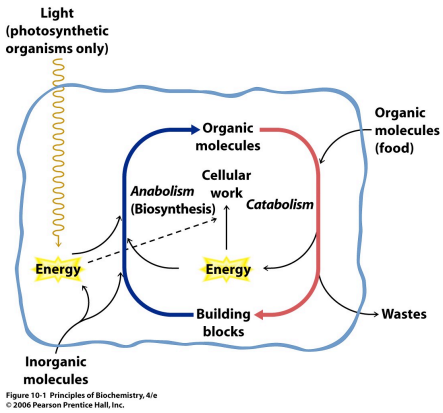
Chapters 7 & 10 Bioenergetics

To live, organisms must obtain **energy** from their environment and use it to do the **work** of building and organizing cell components such as proteins, enzymes, nucleic acids, membranes, organelles ……..



Photosynthetic **autotrophs** use the energy of sunlight to convert low-*G* CO_2 and H_2O into energy-rich complex sugar molecules.

$$6CO_2 + 6H_2O \rightarrow (CH_2O)_6 + 6O_2$$

This reaction has a large positive ΔH and a large negative ΔS.

The products have more enthalpy and are more ordered than the reactants.

Heterotrophs extract the chemical potential energy stored in sugars and other organic compounds and release CO_2 and H_2O.

$$(CH_2O)_6 + 6O_2 \rightarrow 6CO_2 + 6H_2O$$

This reaction has a large negative ΔH and a large positive ΔS. The products have lost energy and are less ordered.

The G is released slowly in many steps using several metabolic pathways.

For any (bio)chemical reaction recall that $\Delta G = -RT \ln(K_{eq})$

where $\Delta G = G_P - G_S$

R = Gas Constant = 8.31 J / mol · K. T = Temp in K.

Under "*standard*" conditions: $\Delta G^o = -RT \ln(K_{eq})$

^{o} refers to 25^o C, 55 M H_2O, [reactant] = 1M

Biochemists prefer pH 7 to pH 0 where $[H^+] = 1M$

$$\Delta G'^o = -RT \ln(K'_{eq})$$

where $'$ refers to pH = 7.

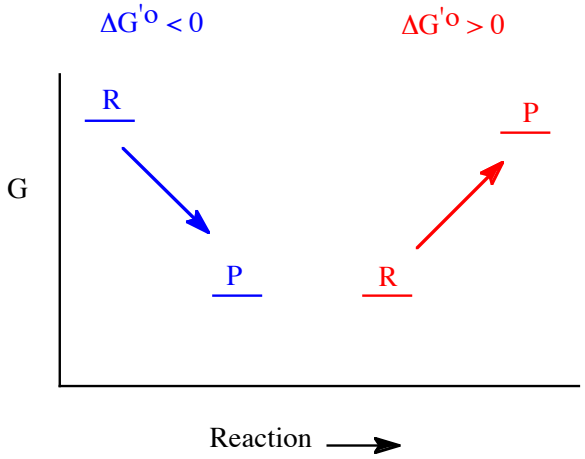
If $K'_{eq} = 19$ at 25^{o}C then, $-RT \ln(K'_{eq}) =$

– (8.315 J/mol K)(298 K)(ln19) = – 7,296 J/mol = – 7.3 kJ/mol

If reactants and products are present at 1M we can predict the direction of the reaction:

K'_{eq}	$\Delta G'^o$	Direction
> 1	negative	Forward
< 1	positive	Reverse
= 1	0	–

When $\Delta G'^{o}$ is **negative**, the products contain less *G* than the reactants.



When $\Delta G'^{o}$ is **positive** the products contain more *G* than the reactants.

For a chemical reaction **at** equilibrium, the rates of the forward and reverse reactions are equal and no net change is occurring but no work can be done and $\Delta G = 0$.

For this reaction, Glc-1-P $\overset{\text{phosphoglucomutase}}{\rightleftharpoons}$ Glc-6-P

$K'_{eq} = \dfrac{[\text{Glc-6-P}]}{[\text{Glc-1-P}]} = 19$ $\quad \Delta G'^{o} = -7.3$ kJ/mol

When [Glc-6-P] = [Glc-1-P] = 1 M, the reaction will spontaneously convert Glc-1-P into Glc-6-P until equilibrium is established.

Glc-1-P ⟶ Glc-6-P

What if we add [Glc-1-P] @ 1 mM to [Glc-6-P] @ 100 mM?

The reaction will convert Glc-6-P into Glc-1-P to re-establish equilibrium.

Glc-1-P ⟵ Glc-6-P

So the ***actual*** ΔG under ***non-standard*** conditions is positive.

The ***actual*** ΔG under **non**-standard conditions for:

$$A + B \rightleftharpoons C + D$$

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

where the concentrations are "*actual*".

Notes:

1. Both ΔG and $\Delta G'^{o}$ are ***theoretical*** maxima. Some G is always lost as heat.

2. Even if $\Delta G'^{o}$ is positive, the reaction can go forward if ΔG is negative. *i.e.* if the second term is negative and bigger than $\Delta G'^{o}$.

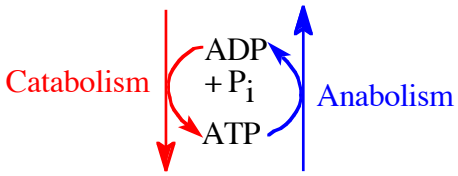
3. ΔG's of **sequential** reactions are additive because ΔG is path-independent. For example,

		$\Delta G'^{o}$
Glc-1-P + H_2O	⇌ Glc + P_i	-21.06 kJ
Glc + P_i	⇌ Glc-6-P + H_2O	+13.76 kJ
Glc-1-P	⇌ Glc-6-P	-7.3 kJ

The *G* released in 1 reaction can be used to drive a 2nd reaction as long as the two share a **common intermediate**. *E.g.* **Glc**

Nearly all of the *G* stored in carbohydrates, lipids, and AA is **temporarily** stored in **ATP**, the cell's "***energy currency***", for use in building cell constituents and doing cellular work such as muscle contraction and active transport of molecules against concentration gradients.

So ATP is the **common intermediate** between **catabolism** and **anabolism**. ATP is **not** an energy storage molecule.



The **synthesis** of ATP is a highly endergonic reaction and the **hydrolysis** of ATP is a highly exergonic reaction.

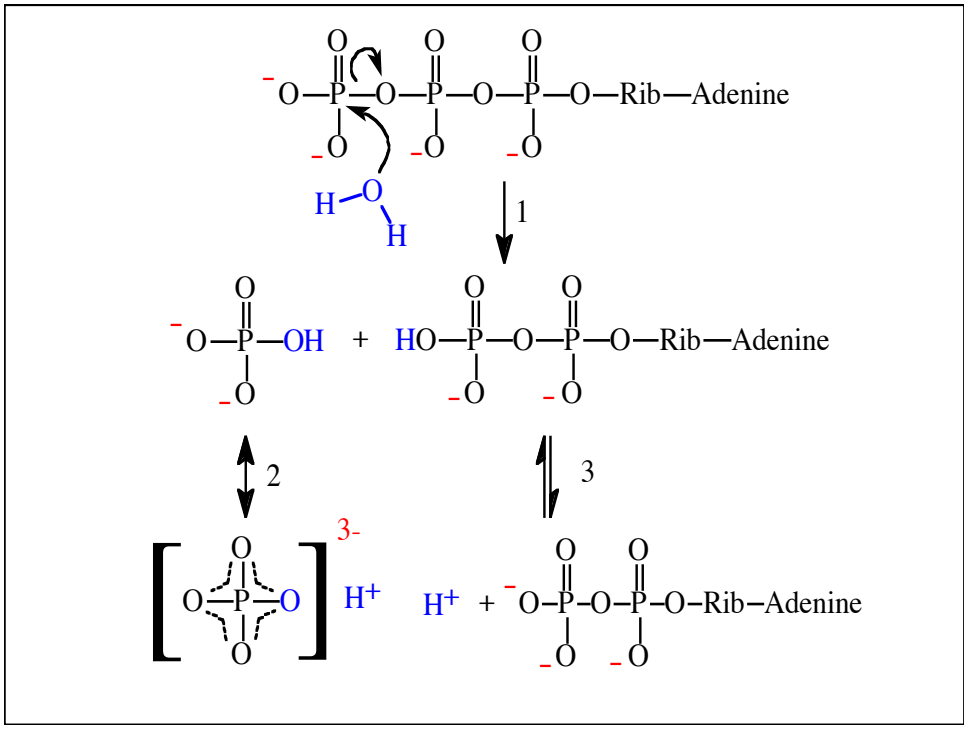
$$ATP + H_2O \rightleftharpoons ADP + P_i$$

$$\Delta G'^{o} = -30 \text{ kJ / mole}$$

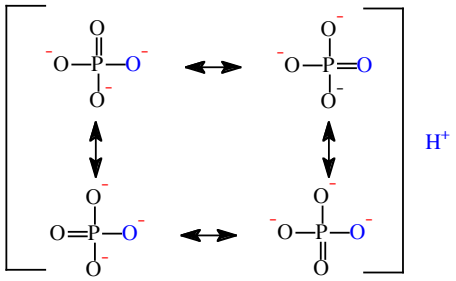
How does ATP store **chemical potential energy** or why does hydrolysis release this energy? There are 4 parts to the answer:

1. Relief of charge repulsion.

One molecule with 4 negative charges is converted into 2 molecules with 2 negative charges each.



2. There are more resonance forms of ADP + P_i than of ATP, so there is an **entropy increase** due to hydrolysis.



3. At the usual pH of cells, ADP ionizes and Le-Chatelier's Principle pulls the reaction forward.

4. Solvation of ADP + P_i > the solvation of ATP which lowers the *G* of the products relative to the reactants.

Notes:

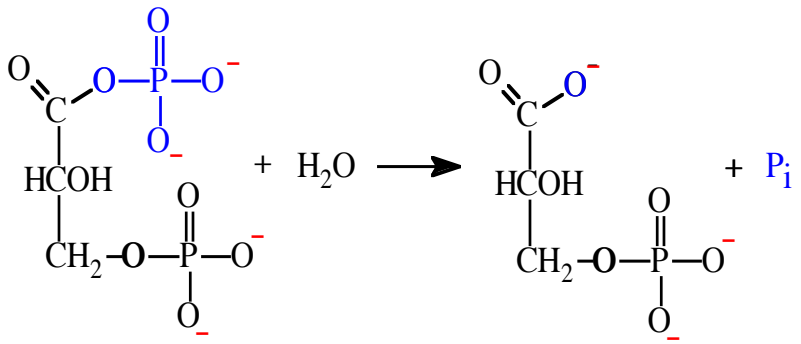
1. In cells, Mg ATP^{2-} and Mg ADP^{-} are present. Hydrolysis of Mg ATP^{2-} has a different $\Delta G'^{o}$ than ATP^{4-}.

2. In cells, [ATP] ~ 2.25 mM; [ADP] ~ 0.25 mM; $[P_i]$ ~ 1.65 mM

These concentrations are far from standard. So "*actual*" ΔG = –50 to –65 kJ / mole.

Other compounds have large ΔG of hydrolysis for reasons similar to ATP:

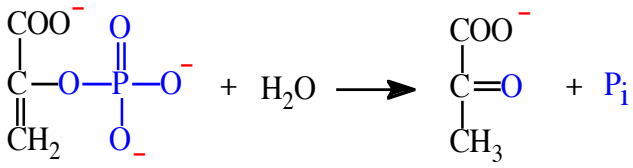
1. 1,3 bis-phosphoglycerate



1,3 bis-phosphoglycerate + H_2O → 3-phosphoglycerate + P_i

$\Delta G'^{o}$ = -49 kJ/mol

2.



Phosphoenolpyruvate Pyruvate

$\Delta G'^{o}$ = -62 kJ/mol

There is more than enough *G* released in the above two reactions to form ATP from ADP + P_i.

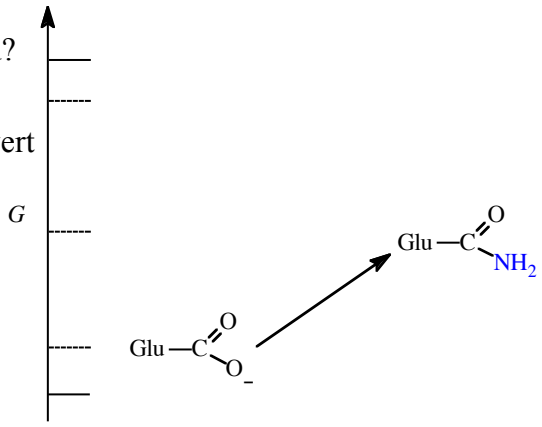
1. So cells use high-G compounds to make ATP. This is called: "**Substrate Level Phosphorylation**".

Two other processes for regenerating ATP are:

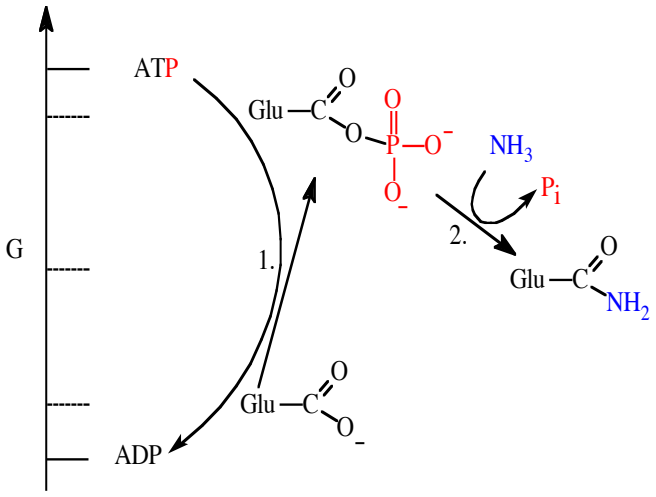
2. **Photophosphorylation** in chloroplasts.

3. **Oxidative phosphorylation** in mitochondria.

How are reactions coupled? For *e.g.* how can ATP hydrolysis be used to convert Glu + NH_3 → Gln, an endergonic process?

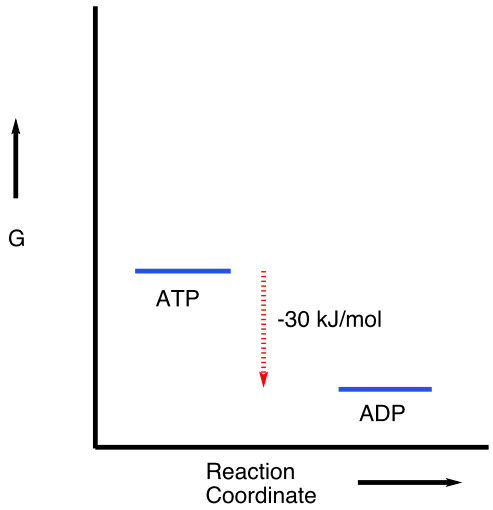


1. The phosphate from ATP is transferred to an enzyme or to a substrate which becomes **activated**. In this case, the *G* of Glu is raised by phosphorylation.

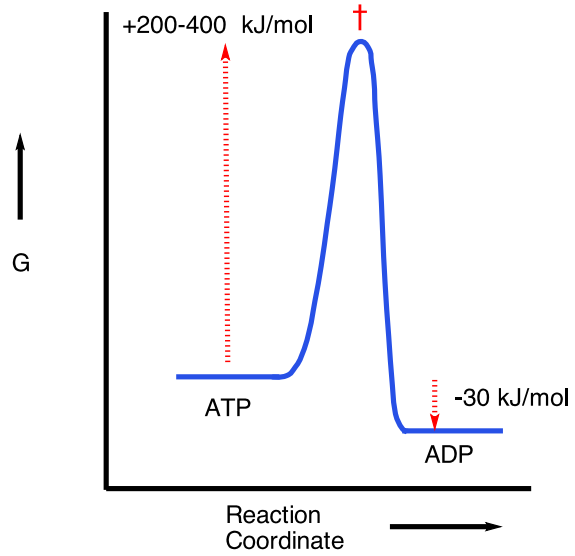


2. The reaction is completed by displacement of the P_i by NH_3^+, which is a "*downhill*" *G*-releasing reaction.

If ATP has so much *G*, why does it not spontaneously decompose?



The reaction is spontaneous, but very slow because it has a very high **activation energy** $\Delta G^{\dagger}$.



So ***enzymes*** lower the activation energy and allow the *G* release.

Other nucleoside triphosphates are energetically equivalent to ATP and are also used by cells. *e.g.* GTP

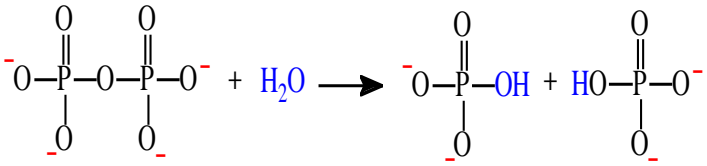
These are usually made by the following reactions catalysed by ***nucleoside diphosphate kinases***:

$$ATP + NDP \rightleftharpoons ADP + NTP \quad \Delta G'^{o} = 0$$

The following reactions release about the same amount of G as ATP hydrolysis and also can be used as energy "*currency*":

$ADP + H_2O \rightarrow AMP + P_i$ $\quad\quad$ $ATP + H_2O \rightarrow AMP + PP_i$

$\Delta G'^{o} \sim -33$ kJ/mole $\quad\quad$ $\Delta G'^{o} \sim -33$ kJ/mole

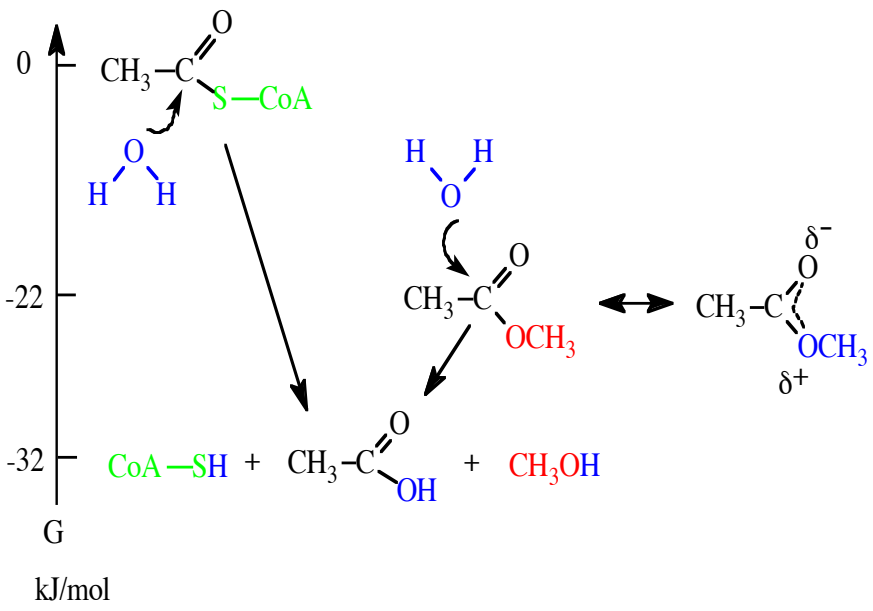


PP_i is inorganic pyrophosphate. $\Delta G'^{o} \sim -29$ kJ/mole

Another type of energy storage is provided by **thioester bonds.**

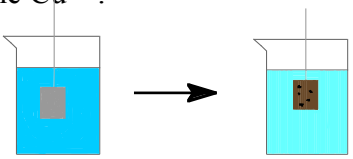
By comparison, oxygen esters are resonance stabilized, and thus store less *G* than thioesters.

E.g. Coenzyme A



Electron flow can also do work.

1. Cu^{2+} has a higher electron affinity than Zn^{2+}, so when Zn is placed in a copper sulphate solution, electrons flow spontaneously from the Zn to the Cu^{2+}:



$$Zn \longrightarrow Zn^{2+} + 2\,e^{-1}$$

$$Cu^{2+} + 2\,e^{-1} \longrightarrow Cu$$

The Zn metal erodes and Zn^{2+} goes into the solution.

Cu deposits on the Zn. $Cu^{2+}SO_4^{2-}$ is blue.

As Cu^{2+} is converted into Cu, the blue colour of the solution fades.

The zinc is "**oxidized**" (loses electrons) and the copper is "**reduced**" (gains electrons). Overall, the Redox reaction is:

$$Cu^{+2} + Zn \rightleftharpoons Cu + Zn^{+2}$$

By separating the two half reactions, and permitting the electrons to flow through a conductor, the *G* released can be used to do work.

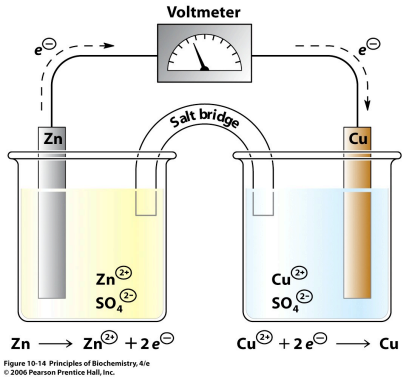


Figure 10-14 Principles of Biochemistry, 4/e
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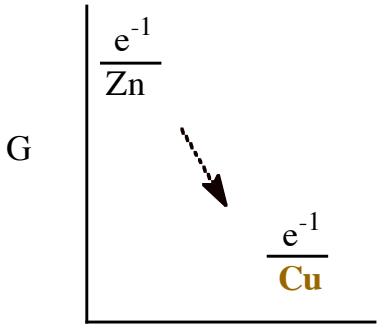
The electrons fall down the *G* hill from Zn to **Cu**.



2. A second type of electron flow involves O_2.

$$C_6H_{12}O_6 + O_2 \longrightarrow CO_2 + H_2O + 2{,}840\ kJ/mole$$

O_2 has a higher electron affinity than glucose, so electrons flow from glucose to O_2 releasing *G*.

The C is oxidized and the O is reduced.

In O_2, bonding e^{-1} are shared equally by the 2 O.

In CO_2 and H_2O the electronegative O pulls e^{-1} away from C and H.

In CO_2, the C's have lost a share of the e^{-1} they had in glucose.

In living cells, this oxidation is a multi-step process involving **glycolysis**, **tricarboxylic acid cycle**, and mitochondrial **respiration**.

At various points, e^{-1} are transferred to electron carriers, and then to O_2.

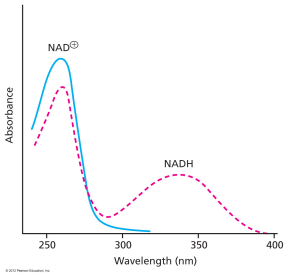
The electron flow is used to do the work of making ATP.

1. A water-soluble, diffusible electron carrier is: NAD^+
Nicotinamide **A**denine **D**inucleotide.

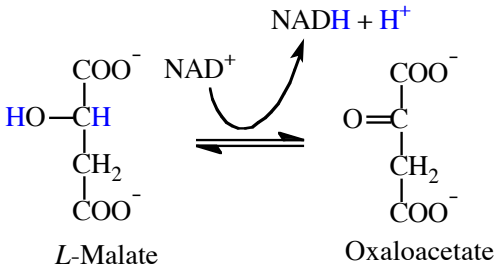
Nicotinamide is also known as Niacin and Vitamin B6.

NAD^+ = Adenine-1'-Ribose-5'-P-P-5'-Ribose-1'-Nicotinamide

Electrons are transferred in the form of **H^-**,
a **hydride** ion.

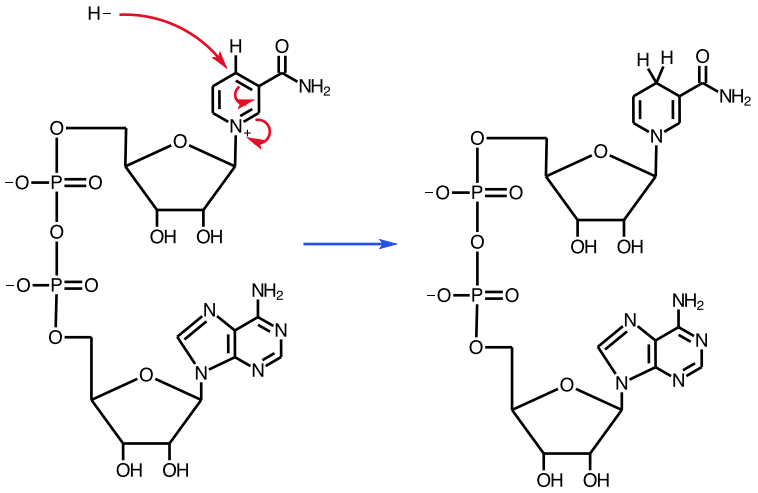


For e.g., the enzyme malate dehydrogenase removes electrons from *L*-malate producing oxaloacetate and reducing NAD^+ to NADH.



Malate is **dehydrogenated** and **oxidized**. It loses 2 electrons in the form of **2H**.

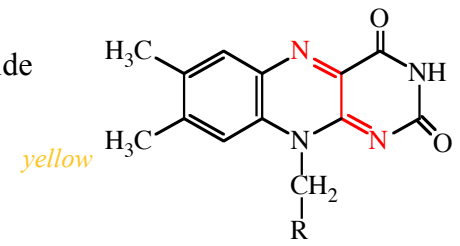
NAD^+ accepts **H^-** and is **reduced**. A **proton** is released.



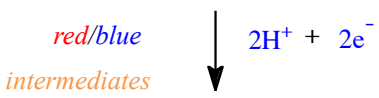
Over 200 **dehydrogenase** enzymes are known that use NAD^+.

2. Two water-soluble, covalently-bound electron carriers are:

FMN, Flavine Mononucleotide and FAD, Flavine Adenine Dinucleotide



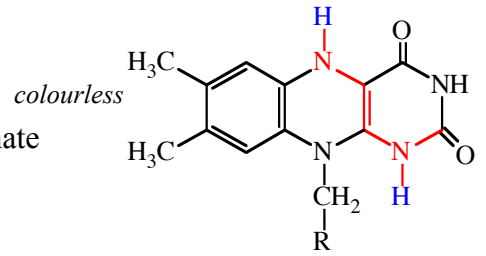
$FAD + 2H \rightarrow FADH_2$



Here, 1 or 2 H are transferre

Flavin + Ribitol = Riboflavin = Vit B_2

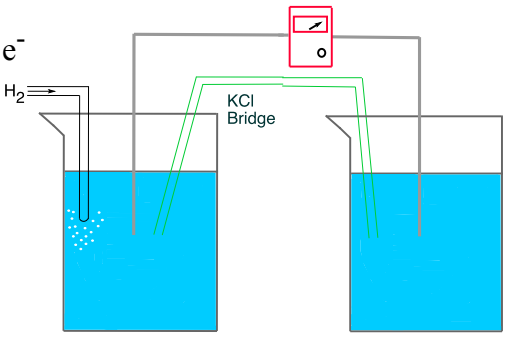
FMN = Riboflavin + phosphate



The relative affinity of an atom or molecule for electrons can be measured.

The **reference** point is a solution containing:
H_2 at 101 kPa + 1 M H^+ (pH = 0)

$$1/2\ H_2 \longrightarrow H^+ + e^-$$

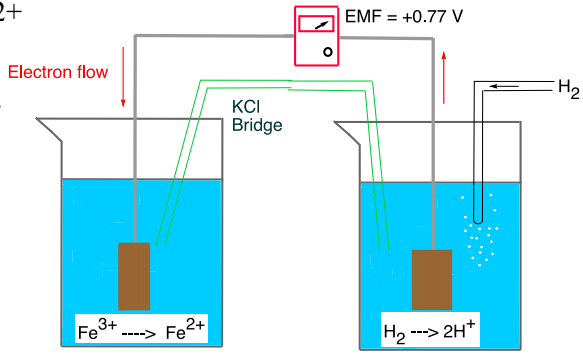


If the reference cell and test cell both contain this solution, no electrons will flow so the electromotive force, emf, will be 0 Volts.

If 1 M Fe^{3+} + Fe^{2+} are in one cell, Fe^{3+} will draw electrons from H_2 and the emf will be +0.77 Volts. This is called the **standard reduction potential, $\mathcal{E}^0$**.

$$Fe^{3+} + e^- \longrightarrow Fe^{2+}$$

$$1/2\ H_2 \longrightarrow H^+ + e^-$$



The net reaction is:

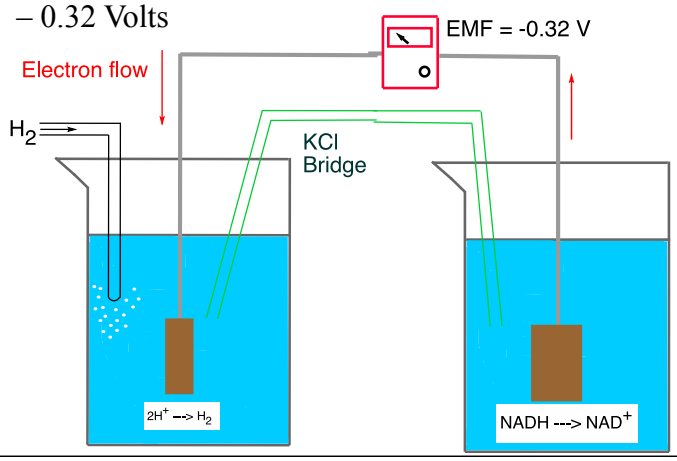
$$1/2\ H_2 + Fe^{3+} \longrightarrow H^+ + Fe^{2+}$$

If the iron is replaced by 1 M NADH + NAD^+, e^- will flow from the test cell to the reference cell. H^+ will draw e^- from NADH:

$NADH \longrightarrow NAD^+ + H^+ + 2e^-$

$2H^+ + 2e^- \longrightarrow H_2$

$\mathcal{E}^0$ = emf = – 0.32 Volts



For biochemists, the test cells are always kept at pH 7 and reported as: $\mathcal{E}'^0$

A Table of $\mathcal{E}'^0$ values, where the half reactions are written as reductions, can be used to predict the directions of Redox reactions.

The more positive is the $\mathcal{E}'^{o}$ (bottom of the Table) the stronger is the oxidizing agent. *i.e.* The reactions go as written in the Table.

In combining pairs of half reactions the more positive $\mathcal{E}'^{o}$ reactions are written as reductions (e^- on the left) and the more negative $\mathcal{E}'^{o}$ are written as oxidations (e^- on the right).

TABLE 10.4 Standard reduction potentials of some important biological half-reactions

Reduction half-reaction	$E^{o\prime}$ (V)
Acetyl CoA + CO_2 + H^+ + $2e^-$ → Pyruvate + CoA	−0.48
Ferredoxin (spinach), Fe^{3+} + e^- → Fe^{2+}	−0.43
$2H^+$ + $2e^-$ → H_2 (at pH 7.0)	−0.42
α-Ketoglutarate + CO_2 + $2H^+$ + $2e^-$ → Isocitrate	−0.38
Lipoyl dehydrogenase (FAD) + $2H^+$ + $2e^-$ → Lipoyl dehydrogenase ($FADH_2$)	−0.34
$NADP^+$ + $2H^+$ + $2e^-$ → NADPH + H^+	−0.32
NAD^+ + $2H^+$ + $2e^-$ → NADH + H^+	−0.32
Lipoic acid + $2H^+$ + $2e^-$ → Dihydrolipoic acid	−0.29
Glutathione (oxidized) + $2H^+$ + $2e^-$ → 2 Glutathione (reduced)	−0.23
FAD + $2H^+$ + $2e^-$ → $FADH_2$	−0.22
FMN + $2H^+$ + $2e^-$ → $FMNH_2$	−0.22
Acetaldehyde + $2H^+$ + $2e^-$ → Ethanol	−0.20
Pyruvate + $2H^+$ + $2e^-$ → Lactate	−0.18
Oxaloacetate + $2H^+$ + $2e^-$ → Malate	−0.17
Cytochrome b_5 (microsomal), Fe^{3+} + e^- → Fe^{2+}	0.02
Fumarate + $2H^+$ + $2e^-$ → Succinate	0.03
Ubiquinone (Q) + $2H^+$ + $2e^-$ → QH_2	0.04
Cytochrome *b* (mitochondrial), Fe^{3+} + e^- → Fe^{2+}	0.08
Cytochrome c_1, Fe^{3+} + e^- → Fe^{2+}	0.22
Cytochrome *c*, Fe^{3+} + e^- → Fe^{2+}	0.23
Cytochrome *a*, Fe^{3+} + e^- → Fe^{2+}	0.29
Cytochrome *f*, Fe^{3+} + e^- → Fe^{2+}	0.36
Plastocyanin, Cu^{2+} + e^- → Cu^+	0.37
NO_3^- + $2H^+$ + $2e^-$ → NO_2^- + H_2O	0.42
Photosystem I (P700)	0.43
Fe^{3+} + e^- → Fe^{2+}	0.77
$\frac{1}{2}O_2$ + $2H^+$ + $2e^-$ → H_2O	0.82
Photosystem II (P680)	1.1

Table 10-4 Principles of Biochemistry, 4/e

e.g. Acetaldehyde / NAD^+ From the Table:

Acetaldehyde + $2H^+$ + $2e^-$ ⟶ Ethanol $\quad \mathcal{E}'^{o} = -0.197$ V

NAD^+ + $2e^-$ + H^+ ⟶ NADH $\quad \mathcal{E}'^{o} = -0.32$ V

So the coupled reaction is:

NADH ⟶ NAD^+ + $2e^-$ + H^+

Acetaldehyde + $2H^+$ + $2e^-$ ⟶ Ethanol

Acetaldehyde + H^+ + NADH ⟶ Ethanol + NAD^+

$\Delta\mathcal{E}'^{o} = -0.197\text{ V} - -0.32\text{ V} = +0.123\text{V}$ (acceptor – donor)

Another way to do this is to reverse the sign of the $\mathcal{E}'^{o}$ value for the reaction that is written as an oxidation, and then add the $\mathcal{E}'^{o}$ values.

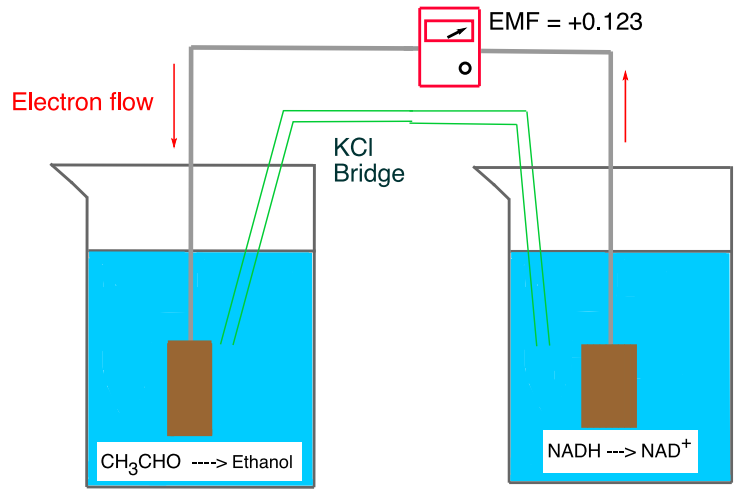
$$\text{NADH} \longrightarrow \text{NAD}^+ + 2e^- + \text{H}^+ \quad \mathcal{E}'^{o} = +0.32$$

$$\text{Acetaldehyde} + 2\text{H}^+ + 2e^- \longrightarrow \text{Ethanol} \quad \mathcal{E}^{o} = -0.197$$

$$\text{Acetaldehyde} + \text{H}^+ + \text{NADH} \longrightarrow \text{Ethanol} + \text{NAD}^+$$

$$\Delta\mathcal{E}'^{o} = +0.32\ \text{V} + (-0.197\ \text{V}) = +0.123\ \text{V}$$

$$\Delta\mathcal{E}'^{o} = +0.32\ \text{V} + (-0.197\ \text{V}) = +0.123\text{V}$$

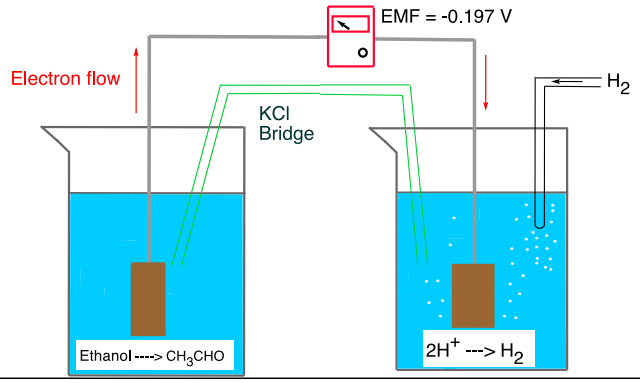


For any pair of 1/2 reactions the ΔG can be calculated from the difference in emf:

$\Delta G'^{o} = -n\mathcal{F}\Delta\mathcal{E}'^{o}$ n = # of moles of e^- transferred

$\mathcal{F}$ = Faraday constant = 96.48 kJ / Volt mole

$\Delta G'^{o} = -2\ (96.5)\ (0.123) = -23.7$ kJ / mole



Under non-standard conditions:

$$\mathcal{E} = \mathcal{E}'^{o} + \frac{RT}{n\mathcal{F}} \ln \frac{[\text{electron acceptor}]}{[\text{electron donor}]}$$

Remember, $\Delta\mathcal{E}'^{o} = \mathcal{E}'^{o}_{\text{acceptor}} - \mathcal{E}'^{o}_{\text{donor}}$