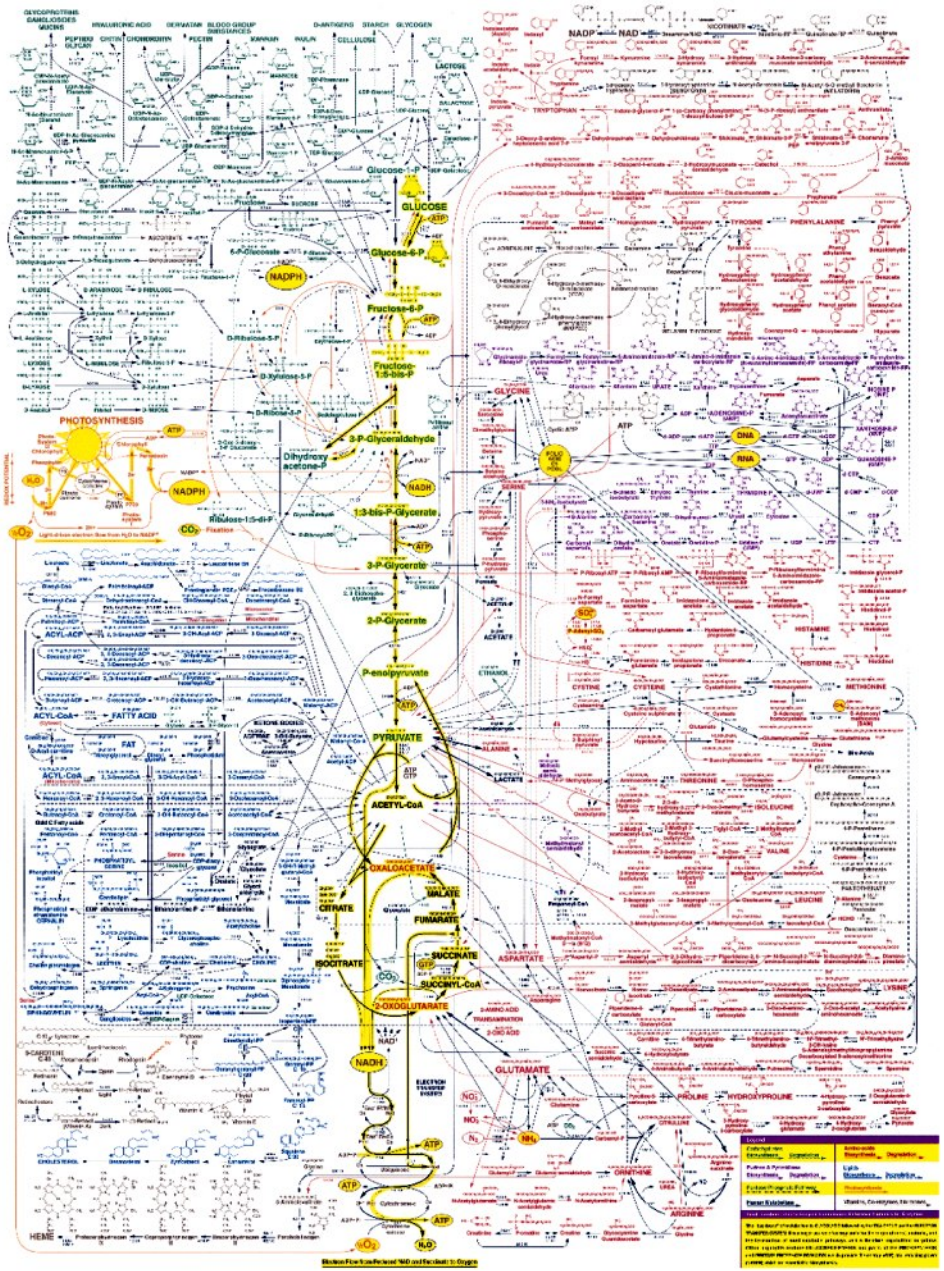
Introduction to Metabolism



Chapter 13 Opener Fundamentals of Biochemistry, 2/e

Trophic strategies: nutritional requirements

Autotrophs
- chemolithotrophs
- photoautotrophs

Heterotrophs

Energy

Obligate aerobes

Anaerobes
- facultative anaerobic
- obligate anaerobic

Electron acceptor (oxidizing agent)

Metabolic pathway

Catabolic & anabolic

Enzymes & metabolites

Roles of ATP and NADP+ in metabolism

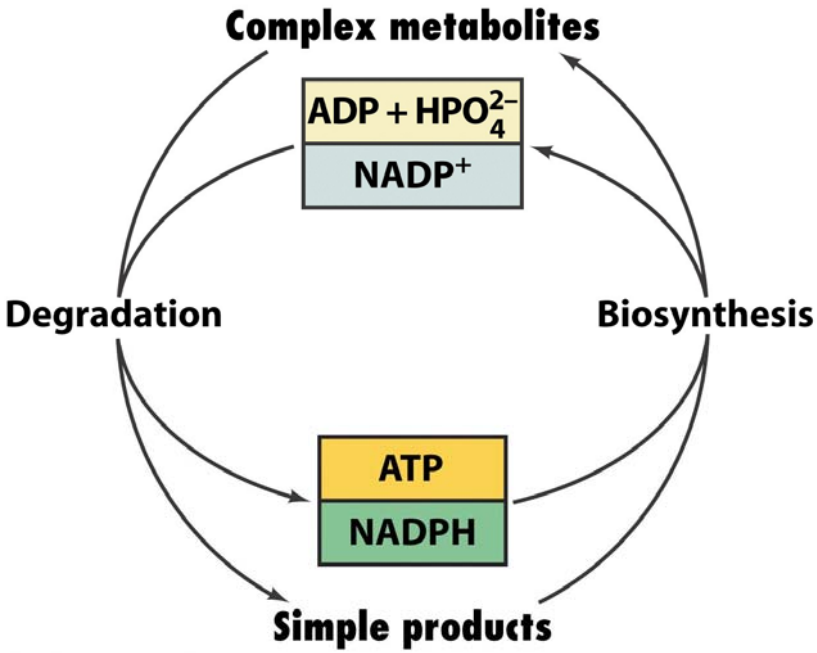


Figure 13-1 Fundamentals of Biochemistry, 2/e
© 2006 John Wiley & Sons

Overview of catabolism

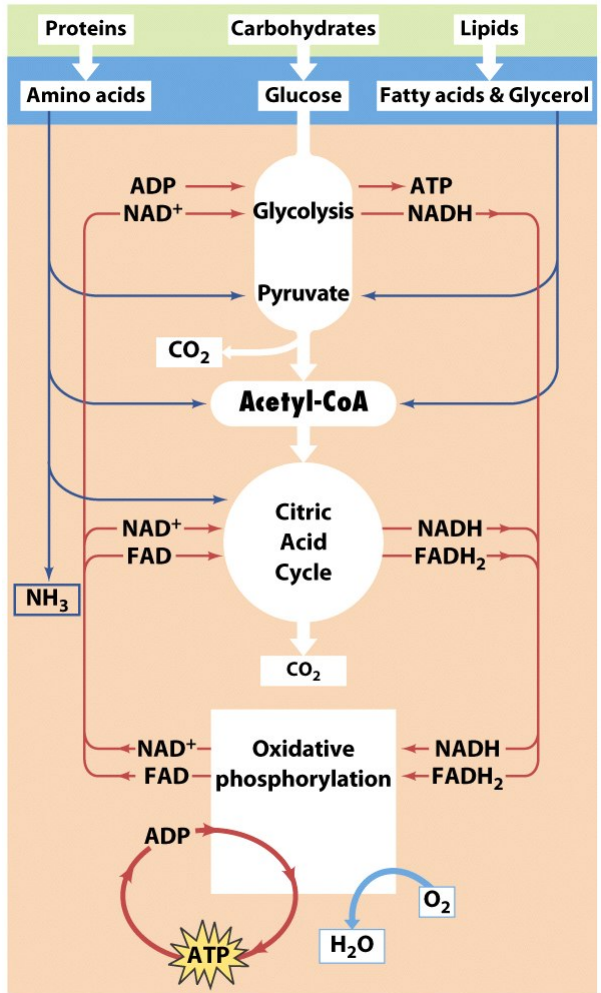
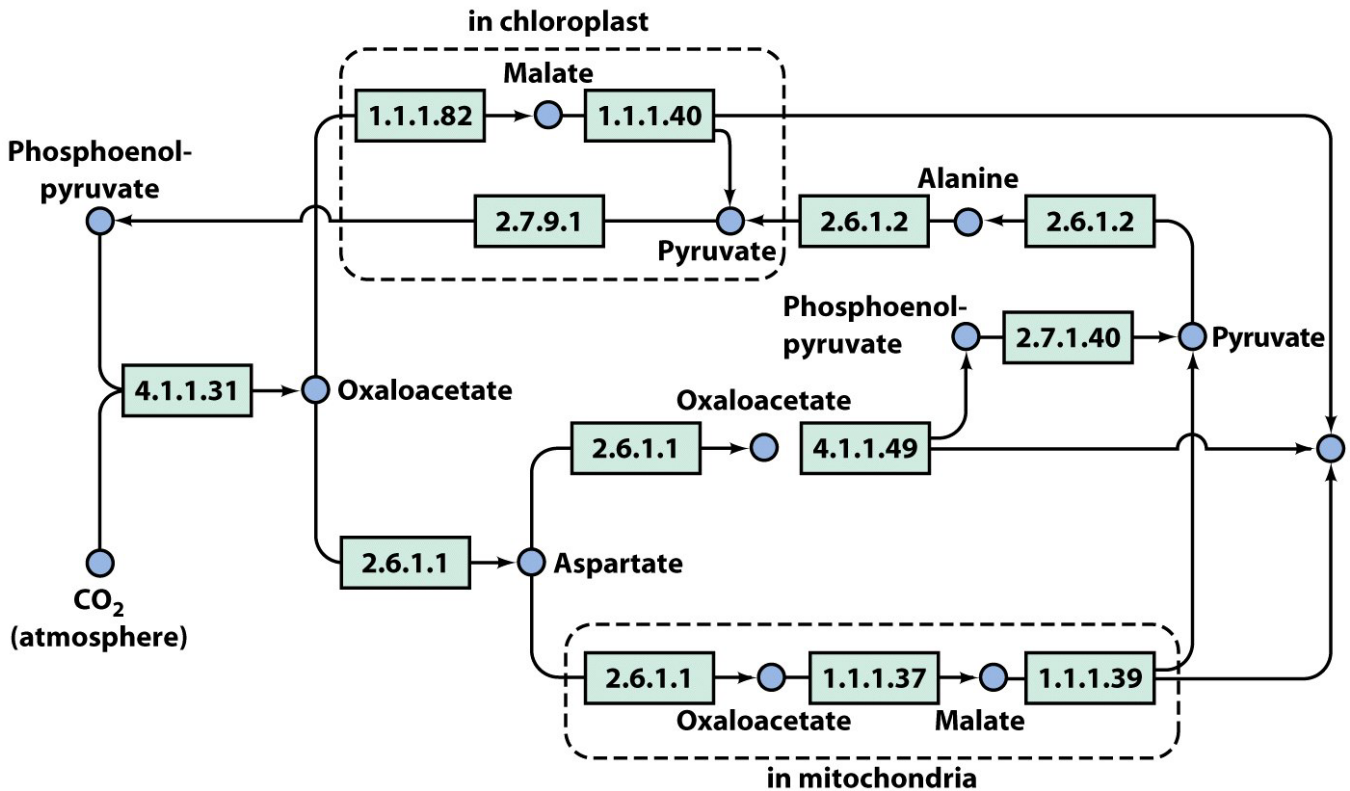


Figure 13-2 Fundamentals of Biochemistry, 2/e
© 2006 John Wiley & Sons

Converge to common intermediates

Mapping metabolic pathways catalyzed by enzymes



Box 13-2 Fundamentals of Biochemistry, 2/e

Enzyme reactions
fall into 4 major types

Oxidations and reductions (oxidoreductases)
Group-transfer reactions (transferases and hydrolases)
Eliminations, isomerizations, and rearrangements (isomerases and mutases)
Reactions that make or break C-C bonds (hydrolases, lyases, and ligases)

Compartmentation

Metabolic pathways occur in specific cellular locations

Table 13-1 Metabolic Functions of Eukaryotic Organelles

Organelle	Major functions
Mitochondrion	Citric acid cycle, oxidative phosphorylation, fatty acid oxidation, amino acid breakdown
Cytosol	Glycolysis, pentose phosphate pathway, fatty acid biosynthesis, many reactions of gluconeogenesis
Lysosomes	Enzymatic digestion of cell components and ingested matter
Nucleus	DNA replication and transcription, RNA processing
Golgi apparatus	Posttranslational processing of membrane and secretory proteins; formation of plasma membrane and secretory vesicles
Rough endoplasmic reticulum	Synthesis of membrane-bound and secretory proteins
Smooth endoplasmic reticulum	Lipid and steroid biosynthesis
Peroxisomes (glyoxysomes in plants)	Oxidative reactions catalyzed by amino acid oxidases and catalase; glyoxylate cycle reactions in plants

Table 13-1 Fundamentals of Biochemistry, 2/e

Metabolic pathways depends on tissues and organs

liver, muscle, adipocyte

isozymes: LDH (take-home work)

Thermodynamic considerations

metabolic flux (rate of flow): analogous to dam

near-equilibrium reaction (reversible):

depends on the relative concentrations of substrates and products

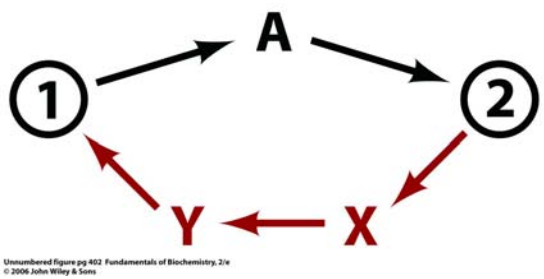
<u>far-equilibrium reaction (irreversible):</u>

accumulation of substrate (insufficient catalytic efficiency)

controlled by allosteric effector

$$\Delta G = \Delta G^{o} + RT\ln \frac{[C]^{c}[D]^{d}}{[A]^{a}[B]^{b}}$$

1. Metabolic pathways are irreversible: confers directionality
2. Every metabolic pathway has a first committed step
3. Catabolic and anabolic pathways differ



Unnumbered figure pg 402 Fundamentals of Biochemistry, 2/e
© 2006 John Wiley & Sons

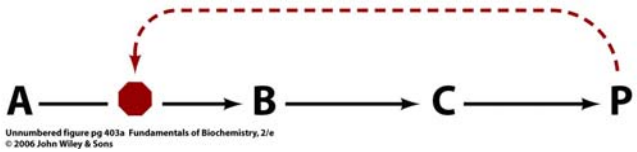
Control of metabolic flux

J = v(f) – v(r)

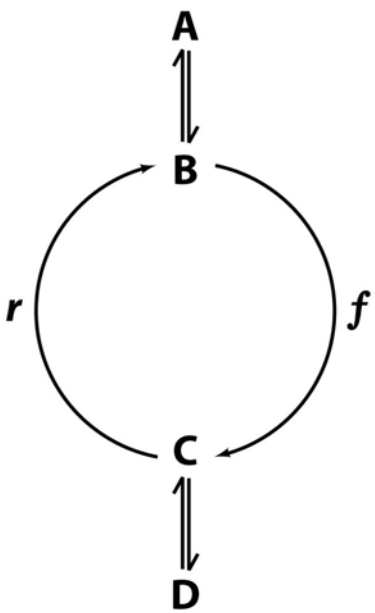
At equilibrium J=0, although v(f) and v(r) may be quite large

Flux is determined by the slowest step (rate-determining step) *** committed step

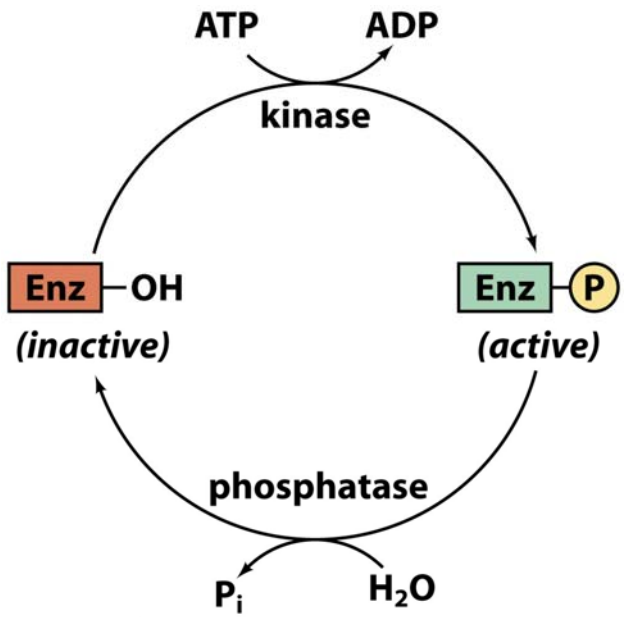
Allosteric control
Covalent modification
Substrate cycles
Genetic control



Unnumbered figure pg 403a Fundamentals of Biochemistry, 2/e



Unnumbered figure pg 404 Fundamentals of Biochemistry, 2/e



Unnumbered figure pg 403b Fundamentals of Biochemistry, 2/e

High-energy compounds

High-energy intermediates: phosphorylated compounds, NADH
A sort of free energy currency

ATP and phosphoryl group transfer:
thermodynamically favored
but kinetically disfavored

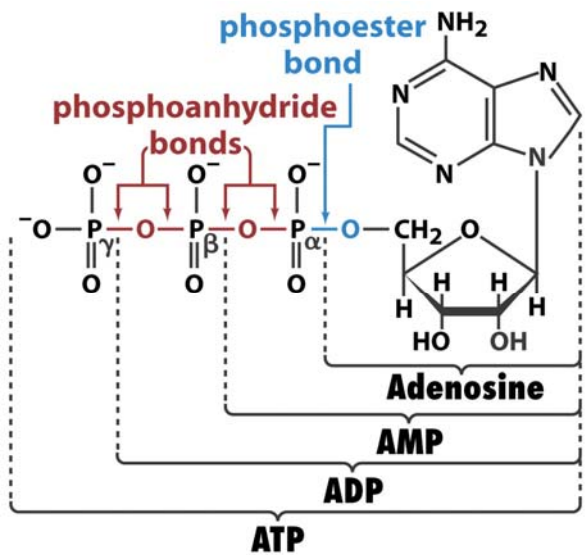


Figure 13-3 Fundamentals of Biochemistry, 2/e
© 2006 John Wiley & Sons

Table 13-2 Standard Free Energies of Phosphate Hydrolysis of Some Compounds of Biological Interest

Compound	$\Delta G^{\circ\prime}$ (kJ · mol^{-1})
Phosphoenolpyruvate	−61.9
1,3-Bisphosphoglycerate	−49.4
ATP (→ AMP + PP_i)	−45.6
Acetyl phosphate	−43.1
Phosphocreatine	−43.1
ATP (→ ADP + P_i)	−30.5
Glucose-1-phosphate	−20.9
PP_i	−19.2
Fructose-6-phosphate	−13.8
Glucose-6-phosphate	−13.8
Glycerol-3-phosphate	−9.2

Source: Mostly from Jencks, W.P., *in* Fasman, G.D. (Ed.), *Handbook of Biochemistry and Molecular Biology* (3rd ed.), Physical and Chemical Data, Vol. I, pp. 296–304, CRC Press (1976).

Table 13-2 Fundamentals of Biochemistry, 2/e
© 2006 John Wiley & Sons

Coupled reactions

coupling of exergonic and endergonic process

Not actual process in the catalyzing enzyme

ATP coupling to conformational changes

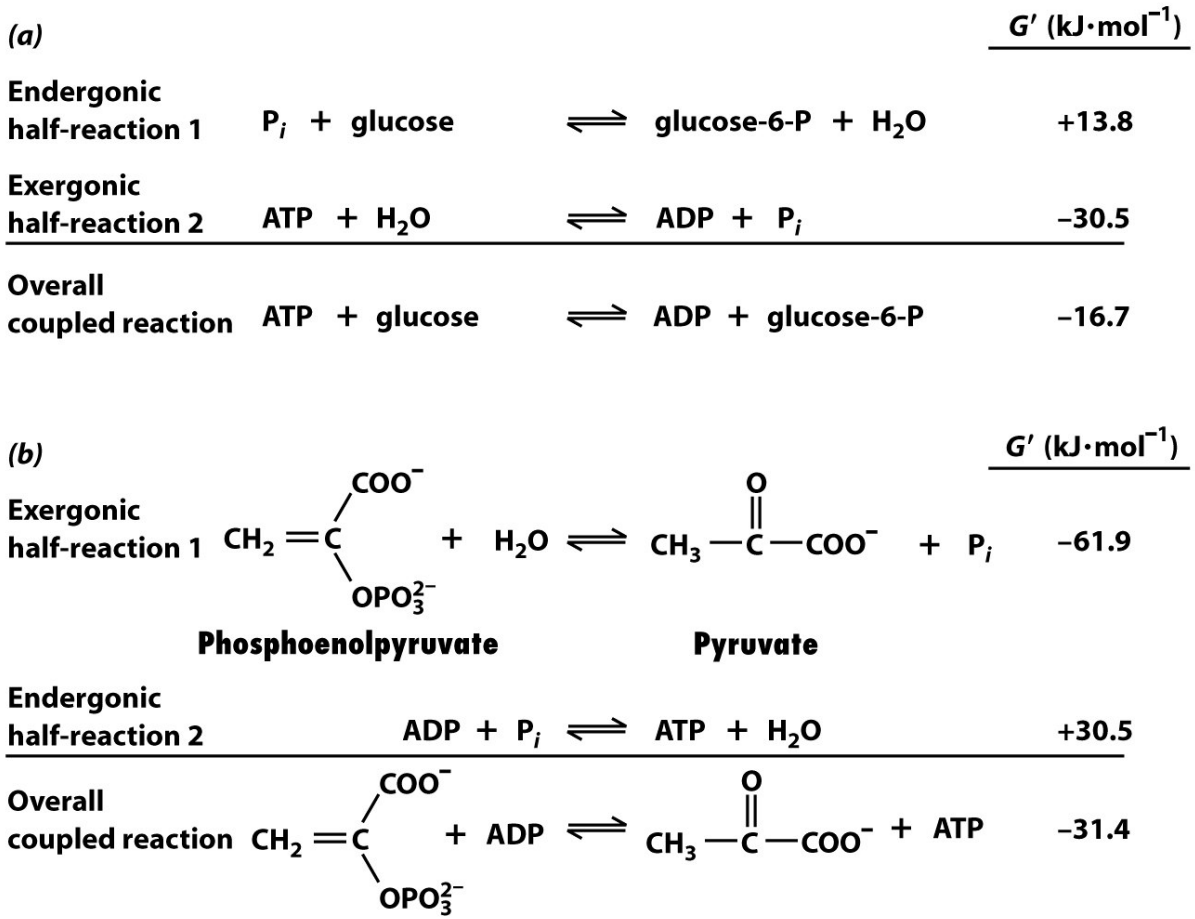


Figure 13-5 Fundamentals of Biochemistry, 2/e

Other phosphorylated compounds

ATP is continually being hydrolyzed and regenerated
metabolic half-life: from seconds to minutes

Substrate level phosphorylation
Oxidative phosphorylation
Photophosphorylation

Interconversion of nucleoside triphosphates
Kinases & phosphatases

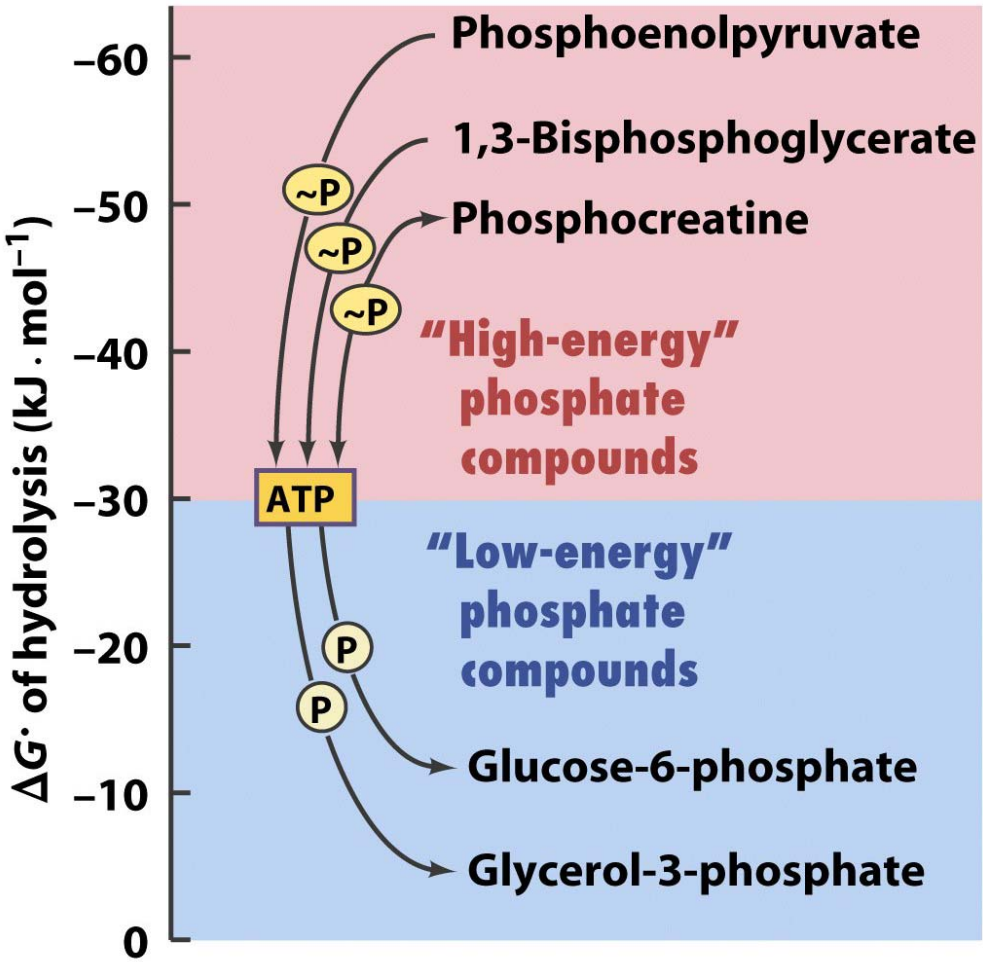
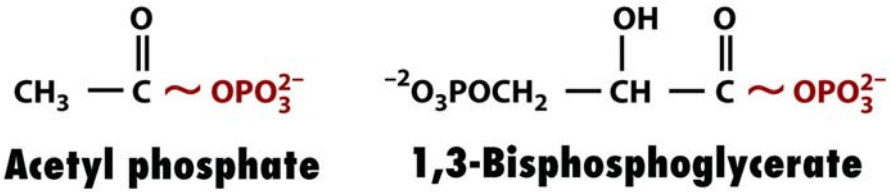
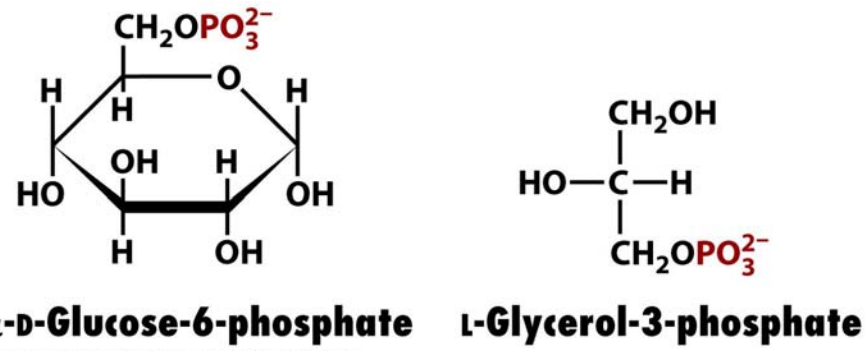


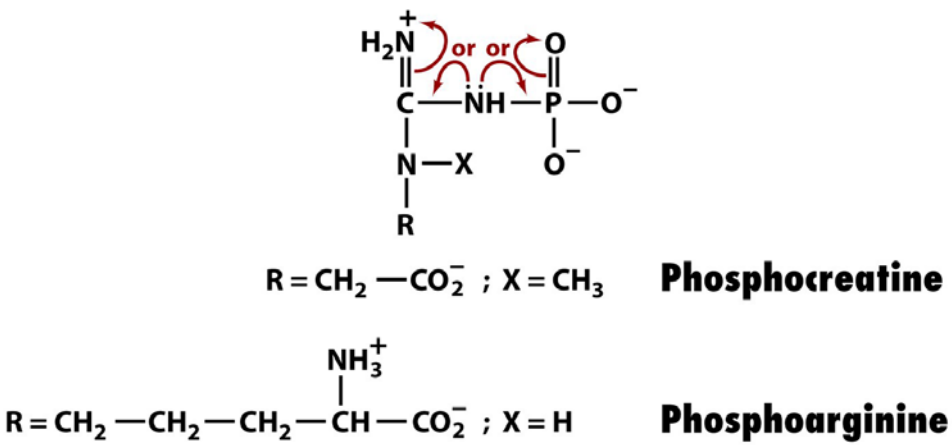
Figure 13-7 Fundamentals of Biochemistry, 2/e



Unnumbered figure pg 411a Fundamentals of Biochemistry, 2/e



Unnumbered figure pg 411b Fundamentals of Biochemistry, 2/e



Unnumbered figure pg 411c Fundamentals of Biochemistry, 2/e

Thioester: primitive compound

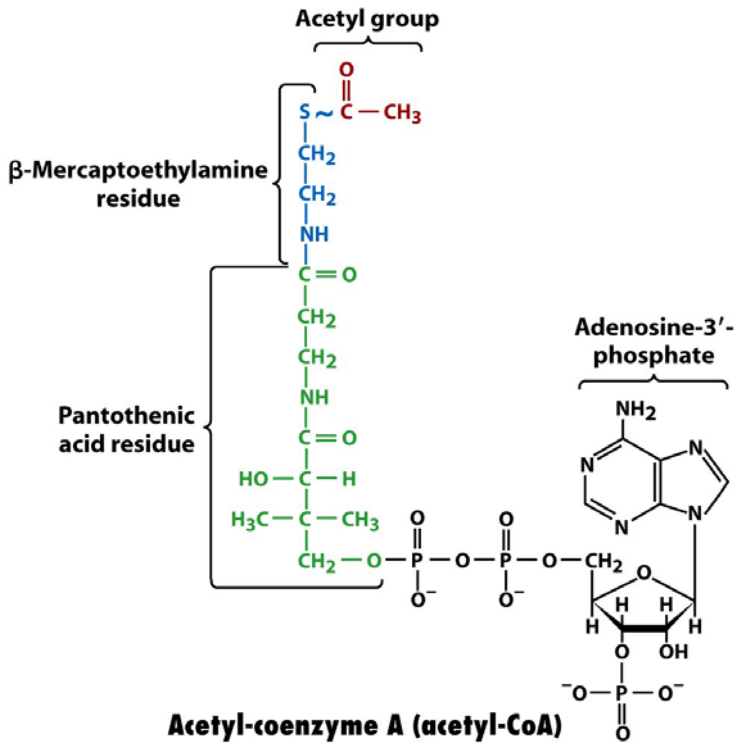


Figure 13-9 Fundamentals of Biochemistry, 2/e

Oxidation-reduction reactions

- One electron transfer
- Two electron transfer
- Reversible reaction

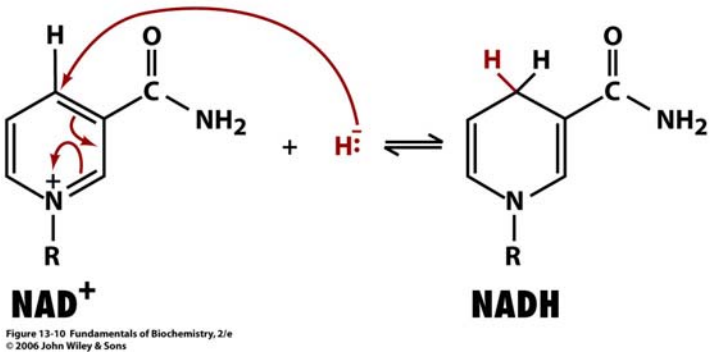


Figure 13-10 Fundamentals of Biochemistry, 2/e
© 2006 John Wiley & Sons

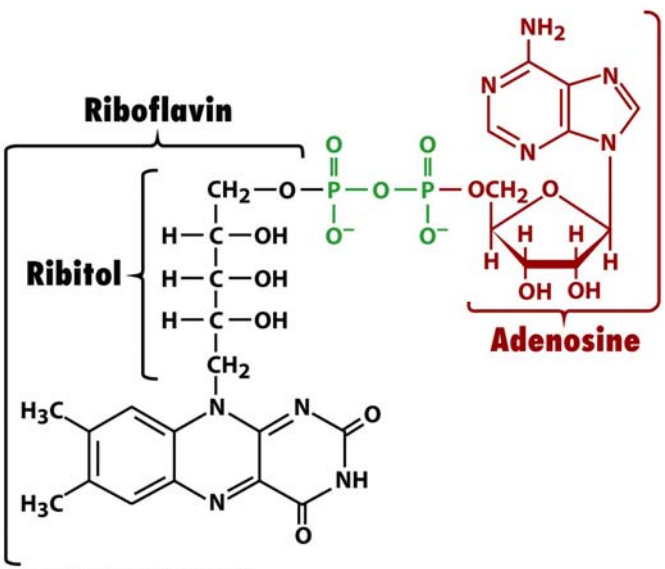


Figure 13-11 Fundamentals of Biochemistry, 2/e
© 2006 John Wiley & Sons

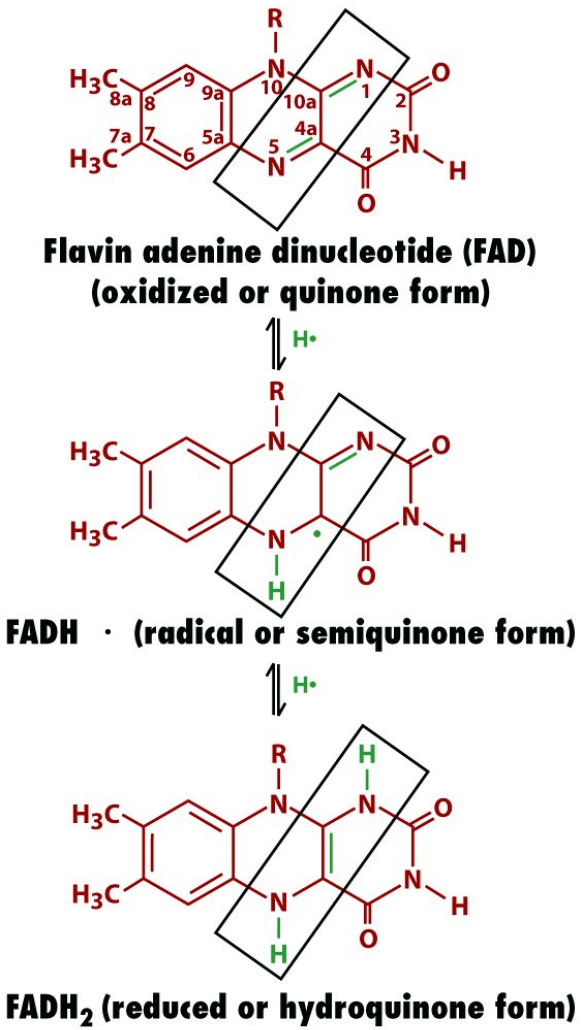


Figure 13-12 Fundamentals of Biochemistry, 2/e
© 2006 John Wiley & Sons

The Nernst equation

Oxidation-reduction reactions: electron transfer reaction
Electron donor & acceptor
Electrochemical cells:

redox pair (analogous to acid-base pair)

a half-reactions: electron donor and its conjugate electron acceptor

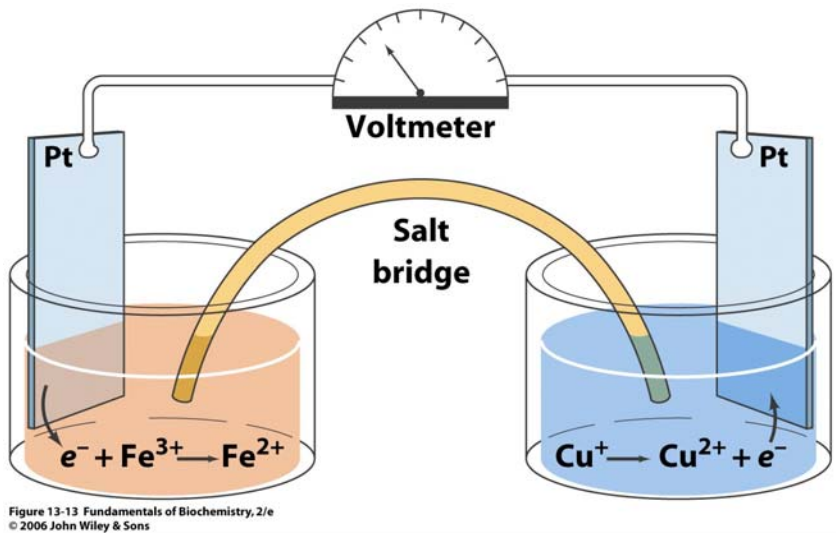


Figure 13-13 Fundamentals of Biochemistry, 2/e

Table 13-3 Standard Reduction Potentials of Some Biochemically Important Half-Reactions

Half-Reaction	$\mathscr{E}^{\circ\prime}$ (V)
$\frac{1}{2}\,O_2 + 2\,H^+ + 2\,e^- \rightleftharpoons H_2O$	0.815
$SO_4^{2-} + 2\,H^+ + 2\,e^- \rightleftharpoons SO_3^{2-} + H_2O$	0.48
$NO_3^- + 2\,H^+ + 2\,e^- \rightleftharpoons NO_2^- + H_2O$	0.42
Cytochrome a_3 $(Fe^{3+}) + e^- \rightleftharpoons$ cytochrome a_3 (Fe^{2+})	0.385
$O_2(g) + 2\,H^+ + 2\,e^- \rightleftharpoons H_2O_2$	0.295
Cytochrome a $(Fe^{3+}) + e^- \rightleftharpoons$ cytochrome a (Fe^{2+})	0.29
Cytochrome c $(Fe^{3+}) + e^- \rightleftharpoons$ cytochrome c (Fe^{2+})	0.235
Cytochrome c_1 $(Fe^{3+}) + e^- \rightleftharpoons$ cytochrome c_1 (Fe^{2+})	0.22
Cytochrome b $(Fe^{3+}) + e^- \rightleftharpoons$ cytochrome b (Fe^{2+}) (*mitochondrial*)	0.077
Ubiquinone $+ 2\,H^+ + 2\,e^- \rightleftharpoons$ ubiquinol	0.045
Fumarate$^- + 2\,H^+ + 2\,e^- \rightleftharpoons$ succinate$^-$	0.031
FAD $+ 2\,H^+ + 2\,e^- \rightleftharpoons FADH_2$ (*in flavoproteins*)	~0.
Oxaloacetate$^- + 2\,H^+ + 2\,e^- \rightleftharpoons$ malate$^-$	−0.166
Pyruvate$^- + 2\,H^+ + 2\,e^- \rightleftharpoons$ lactate$^-$	−0.185
Acetaldehyde $+ 2\,H^+ + 2\,e^- \rightleftharpoons$ ethanol	−0.197
FAD $+ 2\,H^+ + 2\,e^- \rightleftharpoons FADH_2$ (*free coenzyme*)	−0.219
$S + 2\,H^+ + 2\,e^- \rightleftharpoons H_2S$	−0.23
Lipoic acid $+ 2\,H^+ + 2\,e^- \rightleftharpoons$ dihydrolipoic acid	−0.29
$NAD^+ + H^+ + 2\,e^- \rightleftharpoons NADH$	−0.315
$NADP^+ + H^+ + 2\,e^- \rightleftharpoons NADPH$	−0.320
Cystine $+ 2\,H^+ + 2\,e^- \rightleftharpoons 2$ cysteine	−0.340
Acetoacetate$^- + 2\,H^+ + 2\,e^- \rightleftharpoons$ β-hydroxybutyrate$^-$	−0.346
$H^+ + e^- \rightleftharpoons \frac{1}{2}\,H_2$	−0.421
Acetate$^- + 3\,H^+ + 2\,e^- \rightleftharpoons$ acetaldehyde $+ H_2O$	−0.581

Source: Mostly from Loach, P.A., *In* Fasman, G.D. (Ed.), *Handbook of Biochemistry and Molecular Biology* (3rd ed.), Physical and Chemical Data, Vol. I, pp. 123–130, CRC Press (1976).

Table 13-3 Fundamentals of Biochemistry, 2/e

Experimental approaches to the study of metabolism

Understanding the sequence, mechanism, and regulation

Approaches

- tracing metabolic fates
- perturbing the system
  - metabolic inhibitors, genetic defects, genetic manipulation
- DNA microarrays (DNA chips): transcriptomics
- proteomics

13C NMR spectrum of rat liver

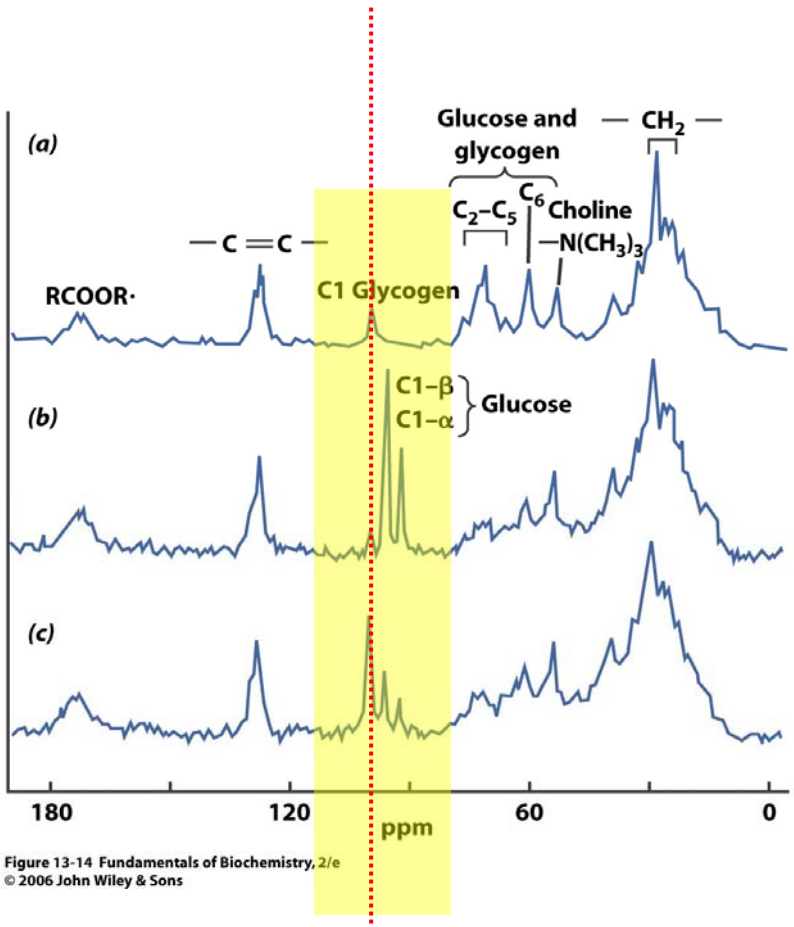


Figure 13-14 Fundamentals of Biochemistry, 2/e
© 2006 John Wiley & Sons

(a) feeding with [1-^{13}C]glucose
(b) after 5 min
(c) after 30 min

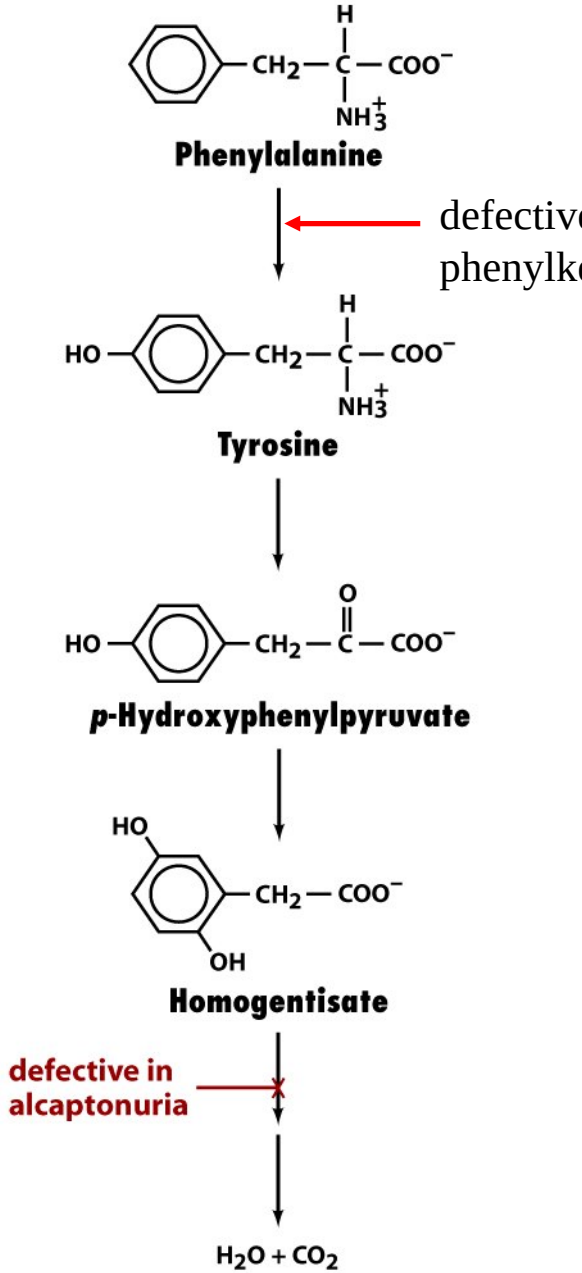


Figure 13-15 Fundamentals of Biochemistry, 2/e
© 2006 John Wiley & Sons

PCR amplified yeast cDNAs

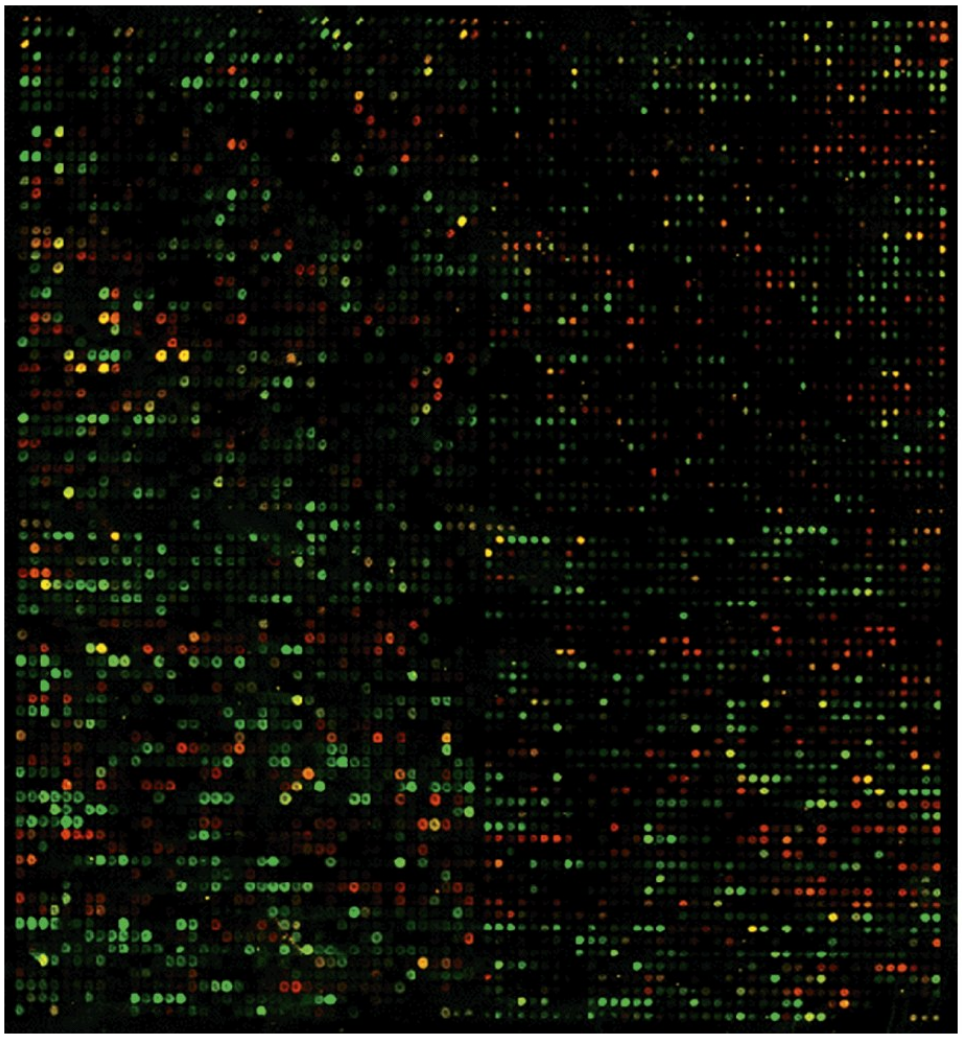


Figure 13-16 Fundamentals of Biochemistry, 2/e

Red spots: cDNAs from the cells with glucose
Green spots: cDNAs from the cells without glucose

hepatocarcinoma

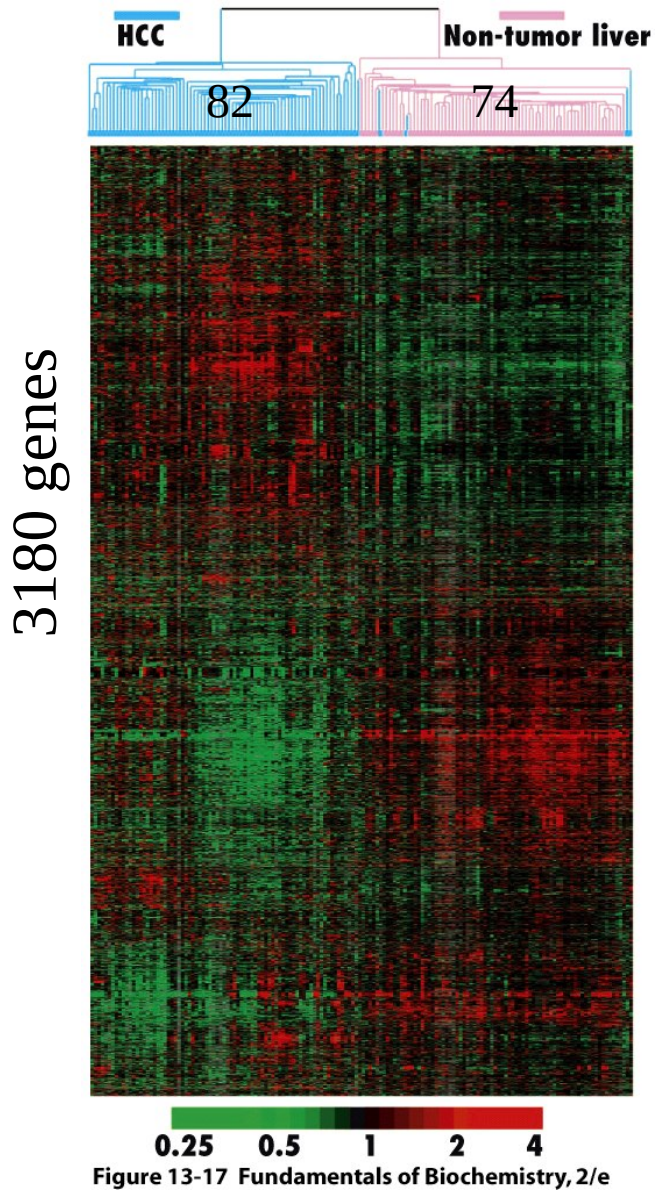


Figure 13-17 Fundamentals of Biochemistry, 2/e