HNS 103: BIOCHEMISTRY

LECTURE 1: Bioenergetics

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Course Outline

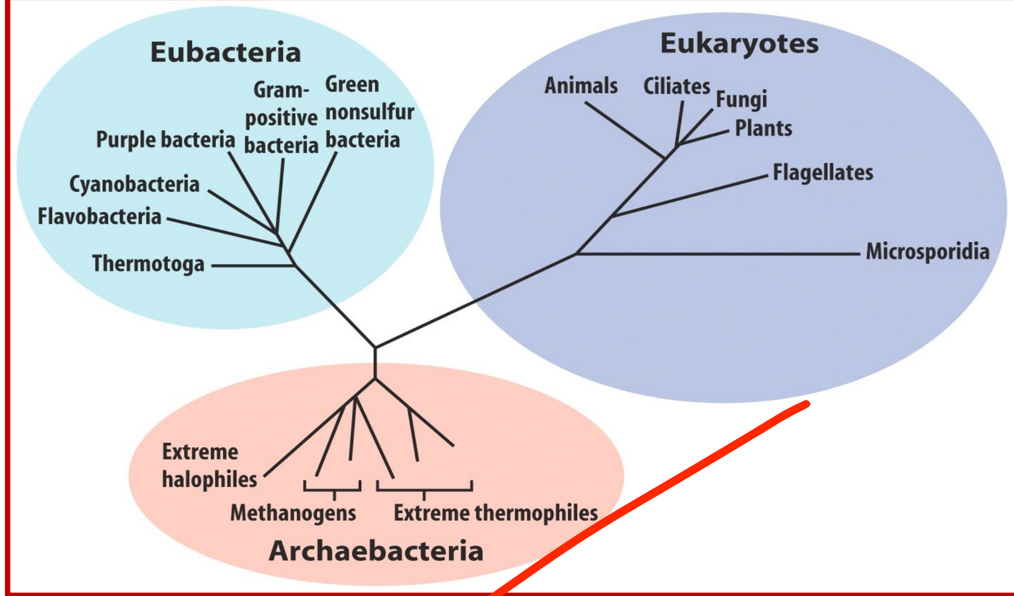
WEEK	DATE	TOPIC
1	21.02	**BIOENERGETICS:** standard free energy in chemical reactions, exergonic and endergonic reactions; Standard free energy of hydrolysis of ATP, Enzymatic transfer of phosphate groups to ATP; Properties of ATP and high energy phosphate compounds
2	28.02	**CARBOHYDRATE METABOLISM:** Carbohydrate digestion & mobilization; Glycolysis and Its regulations, Substrate Level Phosphorylation; pyruvate oxidation.
3	07.03	**KREBS CYCLE:** Krebs cycle and regulation; Anaploretic reactions; phosphogluconate pathway.
4	14.03	**MITOCHODRIAL STRUCTURE & FUNCTION:** Electron Transfer Chain; Oxidative Phosphorylation; Mechanisms of ATP generation; Uncouplers; inhibitors of ATP generation
5	21.03	**DISACCHARIDE METABOLISM:** Phosphogluconate pathway; Glycogen metabolism; Glycogenolysis and gluconeogenesis; Regulation of glycogen metabolism; Covalent modification; cAMP and hormonal regulation; Glycogen storage disease
	29.03	**CAT I**

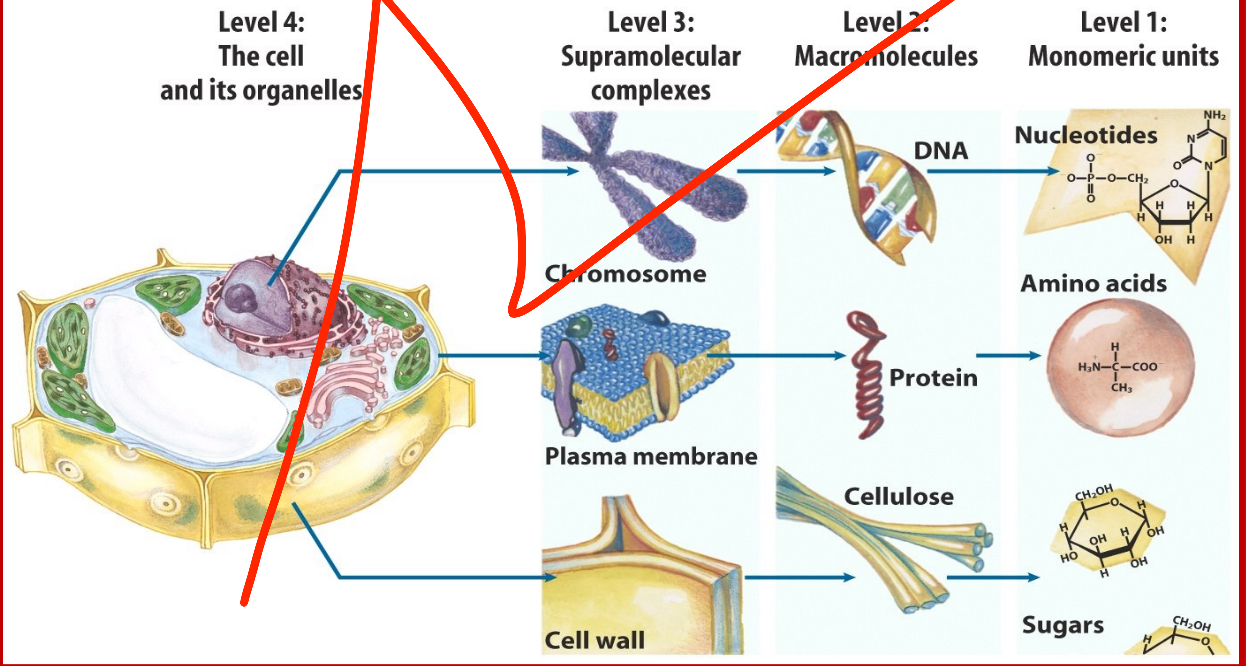
Introduction

What is BIOCHEMISTRY

- Deals with the chemical processes and compounds of living organisms, from viruses, (archaea, bacteria, fungi, plants, to invertebrates and vertebrates
- Applies the tools and concepts of chemistry to living systems
- Analyzes the structure and physical properties and the function of biological molecules (proteins, carbohydrates, lipids, and specialized chemical compounds that make up living cells)

- Also studies the interaction between molecules in living organisms
- Analyzes the mechanisms and products of all reactions involved





Bioenergetics and Thermodynamics

- Bioenergetics is the quantitative study of energy transductions that occur in living cells, and of the nature and function of the chemical processes underlying these transductions.
- Biological energy transformations obey the laws of thermodynamics

"Now, in the *second* law of thermodynamics..."

The three thermodynamic quantities:

- Gibbs free energy, ***G***

➔ Is the amount of energy capable of doing work during a reaction at constant temperature and pressure.

➔ When a reaction proceeds with a release of free energy, Δ*G* (the change in free-energy) is negative; such a reaction is said to be exergonic.

➔ When a reaction requires free-energy, Δ*G* is positive; this reaction is said to be endergonic.

- Enthalpy, ***H***

➔ Is the heat content of the reacting system.

➔ When a chemical reaction releases heat, it is said to be exothermic: the heat content of the product(s) is less than that of the reactant(s) and Δ*H* by convention is negative. Reacting systems taking up heat from the surroundings are endothermic and have a positive value of Δ*H.*

- Entropy, ***S***

➔ Is a quantitative expression for the randomness or disorder in a system.

When the products of a reaction are less complex and more disordered than the reactants, the reaction is said to proceed with a gain in entropy ($\Delta S > 0$).

A very important relationship between these thermodynamic quantities:

The free-energy change Δ*G* of a spontaneously reacting system is always negative; a reacting system for which Δ*G* > 0 is one that requires an input of energy

$$\boxed{\Delta G = \Delta H - T^{*}\Delta S}$$

The standard free-energy change Δ*G* is directly related to the equilibrium constant of a reaction

G = Gibbs free energy

H = Enthalpy

S = Entropy

TABLE 13–1 Some Physical Constants and Units Used in Thermodynamics

Boltzmann constant, $\mathbf{k} = 1.381 \times 10^{-23}$ J/K
Avogadro's number, $N = 6.022 \times 10^{23}$ mol^{-1}
Faraday constant, $\mathcal{F} = 96{,}480$ J/V · mol
Gas constant, $R = 8.315$ J/mol · K
(= 1.987 cal/mol · K)

Units of ΔG and ΔH are J/mol (or cal/mol)
Units of ΔS are J/mol · K (or cal/mol · K)
1 cal = 4.184 J

Units of absolute temperature, T, are Kelvin, K
25 °C = 298 K
At 25 °C, $RT = 2.479$ kJ/mol
(= 0.592 kcal/mol)

Relationship between ΔG and K_{eq}

Imagine a reaction: $a\,\mathrm{A} + b\,\mathrm{B} \rightleftharpoons c\,\mathrm{C} + d\,\mathrm{D}$

The composition of such a reacting systems continues changing until equilibrium is reached. The equilibrium is defined by:

$$K_{eq} = \frac{[\mathrm{C}_{eq}]^{c} * [\mathrm{D}_{eq}]^{d}}{[\mathrm{A}_{eq}]^{a} * [\mathrm{B}_{eq}]^{b}}$$

At equilibrium, the rates of the forward and the reverse reactions are exactly equal. Before equilibrium is reached, the tendency to move towards equilibrium is the driving force for the reaction, and its magnitude can be expressed as the free-energy change, **Δ*G***.

Under standard conditions (25°C = 298K) and when reactants and products are **initially** present at 1M concentrations (for gases at a partial pressure of 1atm = 101.3kPa), the force driving the reaction is defined as the standard free-energy change, **Δ*G*°**.

Gibbs showed that:

$$\Delta G = \Delta G° + RT\ \ln \frac{[C_i]^c * [D_i]^d}{[A_i]^a * [B_i]^b}$$

in which A_i, B_i,... are the initial concentrations of A, B,...

When a reaction is at equilibrium, no driving force remains and it can do no work: $\Delta G = 0$.

➡ $[A_i] = [A_{eq}]$, etc...

➡ $$\frac{[C_i]^c * [D_i]^d}{[A_i]^a * [B_i]^b} = \frac{[C_{eq}]^c * [D_{eq}]^d}{[A_{eq}]^a * [B_{eq}]^b}$$

➡ $0 = \Delta G° + RT\ \ln K_{eq}$

⇨ $$\Delta G° = -RT\ \ln K_{eq}$$

K_{eq} can be determined experimentally

when $K_{eq} >> 1$, then $\Delta G° < 0$, and
when $K_{eq} << 1$, then $\Delta G° > 0$

Bioenergetics

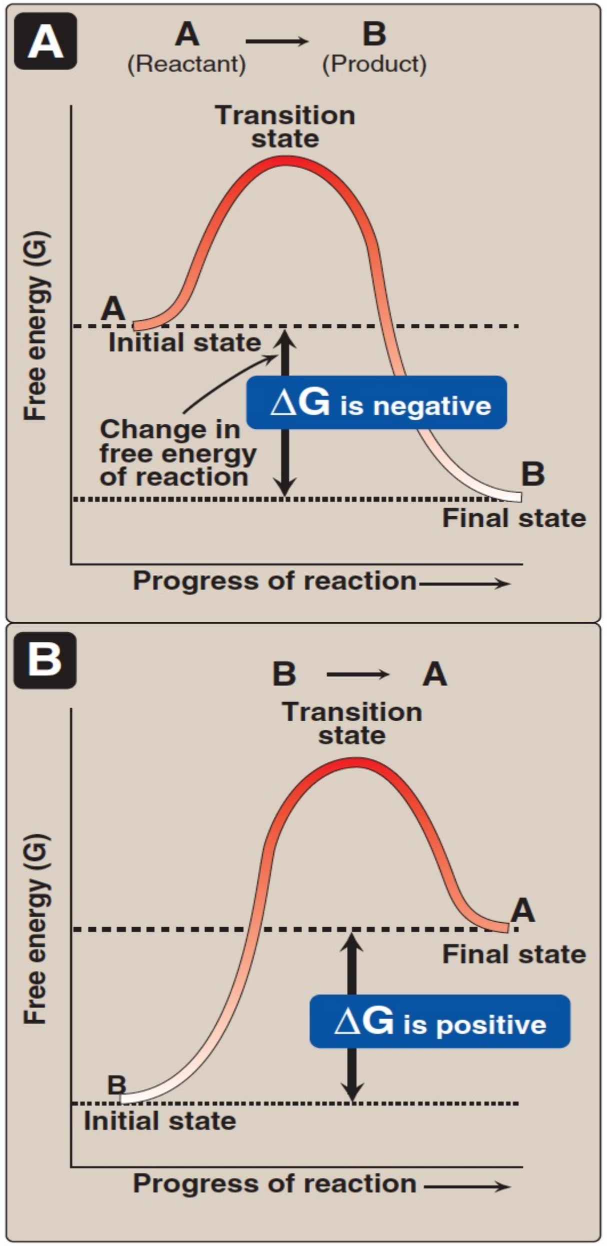


Fig 6.2

Sign of ΔG predicts the direction of a reaction

The change in free energy, ΔG, can be used to predict the direction of a reaction at constant temperature and pressure. Consider the reaction:

$$A \rightleftarrows B$$

1. **Negative ΔG:** If ΔG is a negative number, there is a net loss of energy, and the reaction goes spontaneously as written—that is, A is converted into B (Figure 6.2A). The reaction is said to be exergonic.
2. **Positive ΔG:** If ΔG is a positive number, there is a net gain of energy, and the reaction does not go spontaneously from B to A (see Figure 6.2B). Energy must be added to the system to make the reaction go from B to A, and the reaction is said to be endergonic.
3. **ΔG is zero:** If ΔG = 0, the reactants are in equilibrium. [Note: When a reaction is proceeding spontaneously—that is, free energy is being lost—then the reaction continues until ΔG reaches zero and equilibrium is established.]

ΔG: CHANGE IN FREE ENERGY

- Energy available to do work.
- Approaches zero as reaction proceeds to equilibrium.
- Predicts whether a reaction is favorable.

ΔH: CHANGE IN ENTHALPY

- Heat released or absorbed during a reaction.
- Does not predict whether a reaction is favorable.

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

ΔS: CHANGE IN ENTROPY

- Measure of randomness.
- Does not predict whether a reaction is favorable.

Since most biolological systems do not operate at a $[H^+]$ = 1M (pH=0) but around $[H^+] = 10^{-7}$M (pH=7), a new value for the free-energy is defined, also called the standard transformed constant, *i.e*. **$\Delta G'^{\circ}$**.

By definition, also the $[H_2O]$ = 55.5M, and
for reactions in which Mg^{2+} is involved (*e.g*. those involving ATP), its concentration is taken to be Mg^{2+} = 1mM.
The prime distinguishes this value from the untransformed value that is used by chemists and physicists.

By convention, when H_2O, H^+ and/or Mg^{2+} are reactants or products of a biochemical reaction, their concentrations are not taken up in the equations but they are incorporated in the constants K'_{eq} and $\Delta G'^{\circ}$.

There is a simple relationship between K'_{eq} and $\Delta G'^{\circ}$, *i.e*.:

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

K'_{eq} and $\Delta G'^{\circ}$ are constants characteristic of a reaction

TABLE 13–2 Relationship between the Equilibrium Constants and Standard Free-Energy Changes of Chemical Reactions

	$\Delta G'^\circ$	
K'_{eq}	(kJ/mol)	(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 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 13–4 and 13–6. To convert kilojoules to kilocalories, divide the number of kilojoules by 4.184.

TABLE 13–3 Relationships among K'_{eq}, $\Delta G'^\circ$, and the Direction of Chemical Reactions under Standard Conditions

When K'_{eq} is . . .	$\Delta G'^\circ$ is . . .	Starting with all components at 1 M, the reaction . . .
>1.0	negative	proceeds forward
1.0	zero	is at equilibrium
<1.0	positive	proceeds in reverse

Example:

$$\text{Glucose-1-phosphate} \quad \overset{\text{phosphoglucomutase}}{\rightleftharpoons} \quad \text{Glucose-6-phosphate}$$

Whether we start from 20mM glucose-1P and no glucose-6P, or from 20mM glucose-6P and no glucose-1P, at equilibrium (25°C, pH 7) the final mixture will contain 1mM glucose-1P and 19mM glucose-6P.

Important: an enzyme does NOT change the equilibrium, it speeds up the rate at which this equilibrium is reached...!!!

$$K'_{eq} = \frac{[\text{glucose-6P}]_{eq}}{[\text{glucose-1P}]_{eq}} = \frac{19\ \text{mM}}{1\ \text{mM}} = 19$$

$$\begin{aligned} \Delta G'^{\circ} &= -RT\ \ln K'_{eq} = -(8.315\ \text{J/mol*K}) * 298\text{K} * \ln(19) \\ &= -\ 7.3\ \text{kJ/mol} \end{aligned}$$

Under standard conditions, the reaction proceeds spontaneously when it starts with 20mM glucose-1P.

Other examples

TABLE 13–4 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 + $H_2O \longrightarrow$ 2 acetate	−91.1	−21.8
ATP + $H_2O \longrightarrow$ ADP + P_i	−30.5	−7.3
ATP + $H_2O \longrightarrow$ AMP + PP_i	−45.6	−10.9
PP_i + $H_2O \longrightarrow$ 2P_i	−19.2	−4.6
UDP-glucose + $H_2O \longrightarrow$ UMP + glucose 1-phosphate	−43.0	−10.3
Esters		
Ethyl acetate + $H_2O \longrightarrow$ ethanol + acetate	−19.6	−4.7
Glucose 6-phosphate + $H_2O \longrightarrow$ glucose + P_i	−13.8	−3.3
Amides and peptides		
Glutamine + $H_2O \longrightarrow$ glutamate + NH_4^+	−14.2	−3.4
Glycylglycine + $H_2O \longrightarrow$ 2 glycine	−9.2	−2.2

Actual free-energy changes depend on reactant and product concentrations

$\Delta G'^\circ$: free-energy change when initial concentrations of reactants and products is 1M, at pH 7, 25°C, 1 atm (101.3 kPa).

determines the direction and how far a reaction will go to reach equilibrium under standard conditions.

$\Delta G'^\circ$ is a characteristic, unchangeable value of a given reaction.

ΔG: the actual free-energy change. This value depends on:
- temperature prevailing during the reaction
- reactant and product concentrations.

ΔG changes during the course of the reaction from strongly negative to zero at equilibrium.

At equilibrium, no more work can be done by the reaction.

In thermodynamic calculations, the route used between the initial and final state is unimportant.

Standard free-energy are additive

$$\begin{array}{lll} \mathrm{A} \rightleftharpoons \mathrm{B} & & \Delta G_1{'^\circ} \\ \mathrm{B} \rightleftharpoons \mathrm{C} & & \Delta G_2{'^\circ} \\ \hline \mathrm{A} \rightleftharpoons \mathrm{C} & & \Delta G_1{'^\circ} + \Delta G_2{'^\circ} = \Delta G_{\mathrm{total}}{'^\circ} \end{array}$$

By coupling reactions, a thermodynamically unfavourable reaction can be driven in the forward direction.

Example:

1. Glucose + P_i $\longrightarrow$ Glucose-6P + H_2O $\Delta G'^\circ = +13.8$ kJ/mol
   $\Delta G'^\circ >> 0$ so, the reaction will not proceed as written.
2. ATP + H_2O $\longrightarrow$ ADP + P_i $\Delta G'^\circ = -30.5$ kJ/mol

Reactions (1) and (2) have common intermediates, *i.e*. P_i and H_2O.
Couple (1) and (2):

$$\text{Glucose} + \cancel{P_i} + \text{ATP} + \cancel{H_2O} \longrightarrow \text{Glucose-6P} + \cancel{H_2O} + \text{ADP} + \cancel{P_i}$$

$$\Delta G'^\circ = (+13.8\ \text{kJ/mol}) + (-30.5\ \text{kJ/mol}) = -16.7\ \text{kJ/mol}$$

Energy stored in ATP is used to drive the formation of glucose-6P.

$$\text{Glucose} + P_i \longrightarrow \text{Glucose-6P} + H_2O$$

$$K'_{eq},1 = \frac{[\text{glucose-6P}]}{[\text{glucose}]*[\text{Pi}]} = -3.9*\ 10^{-3}\ M^{-1}$$

Note that H_2O is not included in the expression: its concentration is considered to remain unchanged during the reaction!!!

$$\text{ATP} + H_2O \longrightarrow \text{ADP} + P_i$$

$$K'_{eq},2 = \frac{[\text{ADP}]*[\text{Pi}]}{[\text{ATP}]} = 2*\ 10^{5}\ M$$

$$K'_{eq},\ \text{overall} = \frac{[\text{glucose-6P}]*[\text{ADP}]*[\text{Pi}]}{[\text{glucose}]*[\text{ATP}]*[\text{Pi}]} = K'_{eq},1 * K'_{eq},2$$

$$= (3.9*\ 10^{-3}\ M^{-1})*(2*10^{5}\ M)$$

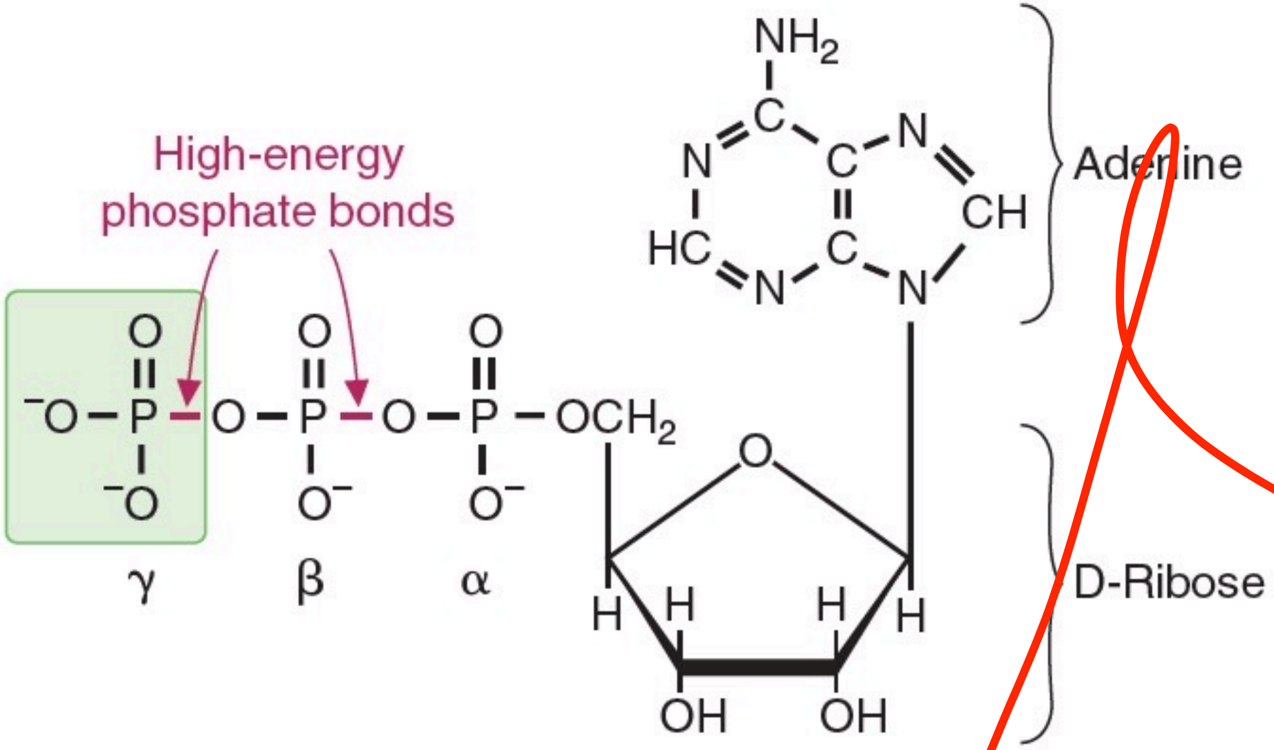
$$= 780$$

$\Delta G'°$ values are additive

K'_{eq} for a reaction that is the sum of two reactions is the product of their individual K'_{eq}, *i.e.* equilibrium constants are multiplicative

ATP

Structure of ATP

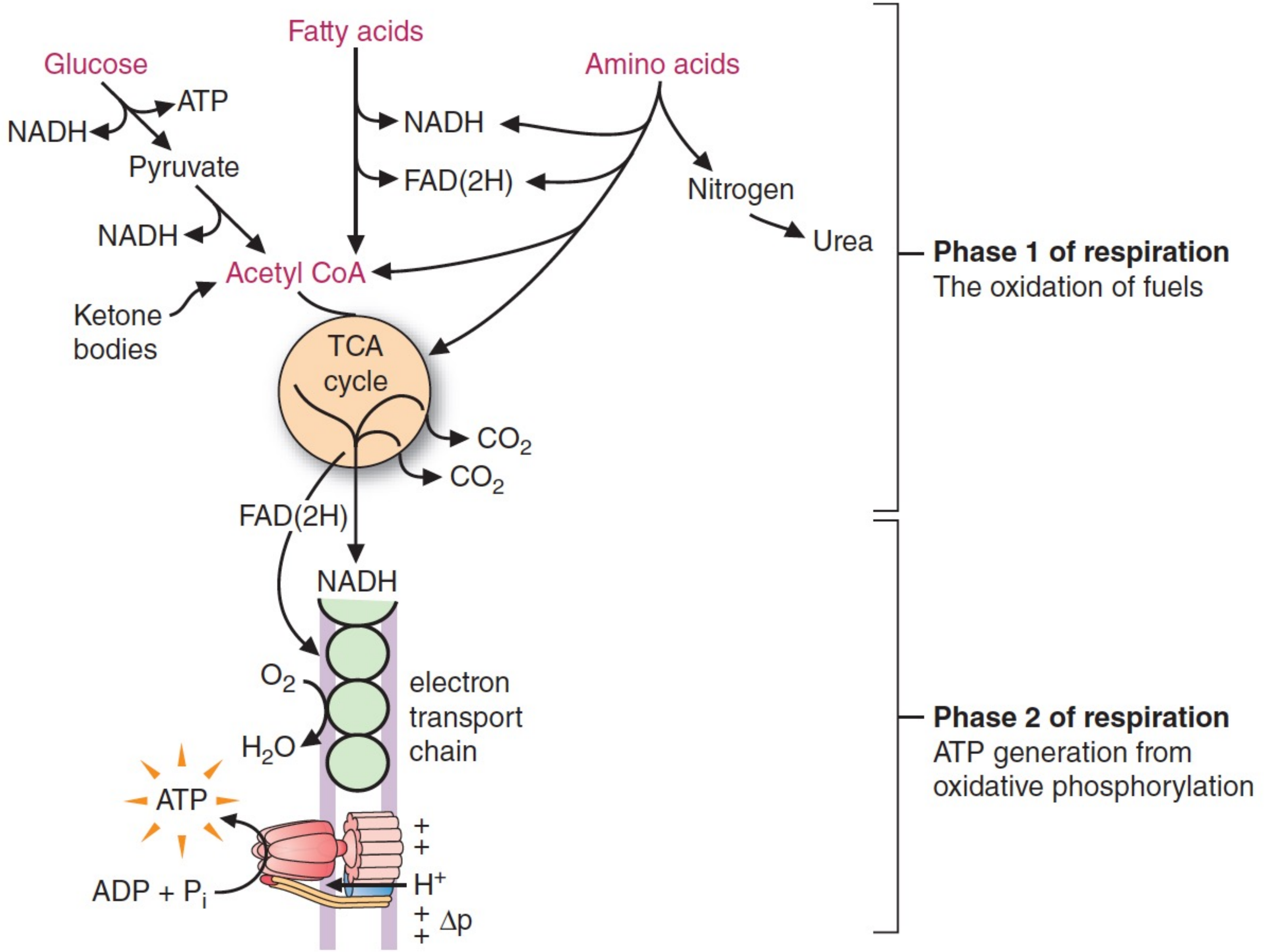


Adenosine 5'-triphosphate

ATP

The structure of adenosine triphosphate (ATP), and the location of the two high energy bonds in the molecule.

Generation of ATP from fuels in the blood, and cellular respiration



Phase 1 of respiration
The oxidation of fuels

Phase 2 of respiration
ATP generation from oxidative phosphorylation

function of ATP

The functions of ATP

ATP plays a central role in **energy exchanges** in the body.

1. ATP is constantly being consumed and regenerated.
   - It is consumed by processes such as muscular contraction, active transport, biosynthetic reactions etc etc
   - It is regenerated by the oxidation of foodstuffs.

2. The free energy released when ATP is hydrolyzed is used to drive reactions that require energy.
   - ATP can be hydrolyzed to ADP and inorganic phosphate (Pi) or to AMP and pyrophosphate (PPi).
   - Other nucleoside triphosphates (GTP, UTP, and CTP) are sometimes used to drive biochemical reactions. They can be derived from ATP, and have the same Gibbs free energy of hydrolysis as do the two high-energy bonds in ATP.

3. For the hydrolysis of ATP to ADP + Pi, $\Delta G^{o\prime}$ **= –7.3 kcal/mol.**
   - The anhydride bonds of ATP are often called “high-energy bonds.”
   - **$\Delta G^{o\prime}$** is large, however, not because a single bond is broken, but because the products of hydrolysis are more stable than ATP.

4. ATP can transfer phosphate groups to compounds such as glucose, forming ADP.

5. ADP can accept phosphate groups from compounds such as phosphoenolpyruvate, phosphocreatine, or 1,3-bisphosphoglycerate, forming ATP.

relevance of free energy changes to biologic systems

1. The rate of a reaction is not related to its free energy change.
    a. A reaction with a large negative free energy change does not necessarily proceed rapidly.
    b. The speed of a reaction depends on the properties of the enzyme that catalyzes the reaction.
        (1) An enzyme increases the rate at which a reaction reaches equilibrium. It does not affect Keq (the relative concentrations of the substrates and products at equilibrium).

2. Most biochemical reactions exist in pathways; therefore, other reactions are constantly adding substrates and removing products.
    a. The relative activities of the enzymes that catalyze the individual reactions of a pathway differ.
    b. Some reactions are near equilibrium ($\Delta G = 0$). Their direction can be readily altered by small changes in the concentrations of their substrates or products.
    c. Other reactions are far from equilibrium. Allosteric factors that alter the activity of these enzymes can change the overall flux through the pathway.

Questions