc trisaccharide.O-linked oligosaccharides are negatively charged (due to presence of sialic acid), repel each other to prevent protein folding and assume an extended state, yielding a highly viscous (mucous) solution
LDL receptor	Found in plasma membrane of smooth muscle cells and fibroblasts Contain two N-linked biantennary complex chains in addition to a cluster of O-linked chains attached to membrane Attached to hydrophobic amino acid region which keep LDL in extended state **Biantennary:**Terminal trisaccharide sequences, one attached to each mannose 
Protein folding	N-linked oligosaccharides help in protein folding in ER
Cell-cell interaction	Provide specific cell recognition and is a key factor in fertilization, inflammation, development and differentiation, virus infectivity
Targeting of lysosomal enzymes	Lysosomal enzymes are n-linked glycoproteins and sorted by exposing Man-6-P structures (by hexosaminidase) which are recognized by Man-6-P receptor in golgi
Protein stability and solubility	Proteins secreted from cells such as plasma proteins are conferred increased solubility and stability by oligosaccharide chains

Mucopolysaccharidosis

Genetic defects of proteoglycan metabolism			
	Syndrome	*Defect*	*Products accumulated in lysosome*
I	Hurler	α-1-iduronidase[4]	Heparan sulfate (HS), Dermatan sulfate (DS)
II	Hunter	Iduronate sulfatase[3]	HS, DS
III	Sanfilipino A	Heparin sulfatase	HS
	Sanfilipino B	NAc glucosaminidase	
	Sanfilipino C	NAc Gln-6-sulfatase[1]	
IV	Morquio's	Galactose-6-sulfatase	KS
V	Maroteaux-Lamy	NAc Gal-4-sulfatase	DS
VI	Sly	β-glucuronidase	DS, HS

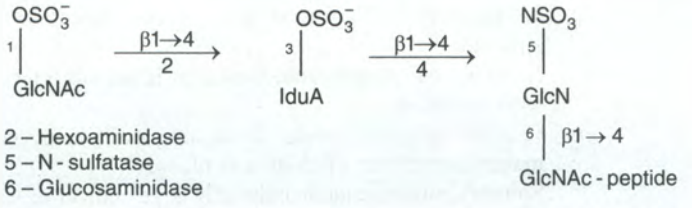


Fig 2.6. Lysosomal degradation of HS.

between N-acetyl galactosamine (GalNAc) and hydroxyl group of serine or threonine residues.

N-linked oligosaccharides

Always attached by glycosylamine linkage of N-acetyl glucosamine (GlcNAc) to amide nitrogen of asparagine residue; characteristic of plasma and membrane proteins; Mucopolysaccharidosis.

Clinical features of lysosomal storage disease

- Skeletal deformity
- Mental retardation
- Early death in severe cases

Diagnosis

- Urine GAG
- Leukocyte/fibroblast assay for specific enzymes

II. Lipids: Structure And Function

LIPID CHEMISTRY

Lipids of major physiologic significance are fatty acids, their ester, cholesterol and steroids.

Fact file: Lipids

- Hydrophobic
- Insoluble in water, soluble in polar solvents.

- Composed of saturated or unsaturated long chain hydrocarbons with a carboxyl group at end of chains.
- Important dietary constituent.
- Serve as:
  - Source of energy
  - Thermal insulator
  - Important component of cell membrane
  - Lipoprotein serves transport function
  - Storage: TAG, stored in adipose tissue
  - Precursor for steroid hormones

General formula

Saturated fatty acid:

$CH_3 - [CH_2]_n - COOH$

N = number of methylene groups

Systemic name gives number of carbons followed by acid suffix.

- anoic for saturated FA
- enoic for unsatured FA

e.g. Palmitic acid, 16C: hexadecanoic acid.

Important points

- Carbons are numbered from carboxyl carbon (carbon no. 1).
- Carbon adjacent to it is carbon no. 2 (α-carbon).
- Terminal methyl carbon is ω-carbon or n-carbon.
- Position of double bond:
- Δ^9: double bond between C9 and 10
- ω^9: double bond on 9th carbon

Fatty acids

- Exist as free or esterified to glycerol.
- Humans have FA with even number of carbon atoms: 16-20 carbon in length, saturated or unsaturated. The aliphatic chain may be saturated or unsaturated (containing one or more double bonds).

Neutral lipids

Being uncharged, acyl glycerols, cholesterol and cholesterol esters are termed neural lipids.

CLASSIFICATION

- Simple
- Complex
- Precursor and derived lipids

1. Simple lipids

Esters of FA with alcohol:

i. Fats – esters of FA with glycerol
ii. Waxes – esters of FA with higher molecular weight alcohols.

Name	*No. of carbon atom*	*Systemic name*	*Double*
Lauric acid	12	Dodecanoic acid	-
Myristic acid	14	Tetradecanoic acid	-
Palmitic acid	16	Hexadecanoic acid	-
Stearic acid	18	Octadecanoic acid	-
Palmitoleic acid	16	Cis-Hexadecenoic acid	1:9 (ω9)
Oleic acid	18	Cis-Octadecenoic acid	1:9 (ω9)
Elaidic acid	18	Trans-octadecenoic acid	1:9 (ω9)
Linoleic acid	18	Cis-9, 12 octadecoidenoic acid	2:9, 12 (ω6)
Linolenic acid	18	Cis-9, 12, 15 octadecatrienoic acid	3:9, 12, 15 (ω3)
Arachidonic acid	20	Cis 5, 8, 11, 14 Eicosatetraenoic acid	4:5, 8, 11, 14 (ω6)

2. Complex lipids

Esters of FA with alcohol and containing an additional group

i. **Phospholipids:**
Esters of FA with alcohol and phosphoric acid residue.
Alcohol can be glycerol : glycerophospholipids.
Alcohol can be sphingosine: sphingophospholipids.

ii. **Glycolipids (Glycosphingolipids):**
Esters of FA, contain sphingosine and a carbohydrate

iii. **Other complex lipids:**
Sulfolipids, aminolipids, lipoproteins

3. Precursor and derived lipids

Include fatty acids, glycerol, steroids, alcohol in addition to glycerol and sterols, fatty aldehydes, and ketone bodies, hydrocarbons, lipid-soluble vitamins and hormones.

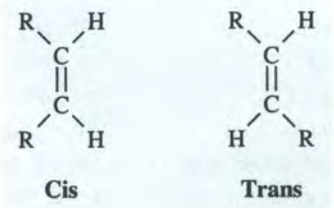
Unsaturated fatty acid

May contain one or more double bonds

- Mono unsaturated: One double bond
- Polyunsaturated: Two or more double bond
- Eicosanoids: Prostanoid, leukotriene, prostacyclin, thromboxane

Isomerism in unsaturated fatty acid

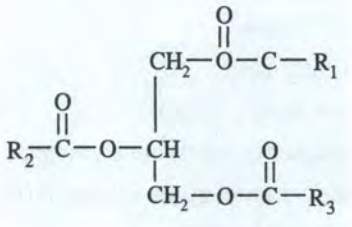
- Naturally occurring unsaturated FA have cis double bonds.
- Display geometric isomerism



- In cis configuration, molecule bent at 120° in double bond and produces kinks.
- In trans configuration, molecule remains straight at double bond.
- With increase in chain length, melting point of even- numbered fatty acids increases and it decreases with unsaturation.

Triacylglycerol

- Ester of fatty acid and glycerol
- Naturally occurring fat, storage form of fat

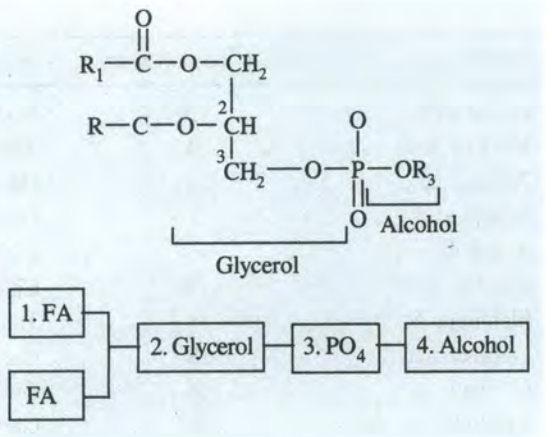


R_1 and R_3: saturated FA
R_2: unsaturated

1. Glycerophospholipids (GPL)

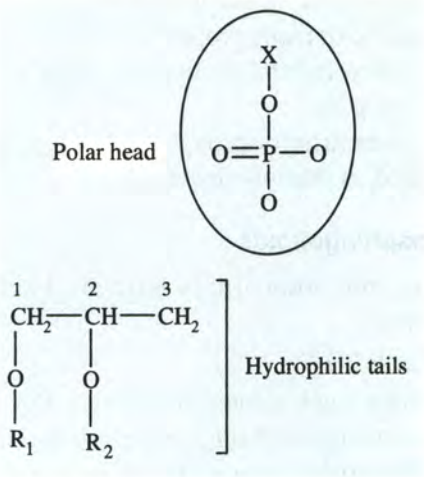
- Major class of membrane lipids
- Abundant in all biological membrane.

Phospholipid



1. Fatty acid
2. Platform back bone with FA attached
3. PO_4
4. Alcohol attached to PO_4

FA components provide hydrophobic barrier and remainder is hydrophilic



Thus GPL contain:

- phosphorylated head
- 3-C glycerol back bone
- 2 hydrocarbon fatty acid chains

Simplest GPL is phosphatidic acid or diacylglycerol

GPL include:

- Phosphatidylcholine (PC)
- Phosphatidyl ethanolamine (PE)
- Phosphatidyl serine (PS)
- Phosphatidylcholine glycerol
- Phosphatidylcholine inositol
- Diphosphatidyl glycerol (Cardiolipin):

Present exclusively in inner mitochondrial membrane.

GPL contain two fattty acyl groups esterified to C-1 and C-2 of glycerol.

	C_1	C_2
PC	Palmitic/stearic acid	Oleic, linoleic, linolenic acid
PE	Contains palmitic acid on C_1 or oleic acid	Arachidonic acid

Usually, on C-1, saturated FA is found: e.g. palmitic, stearic acid and on C-2, unsaturated FA is present e.g. oleic, linoleic, linolenic acid.

Designation of GPL does not specify which FA is present as seen in case of P.C. and P.E.

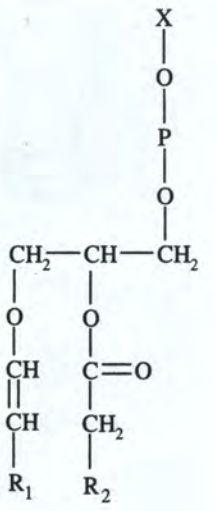
Saturated FA: is a straight chain.

Unsaturated FA: can be trans, however, usually cis occurs naturally and produces a kink.

There is a high degree of coiling of hydrocarbon chain in a glycerophospholipid that is disrupted by a double bond.

Glycerol ether phospholipids

- α-β unsaturated.
- Plasmalogen is ethanolamine ether/ choline esterified to PO_4.



Ethanolamine plasmalogen – abundant in nervous tissue and heart.

Thus, glycosphingolipids are amphipathic, contains:

- Polar end or head group because of charged PO_4 and substitution on PO_4
- Non-polar tail due to hydrophobic carbohydrates chains of fatty acyl groups.

Every tissue and cellular membrane has a distinct composition of GPL and a definite pattern of FA composition.

2. Sphingolipids

In sphingomyelin, terminal OH group of sphingosine is esterified to phosphorylcholine, so that its polar head similar to P.C. (Fig. 2.7).

- 2nd major class
- Contains sphingosine back bone rather than glycerol
- Sphingosine (amino alcohol) is basis of sphingolipids
- Sphingosine is linked to FA by amide bond forming CERAMIDE
- Sphingomyelin has P.C. esterified to 1-OH group: Most abundant sphingolipid in mammalian tissues.

Sphingomyelin has structural similarity to GPL and have many properties in common, e.g. Sphingomyelin are amphipathic with charged head group.

Glycosphingolipids, sphingomyelin are classified as phospholipids.

Glycosphingolipids

Sugar containing lipids built on backbone ceramide.

- Lack PO_4
- Have sugar moiety or primary OH group of sphingosine (in ceramide), e.g. Glucocerebroside –present in non neuronal tissue

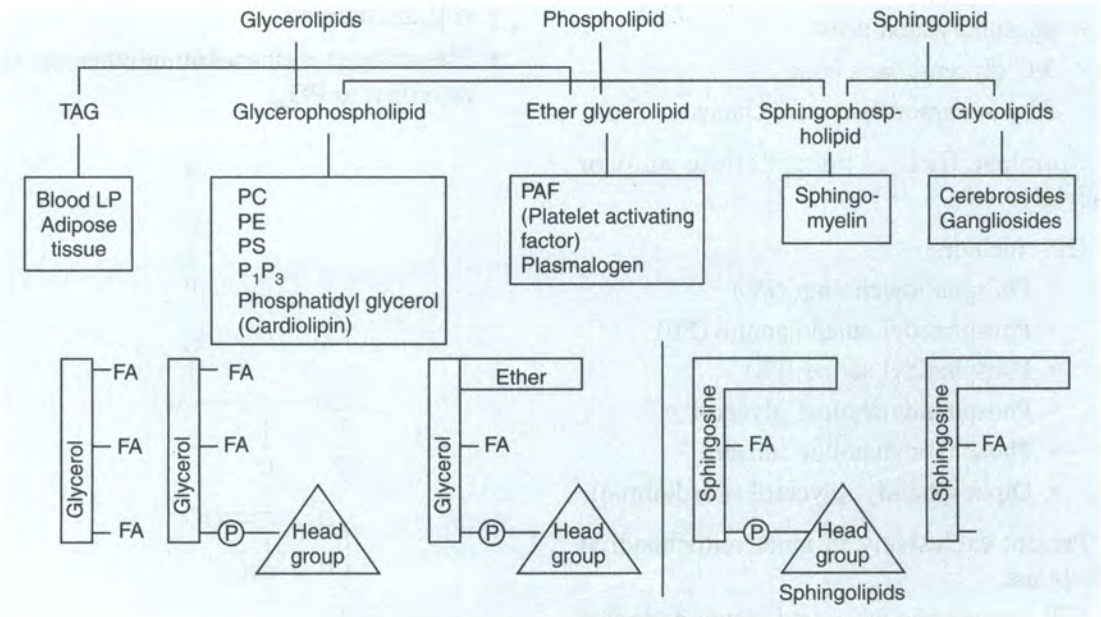


Fig 2.7. Types of glycolipids and sphingolipids.

- Galactocerebroside – present in brain and nervous tissue
- Ganglioside contain sialic acid residue in head groups. Present in brain (represent 5-8% of total lipid in brain).

III. Proteins: Structure and Function

PROTEINS

Table 2.1. Functions of proteins

Structural: make up cytoskeleton to provide structure and strength to cells
* Component of: – Collagen
– Elastin
– Keratin
Enzyme catalysis
Transport
Storage
Hormone
Blood coagulation
Immunity
Control of gene expression

There are about 300 amino acids present in nature and only 20 α-amino acids coded by genetic code appear in proteins. Additional amino acids occur in specific protein by "post translational" modifications of these 20 common amino acids, e.g. Peptidyl proline-4-hydroxy proline, Peptidyl lysine-5 hydroxy lysine, Peptidyl glutamate-γ-carboxy glutamate.

Amino Acids

Amino acids are functional units of proteins. Amino acids are composed of two **functional groups** – amino ($-NH_2$), carboxyl (–COOH); a hydrogen atom (–H) and a **distinctive side**-chain (–R), attached to a central carbon termed **α-carbon**.

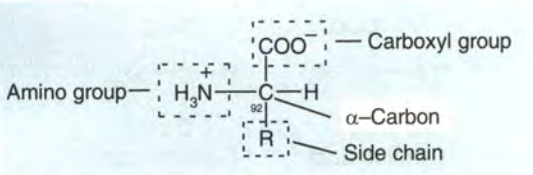


Fig. 2.8. General structure of amino acid chain.

Except for glycine, all acids contain at least one asymmetric carbon atom (α-carbon atom).

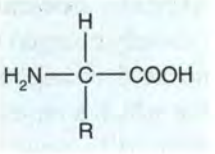


Fig. 2.9. Structure of amino acid.

- NH_2 = amino group (basic)
- COOH = carboxyl group (acidic)
- R = side chain

Of these 20 amino acids, only **proline** is an imino acid (–NH–), and not an α-amino acid.

Optical Activity

Due to tetrahedral orientation of four different group about α-carbon atom, amino acids exhibit optical isomerism. (i.e. ability to rotate plane polarized light).

These isomers are non-superimposable mirror images and referred to as **enantiomers**.

The two amino acid configuration (based on configuration of D and L-glyceraldehyde) are D-(dextro or right) and L-(levo or left) (Fig. 2.10).

Only L-α-amino acids occur in proteins.

Amphoteric properties of amino acids

Amino acids have two ionizable weak acid groups, –COOH and an NH_3^+. In aqueous solution, amino acids exist as **Zwitterions** i.e., they have both positive and negative charges.

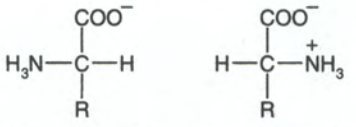


Fig. 2.10. Amino acid configuration.

The α-carboxyl group is negatively charged and α-amino group is positively charged and overall molecule is **electrically neutral**. At **low pH**, amino acid is positively charged (cation) and at **high pH**, amino acid is negatively charged (anion). The pH at which a molecule exists as a Zwitterion (electrically neutral) is termed **isoelectric pH** and is electrically neutral.

Classification of amino acids

(a) Based on Structure

On the basis of side chain (–R) attached to α-carbon atom, amino acids can be divided into polar and non polar amino acids.

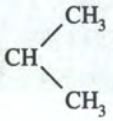
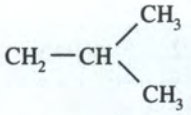
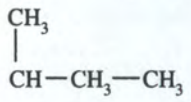
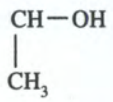
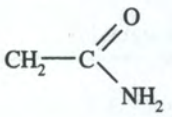
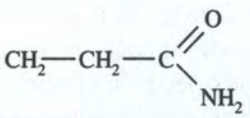
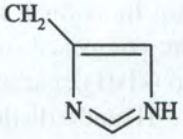
Table 2.2.

Polar (hydrophilic)	*Non polar (hydrophilic)*
Glycine	Alanine
Serine	Leucine
Threonine	Isoleucine
Cysteine	Valine
Arginine	Methionine
Histidine	Proline
Lysine	Tyrosine
Aspartate	Phenylalanine
Asparagine	Tryptophan
Glutamate	
Glutamine	

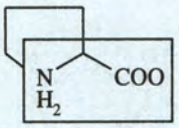
(b) Based on side chain

Polar amino acids are exposed on surface of proteins and non-polar one are buried in hydrophobic core of a protein.

Table 2.3. L-/α-amino acids

Amino acid	*Structure of R moiety*
Aliphatic amino acids	
Glycine [Gly, G]	H
Alanine [Ala, A]	CH_3
Valine [Val, V]	
Leucine [Leu, L]	
Isoleucine [Ile, I]	
Neutral amino acids	
Serine [Ser, S]	CH_2–OH
Threonine [Thr, T]	
Asparagine [Asn, N]	
Glutamine [Gln, Q]	
Acidic amino acids	
Aspartic acid [Asp, D]	CH_2 – COOH
Glutamic acid [Glu, E]	CH_2 – CH_2 - COOH
Basic amino acids	
Histidine [His, H]	

(Contd.)

Amino acid	*Structure of R moiety*
Lysine [Lys, K]	CH_2–CH_2–CH_2–CH_2–NH_2
Arginine [Arg, R]	CH_2–CH_2–CH_2–NH–C–NH_2
Sulfur-containing amiuo acid	
Cysteine [Cys, C]	CH_2-SH
Methionine [Met, M]	$CH_2 - CH_2 - S - CH_3$
Imino acid	
Proline (Pro, P)	

c. Based on nutritional requirement

(i) Essential Amino acid
(ii) Non-essential Amino Acid

(i) **Essential amino acids:** Amino acids that cannot be synthesized by body and must supplied in diet are called essential amino acids. They are ten in number and include the following.

Methionine, Arginine, Threonine, Tryptophan, Valine, Isoleucine, Leucine, Phenylalanine, Lysine, Histidine

(ii) **Non-essential amino acids:** Amino acids that can be synthesized by the body to meet its demands. They include-glycine, alanine, serine, cysteine, aspartate, asparagine, glutamate, glutamine, tyrosine, proline.

d. Based on their metabolic fate

Amino acids can be classified based on fate of their carbon skeletons

Glycogenic		*Ketogenic*	*Both*
Ala	Met	Leu	ILe
Arg	Pro		Lys
Asp	Ser		Phe
Cys	Thr		Trp
Glu	Val		Tyr
Gly			
His			

IV. Nucleic Acid

STRUCTURE AND METABOLISM

Nucleotide

- Combination of heterocyclic amine, a pentose and phosphoric acid

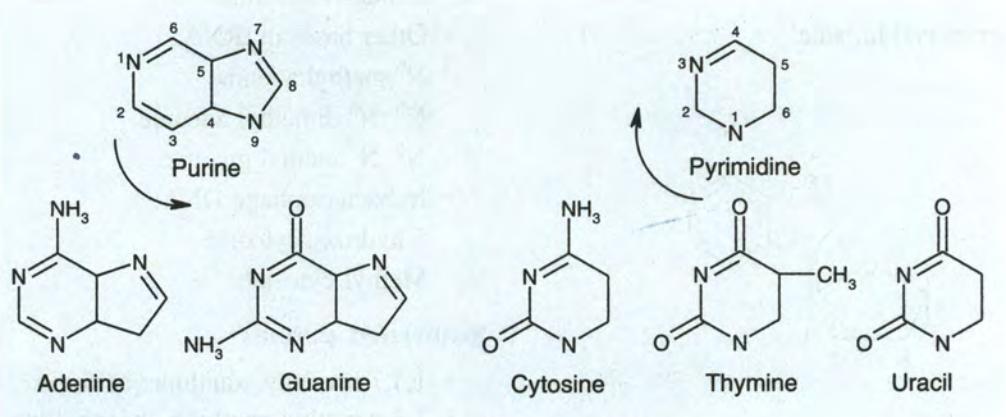


Fig. 2.11. Structure of bases.

- Monomeric unit of nucleic acids
- Purine and pyrimidines supply building blocks of nucleic acid
- Also, they are high energy intermediates
- Form part of coenzyme: FAD, NAD, NADP, CoA, SAM
- Have regulatory function: signal transduction, second messenger (cAMP, cGMP).

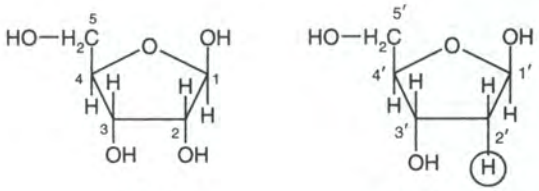


Fig. 2.12. Numbering of sugars is primed.

Nucleoside (Fig. 2.12)

Nucleotides

Linking one or more phosphates with a nucleoside onto 5′ end of molecule through esterification

NAMING-CONVENTIONS

Nucleoside

- Purine: end in 'sine'

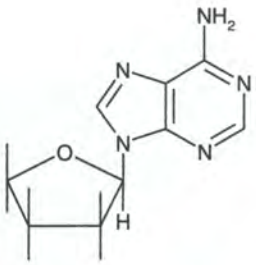


Fig. 2.13. N glycosidic linkage. Pentose sugar added to N_9 or N_1 by N–glycosidic bond.

Adenosine, guanosine

- Pyrimidine end in 'dine'
  Thymidine, cytidine, uridine

Nucleotides

Start with nucleotide name and add mono-, di-, or triphosphate to it.

- Adenosine monophosphate
- Deoxythymidine diphosphate

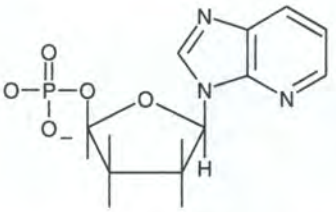


Fig. 2.14. Nucleotides.

PSEUDOURIDINE

TMP

- Contains ribose rather than deoxyribose
- Arises when UMP of preformed tRNA is methylated by SAM

UNUSUAL BASES OR MINOR BASES

- 5 methyl cytosine
- Other bases in tRNA:
  N^6 methyl adenine
  N^6, N^6 dimethyl adenine
  N^6, N^7 methyl guanine
- In bacteriophage DNA:
  5 hydroxy cytosine
  Methyl cytosine

Methylated purines

- 1,3,7 trimethyl xanthine: caffeine
- 1,3 dimethyl xanthine: theophylline
- 3,7 dimethyl xanthine: theobromine

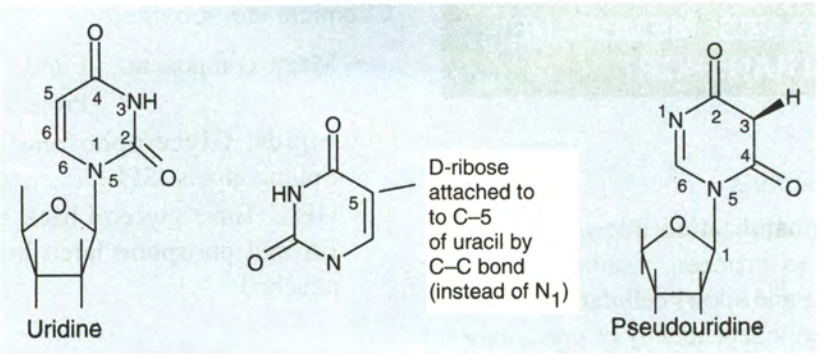


Fig. 2.15. Pseudouridine.

Free nucleotides

Xanthine, hypoxanthine, uric acid.

Nucleotide	*Function*
I. Nucleotide	
1. Adenosine nucleotides	
ATP	Source of energy
cAMP	Second messenger
Active sulfate (adenosine 3'phosphate 5'phospho sulfate PAPS)	Sulfur donor (proteoglycans) Sulfur conjugation of drugs
Active methionine (5-adenosyl methionine SAM)	Methyl donor, source of propyl-amine, in poly-amines
2. Guanosine derivative	
GDP	Coupled to substrate level phosphorylation
GTP	Allosteric regulation, energy source
cGMP	Intracellular signal second messenger (NO)
3. Hypoxanthine IMP	Purine salvage pathway
4. Uracil	Glycogen synthesis
UDP-G	Glycoprotein synthesis
UDP-Gln	Glucuronide conjugation reaction of bilirubin, drugs
5. Cytosine	
CTP	Phosphoglycerate synthesis
CDP	CDP choline: formation of sphingomyelin with ceramide
II. Coenzyme	NAD, FAD, NADP, CoA, SAM
III. Monomeric precursors	Monomeric unit of RNA, DNA
Energy metabolism	ATP, muscle contraction, active transport, ion gradient, phosphate donor
Monomeric unit	NTP, dNTP (for RNA, DNA)
Physiological mediators	Adenosine (coronary blow flow), cAMP, cGMP (second messenger), signal transduction (GTP binding protein)
Precursor function	GTP (mRNA capping)
Activate intermediates	UDP-G (glycogen), CDP-choline (phospholipid), SAM (methylation), PAPS (Sulfation)
Allosteric affects	ATP (negative for PFK), AMP (positive for phosphorylase B), dATP (negative effector RNP reductase)

(Contd.)

MEMBRANES, MEMBRANE TRANSPORT

MEMBRANES

Membrane functions

1. **Compartmentalization**: allows specialized activities to proceed without external interference and allows cellular activities to be regulated independently of one another.
2. **Frame work for biological activities**: Provide frame work within which components can be ordered for effective interaction.
3. **Provide selective permeability barrier**: To prevent unrestricted exchange of molecules from one side to the other.
4. **Transport of solutes:** From one side of membrane to another e.g. sugar, amino acids, ions.
5. **Signal transduction**: Membrane possess receptors to which various ligands can bind to generate a signal that stimulates or inhibits internal activities.
6. **Cell to cell interaction.**
7. **Energy transduction.**

Biological membranes

- Highly viscous and plastic structures, form boundaries around cell and sub-cellular compartments.
- Acts as selective permeability barrier
- Involved in signaling processes
- Contain varying amount of lipid and protein and carbohydrates
- Thickness 60-100 Å..
- Dynamic structures
- Thermodynamically stable, metabolically active
- Show chemical asymmetry: two faces of biological membrane differ from one another

Chemical composition

- Major component: Lipid
  Protein
- Lipids: Glycerophospholipids (GPL), Sphingolipids (SPL), cholesterol
- GPL: Have glycerol back bone to which FA and phosphorylated head group are attached

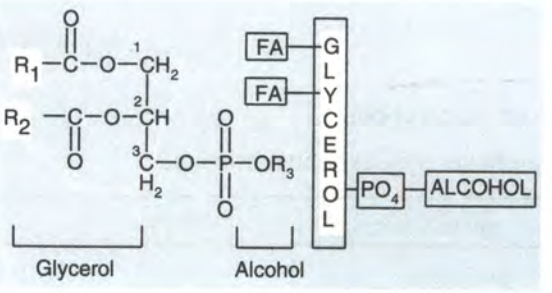


Fig. 2.16. Glycerophospholipid.

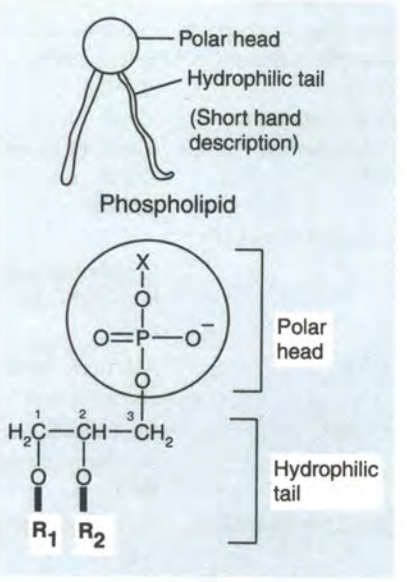


Fig. 2.17. Glycerophospholipid.

Phosphatidate, phosphatidylcholine, phosphatidylethanolamine, phosphatidylgly-

cerol, phosphaditylinositol, phosphatidyl-serine, and diphosphatidyl glycerol.

Present exclusively in IMM.

GPL contain two fatty acyl groups esterified to C-1 and C-2 of glycerol:

- C1: Usually saturated FA is found e.g. palmitic, stearic acid and form a straight chain.
- C2: Unsaturated FA is present e.g. oleic, linoleic, linolenic acid; usually cis and produces a kink.

	C_1	C_2
PC	Palmitic/Stearic acid	Oleic, linoleic, linolenic acid
PE	Palmitic/oleic acid	Arachidonic acid

Plasminogen

- Ethanolamine, ether/choline esterified to phosphate
- Abundant in nervous tissue and heart.

Every tissue and cellular membrane has a distinct composition of GPL and a definite pattern of FA composition (Fig. 2.18).

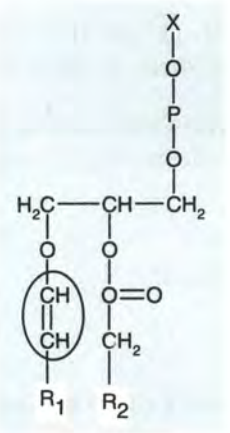


Fig. 2.18. Plasmalogen (α-β unsaturated ether).

SPL

In sphingomyelin, terminal-OH group of sphingosine is esterified to phosphorylcholine, so that its polar head is similar to PC.

Features

- Contain sphingosine back bone
- Sphingosine amino alcohol
- Sphingosine linked to FA by amide bond forming ceramide
- Sphingomyelin has PC esterified to 1-OH group
- Most abundant sphingolipid in mammalian tissues
- Lack phosphate, have sugar or primary group of sphingosine

Similarities between GPL and SPL

- Amphipathic with charged head group
- Both are classified under phospholipids

SPL	*Location*
Glucocerebroside	Present in non-neuronal tissue
Galactocerebroside	Present in brain and nervous tissue
Ganglioside	Contain sialic acid in head groups, present in brain

Cholesterol in membrane

- Most common sterol in membrane
- Also found in golgi apparatus, mitochondria and nuclear membrane
- Intercalates among PL with its OH group at interface and remainder in leaflet
- Amphipathic
- Ring system gives rigidity to membranes

Lipid composition varies in different membranes

- Highest concentration of neutral lipids and SPL in plasma membrane

- Myelin membrane rich in SPL and GPL
- Intracellular membrane contain primarily GPL
- In mitochondria, nucleus and RER lipid composition is similar
- Cardiolipin present exclusively in IMM
- Phosphatidylcholine, SPL predominant and ethanolamine, GPL are second.
- In mitochondria, there is no sphingosine
- Golgi apparatus has high concentration of neutral lipids especially cholesterol

Carbohydrates in membrane

- Oligosaccharides covalently attached to
  - Proteins to form glycoproteins
  - Lipids in lesser amount to form glycolipids

Sugar in glycoprotein and glycolipid: glucose, galactose, mannose, fructose, n-acetyl glucosamine (NAc Gln), n-acetyl galactosamine (NAc Galn), Sialic acid.

Location

Carbohydrates are located on:

- External side of plasma membrane
- Luminal side of ER.

Role

- Cell-cell recognition, adhesion, receptor action
- Carbohydrates of glycolipids of RBC plasma membrane determine whether person's blood type is A, B, AB or O.

Amphipathic nature of membrane lipids

- Membrane lipids contain hydrophobic and hydrophilic regions termed: **Amphipathic.**
  - Polar head of phospholipids and –OH group of cholesterol are:
    Hydrophilic: favor contact with water
    Hydrophobic tail: Saturated fatty acids straight chain
- Unsaturated FA – make kinked tails.

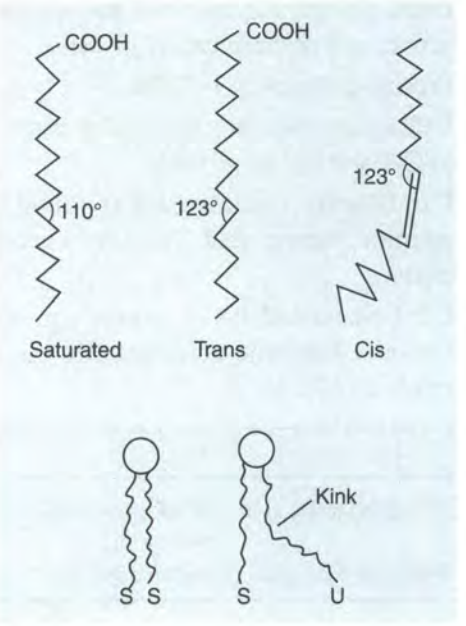


Fig. 2.19. Saturated and unsaturated FA in phospholipidds.

Role of kinking

Unsaturated FA make kinked tails and as more kinks are inserted in tails, membranes become less tightly packed and becomes more fluid.

- Membrane formation is consequence of amphipathic nature of molecules
- PL and GL (glycolipid) readily form bimolecular sheets in aqueous media.

Molecules with above mentioned preferences can thus arrange themselves in aqueous solution in two ways:

- Micelle
- Lipid bilayer

Micelle

A globular structure of 200nm size with polar head surrounded by water and carbohydrate tails sequestered inside.

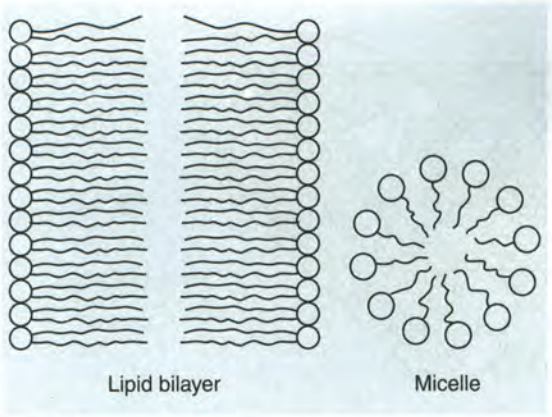


Fig. 2.20. Phospholipids and glycolipids form bi-molecular sheets in aqueous media.

Lipid bilayer

Lipid bilayer in formed due to hydrophilic and hydrophobic moieties of membrane lipids resulting in hydrophobic interior and hydrophilic exterior interacting with aqueous medium on each side of the bilayer.

Formation of lipid bilayer

- Self assembly process
- Rapid and spontaneous in water
- Dividing force is hydrophobic interactions.

Permeability

- Steroids traverse bilayer more readily than electrolytes
- Non lipid soluble molecules can't pass through bilayer and require pores and channels for them

Liposomes

- They are lipid vesicles with aqueous compartments enclosed by a lipid bilayer.
- Produced by suspending phospholipids (PC) in aqueous medium and sonicating to give closed vesicles of 500Å.
- Important experimental and clinical tool

Uses

1. To study effect of varying lipid composition on certain membrane functions.
2. Drugs, antibodies or isolated gene/DNA can be entrapped inside them to target them to specific tissues

Biological membrane structure

Fluid mosaic model

Proposed by Singer Nicolson (1972), widely accepted.

According to this model, membranes are two dimensional solutions of oriented lipids and globular proteins (Fig. 2.21)..

Role of biological membrane

- Permeability barrier
- Solvent for integral membrane proteins

Translational diffusion

PL undergoes redistribution in the plane of biological membrane, termed **translational diffusion**. It is several mm/sec.

Flippases-proteins that allow phospholipids to flip from one membrane surface to the other.

Transverse diffusion

A transition of molecule from one membrane surface to other is transverse diffusion or flips flop.

The characteristics of lipid bilayer explain many of the observed cellular membrane properties:

- Fluidity
- Flexibility
- Ability to self anneal
- Impermeability
- The model continues to undergo modification and refinement. It allows lateral movement, but not rotation.

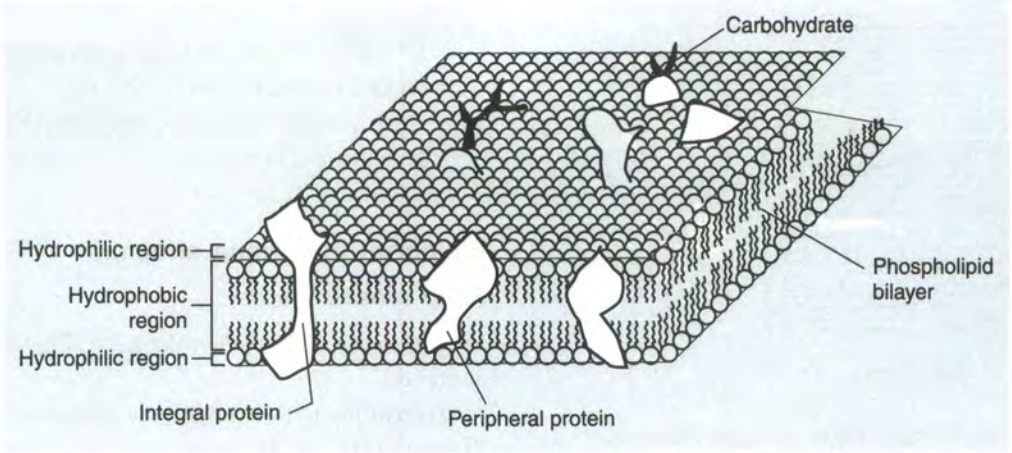


Fig. 2.21. Membrane structure.

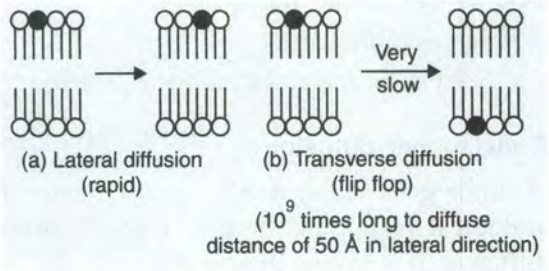


Fig. 2.22. (a) Lateral diffusion, (b) Transverse diffusion.

Membrane fluidity

Depends on:

- Temperature
- Composition
- Lipid composition
- Ordered/disordered
- Rigid/fluid

Lipids with short or unsaturated FA chain undergo phase transition.

When transition temperature (T_m) raised above melting temperature, rigid to fluid state occurs.

T_m depends on length of fatty acyl chain, degree of unsaturation.

Facts

- Saturated FA favor rigid membrane
- Double bond produces bend in hydrocarbon chain interferes high ordered packing and lowers T_m.
- Higher T_m required to increase fluidity
- Longer, saturated FA chains increase T_m, decrease fluidity; cis unsaturated bond lowers T_m and increase fluidity
- Cholesterol insertion in bilayer limits fluidity owing its rigid ring.

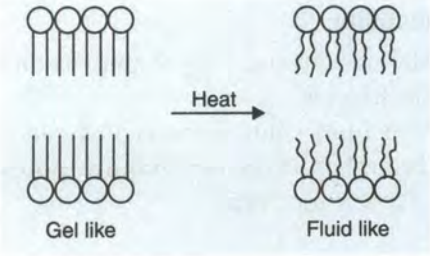


Fig. 2.23. Effect of temperature on membrane fluidity.

Lipid rafts and caveolae

The presence of the lipid ordered micro domains in cells has transformed fluid mosaic model into more complex system.

Lipid rafts

- Dynamic assembly of proteins and lipids that float freely in lipid bilayer.
- Dynamic areas of exoplasmic leaflet of lipid bilayer enriched in cholesterol, SPL

Caveolae

- Small surface invaginations seen in many cells and postulated to be formed by rafts on cell surface.
- Serves to store and down regulate raft proteins or acts as reservoir of rafts
- Several groups of pathogens, bacteria, virus, prion, parasites hijack lipid rafts for their purposes
- Lipid rafts play a role in signal transduction.

Fluidity of membrane affects its functions

- As membrane fluidity increases, its permeability to water and hydrophilic molecules increases
- Membrane fluidity can control activity of membrane bound enzyme, membrane functions such as phagocytosis and cell growth.
- Examples
  - RBCs in spur cell anaemia have a high cholesterol content, also seen in cirrhotic liver and premature RBC destruction in spleen
  - Insulin receptor: Increased unsaturated FA concentration in membrane increases fluidity and alters the receptor to bind more insulin.
  - Calcium ions decrease fluidity causing decrease in repulsion between polar groups and increasing packing of lipid molecules, thus decreasing fluidity.

Asymmetry

- Occurs in membrane bilayer
- Important for function of biological membranes
- Established during its biosynthesis
- Have different components and enzyme activities on both the surfaces
- Maintained throughout the lifetime of protein.

For example RBC membrane: SPL and PC on outer leaflet and PE, PS on inner leaflet and cholesterol on both leaflets.

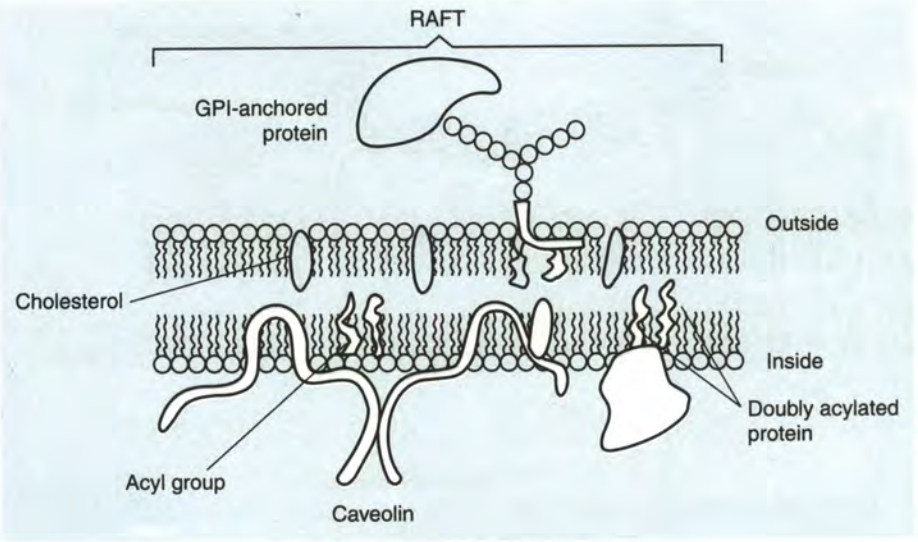


Fig. 2.24. Membrane raft group.

Regional asymmetries

- Villous border of mucosal cells
- Gap junction, tight junction, synapses occupy much smaller regions of membrane and generate local asymmetry
- PL: have inside-outside asymmetry (transverse asymmetry)
- PC, SM: outer leaflet
- PS, PE: Inner leaflet

GP and GL

- Abundant in RBC membrane
- Absent in mitochondrial membrane

Membrane proteins

- Features of proteins in membrane: myelin has low protein content (18%)
- Mitochondria has higher protein content (75%)
- Plasma membrane has intermediate protein content.

Function of proteins in membrane

1. Form a permeability barrier
2. Associated with bilayer as membrane proteins (integral and peripheral)

Proteins in membrane

Proteins in membrane are classified as:

- Peripheral (extrinsic)
- Integral (intrinsic)

Integral membrane proteins

- Span the lipid bilayer
- Present in membrane
- Amphipathic and globular e.g. transporter, receptors, G protein
- Distributed asymmetrically across bilayer
- May span bilayer many time
- Require detergents for their release

Peripheral proteins

- Bound to membrane by electrostatic and H-bond interaction with the head groups of lipids and integral proteins
- Do not interact with PL of bilayer
- Do not require detergents for their release
- Can be bound on cytosolic or extracellular side of membrane
- May be anchored to bilayer

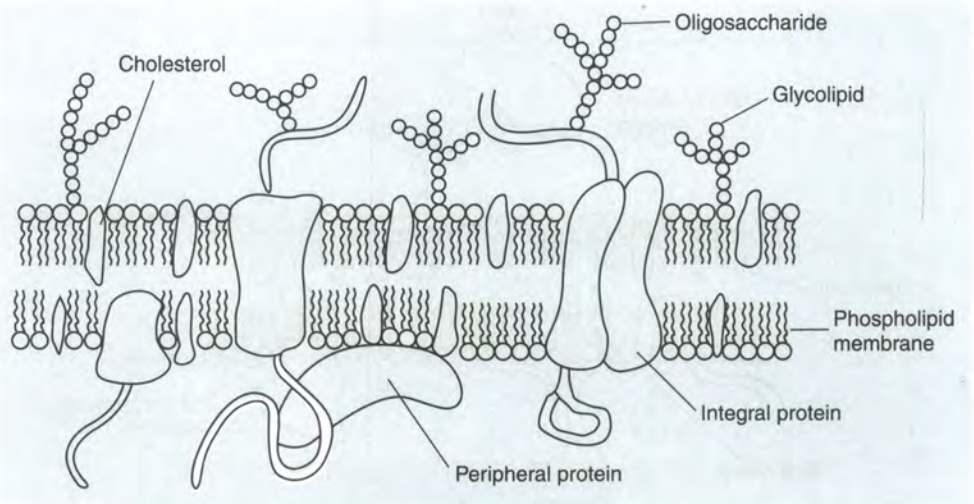


Fig. 2.25. Membrane structure.

Interaction of proteins with membrane

- Membrane-spanning: α-helices: most common
- Channel forming: β-strands: porin protein
- Bound to other membrane protein
  For example, Ankyrin: peripheral protein is bound to spectrin (a cytosolic structure) to maintain biconcave shape of RBC
- PGH_2 synthase :
  Lies along outer surface of membrane
  Bound by a set of α-helices
  Integral protein

Its localization is crucial for its function, as arachidonic acid, a substrate generated by hydrolysis of membrane lipids and reaches the enzyme through a hydrophobic channel.

Aspirin, ibuprofen block this channel, thus inhibiting PG synthesis.

MEMBRANE TRANSPORT

Membrane permeability

Plasma membrane is:

- Selectively permeable
- Water, gases (O_2, CO_2) can pass directly through it
- Other molecules (sugar, amino acid, ions etc.) require presence of transporter
- Permeability is conferred by two classes of membrane proteins: pumps and channels.

Types of transport

1. Passive transport
2. Active transport

Passive transport

- Molecules move from a higher to lower concentration
- Movement of molecules across a membrane does not require energy.
- Permeability property of membrane ensures that:
  - Essential molecules such as glucose, amino acids, and lipids readily enter the cell
  - Metabolic intermediate remain in cell
  - Waste compounds leave the cell, allowing cell to maintain a constant internal environment.

Simple diffusion

- Type of passive transport
- Does not require transport protein e.g. Water, gases, urea, ethanol
- Rate of diffusion is directly proportional to concentration gradient
- Not saturable

Terminology

Transporter

Translocation of molecule or ion across membrane by binding or physically moving the substance.

Group translocation

Involves movement as well as chemical modification of substrate, e.g. uptake of glucose by cell and its phosphorylation before its release into cytosol.

Pores

Tertiary and quaternary structures of intrinsic membrane proteins create an aqueous hole that permits diffusion of substrate through membrane in direction of a lower concentration.

Site of transporters

- Epithelial lining of all body cavities
- Stomach secrete acid
- Intestine secrete glucose, amino acid to blood
- Urinary bladder
- Skin

Membrane translocations

Type	*Class*	*Example*
Channel (pores)	Voltage regulated Agonist regulated cAMP regulated Others	Na channel AcH ® Cl channel pressure, stretch, heat sensitive
Transporter	Facilitated diffusion Active, mediated by: (i) Primary ATPase ATP binding cassette (ii) secondary	G-transporter Respiratory chain Na/K ATPase Multidrug resistance Na dependent G-transporter
Group translocation	-	Amino acid translocation

Channel and pores in membrane function differently

Membrane channels and pores are intrinsic membrane proteins and are differentiated based on their degree of specificity for molecules moving across the membrane.

Channel

Selective for inorganic ions and anions, while pores are not. They are:

- Specific
- Amphipathic
- α-helical

Common modification

- Amphipathic, α helices of associated protein sub units or domains within a single polypeptide
- Create an aqueous space

Exception: Porin in gram negative bacteria: β-sheet:

- Transport proteins are specific for particular molecule or group of structurally similar molecule
- Display Michalis-Mentin type of binding kinetics: V_{max}, K_m influenced by pH,

Comparison

Pore/channel	*Transporter*
1. Substrate move/drive down hill in direction of low concentration. Can conduct millions of ions per second	1. Uphill can move only hundreds to thousands molecules per second
2 Do not bind molecule ion to other side of membrane	2. Translocate ion/molecule by binding and physically moving the substance
3. Can be inhibited	3. Inhibited: Competitive Non-completive Defined reaction kinetics
4. Some degree of specificity	4. Specific to substrate
5. Rate faster: 10^7 ion s^{-1}	5. 10^2–10^3 molecule s^{-1}
6. Activity can be modulated	6. Activity can be modulated
7. Different drugs for specific channels have been developed	7. Different drugs for specific transporters have been developed

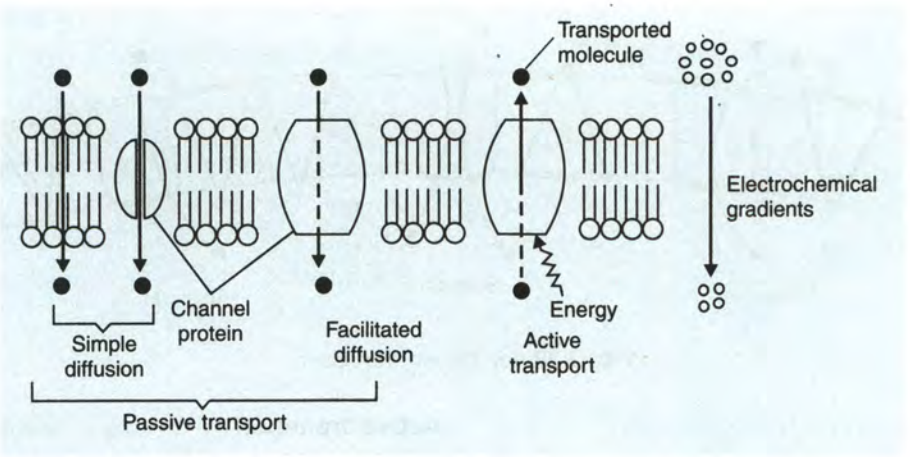


Fig. 2.26. Membrane transport.

temperature, inhibitor molecule in similar manner to enzyme.

Facilitated diffusion (or passive mediated transport)

- Facilitated diffusion is translocation of solutes through cell membrane without expenditure of energy.
- It requires presence of specific integral membrane protein to facilitate movement of molecules known as **uniport**.

Uniport system

- Moves one type of molecule bidirectionally
- Molecule binds to protein on one side of membrane, protein undergoes conformational change and transports molecule across the membrane and releases it on the other side.

Ping pong mechanism explains facilitated diffusion

Protein carrier in lipid bilayer associates with a solute undergoes a conformation change (pong to ping) and discharges solute on the other side. Empty carrier then reverts to original conformation (ping to pong) to complete the cycle.

Features

- Reversible
- Regulated by hormones

Role of hormones in facilitated diffusion

By changing the number of transporter available.

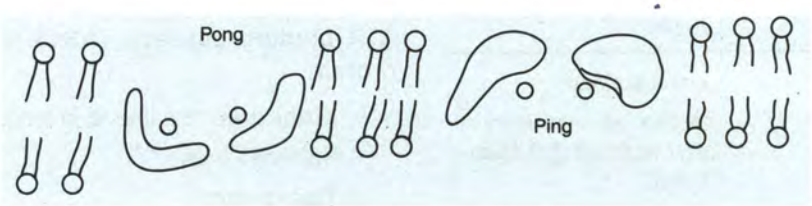


Fig. 2.27. Ping Pong mechanism.

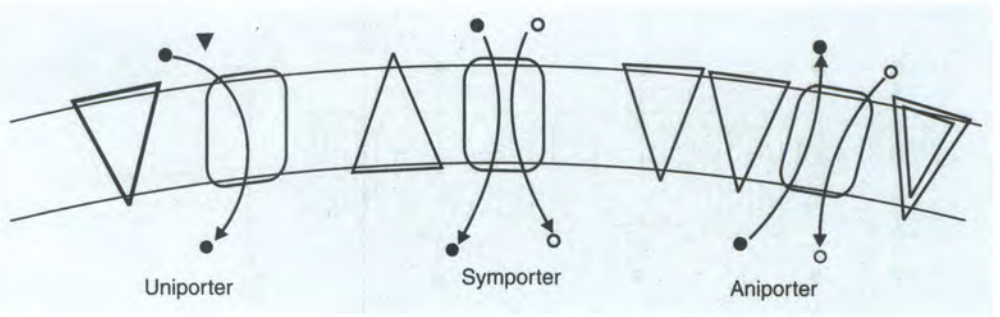


Fig. 2.28. Ion driven transport.

Examples

i. Insulin: increase transport of glucose and fat in muscle
ii. Glucocorticoids: enhance amino acid transport in all cells
iii. Estrogen: increase amino acid transport in liver

Facilitative *vs* active transport

Similarities

Both have:

- Specific binding sites for solute
- Carrier is saturable
- Carrier follows enzyme kinetics
- No covalent interaction occurs
- There is K_m (binding constant) for the solute
- Transport can be blocked by competitive inhibitors

Differences

Facilitative	*Active*
Bidirectional	Unidirectional
Occurs down hill	Occurs against electrical or chemical gradient-Uphill
Does not require energy	Requires energy

Active transport

Requires an input of metabolic energy that comes from ATP hydrolysis or light.

i. ATP driven ions:
   Na^+, K^+, Ca^{+2}, H^+
   Uses Na^+/K^+ ATPase.
   - Provides energy required for transport of molecule across membrane
   - Maintains Na^+/K^+ gradient and membrane electrical potential across plasma membrane: all cells maintain a high internal concentration of K^+ and low concentration of Na^+.

ii. Ion driven active transport
   Na^+/K^+ ATPase
   - Tetramer: 2α, 2β subunits.
   - Integral membrane protein
   - Coupled to ATP hydrolysis
   - 2 Na^+ ions pumped out and 3 K^+ ions pumped inside.

ATP binding cassette (ABC) transport protein

- Multi drug resistance protein
- Sulfonyl urea
- Transporter
- Xenobiotics

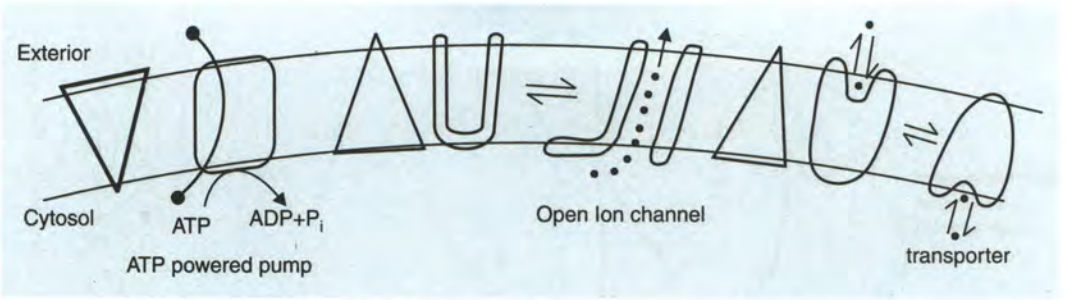


Fig. 2.29. Pumps, channels and transporter.

Glucose transport

- Glucose transport is facilitated transport
- Five glucose transporters have been reported: GLUT 1-5.
- GLUT1 has 12 hydrophobic segments in transporter region.
- GLU2 - Exports glucose from liver cells.

Rest of the transporters are responsible for movement of glucose into cell.

Transporter	*Location*
GLUT 1	Muscle, heart, blood brain barrier
GLUT 2	Liver, Pancreas, intestine, kidney
GLUT 3	Neuron, kidney
GLUT 4	Muscle, adipose tissue, heart (insulin-sensitive)
GLUT 5	Muscle, sperm

SGLT-1

- Sodium-dependent glucose transporter-1
- Secondary active transport of glucose
- Binds sodium and glucose at separate sites and transports them through plasma membrane of intestine
- Inhibited by Ouabain, phlorhizin
- The movement of Na^+ and glucose across the cell sets up a difference in osmotic pressure causing water to follow simple diffusion. This forms the basis of *glucose rehydration therapy*.

Ion driven active transport

It is movement of molecule across a membrane coupled with movement of ion, e.g. Na^+, K^+

Types

i. Symport

- Both molecule and ion move in same direction, e.g. Na^+/glucose transporter SGLT-1
- Cl/HCO_3 exchange transporter
- RBC band 3 anion transporter

ii. Antiport

- Ions move in opposite direction
- Concentration gradient of one compound can drive the movement of other solute, e.g. Cl/HCO_3 exchanger: in RBC, kidney
- In RBC: adjust HCO_3 concentration in arterial and venous blood
- In Kidney: responsible for base (HCO_3) efflux to balance ATP-driven H^+ efflux.
  - ATP-ADP transporter antiport: in inner mitochondrial membrane
  - PO_4-H^+ symport
  - Dicarboxylate carrier: exchanges malate for phosphate

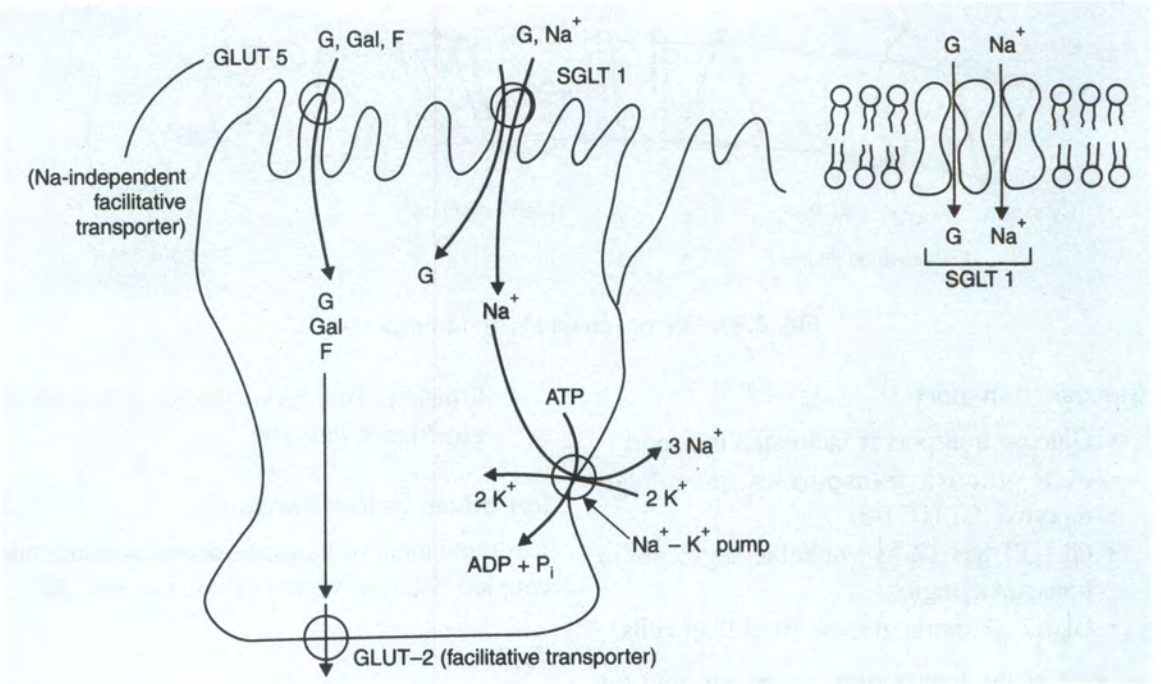


Fig. 2.30. Glucose transporters.

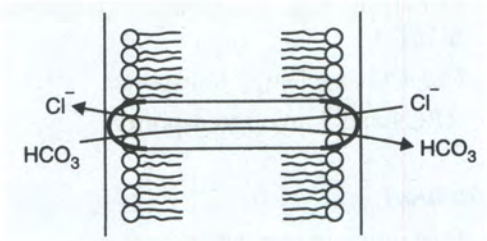


Fig. 2.31. Cl^-/HCO_3 exchange.

– Glutamate transport: exchange of aspartate

CALCIUM TRANSLOCATION

Ca^{+2} transporting ATPase

- This movement is coupled with ATP hydrolysis along concentration gradient.
- Ca is present in membranes of ER where it transports calcium ions out of the cytosol into lumen of ER.

CFTR

Cystic fibrosis trans membrane conductance regulator (CFTR)

- Type of ABC transporter
- Serves as both conductance regulator and chloride channel.

H^+/K^+ ATPase

- Present in epithelial lining of stomach (parietal cells)
- Secretes a solution of concentrated acid into stomach chamber
- At resting state, these pump molecules are situated in cytoplasmic membrane of parietal cell and are non-functional.
- When food enters stomach: a hormone message is transmitted to parietal cells. Causing pump containing membranes to move to

Transport mechanisms of various solutes

Sr. No.	*Solute*	*Mechanism*	*Tissues*
1.	Sugars		
	Glucose	Passive, active symport with Na^+	Widespread, renal tubules, small intestine
	Fructose	Passive	Intestine, liver
2.	Amino acids		
	Amino acid specific transporter	Active transport with Na^+	Intestine, kidney, liver
	All amino acid except proline	Active group translocation	Liver
	Specific amino acids	Passive	Intestine
3.	Dicarboxylic acid	Active symport with Na^+	Kidney
4.	Lactate and monocarboxylic acid	Active symport with H^+	Widespread
5.	Neurotransmitter: GABA, norepinephrine, glutamate, dopamine	Active symport with Na^+	Brain
6.	Urea	Passive	RBC, kidney
7.	Inorganic ions	Active	Mitochondria
	H^+	Active	Lysosome, endosome, golgi complex
8.	Na^+	Passive	Distal tubular
9.	Na, H^+	Active, antiport	PCT, small intestine
10.	Na^+, K^+	Active ATP driven	All cell plasma membrane
11.	Ions:		
	Ca^{+2}	Active ATP driven	Plasma membrane and ER
	Ca^{+2}, Na^+	Active antiport	Widespread
	H^+, K^+	Active antiport	Gastric, parietal cell
	Cl^-, HCO_3^-	Passive antiport	RBC, tissues

apical cell surface, where they fuse with plasma membrane and begin acid secretion.

- Drugs:
  - Prilosec inhibit H^+/K^+ ATPase to prevent heart burn
  - Zantac, Pepcid and Tagomet:
  - Block receptor on surface of parietal cell stopping cell from getting activated by hormone
  - Does not inhibit H^+/K^+ ATPase pump.

Types: Active transport

- Primary
- Secondary

Primary

- Requires ATP as energy source
- 3 types: P-, V-, F- type

P-type: Protein is phosphorylated and dephosphoryated during transport. Eg. Na^+-K^+ translocation

Diseases due to loss of membrane transport system

Sr. No.	*Transport defect*	*Outcome*
1.	Glucose-galactose transport	Decreased uptake in intestine
2.	Fructose transport system alteration	Fructose malabsorption
3.	Decreased neutral amino acids in epithelial cells	Hartnup disease symptoms in intestine and kidney
4.	Cystinuria	Renal absorption of cysteine and basic amino acid arginine is abnormal, cystine renal stones Tryptophan deficiency: pellagra-like syndrome
5.	Hypophosphatemia	Vitamin-D resistant rickets: renal absorption of phosphate abnormal
6.	Cystic fibrosis (CF)	cAMP regulated Cl channel affected increasing viscosity of body secretions

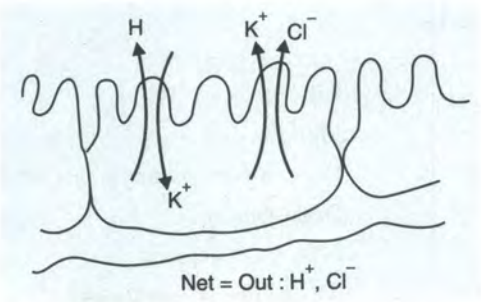


Fig. 2.32. H^+/K^+ ATPase in gastric mucosal cell.

V-type: Responsible for acidification (proton pumps) of:

- Interior of lysosome
- Endosomes
- Golgi apparatus
- Vesicles

Secretary vesicle:

- Utilize energy of ATP without forming a phosphorylated protein
- Cause active transport of hydrogen
- V pumps are present in plasma membrane of kidney tubules: to maintain acid-base balance by secreting proton into urine.

F-type:

- Involved in ATP synthesis
- Present in mitochondria (F_1/F_0 ATPase or ATP synthase)

Secondary

For translocation of solutes transmembrane chemical gradient of Na^+ or H^+ is required, e.g. sugar, amino acids

Ionophores

- Certain bacteria synthesize small organic molecules, ionophores that function as shuttles for movement of ions across membranes.
- They contain hydrophilic centers that bind specific ions and are surrounded by hydrophobic regions
- Each ionophore has definite ion sensitivity

Types

1. **Mobile carriers:** These readily diffuse in a membrane and can carry an ion across the membrane.
2. **Channel former:** These create a channel that traverses the membrane through which ions can diffuse.

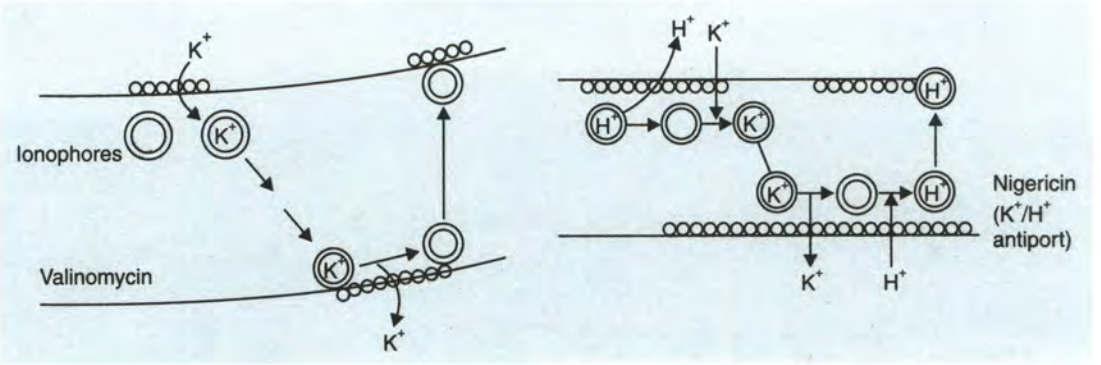


Fig. 2.33. Ionophores.

Examples

Valinomycin K^+ uniport:

- 1000 times more affinity for K^+ than Na^+
- 10 times more affinity for Ca^{+2} than Mg^{+2}.

A23187:

- $Ca^+/2H^+$ antiport

Nigericin:

- K^+/H^+ antiport (neutral)

Gramicidin:

- H^+, Na, K^+, Rb^+ forms channels

Alamethicin:

- K^+, Rb^+ forms channels

Ionophores are valuable experimental tools studying ion translocation in biological membranes and manipulation of ionic composition of cells.

Aquaporins (AP)

- Proteins that form water channels in certain membrane e.g. RBC, collecting ducts in kidney.
- Tetramer transmembrane proteins.
- Mutation in gene coding AP-2 causes diabetes insipidus.

Movement of large molecules

Large molecule can enter or leave cells through mechanisms such as exocytosis and endocytosis

Cotransport (Coupling active transport to existing ion gradient)

Na^+, K^+, H^+ ions produce concentration gradient by which free energy can be stored in a cell, e.g. glucose.

Its movement across apical plasma membrane is against concentration gradient and occurs by a cotransport with sodium.

Na^+ concentration is kept very low in the cell by primary active transport (Na^+/K^+ ATPase).

Tendency of Na^+ to diffuse back into cell down the concentration gradient is tapped by plasma cells to drive glucose into cells against concentration gradient termed as secondary active transport with help of sodium/glucose cotransporter.