

Unit 2: Bioenergetics

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What is Bioenergetics?

- The study of **energy** in **living systems** (environments) and the **organisms** (plants and animals) that utilize them.
- *Energy*
 - **Required by all organisms**
 - **May be Kinetic or Potential energy**

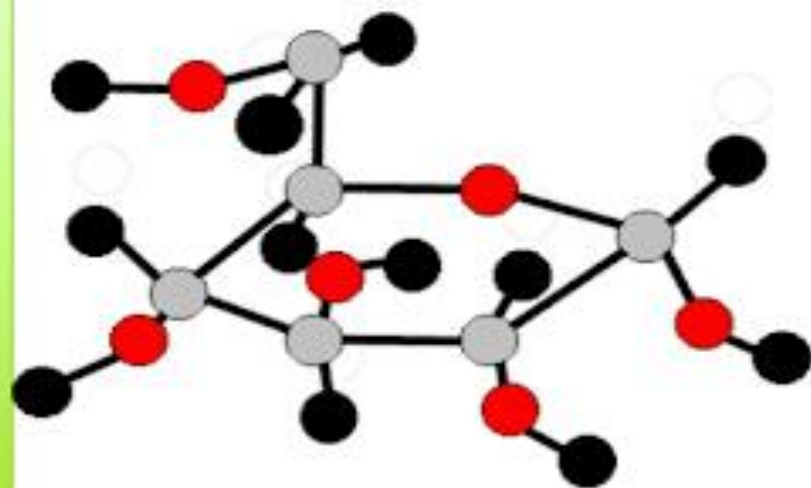
Kinetic Energy

- Energy of Motion
- Heat and light energy are examples



Potential Energy

- Energy of position
- Includes energy stored in chemical bonds



Bioenergetics

- Bioenergetics is the part of biochemistry concerned with the energy involved in making and breaking of chemical bonds in the molecules found in biological organisms.
- Growth, development and metabolism are some of the central phenomena in the study of biological organisms. The role of energy is fundamental to such biological processes. The ability to harness energy from a variety of metabolic pathways is a property of all living organisms. Life is dependent on energy transformations; living organisms survive because of exchange of energy within and without.

Bioenergetics

- In a living organism, chemical bonds are broken and made as part of the exchange and transformation of energy. Energy is available for work (such as mechanical work) or for other processes (such as chemical synthesis and anabolic processes in growth), when weak bonds are broken and stronger bonds are made. The production of stronger bonds allows release of usable energy.

Bioenergetics

- It is the quantitative study of the energy transductions that occur in living cells and of the nature and function of the chemical processes underlying these transductions.

Law of Thermodynamics

- The laws of thermodynamics are important unifying principles of biology. These principles govern the chemical processes (metabolism) in all biological organisms.

Law of Thermodynamics

- The First Law of Thermodynamics, also known as the law of conservation of energy, states that energy can neither be created nor destroyed. It may change from one form to another, but the energy in a closed system remains constant.
- In any physical or chemical change, the total amount of energy in the universe remains constant, although the form of energy may change.

Law of Thermodynamics

- The Second Law of Thermodynamics states that when energy is transferred, law of entropy, there will be less energy available at the end of the transfer process than at the beginning. Due to entropy, which is the measure of disorder in a closed system, all of the available energy will not be useful to the organism. Entropy increases as energy is transferred.
- In all natural processes, the entropy of the universe increases.

Free energy of a reaction

The **free energy change** (ΔG) of a reaction determines its spontaneity. A reaction is spontaneous if ΔG is negative (if the free energy of products is less than that of reactants).

For a reaction $A + B \leftrightarrow C + D$

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

$\Delta G^{\circ'}$ = standard free energy change (at pH 7, 1M reactants & products); R = gas constant; T = temp.

For a reaction $A + B \leftrightarrow C + D$

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

$\Delta G^{\circ'}$ of a reaction may be positive, & ΔG negative, depending on cellular concentrations of reactants and products.

Many reactions for which $\Delta G^{\circ'}$ is positive are spontaneous because other reactions cause depletion of products or maintenance of high substrate concentration.

At **equilibrium**

$$\Delta G = 0.$$

K'_{eq} , the ratio $[C][D]/[A][B]$ at equilibrium, is the **equilibrium constant**.

An equilibrium constant (**K'_{eq}**) **greater than one** indicates a **spontaneous** reaction (negative $\Delta G^{\circ'}$).

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

$$0 = \Delta G^{\circ'} + RT \ln \left(\frac{[C][D]}{[A][B]} \right)$$

$$\Delta G^{\circ'} = -RT \ln \left(\frac{[C][D]}{[A][B]} \right)$$

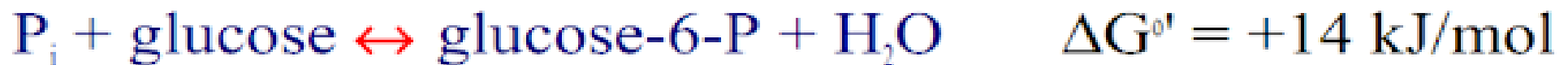
$$\text{defining } K'_{eq} = \left(\frac{[C][D]}{[A][B]} \right)$$

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

Energy coupling

- ♦ A spontaneous reaction may drive a non-spontaneous reaction.
 - ♦ **Free energy changes** of coupled reactions are **additive**.
- A.** Some enzyme-catalyzed reactions are interpretable as **two coupled half-reactions**, one spontaneous and the other non-spontaneous.
- ♦ At the enzyme active site, the coupled reaction is kinetically facilitated, while individual half-reactions are prevented.
 - ♦ Free energy changes of half reactions may be summed, to yield the free energy of the coupled reaction.

For example, in the reaction catalyzed by the Glycolysis enzyme **Hexokinase**, the half-reactions are:



Coupled reaction:



The structure of the enzyme active site, from which H_2O is excluded, prevents the individual hydrolytic reactions, while favoring the coupled reaction.

B. Two separate reactions, occurring in the same cellular compartment, one spontaneous and the other not, may be coupled by a **common intermediate** (reactant or product).

A hypothetical, but typical, example involving PP_i :

Enzyme 1:



Enzyme 2:



Overall spontaneous reaction:



Pyrophosphate (PP_i) is often the product of a reaction that needs a driving force.

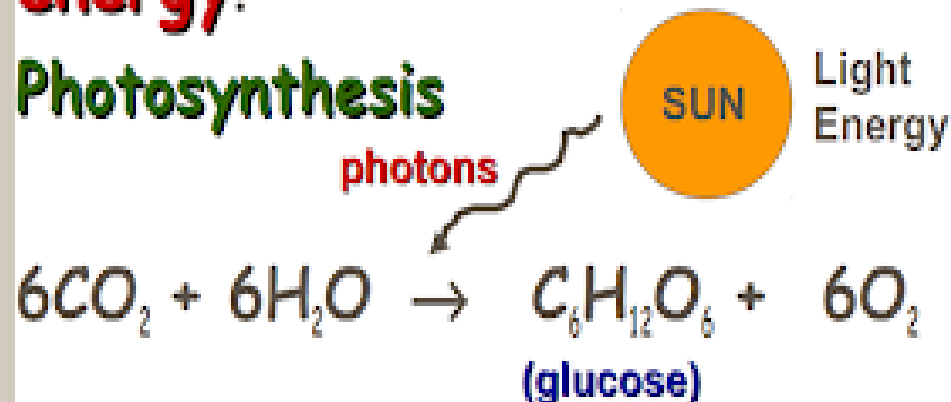
Its spontaneous hydrolysis, catalyzed by Pyrophosphatase enzyme, drives the reaction for which PP_i is a product.

Two Types of Energy Reactions

Endergonic Reactions

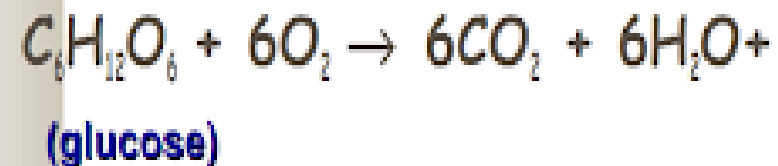
- **Chemical reaction** that requires a net input of **energy**.

- **Photosynthesis**



Exergonic Reactions

- **Chemical reactions** that **releases energy**
- **Cellular Respiration**

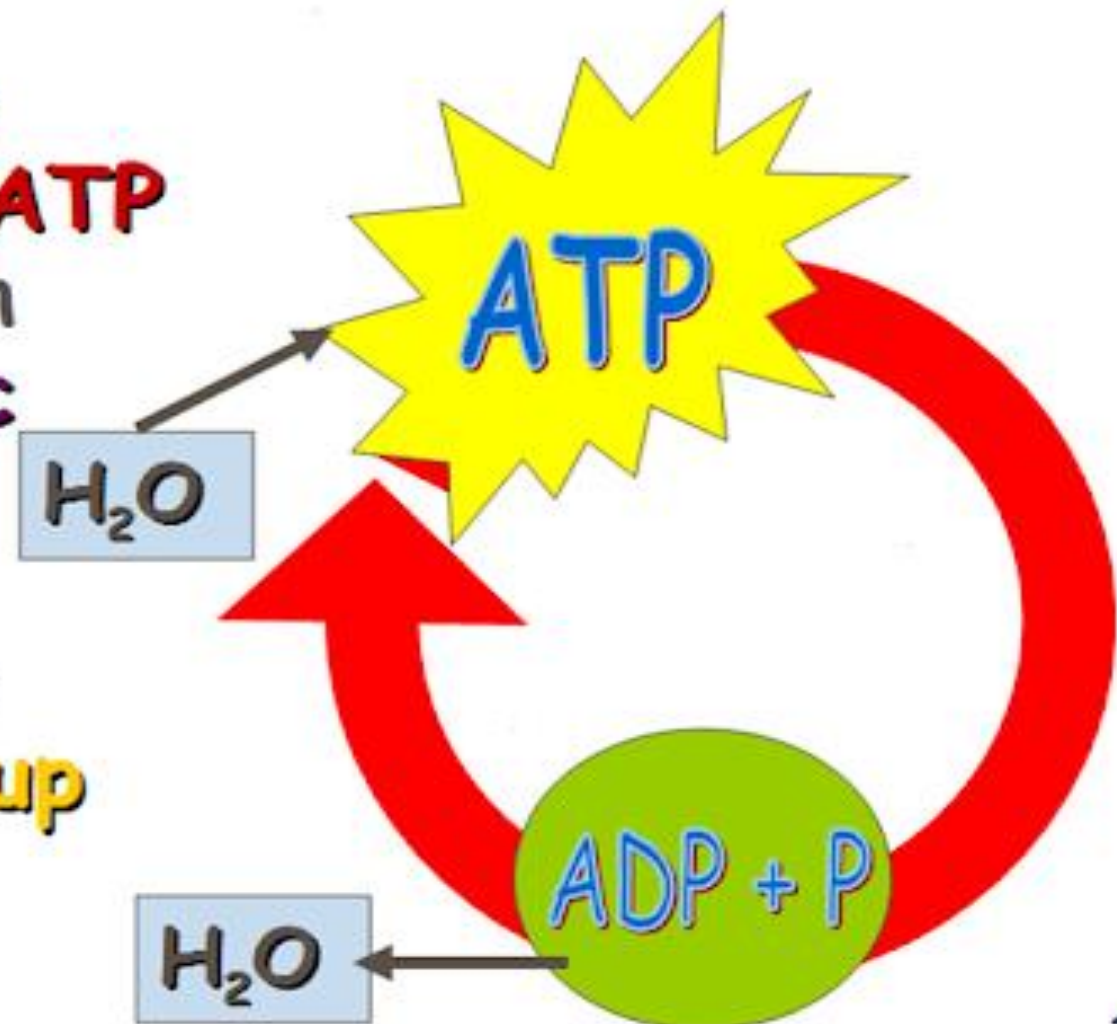


Energy

ATP

Coupled Reaction - ATP

- The **exergonic hydrolysis** of **ATP** is coupled with the **endergonic dehydration** process by **transferring a phosphate group** to another molecule.

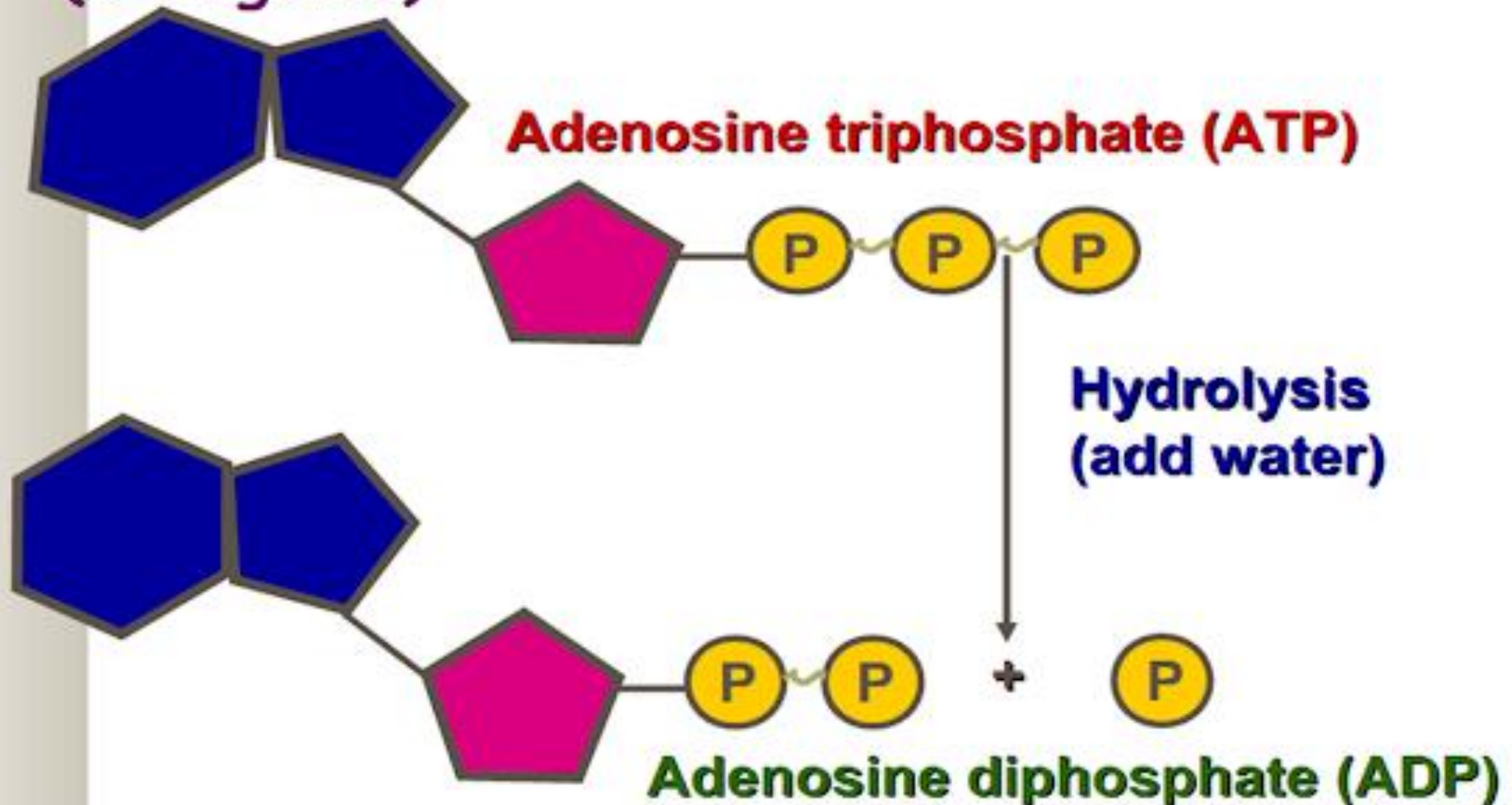


Hydrolysis of ATP

Energy
Used
by
Cells



(exergonic)

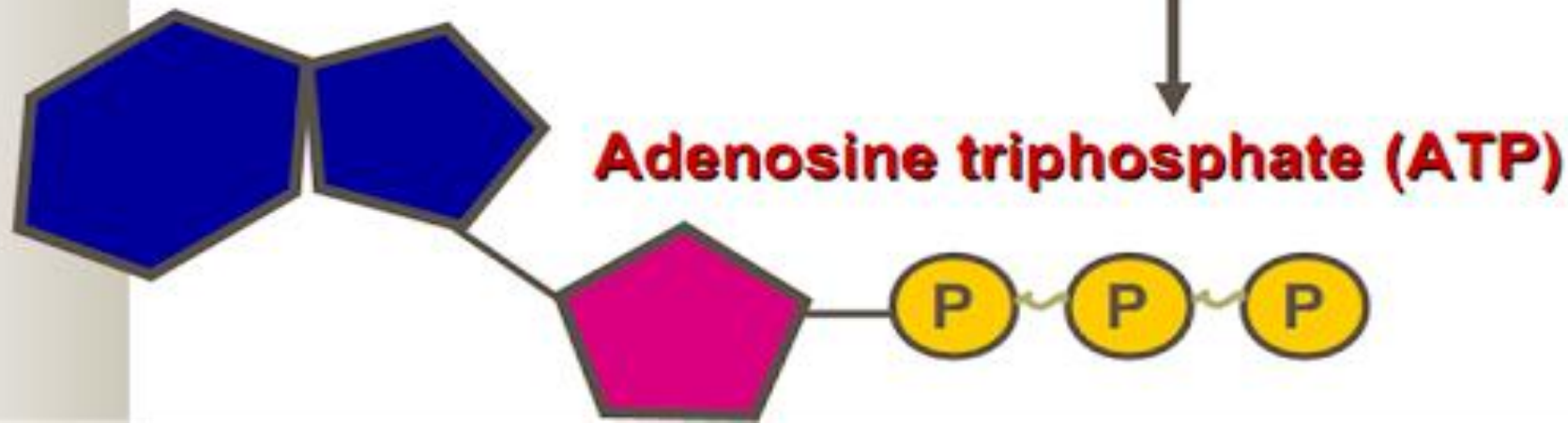
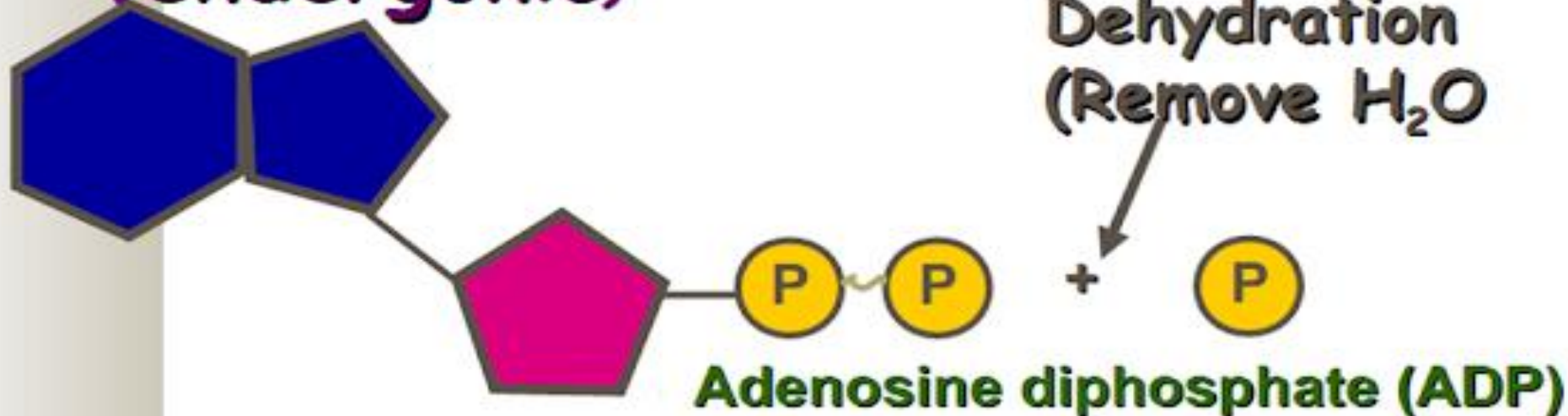


Dehydration of **ATP**



(endergonic)

Dehydration
(Remove H_2O)



Redox Potential:

Capacity of a molecule to accept or donate electrons.

Redox potentials provide a quantitative measure of the oxidizing or reducing power of a molecule.

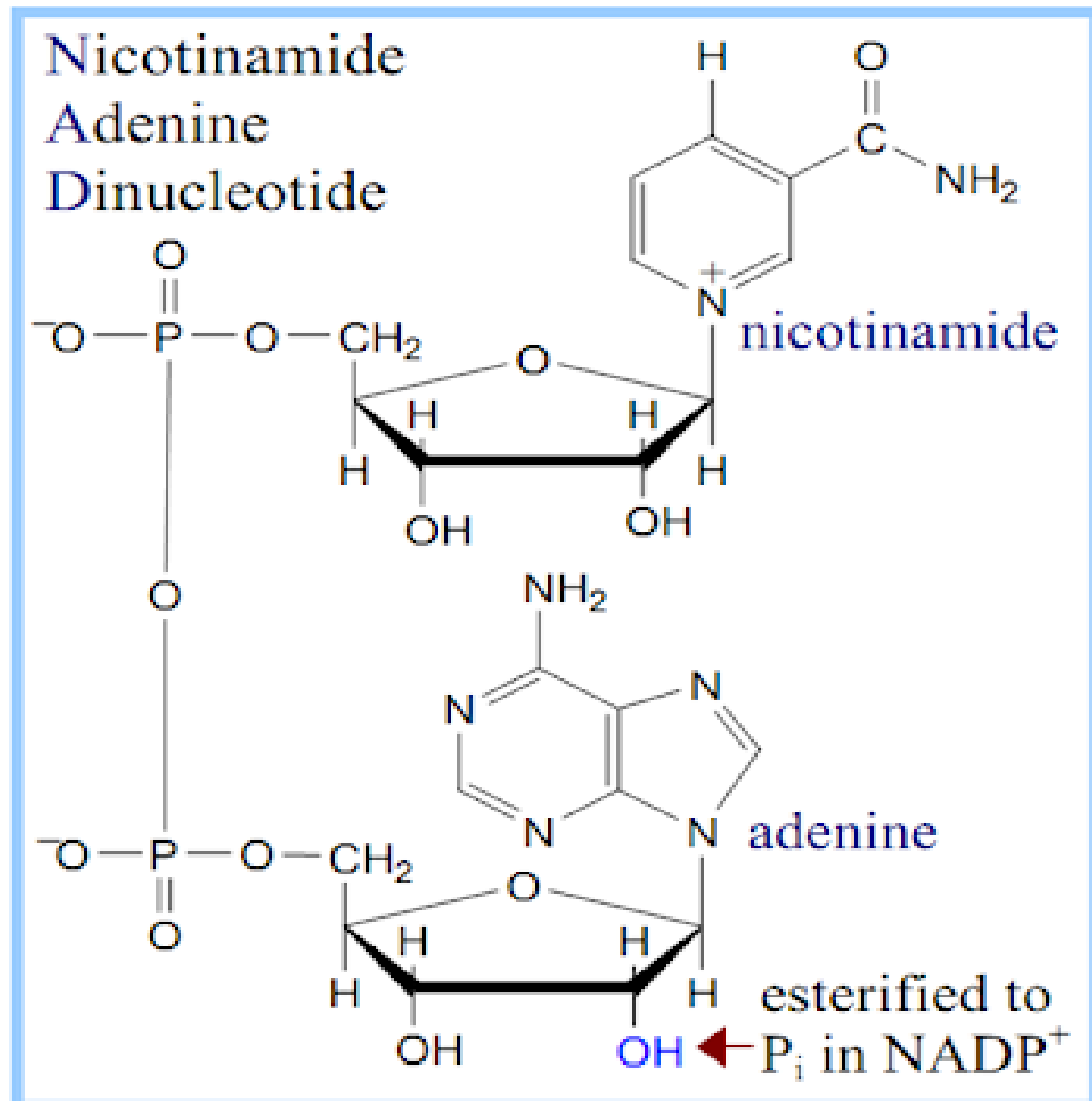
The effective redox potential depends on the proportion of the oxidized and reduced forms.

NADPH/NADP couple (maintained by cells at almost 100% in the reduced form) is much better biological reducing agent than NADH/NAD (no more than 30% reduced under normal conditions) despite the fact that the standard redox potentials are identical.

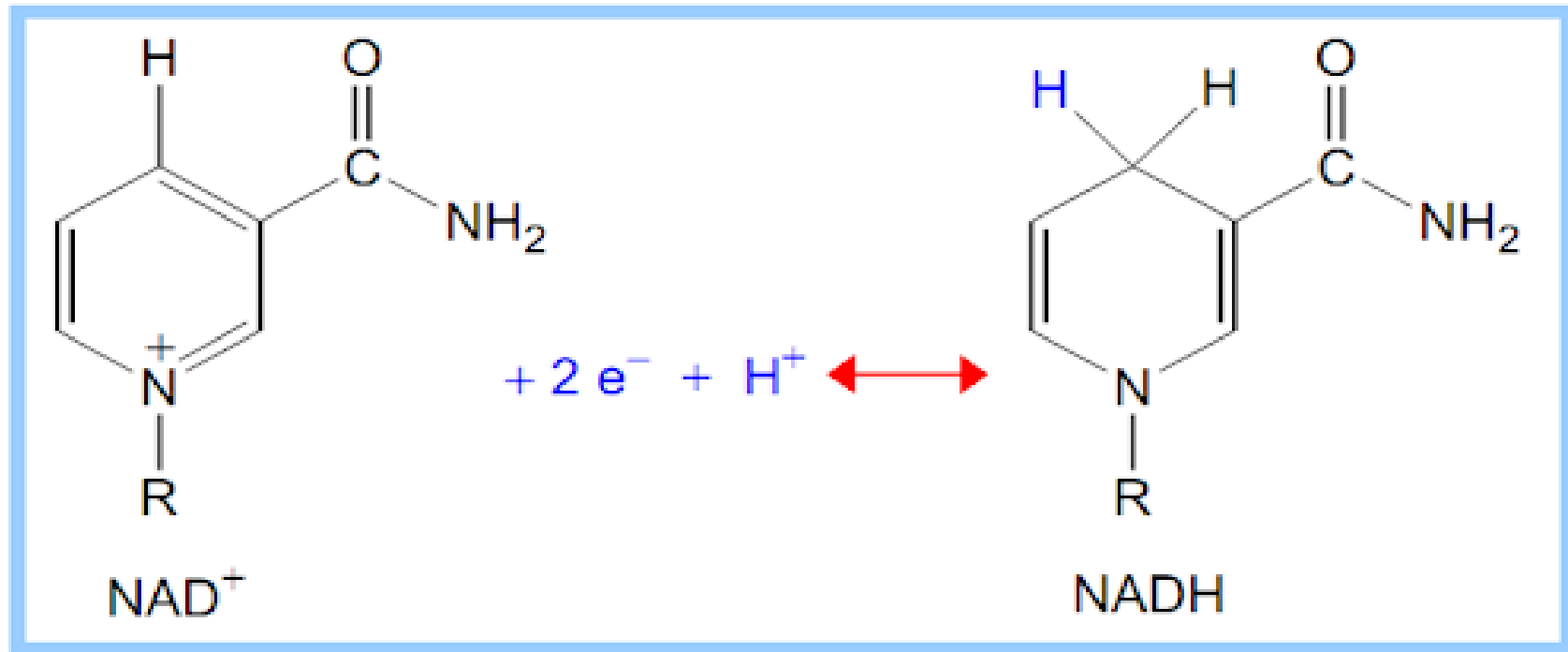
NAD⁺, Nicotinamide Adenine Dinucleotide, is an electron acceptor in catabolic pathways.

The nicotinamide ring, derived from the vitamin **niacin**, accepts 2 e⁻ & 1 H⁺ (a hydride) in going to the reduced state, **NADH**.

NADP⁺/NADPH is similar except for P_i. NADPH is e⁻ donor in synthetic pathways.



NAD⁺/NADH



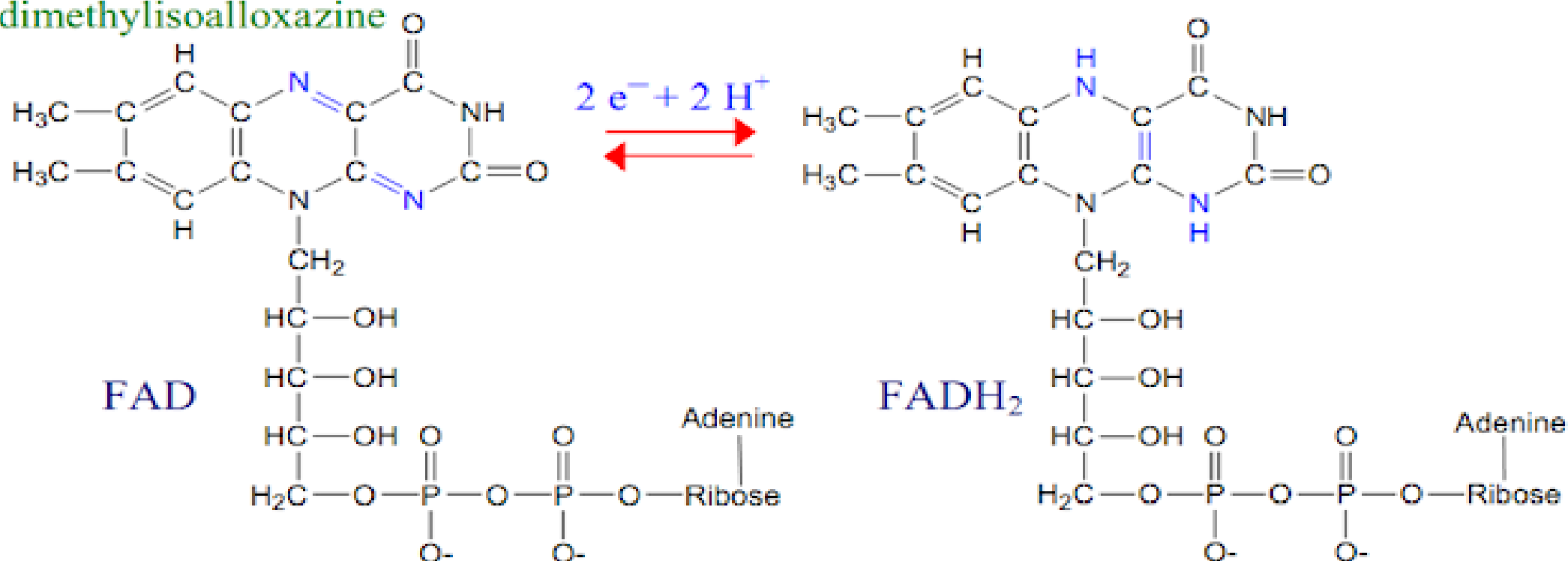
The electron transfer reaction may be summarized as :



It may also be written as:



dimethylisoalloxazine



FAD (**F**lavin **A**denine **D**inucleotide), derived from the vitamin **riboflavin**, functions as an e^- acceptor. The dimethylisoalloxazine ring undergoes reduction/oxidation. FAD accepts $2 e^- + 2 H^+$ in going to its reduced state:



Electron transfer via oxidation-reduction reactions generates biological energy

- When electrons flow from a low affinity carrier (e.g., glucose) to a high affinity carrier (e.g., O_2), an electromotive force (emf) will be generated (with energy released and work done).
- Oxidation of energy-rich biological fuels *often* means dehydrogenation (catalyzed by *dehydrogenases*) from carbons having various oxidation states.

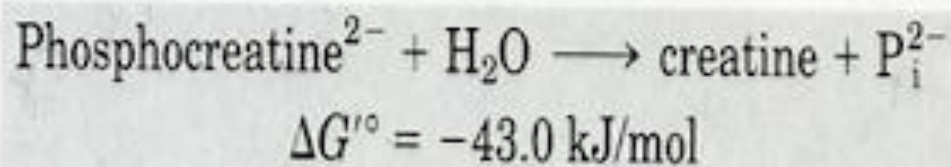
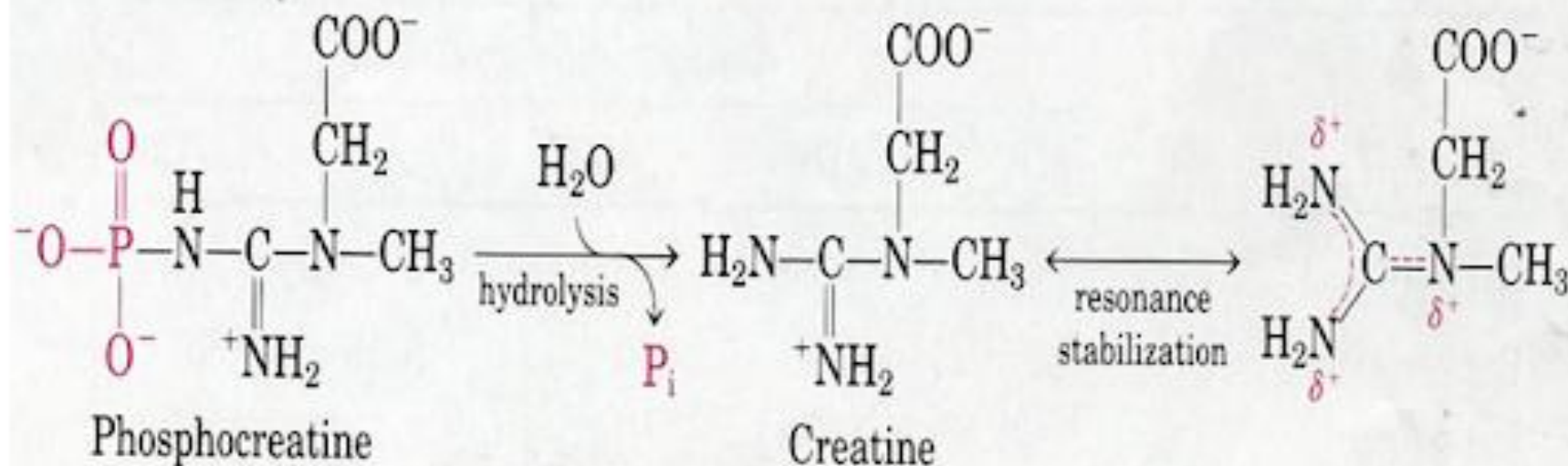
- NAD and NADP can easily diffuse out of the enzymes, but FMN and FAD are tightly bound to the enzymes (thus being called *prosthetic groups*, and the complex proteins being called *flavoproteins*).
- NADH and FADH₂ will be further oxidized via the respiratory chain for ATP production.
- ADP is commonly present in all these universal electron carriers (as well as in Coenzyme A and ATP).

ATP is the universal currency for biological energy

- This was first perceived by Fritz Lipmann and Herman Kalckar in 1941 when studying glycolysis.
- Hydrolysis of the two **phosphoanhydride** bonds in ATP generate *more stable products* releasing large amount of free energy (ΔG° is -30.5 kJ/mol; ΔG_p in cells is -50 to -65 kJ/mol).
- The ATP molecule is **kinetically stable** at pH 7 and enzyme catalysis is needed for its hydrolysis.
- ATP actually exists as **a sum** of various species in cells (e.g., **ATP⁺** **MgATP²⁻**).

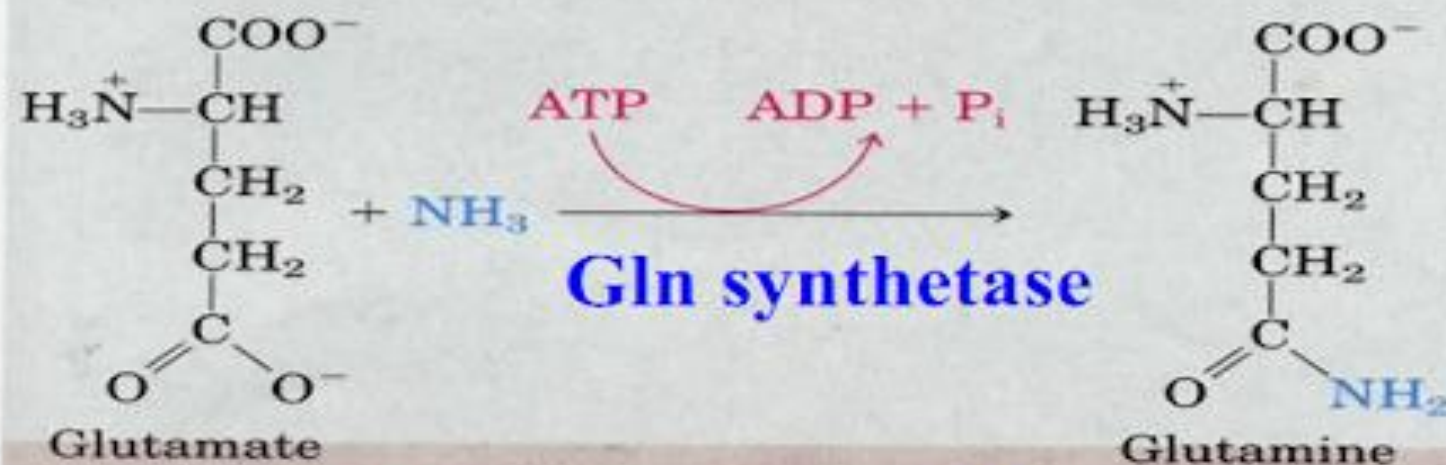
(+ ADP)

(+ ATP)

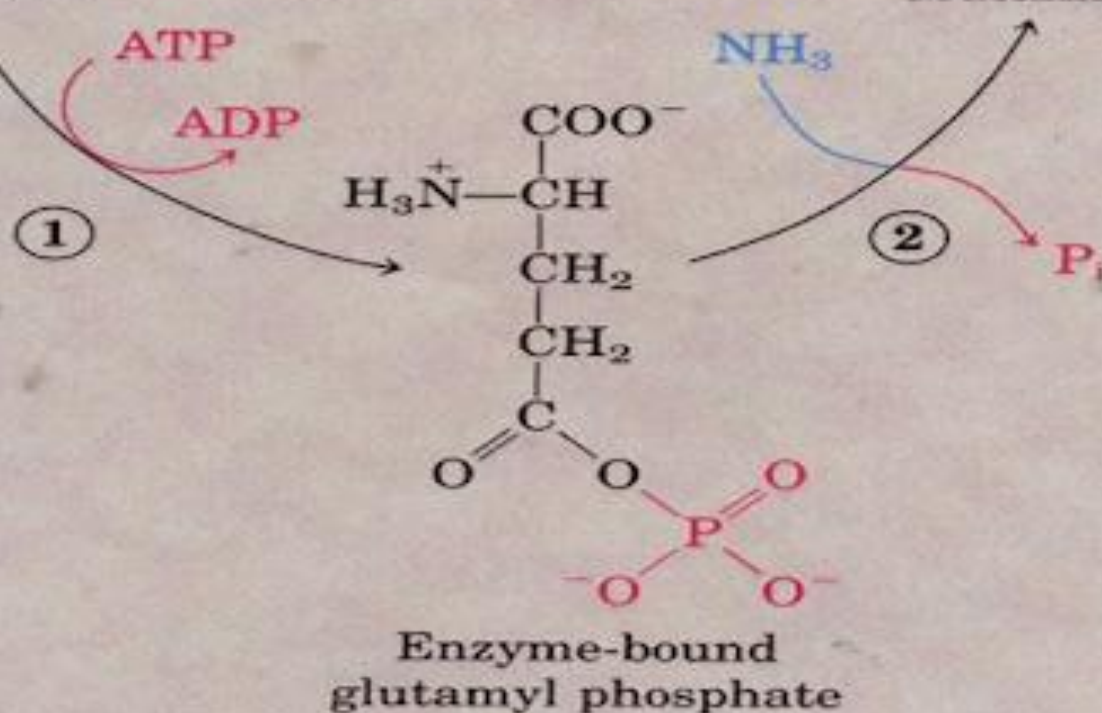


ATP is not a long-term storage form of free energy in living cells, but .

(a) Written as a one-step reaction

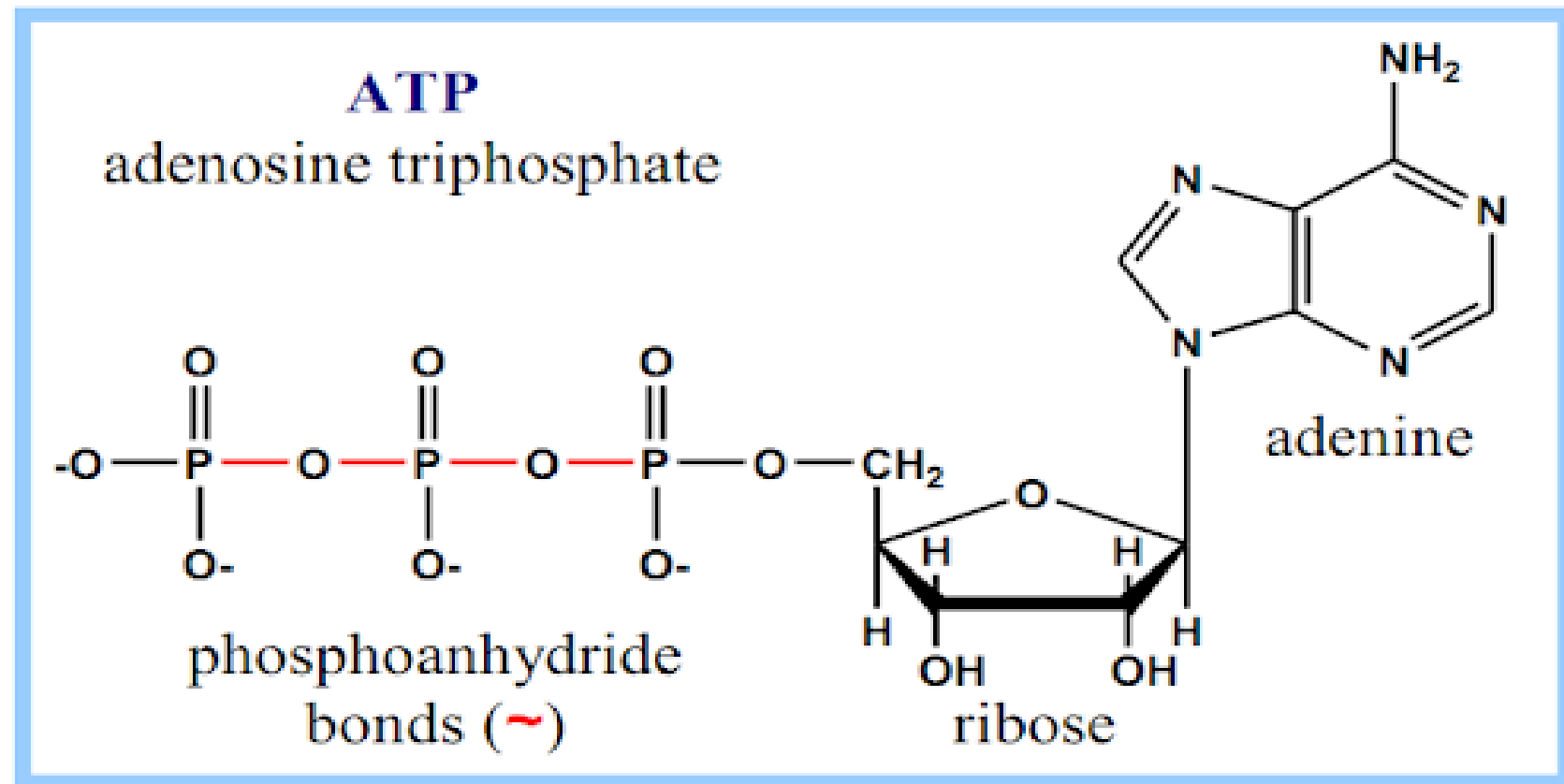


ATP provides
energy usually
through **group**
transfer
(protein could
also be such
acceptors)

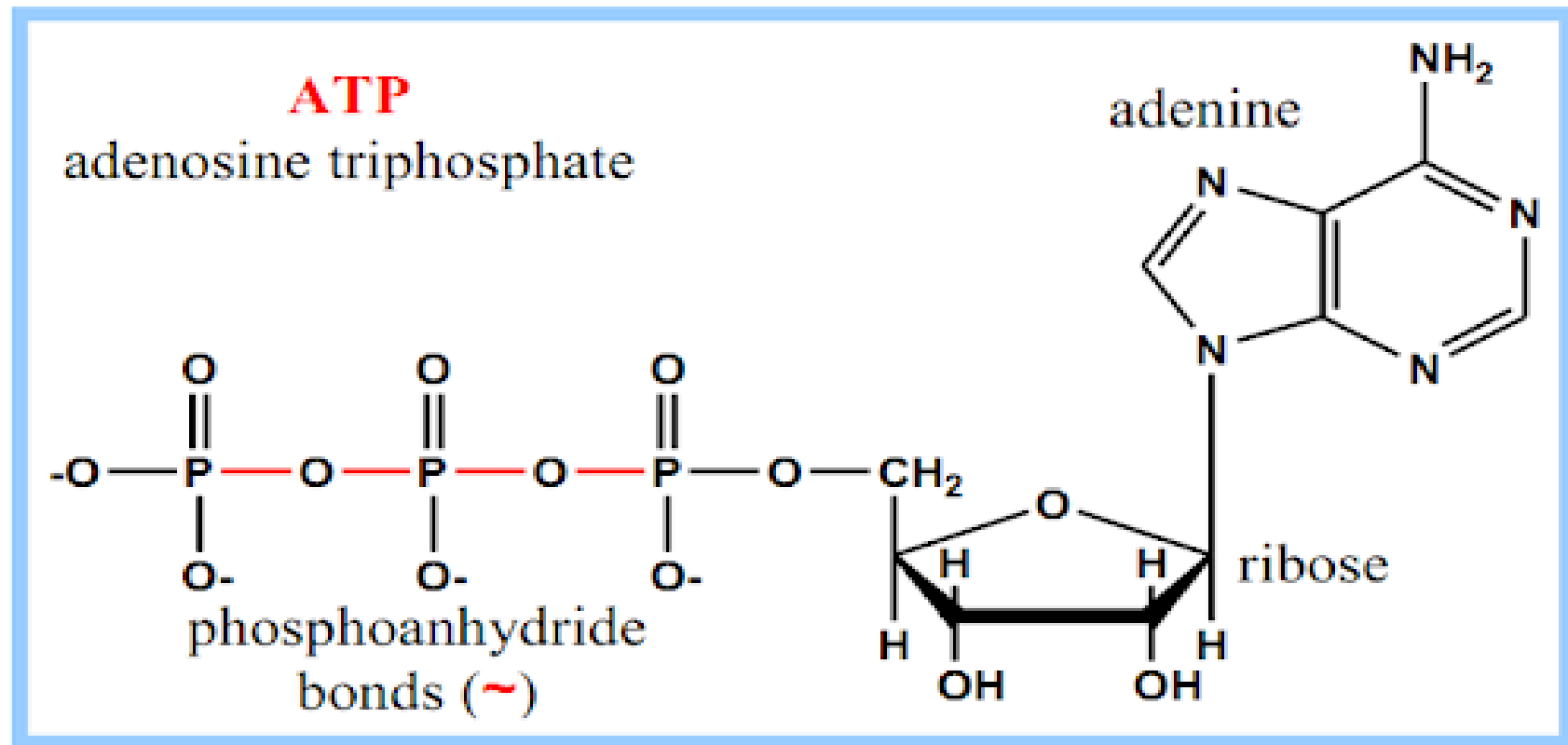


(b) Actual two-step reaction

“High energy” bonds



Phosphoanhydride bonds (formed by splitting out H₂O between 2 phosphoric acids or between carboxylic & phosphoric acids) have a **large negative ΔG of hydrolysis**.



Phosphoanhydride linkages are said to be **"high energy" bonds**. Bond energy is not high, just ΔG of hydrolysis.

"High energy" bonds are represented by the "~" symbol.

~P represents a phosphate group with a large negative ΔG of hydrolysis.

“High energy” bonds

Compounds with “high energy bonds” are said to have **high group transfer potential**.

For example, P_i may be spontaneously cleaved from ATP for transfer to another compound (e.g., to a hydroxyl group on glucose).

Potentially, **2 ~P bonds** can be cleaved, as 2 phosphates are released by hydrolysis from **ATP**.



Alternatively:



- ♦ **ATP** often serves as an **energy source**.

Hydrolytic **cleavage** of one or both of the "high energy" bonds of ATP is **coupled** to an energy-requiring (non-spontaneous) reaction. (Examples presented earlier.)

- ♦ **AMP** functions as an **energy sensor & regulator** of metabolism.

When ATP production does not keep up with needs, a higher portion of a cell's adenine nucleotide pool is AMP.

AMP stimulates metabolic **pathways** that **produce ATP**.

- Some examples of this role involve direct allosteric **activation of pathway enzymes by AMP**.
- Some regulatory effects of AMP are mediated by the enzyme **AMP-Activated Protein Kinase**.

A reaction important for equilibrating ~P among adenine nucleotides within a cell is that catalyzed by **Adenylate Kinase**:



The Adenylate Kinase reaction is also important because the substrate for ATP synthesis, e.g., by mitochondrial ATP Synthase, is **ADP**, while some cellular reactions dephosphorylate ATP all the way to **AMP**.

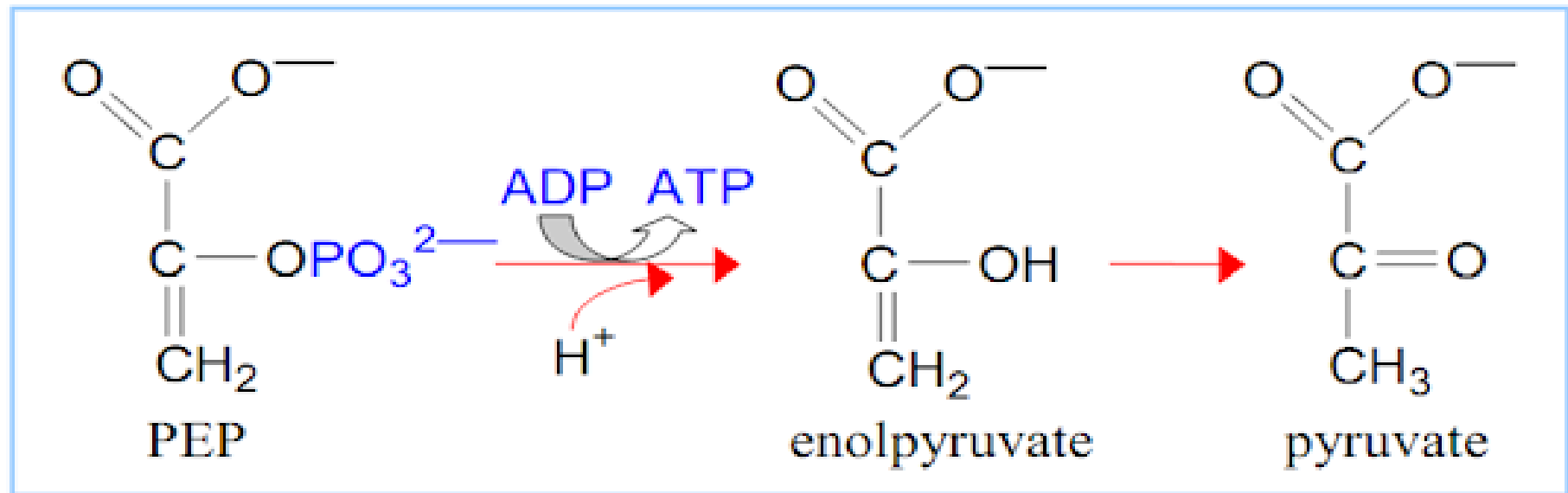
The enzyme **Nucleoside Diphosphate Kinase (NuDiKi)** equilibrates ~P among the various nucleotides that are needed, e.g., for synthesis of DNA & RNA.

NuDiKi catalyzes reversible reactions such as:



Why do phosphoanhydride linkages have a high ΔG of hydrolysis? Contributing factors for ATP & PP_i include:

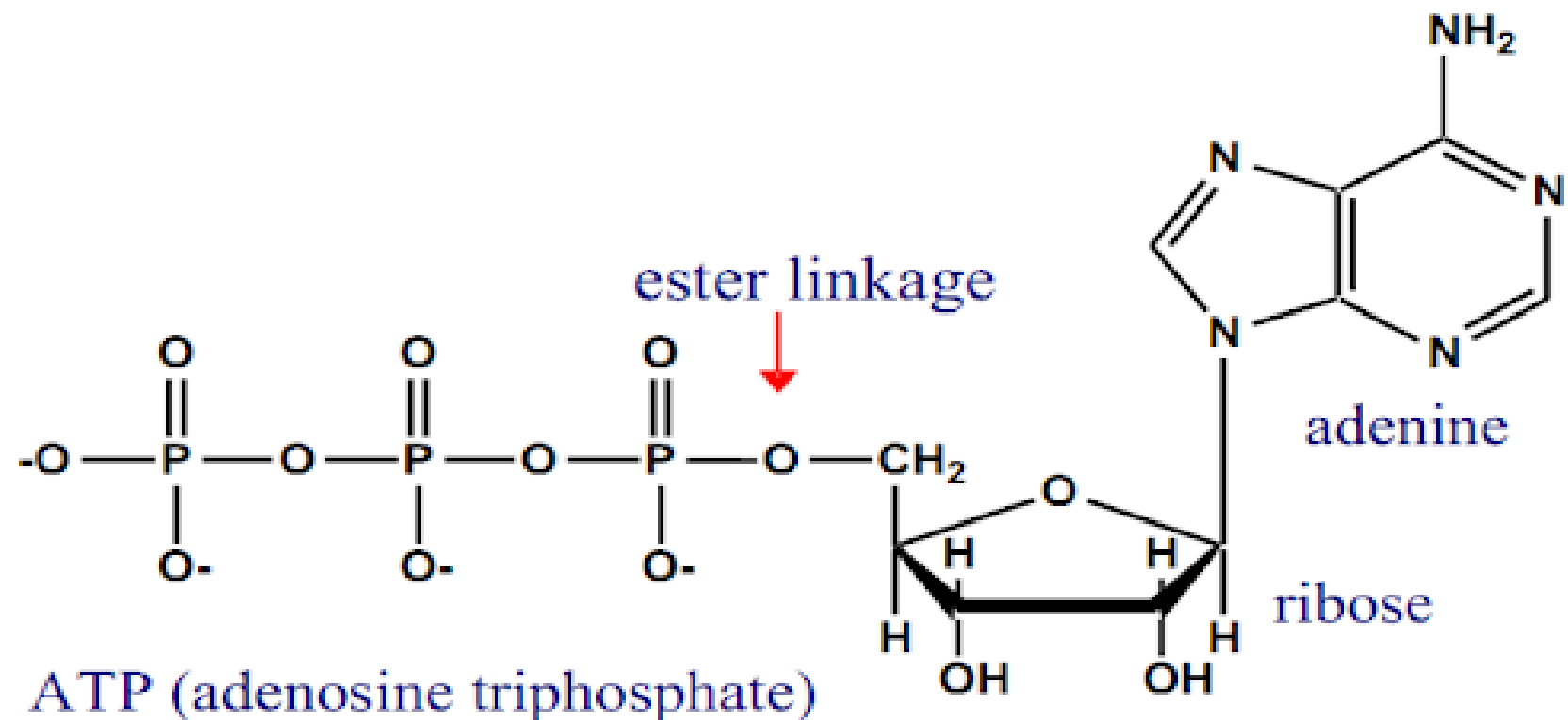
- ♦ **Resonance stabilization** of products of hydrolysis exceeds resonance stabilization of the compound itself.
- ♦ **Electrostatic repulsion** between negatively charged phosphate oxygen atoms favors separation of the phosphates.



Phosphoenolpyruvate (PEP), involved in ATP synthesis in Glycolysis, has a very high ΔG of P_i hydrolysis.

Removal of P_i from ester linkage in PEP is spontaneous because the enol spontaneously converts to a ketone.

The ester linkage in PEP is an exception.



Generally **phosphate esters**, formed by splitting out water between a phosphoric acid and an OH group, have a **low but negative ΔG of hydrolysis**. Examples:

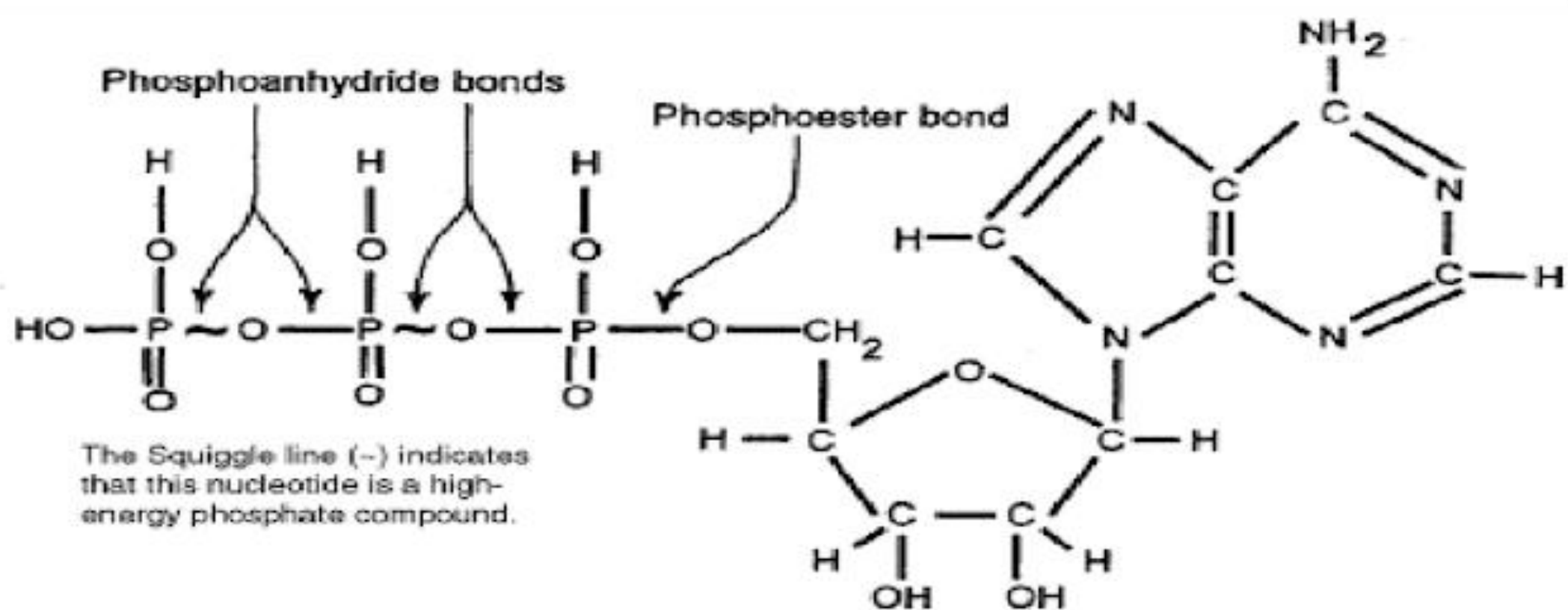
- ♦ the linkage between the first phosphate and the **ribose** hydroxyl of ATP.

ATP has special roles in energy coupling & P_i transfer.

ΔG of phosphate hydrolysis from ATP is **intermediate** among examples below.

ATP can thus act as a P_i donor, & ATP can be synthesized by P_i transfer, e.g., from PEP.

Compound	ΔG° of phosphate hydrolysis, kJ/mol
Phosphoenolpyruvate (PEP)	~~-61.9~~
Phosphocreatine	~~-43.1~~
Pyrophosphate	~~-33.5~~
ATP (to ADP)	~~-30.5~~
Glucose-6-phosphate	~~-13.8~~
Glycerol-3-phosphate	~~-9.2~~



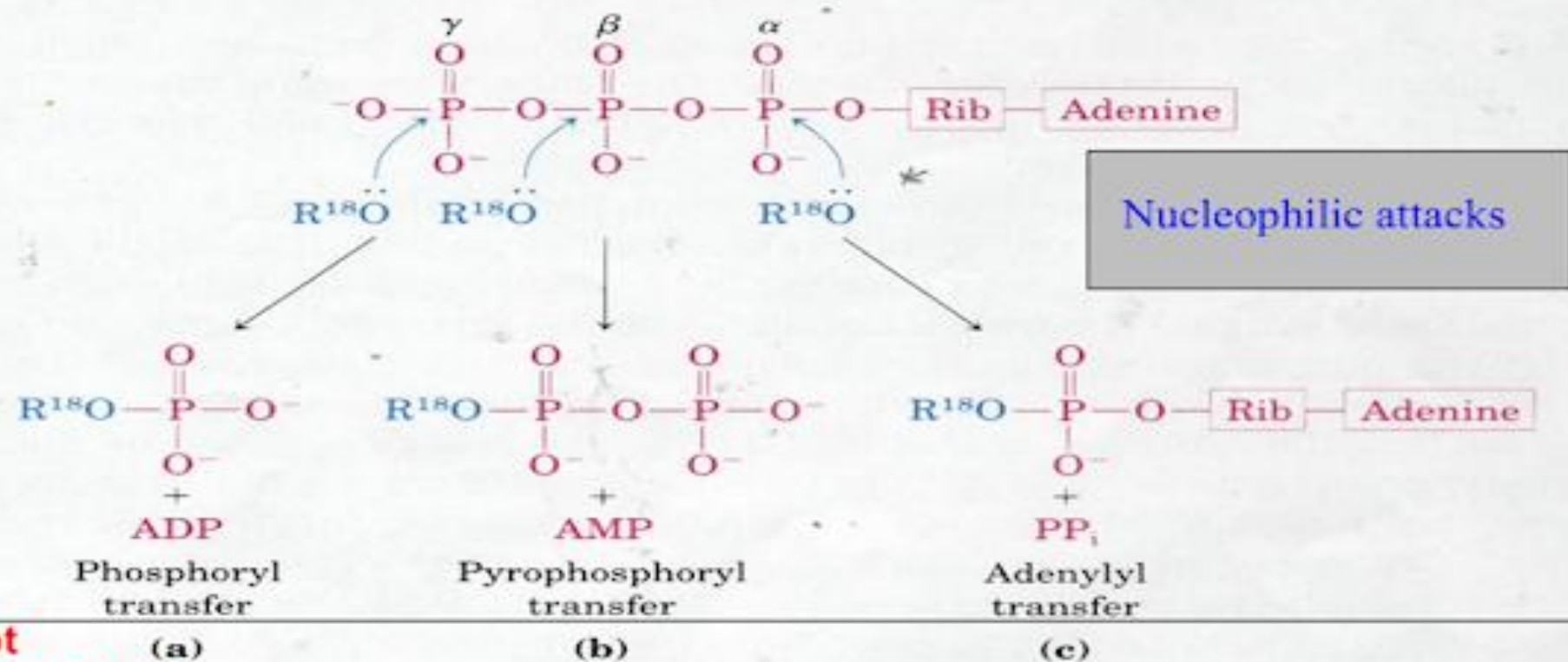
Adenosine (Ribose + Adenine)

Adenosine monophosphate (AMP)

Adenosine diphosphate (ADP)

Adenosine triphosphate (ATP)

Three positions on ATP for attack by the nucleophile $R^{18}O^-$



not
phosphat
e

ATP usually provides energy by group transfer (forming covalent intermediates), not by simple hydrolysis.

List compounds exemplifying the following **roles** of "high energy" bonds:

- ♦ **Energy transfer or storage**

ATP, PP_i, polyphosphate, phosphocreatine

- ♦ **Group transfer**

ATP, Coenzyme A

- ♦ **Transient signal**

cyclic AMP

Kinetics vs Thermodynamics:

A **high activation energy barrier** usually causes hydrolysis of a “high energy” bond to be very **slow** in the absence of an enzyme catalyst.

This **kinetic stability** is **essential** to the role of ATP and other compounds with $\sim$ bonds.

If ATP would rapidly hydrolyze in the absence of a catalyst, it could not serve its important roles in energy metabolism and phosphate transfer.

Phosphate is removed from ATP only when the reaction is coupled via enzyme catalysis to some other reaction useful to the cell, such as transport of an ion, phosphorylation of glucose, or regulation of an enzyme by phosphorylation of a serine residue.

