Welcome to Metabolism-I: 03-59-362

The whole art of teaching is only the art of awakening the natural curiosity of young minds for the purpose of satisfying it afterwards."

Metabolism = Chemistry of Life

6th year of my teaching metabolism.

What other people thought of metabolism course?

Survey of Grad. Students, faculties about their experience with Metabolism Course in 2001.

90%	It is too dry, and boring, too many chemical structures to remember
5%	It is lot of chemistry, but if you like chemistry , it is OK
5%	It is dry but it was good course

What can I do?

Make it less dry, less boring or make it wet, interesting and exciting

I will try my best but need your participation.

Bioenergetics of life

Thermodynamics of energy conversions in living systems

Free energy from ATP and other high energy compound

Free energy from electron transfer from one molecule to other i.e. from oxidation reduction reactions

Metabolic pathways
Organic reaction mechanisms

Experimental approaches in the study of metabolism

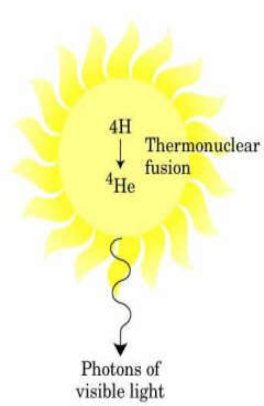
States of Matter: 3 or 4? **For example water**

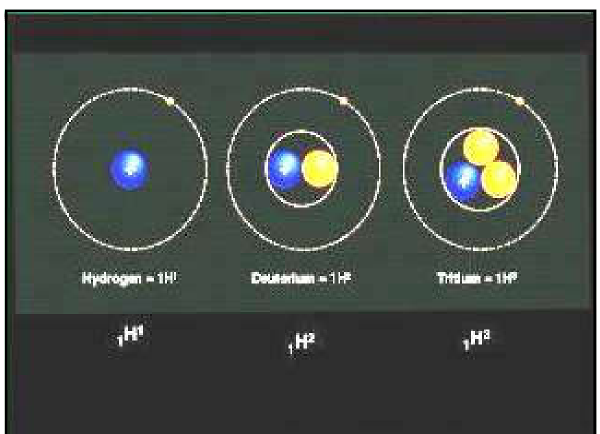
Solid:	Cold	Ice Crystal Liquid:
Liquid:	Warm	Water
Gas:	Hot	Steam
Plasma:	Very hot	Nuclei and electrons separated

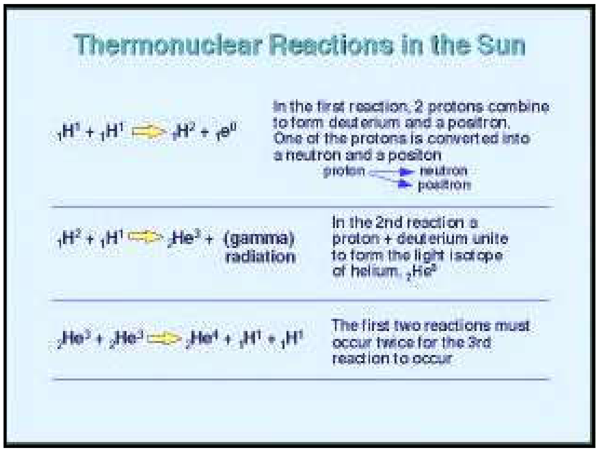
Hydrogen exists as a proton in plasma state

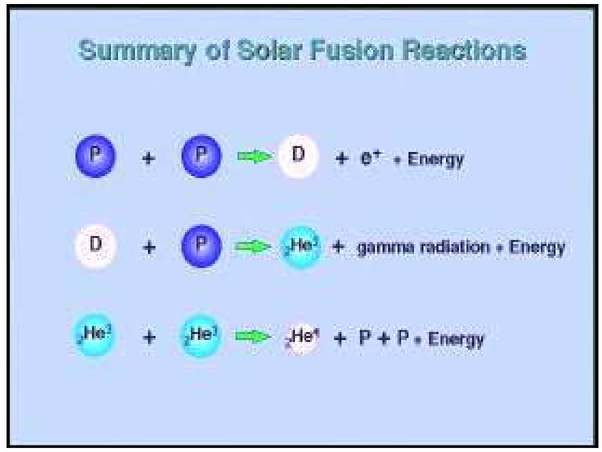
Energy need of all organisms are provided directly or indirectly by solar energy

Conversion of mass into energy is a very large increase in disorder.









Energy and Mass relationship: E = mc^2

m= mass and c = velocity of light

Nuclear reactions involve mass conversions into energy

Example: 4 H = He

Formation of helium from four hydrogen atoms is one of the most common nuclear reaction in solar plasma.

4X1.008= 4.032, whereas atomic mass of He is 4.003

Net loss of mass = 0.029 Daltons/He nuclei formation, this mass is converted into huge amount of energy. 1 Dalton = $1.66X10^{-24}$ gm or $1.66X10^{-27}$ Kg

E = 0.029 X $1.66X10^{-27}$ Kg X $(3X10^8$ m/sec$)^2$ = $4.54X10^{-12}$ Joules/ He

1 Mole of He is 4 gm, and it contains $6.023X10^{23}$ atoms

1 mole He (4gm) formation will produce $(4.54\ X10^{-12}).(6.023X10^{23})$

= $2.7X10^{12}$ Joules

Different forms of energy: Mechanical (kinetic and potential) Light, sound, heat, electrical (including magnetic) and chemical energy

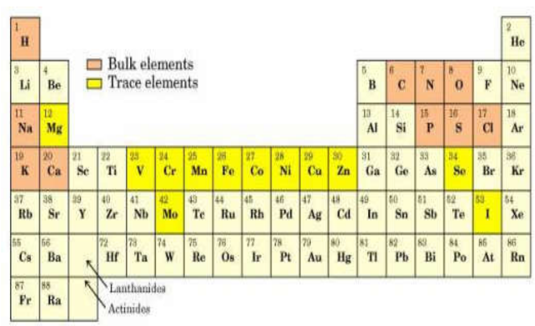
Conversion of mass and energy Inter-conversion of subatomic particle. Various energy forms: Light, electromagnetic, Kinetic, heat, potential, sound and wave	**Physics**
Interactions between atoms through electrons in outer orbit of atoms at various temperature, pressure and pH and solvent condition.	**Chemistry**
Interaction between macromolecules, atoms in similar way as in chemistry except at normal temperature, pressure, around neutral pH and strictly in aqueous Media.	Biochemistry

First law of thermodynamics:

For any chemical or physical change the total energy of the universe remains constant, in other words, energy may change form, or transported from one place to other but it cannot be destroyed or created.

Second law of thermodynamics:

Any spontaneous chemical or physical change is always accompanied by increase in the disorder of the universe. In other words disorder or entropy of the universe increases in all natural processes.



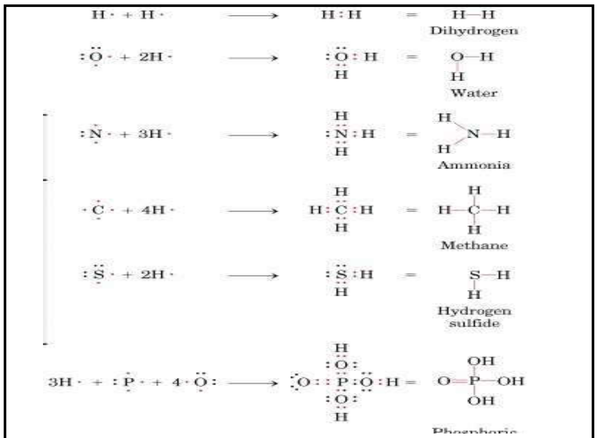
Elemental composition of the human Body

Consider a human with body weight of 60 kg
70% of total body weight is water, i.e 42 kg water
The dry weight of the 60 Kg body = 18 kg

Element	% of dry wt	dry wt (kg)	Trace Elements
C	61.7	11.0	B
N	11.0	2.0	F
O	9.3	1.6	Si
H	5.7	1.0	V
Ca	5.0	0.9	Cr
P	3.3	0.6	Mn
K	1.3	0.23	Fe
S	1.0	0.18	Co
Cl	0.7	0.12	Cu
Na	0.7	0.12	Zn
Mg	0.3	0.054	Se, Mo, Sn I

Main players in the chemistry of life: unpaired electrons

Atom	Number of unpaired electrons (in red)	Number of electrons in complete outer shell
H·	1	2
:Ö·	2	8
:N·	3	8
·C·	4	8
:S·	2	8
:P·	3	8



Most of the chemistry in living system is the result of interplay of electrons in the outer orbit of H, C, N, O, P and S.

Electro negativity: It is the tendency of an atom to attract an electron. F is the most electronegative element, but in biological system O is the most electronegative atom.

Many of the reactions in biological systems are catalyzed by enzymes and involve the lone pair of electron on N, O and S and P.

In many complex reactions a particular group of atoms (as part of compounds) are actively involved in the reactions. The are called functional groups.

table 3–2

The Electronegativities of Some Elements

Element	Electronegativity*
F	4.0
O	3.5
Cl	3.0
N	3.0
Br	2.8
S	2.5
C	2.5
I	2.5
Se	2.4
P	2.1
H	2.1
Cu	1.9
Fe	1.8
Co	1.8
Ni	1.8
Mo	1.8
Zn	1.6
Mn	1.5
Mg	1.2
Ca	1.0
Li	1.0
Na	0.9
K	0.8

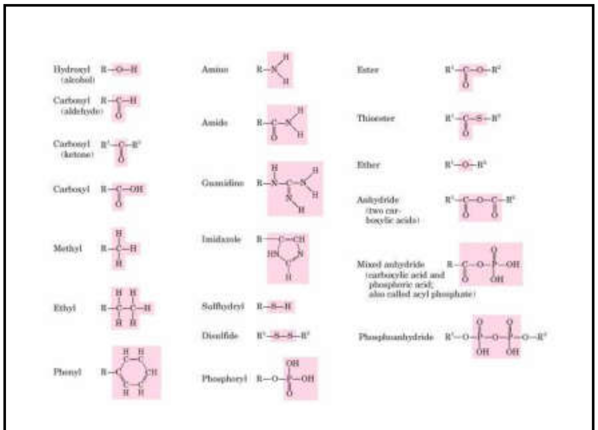
*The higher the number, the more electronegative (the greater the electron affinity of) the element.

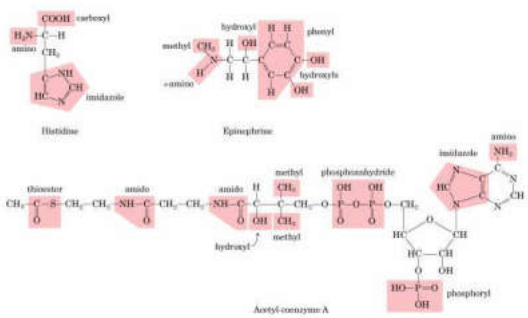
table 3–3

Strengths of Bonds Common in Biomolecules

Type of bond	Bond dissociation energy* (kJ/mol)	Type of bond	Bond dissociation energy (kJ/mol)
Single bonds		Double bonds	
O—H	461	C=O	712
H—H	435	C=N	615
P—O	419	C=C	611
C—H	414	P=O	502
N—H	389		
C—O	352	Triple bonds	
C—C	348	C≡C	816
S—H	339	N≡N	930
C—N	293		
C—S	260		
N—O	222		
S—S	214		

*The greater the energy required for bond dissociation (breakage), the stronger the bond.





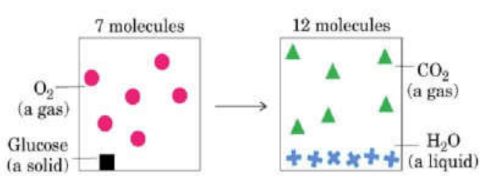
Energy in chemical reactions:

Free energy (G): It is an amount of energy capable of doing work during a reaction at constant temperature and pressure. When a reaction proceeds with release of free energy, the free energy change ΔG has a negative value and the reaction is called exergonic. Opposite is endergonic reaction in which there is gain of free energy.

Enthalpy (H): Represents the heat content of the reacting systems. When there is release of heat during a reaction it is referred as exothermic reaction and the ΔH is negative. Opposite is endothermic reaction with ΔH positive.

Entropy (S): it is a quantitative expression for the randomness or disorder in the system. There is always an increase in entropy or gain of entropy of the overall system in any reaction, i.e ΔS is always +ve.

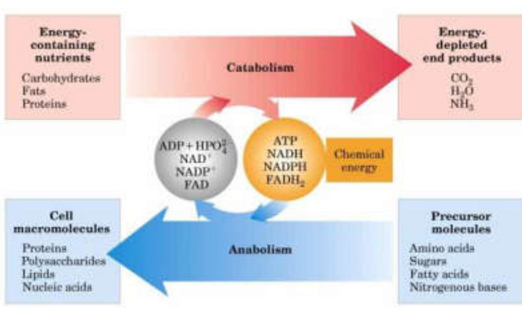
$\Delta G = \Delta H - T\Delta S$



Cells are isothermal systems i.e. they function at constant temperature and pressure. They can only use the free energy to do the work unlike other systems which can use heat released in system to do work (e.g. engines).

Plants acquire free energy from absorbed sun light, and synthesize nutrient. Both plants and heterotropic cells use this nutrients and transform the free energy in nutrient into ATP or other energy-rich compound. This process is called **catabolism**.

The free energy stored in energy-rich compounds is used by the cell to synthesize various compound and ingredients and this process is called **anabolism**.



The standard Free Energy change of a reaction is an alternative mathematical way of expression of equilibrium constant. It is defined as the Free energy change of a reaction at standard conditions (at 298 K and 1 atm pressure when the initial concentrations of reactants and products are at 1M each.

$aA + bB = cC + dD$

equilibrium constant $K_{eq} = [C]^c[D]^d/[A]^a[B]^b$

$$\Delta G'^\circ = -RT \ln K'_{eq}$$

Actual free energy change depends on reactants and product concentration

$$\Delta G = \Delta G'^\circ + RT \ln \frac{[C][D]}{[A][B]}$$

table 14–4

Standard Free-Energy Changes of Some Chemical Reactions at pH 7.0 and 25 °C (298 K)

Reaction type	$\Delta G'^{\circ}$ (kJ/mol)	$\Delta G'^{\circ}$ (kcal/mol)
Hydrolysis reactions		
Acid anhydrides		
Acetic anhydride + H_2O ⟶ 2 acetate	−91.1	−21.8
ATP + H_2O ⟶ ADP + P_i	−30.5	−7.3
ATP + H_2O ⟶ AMP + PP_i	−45.6	−10.9
PP_i + H_2O ⟶ 2P_i	−19.2	−4.6
UDP-glucose + H_2O ⟶ UMP + glucose 1-phosphate	−43.0	−10.3
Esters		
Ethyl acetate + H_2O ⟶ ethanol + acetate	−19.6	−4.7
Glucose 6-phosphate + H_2O ⟶ glucose + P_i	−13.8	−3.3
Amides and peptides		
Glutamine + H_2O ⟶ glutamate + NH_4^+	−14.2	−3.4
Glycylglycine + H_2O ⟶ 2 glycine	−9.2	−2.2
Glycosides		
Maltose + H_2O ⟶ 2 glucose	−15.5	−3.7
Lactose + H_2O ⟶ glucose + galactose	−15.9	−3.8
Rearrangements		
Glucose 1-phosphate ⟶ glucose 6-phosphate	−7.3	−1.7
Fructose 6-phosphate ⟶ glucose 6-phosphate	−1.7	−0.4
Elimination of water		
Malate ⟶ fumarate + H_2O	3.1	0.8
Oxidations with molecular oxygen		
Glucose + $6O_2$ ⟶ $6CO_2$ + $6H_2O$	−2,840	−686
Palmitate + $23O_2$ ⟶ $16CO_2$ + $16H_2O$	−9,770	−2,338

table 14–2

Relationship between the Equilibrium Constants and Standard Free-Energy Changes of Chemical Reactions

K'_{eq}	$\Delta G'^{\circ}$ (kJ/mol)	$\Delta G'^{\circ}$ (kcal/mol)*
10^3	−17.1	−4.1
10^2	11.4	2.7
10^1	−5.7	−1.4
1	0.0	0.0
10^{-1}	5.7	1.4
10^{-2}	11.4	2.7
10^{-3}	17.1	4.1
10^{-4}	22.8	5.5
10^{-5}	28.5	6.8
10^{-6}	34.2	8.2

*Although joules and kilojoules are the standard units of energy and are used throughout this text, biochemists and chemists sometimes express $\Delta G'^{\circ}$ values in kilocalories per mole. We have therefore included values in both kilojoules and kilocalories in this table and in Tables 14–4 and 14–6. To convert kilojoules to kilocalories, divide the number of kilojoules by 4.184.

Standard free energy changes are additive

(1)	A ⟶ B	$\Delta G_1'^{\circ}$
(2)	B ⟶ C	$\Delta G_2'^{\circ}$
Sum:	A ⟶ C	$\Delta G_1'^{\circ} + \Delta G_2'^{\circ}$

Glu + Pi = Glu 6P + H_2O δG = 13.8 kJ/mol

ATP + H_2O = ADP + Pi δG = -30.5 kJ/mol

Glu + ATP = ADP + Glu6P δG = -16.7 kJ/mol

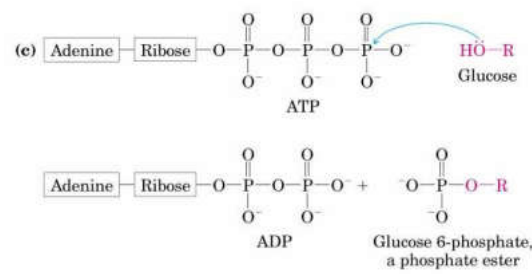
Phosphoryl group transfer and ATP as currency of free energy

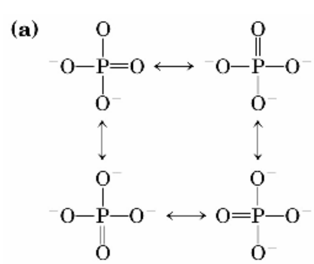
ATP is generated during catabolic reaction and the free energy of ATP hydrolysis is used in synthetic reactions, to do mechanical work or to produce light (in fire fly), sound or to conduct neuronal action potential.

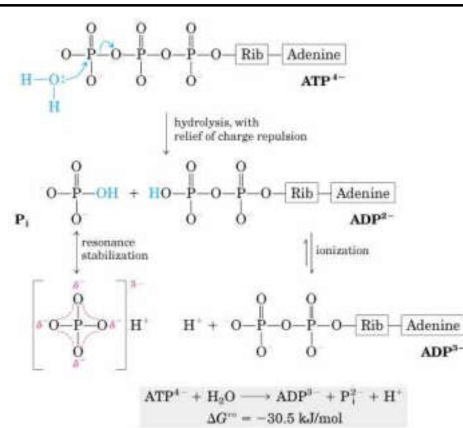
Basis of high energy of hydrolysis of ATP:

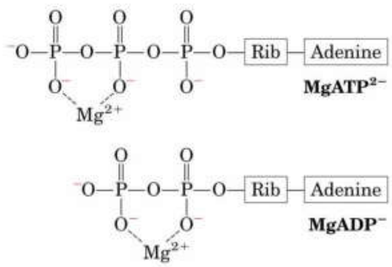
1. Resonance stabilization of products
2. Electrostatic repulsion between negatively charged oxygen atoms in ATP
3. High solvation energy of products

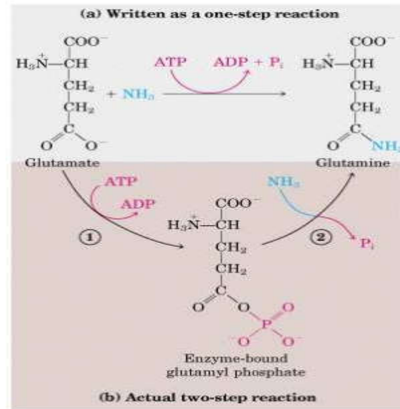
ATP is more stable than other phosphoanhydride compound

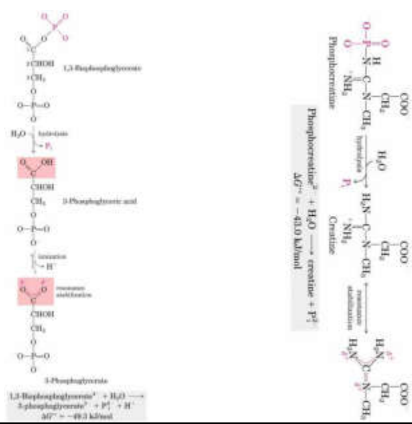


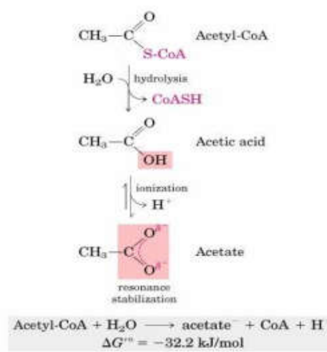


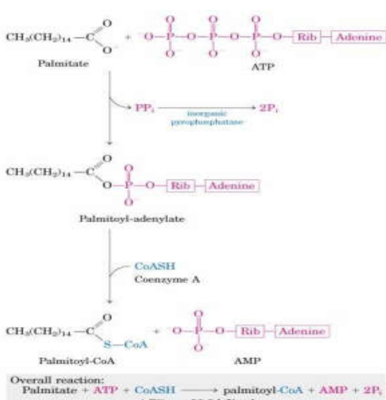












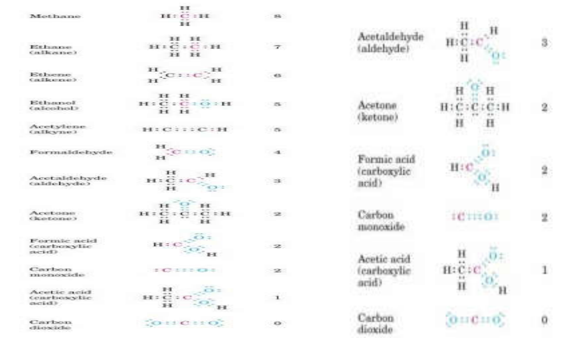
Oxidation-Reduction reactions in biological systems

As we have discussed so far, transfer of phosphoryl group is a central feature of metabolism and in energy transfer (due to the tendency of ATP to get hydrolyzed desperately). An equally important reaction mechanism to transfer free energy in biological systems is the transfer of electron in oxidation reduction reactions (due to tendency of some atoms to accept electron desperately).

Oxygen is the strongest electron acceptor in biological systems, due to it very high electro negativity and hence it is the strongest oxidizing agent. Fluorine is the strongest oxidizing agent but it is present in trace amount in living system.

Oxidizing ability: capacity to accept electrons depend on the electro negativity of the atom.

Flow of electrons can be used to do useful work as is done in battery operated motors, the electromotive force (EMF). In living systems, electron flow from various electron carrier to oxygen and the EMF generated is utilized for various energy transduction reactions.



Oxidation states of Carbon in various compounds; from fully reduced (methane0 to fully oxidized (carbon dioxide)

Electrons are transferred from one molecule (electron donor) to another (electron acceptor) in one of the four different ways;

1. **Direct electron transfers:**
   $Fe^{2+} + Cu^{2+} = Fe^{3+} + Cu^{+}$
2. **As hydrogen atom:** $AH_2 = A + 2e^- + 2H^+$
   $B + 2e^- + 2H^+ = BH_2$

   $AH_2 + B = A + BH_2$
3. **As hydride ion (:H⁻): As in case of NAD-linked dehydrogenases**
   Transfer of two electron.
4. **Through direct combination with oxygen: Covalent incorporation of oxygen into an organic molecule.**

In biological systems oxidation is often referred as dehydrogenation.

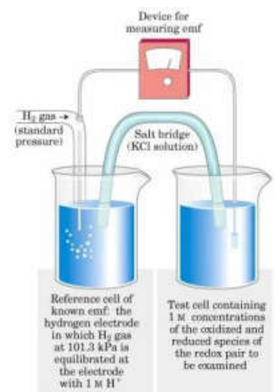
Standard reduction potential: (E^o)

It is the electric potential generated by a redox reaction against hydrogen electrode, when the concentration of reduced and oxidized species are at 1 M concentration.

Reduction potential of a half-cell depends on the the activity of reduced and oxidized species which is approximated by their concentrations.

Nernst Equation:
$E = E^o + RT/nf \ln [\text{electron acceptor}]/[\text{electron donor}]$
N= number of electrons transferred, f= Faraday's constant.
At 25 °C, the equation is;
$E = E^o + 0.026V/n \ln [\text{electron acceptor}]/[\text{electron donor}]$

Standard reduction potential is used for free energy change calculation as follows: $\Delta G = -nf\,\Delta E$ or $\Delta G^o = -nf\,\Delta E^o$



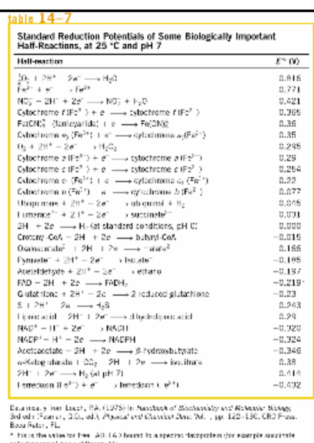


table 14–7

Standard Reduction Potentials of Some Biologically Important Half-Reactions, at 25 °C and pH 7

Half-reaction	E'^o (V)
$\frac{1}{2}O_2 + 2H^+ + 2e^- \longrightarrow H_2O$	0.816
$Fe^{3+} + e^- \longrightarrow Fe^{2+}$	0.771
$NO_3^- + 2H^+ + 2e^- \longrightarrow NO_2^- + H_2O$	0.421
Cytochrome f (Fe^{3+}) + $e^- \longrightarrow$ cytochrome f (Fe^{2+})	0.365
$Fe(CN)_6^{3-}$ (ferricyanide) + $e^- \longrightarrow Fe(CN)_6^{4-}$	0.36
Cytochrome a_3 (Fe^{3+}) + $e^- \longrightarrow$ cytochrome a_3 (Fe^{2+})	0.35
$O_2 + 2H^+ + 2e^- \longrightarrow H_2O_2$	0.295
Cytochrome a (Fe^{3+}) + $e^- \longrightarrow$ cytochrome a (Fe^{2+})	0.29
Cytochrome c (Fe^{3+}) + $e^- \longrightarrow$ cytochrome c (Fe^{2+})	0.254
Cytochrome c_1 (Fe^{3+}) + $e^- \longrightarrow$ cytochrome c_1 (Fe^{2+})	0.22
Cytochrome b (Fe^{3+}) + $e^- \longrightarrow$ cytochrome b (Fe^{2+})	0.077
Ubiquinone + $2H^+ + 2e^- \longrightarrow$ ubiquinol + H_2	0.045
Fumarate^{2-} + $2H^+ + 2e^- \longrightarrow$ succinate^{2-}	0.031
$2H^+ + 2e^- \longrightarrow H_2$ (at standard conditions, pH 0)	0.000
Crotonyl-CoA + $2H^+ + 2e^- \longrightarrow$ butyryl-CoA	−0.015
Oxaloacetate^{2-} + $2H^+ + 2e^- \longrightarrow$ malate^{2-}	−0.166
Pyruvate$^-$ + $2H^+ + 2e^- \longrightarrow$ lactate$^-$	−0.185
Acetaldehyde + $2H^+ + 2e^- \longrightarrow$ ethanol	−0.197
FAD + $2H^+ + 2e^- \longrightarrow FADH_2$	−0.219*
Glutathione + $2H^+ + 2e^- \longrightarrow$ 2 reduced glutathione	−0.23
$S + 2H^+ + 2e^- \longrightarrow H_2S$	−0.243
Lipoic acid + $2H^+ + 2e^- \longrightarrow$ dihydrolipoic acid	−0.29
$NAD^+ + H^+ + 2e^- \longrightarrow$ NADH	−0.320
$NADP^+ + H^+ + 2e^- \longrightarrow$ NADPH	−0.324
Acetoacetate + $2H^+ + 2e^- \longrightarrow$ β-hydroxybutyrate	−0.346
α-Ketoglutarate + CO_2 + $2H^+ + 2e^- \longrightarrow$ isocitrate	−0.38
$2H^+ + 2e^- \longrightarrow H_2$ (at pH 7)	−0.414
Ferredoxin (Fe^{3+}) + $e^- \longrightarrow$ ferredoxin (Fe^{2+})	−0.432

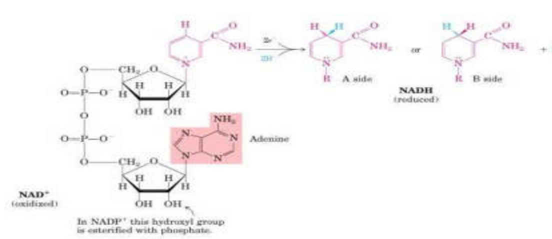
Data mostly from Loach, P.A. (1976) In Handbook of Biochemistry and Molecular Biology, 3rd edn (Fasman, G.D., ed.), Physical and Chemical Data, Vol. I, pp. 122–130, CRC Press, Boca Raton, FL.

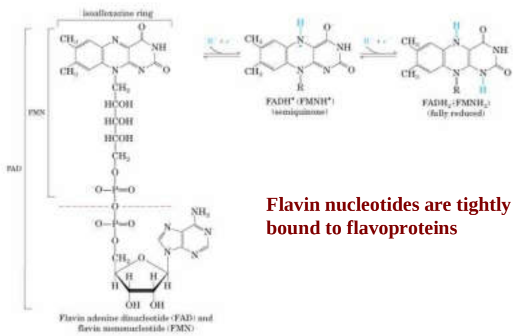
* This is the value for free FAD; FAD bound to a specific flavoprotein (for example succinate dehydrogenase) has a different E'^o.

NADH and NADPH are soluble electron carriers that act with dehydrogenases.

$NAD^+ + 2e^- + 2H^+ = NADH + H^+$

$CH_3CH_2OH + NAD^+ = CH3CHO = NADH + H^+$





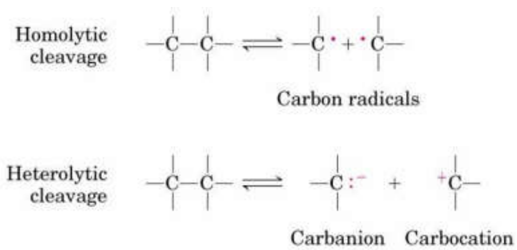
Flavin nucleotides are tightly bound to flavoproteins

Metabolic pathways;

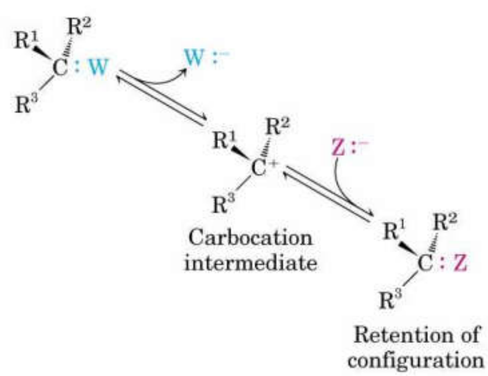
1. Metabolic pathways are irreversible
2. Every pathway has a committed step
3. Metabolic pathways are regulated
4. Some metabolic pathways are compartmentalized

Organic Reaction Mechanism:

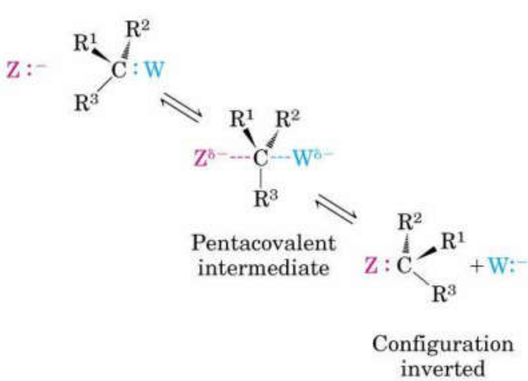
Group transfer: Nucleophilic substitutions
Oxidation reduction
Elimination, isomerization, rearrangement
Hydrolysis and synthesis (making and breaking of C-C bond



(a) SN1 reaction



(b) SN2 reaction



Experimental approaches to study metabolism:

Nutrient → Product

Intermediate steps

Isolation & characterization of specific enzymes

Regulatory mechanism in metabolism.

METABOLIC INHIBITORS

Glucose

G6P

Iodoacetamide

Fructose6p → Fr.1.6.biphosphate → GAP+DHAP

GENETIC DEFECTS:

Mutation in a particular gene

Defective protein enzyme

Blockage of metabolic reaction

Accumulation of an intermediate