

BIOLOGICAL CHEMISTRY

Chapter 4: OVERVIEW OF METABOLISM

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Date of Submission 2/6/2009

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OVERVIEW OF METABOLISM

INTRODUCTION

We gained insights into the major groups of biomolecules in Chapter 2. Let us now look at the reactions they undergo in the body.

The *ultimate* source for all biomolecules is our dietary intake. Digestion of ingested food components in the gastrointestinal tract yields smaller molecules that are absorbed through the epithelial lining of the intestine and sent directly or indirectly into the blood.

CARBOHYDRATES → monosaccharides

LIPIDS → fatty acids and glycerol

PROTEINS → amino acids

The **sum total of all the catabolic and anabolic reactions** that the products of digestion and absorption now undergo in the cells of the body collectively constitute the phenomenon of **metabolism**. Every cell in a living organism works like an industrial organization. It is in a state of *dynamic equilibrium*, constantly taking in substances from its external environment, processing them, synthesizing its own requirements, degrading what is old or harmful, and sending out what it may have produced for other cells, or what is waste. Since all these processes either *require energy* or *release energy*, hence **energy transactions** and transductions are essential features of metabolism.

Remember that a body tends to maintain a state of homeostasis. The cell has to carry out a plethora of reactions, often simultaneously, but almost always under the *same* conditions of temperature, pressure, pH and many such parameters that we can blithely alter when we carry out reactions in test-tubes. To achieve this feat, biochemical evolution has produced **enzymes**, which are biocatalysts, ubiquitous in all prokaryotes and eukaryotes. You have learnt how enzymes facilitate reactions in Chapter 3.

Enzyme-catalyzed reactions within a cell are generally organized as *multi-step sequences* called **pathways**, in which the product of one reaction becomes the substrate in the subsequent reaction. Each consecutive enzyme in a metabolic pathway catalyzes a small, specific, chemical change in a substrate till the final desired product is obtained. Different pathways intersect to form an **integrated and purposeful network** collectively called metabolism.

The network of metabolic pathways in a tiny cell is much more complicated and several times more efficient than any circuit you may have seen on a ‘chip’! The tremendous degree of integration and control required to keep these systems functioning normally and appropriately is one of the most fascinating aspects of cell function. To appreciate this take a look at fig 4.1.

The term **intermediary metabolism** is sometimes used to refer to the “combined activities of all the metabolic pathways that interconvert precursors, metabolites and products of low molecular weight”. A precursor molecule in a catabolic or anabolic pathway is converted into the final product(s) through a series of **intermediates** called metabolites. Many small molecules (generally with $M_r < 1,000$) like glyceraldehyde 3-phosphate, ribose phosphate, pyruvate, acetyl-CoA, succinyl-CoA and oxalacetate are intermediates in more than one metabolic pathway. They function as *links* between one pathway and another and thereby help in the *integration* of carbohydrate, lipid, protein and even nucleic acid metabolism.

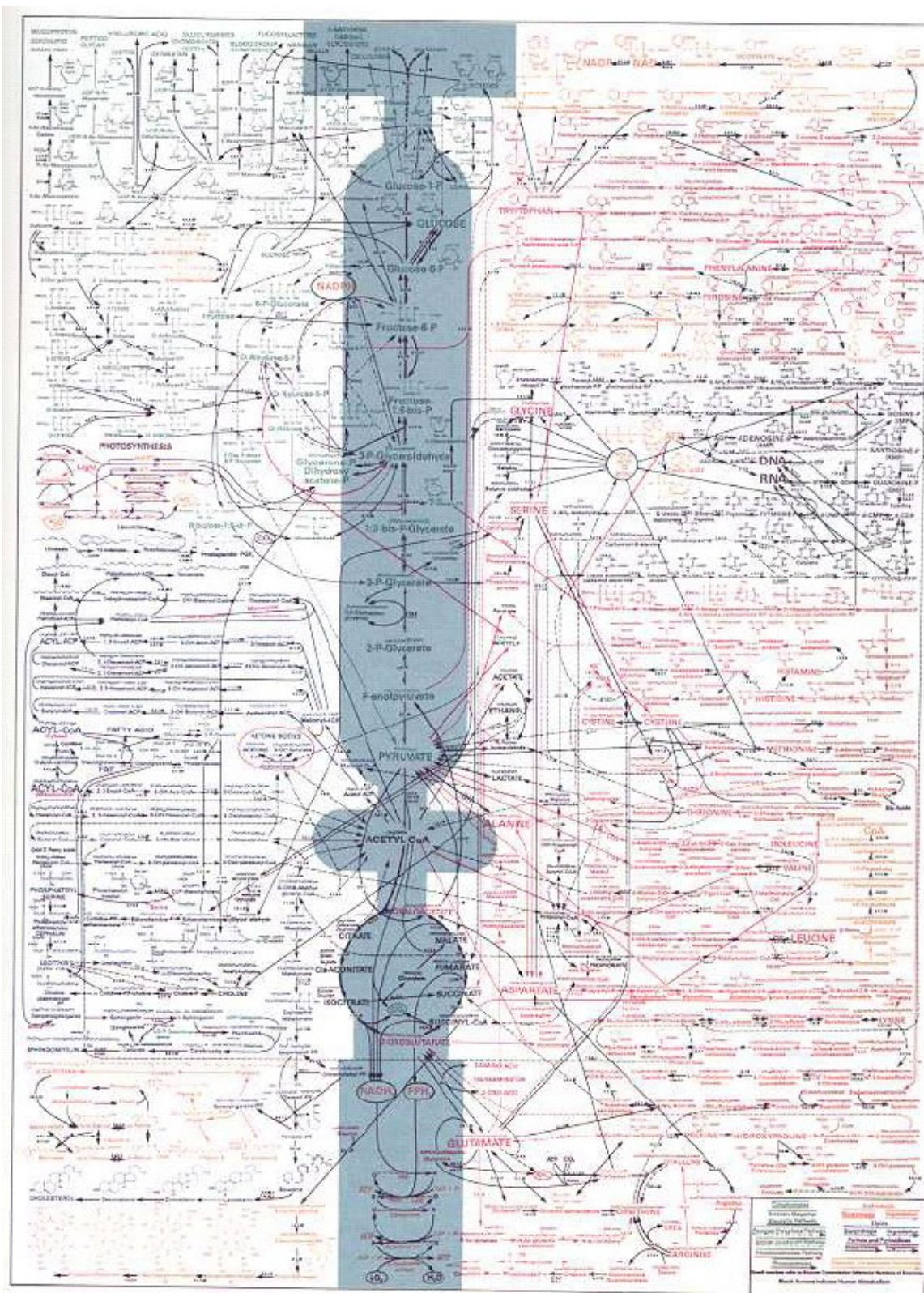


Fig. 4.1. An overview of metabolic pathways, showing its stupendous complexity.
(Source: Voet and Voet, 1995, p 413 fig 15-7)

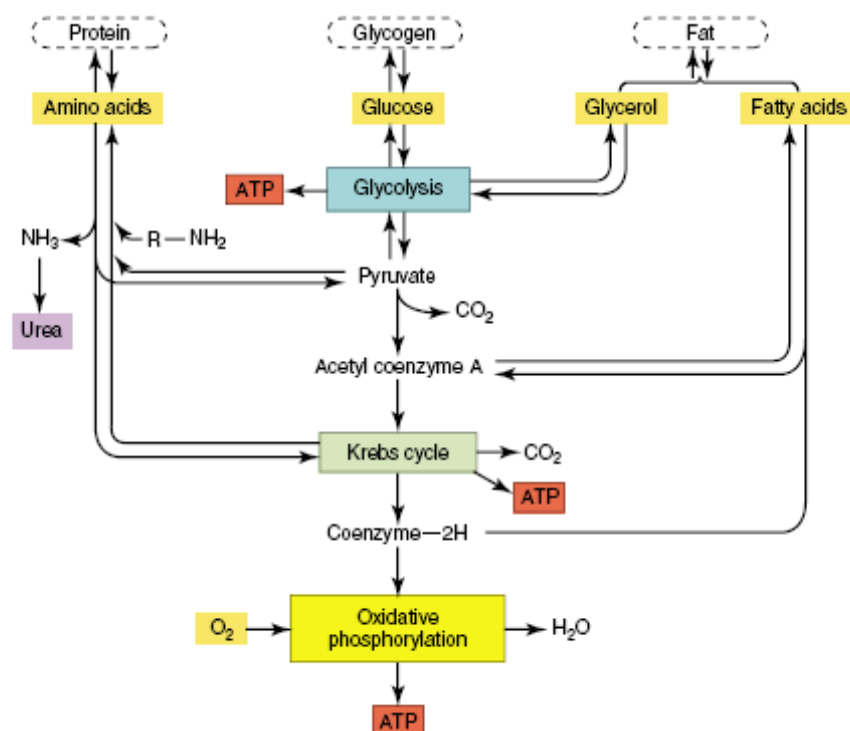


Fig 4.2. Integration of protein, carbohydrate and lipid metabolism through small molecules like pyruvate, acetyl-CoA and reduced coenzymes (e.g. NADH.H⁺, FADH₂ and NADPH.H⁺)
(Source: Vander *et al*, 2001, p 85 fig 4-32)

Since reactions of opposing metabolic pathways can take place simultaneously, it is important that control mechanisms are able to ‘switch on’ and ‘switch off’ pathways as required. The reciprocal regulation of glycolysis and gluconeogenesis (Chapter 5 Section 5.6B) is an excellent example of this control.

The sequence of reactions in a metabolic pathway may be arranged in a linear (e.g. glycolysis), branched (e.g. amino acid synthesis), cyclic (e.g. citric acid cycle) or spiral (e.g. fatty acid degradation) manner as shown in fig 4.2. Suitable examples of these pathways are covered in all the chapters on metabolism (Chapters 5 to 8).

In this chapter we compare catabolism and anabolism in *Section 1* and then list the various central pathways in carbohydrate, lipid, protein and nucleic acid metabolism in *Section 2*. We emphasize the role of the ATP-ADP cycle in energy transactions in *Section 3* and go on to look at some of the other important molecules that serve as carriers of functional groups in *Section 4*. Intracellular localizations of metabolic pathways and the transporters and shuttle systems that help to transfer molecules across intracellular membranes are covered in *Sections 5*. The central role of the liver and the specialized metabolic functions of selected organs are described in *Section 6*. The final topic in this chapter deals with general mechanisms that regulate metabolic pathways (Section 7).

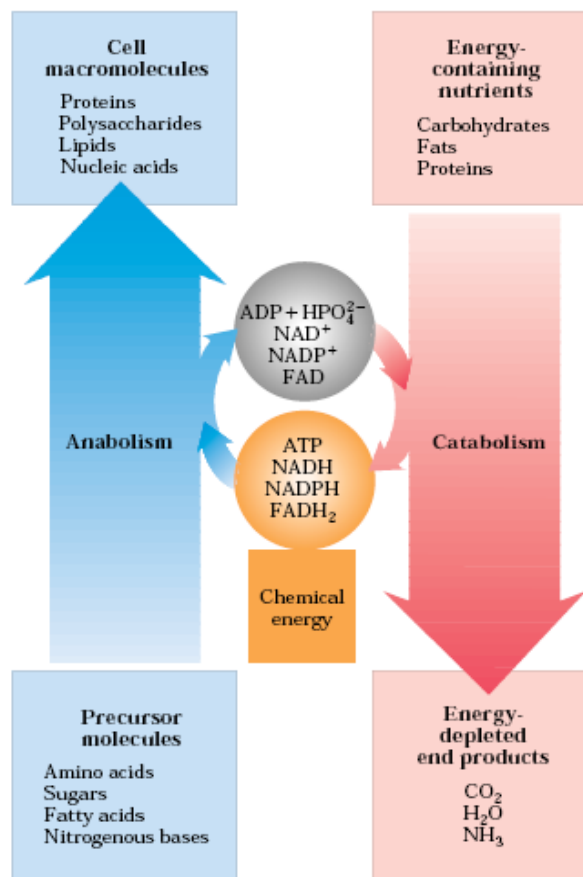


Fig. 4.1.1. Comparison of anabolism and catabolism (Source: Nelson and Cox, 2006, p 48 fig 3)

Catabolism can be broadly divided into three stages. Every stage involves several *co-ordinated* enzyme reactions. Stage I degrades complex polymeric molecules into simple monomeric biomolecules or “building blocks”. Stage II produces crucial intermediates like acetyl-CoA and a few simple molecules from the end-products of Stage I. Stage II either breaks them down further or channelizes them into biosynthetic pathways. Stage III is the final common pathway which completely oxidizes fuel molecules to yield CO₂ and H₂O, and generates ATP.

Anabolism also involves several *co-ordinated* enzyme reactions, but they are diverse and often very specific to the kind of biomolecule desired. Generalizations may therefore be overtly simplistic.

Pathways that link anabolism and catabolism are called **amphibolic** pathways, a superb example of which is the citric acid cycle (Chapter 5, Section 3). Metabolites like acetyl-CoA and α -ketoglutarate serve as intermediates in such connected sequences of anabolic and catabolic reactions.

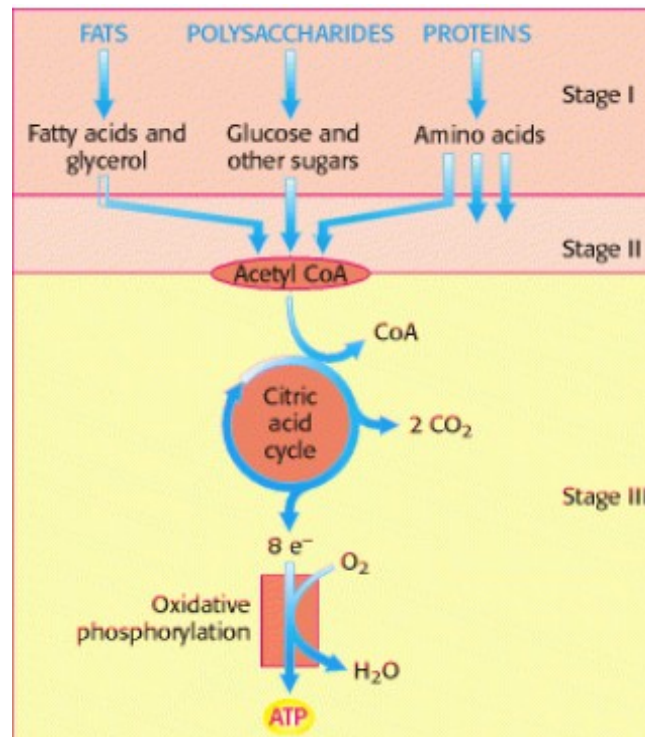


Fig. 4.1.2. The three stages of catabolism: Stage I breaks down lipids, carbohydrates and protein into their basic structural units from which Stage II produces a crucial intermediate acetyl-CoA. Stage III carries out electron transfer and oxidative phosphorylation for final yield of ATP.
(Source: Berg *et al*, 2002, fig 14-12)

Chapter 4 Section 2

OUTLINE OF METABOLIC PROCESSES

Carbohydrate metabolism primarily deals with:

- provision and utilization of glucose as energy fuel
- conservation of excess glucose by synthesis of glycogen/starch/lipids
- synthesis/degradation of sugar derivatives and conjugates for membranes, extracellular matrix, hormones and other physiologically important products

Lipid metabolism mainly concerns:

- synthesis/utilization of fatty acids and ketone bodies as energy fuels
- formation of lipoprotein complexes for lipid transport
- synthesis/degradation of storage triglycerides
- synthesis of membrane lipids
- synthesis/conversions of cholesterol and other physiologically important steroids

Protein metabolism primarily involves:

- synthesis of essential amino acids
- formation of peptides, proteins and specialized non-protein products from amino acids
- post-translational modification of peptides and proteins
- degradation of amino acids, with removal of nitrogen as urea/uric acid/ ammonia.
- degradation of peptides and proteins

Protein metabolism differs from carbohydrate and lipid metabolism in that:

- protein synthesis *directly* depends on activity of the encoding genes, and on transcription and translation
- excess protein is *never* stored

Nucleic acid metabolism involves:

- *de novo* synthesis of purines, pyrimidines, nucleosides and nucleotides
- “salvage” reactions of degraded nucleotides for fresh nucleotide synthesis
- formation of deoxyribonucleotides from ribonucleotides
- catabolism of purines and pyrimidines
- synthesis of DNA by replication of pre-existing DNA template/RNA
- DNA-dependent and RNA-dependent synthesis of RNA
- degradation of DNA and RNA

Chapter 4 Section 3

ENERGY TRANSACTIONS

Autotrophs and heterotrophs differ from each other in the source from which they obtain energy and carbon for the synthesis of biomolecules and for work. Autotrophs (photosynthetic bacteria and plants) synthesize complex biomolecules by using the energy of sunlight, and obtaining carbon from only atmospheric CO₂. Heterotrophs (most animals) obtain energy and carbon from ingested carbohydrates and fats. In spite of this difference, both autotrophs and heterotrophs show a remarkable similarity in many of their major metabolic pathways.

Catabolism involves *exergonic* reactions, while anabolism and many biological functions are *endergonic* processes. The energy released in catabolism is trapped, at least partially, in the chemical bond of a high-energy intermediate from which it is tapped for propelling synthetic pathways and performing cellular functions like active transport, muscle contraction and nerve excitation. The key role in energy transfer is played by “*high-energy phosphates*”, particularly the adenosine nucleotides, ATP and ADP, which constitute the **ATP-ADP system**. ATP is regarded as the “*energy currency*” of the cell. The “**ATP-ADP cycle**” connects those processes that generate ATP to those that utilize ATP and is the fundamental mode of energy transfer in biological systems. It is to be remembered, however, that ATP is the principal *immediate donor* of free energy in biological systems and not a *long-term storage* form of free energy.

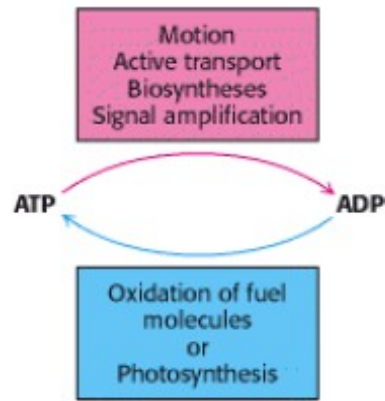


Fig. 4.3.1. The ATP-ADP cycle: The activities enclosed in the pink box utilize ATP, dephosphorylating it to ADP, while those in the blue box phosphorylate ADP to ATP. (Source: Berg *et al*, 2002, fig 14-8)

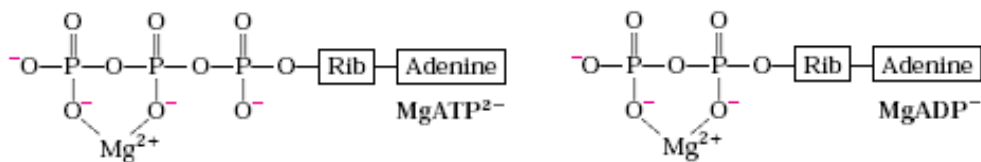


Fig. 4.3.2. Comparative molecular structures of ATP and ADP. (Source: Nelson and Cox, 2003, p 497 fig 13-2)

The terminal and middle phosphoanhydride bonds in ATP and ADP are “high-energy” bonds i.e. their hydrolysis involves a massive and negative free-energy change that is utilized to “drive” other reactions (see Chapter 9).

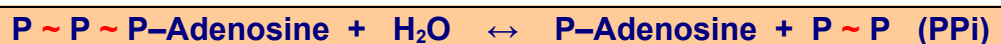
Hydrolysis of ATP is highly exergonic and can proceed in two ways:

- **Orthophosphate cleavage:** which produces ADP and inorganic phosphate



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

- **Pyrophosphate cleavage:** which produces AMP and pyrophosphate; this reaction is driven forward by its immediate coupling to the hydrolysis of pyrophosphate.



$$(\text{Overall } \Delta G'^{\circ} = -64.8 \text{ kJ/mol})$$

A one-step direct hydrolysis of ATP to AMP and 2 Pi never takes place.

Synthesis of ATP is an endergonic reaction and occurs by phosphorylation of ADP. The reaction is carried out by coupling it to energy release or energy transfer processes like:

- an exergonic reaction of the catabolic pathways (e.g. glycolysis: Chapter 5 Section 1)
- electron transfers in the mitochondrial respiratory chain (Chapter 9 Section 6)
- direct transfer from suitable substrates (e.g. creatine phosphate: Chapter 9 Section 2)

Many intermediate compounds in metabolic reactions are phosphorylated by ATP and conserve metabolic energy. Phosphate esters like glucose 6-phosphate, formed by reaction of glucose with ATP, partially conserves the energy which is made available when ATP undergoes hydrolysis during the reaction. Phosphorylated substrates are “activated” compounds and binding to them facilitates the action of enzymes. An added advantage is that *phosphorylated* intermediates remain within the cell even against concentration gradients; there are no membrane transporters to ferry them outside.

Chapter 4 Section 4

CARRIER MOLECULES

Though the massive array of reactions in metabolism is overwhelming, the actual *kinds* of reactions are relatively *few* and the *mechanisms* used are astonishingly *simple*! The reactions that you will come across repeatedly are oxidation-reductions, hydrolytic/non-hydrolytic cleavage of bonds including C-C bonds, hydroxylation/dehydration, formation/saturation of double bonds, transfer of phosphoryl/acyl/carboxylic/amino groups, and isomerizations.

In the course of biochemical evolution, some molecules have been selected to carry out specific tasks in a wide variety of organisms. ADP is an ancient molecule in metabolism, conserved as a fundamental building block in **key molecules** like **ATP**, **NAD⁺**, **NADP⁺**, **FAD**, **FMN** and **CoA** (Coenzyme A).

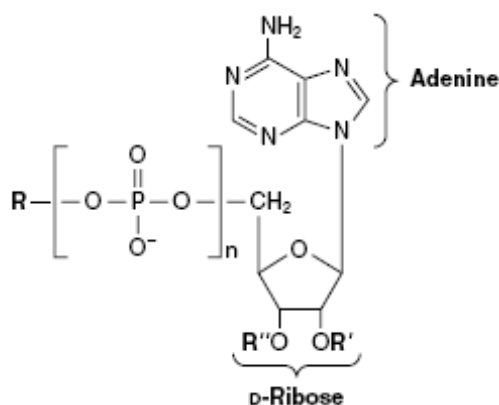


Fig. 4.4.1. Molecular formula of the basic structural unit in the coenzymes NAD, FAD and Coenzyme A. R is derived from niacin in NAD, riboflavin in FAD and pantothenic acid in CoA. R'' is H in NAD and FAD, and PO₃²⁻ in CoA. R' represents hydrogen and n = 2 in all the coenzymes.
(Source: Murray *et al*, 2003, p 290 Table 33-2)

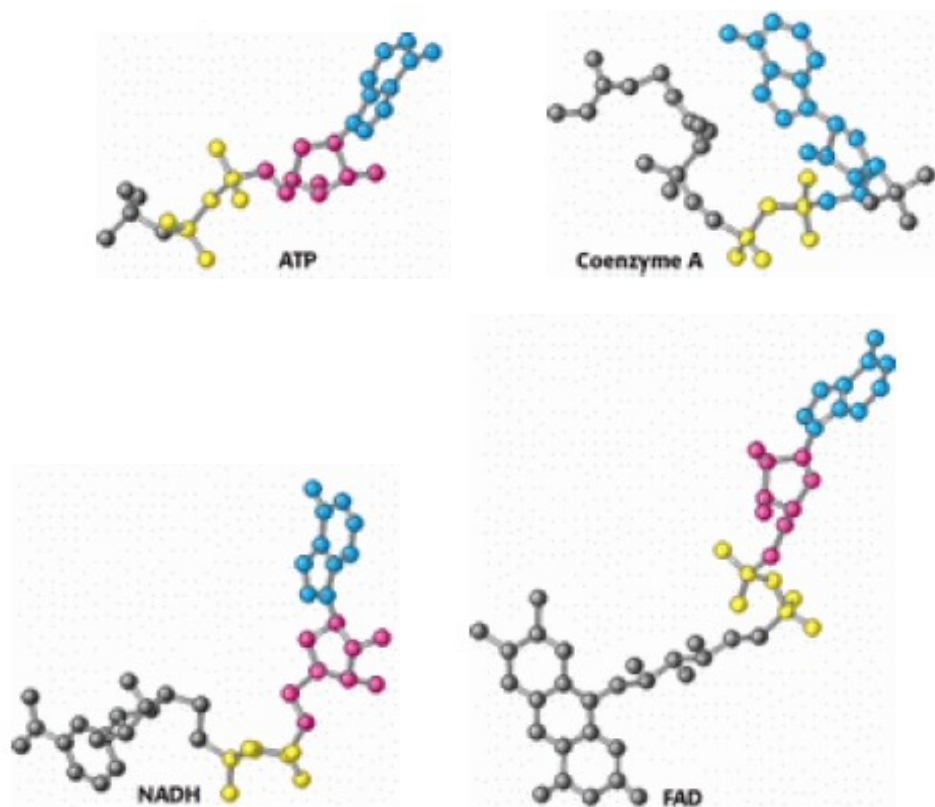


Fig. 4.4.2. Models of key molecules which contain ADP: the colored balls represent common units of the molecules (residues of adenine are blue, ribose is red and phosphate is yellow)
(Source: Berg *et al*, 2002, fig 14-9)

Many small molecules have been conserved to function as **carriers of specific functional groups** in metabolic reactions. The co-enzymes NAD^+ , NADP^+ , FAD and FMN are carriers of H^+ and electrons, and can be reduced and oxidized reversibly. They are reduced in various metabolic reactions and are re-oxidized in other cytosolic reactions or in the mitochondrial electron transport chain. Re-oxidation in the mitochondrion is exergonic, involves transfer of H^+ and electrons, and is coupled to ATP synthesis.

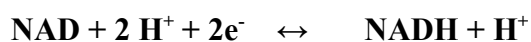
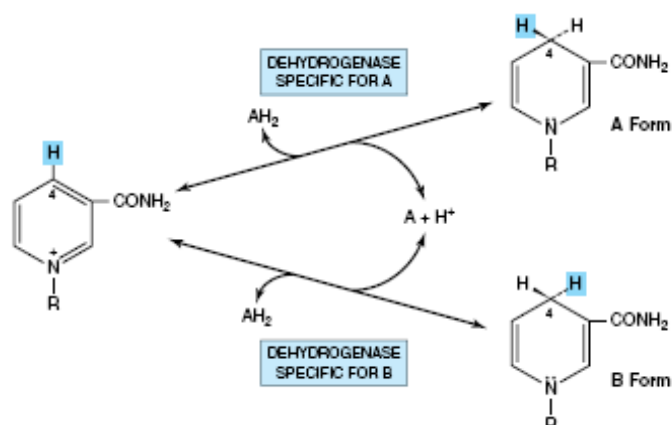


Fig 4.4.3. Reversible reduction of the nicotinamide ring in NAD (Source: Murray *et al*, 2003, p 89 fig 11-4)

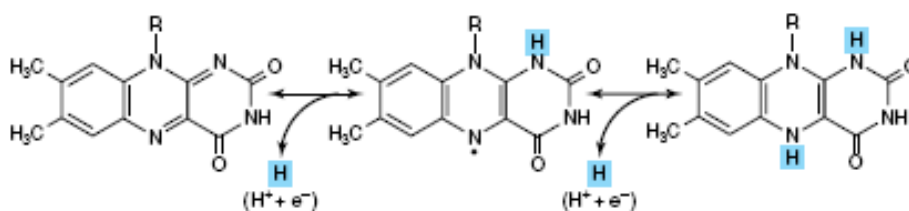


Fig. 4.4.4. Reversible reduction of the isoalloxazine ring of flavin nucleotides
(Source: Murray *et al*, 2003, p 88 fig 11-2)

Among other carrier molecules are **CoA** for the acyl group, **biotin** for COO^- , **UDP** for glucose, **TDP** for aldehyde, and **pyridoxal phosphate** for amino groups. **Acetyl-CoA** is an essential *amphibolic* intermediate in many catabolic and anabolic pathways like the citric acid cycle, fatty acid synthesis and gluconeogenesis where it is a donor of its C_2 unit.

Chapter 4 Section 5

INTRACELLULAR LOCALIZATION OF METABOLIC PATHWAYS

There is a very organized division of labor within the cell. Different metabolic reactions are conducted by different compartments or organelles. Thus, glycolysis and fatty acid synthesis occur only in the **cytosol** while the citric acid cycle and β -oxidation of fatty acids is restricted to the **mitochondrion**. The synthesis of nucleic acids is in the **nucleus** and triglyceride synthesis requires the **endoplasmic reticulum**. Protein synthesis is on the **ribosome** while glycosylation of proteins occurs in the **Golgi apparatus**.

Compartmentalization of metabolic reactions enhances the functional efficiency of a cell. Separation of the sites of anabolism and catabolism ensures that the synthetic and degradative pathways for the same molecule do not get entangled (compare the sites of fatty acid synthesis and oxidation in the above figure). Proper localization also ensures that a key substrate is often produced in the vicinity of the site of its subsequent utilization. Thus, the production of NADH.H^+ in the citric acid cycle and its oxidative phosphorylation in the electron transport chain, both occur within the mitochondrion. Similarly ketogenesis occurs in the mitochondrion at the site where oxidative decarboxylation of pyruvate and β -oxidation of fatty acids generates its precursor, acetyl-CoA.

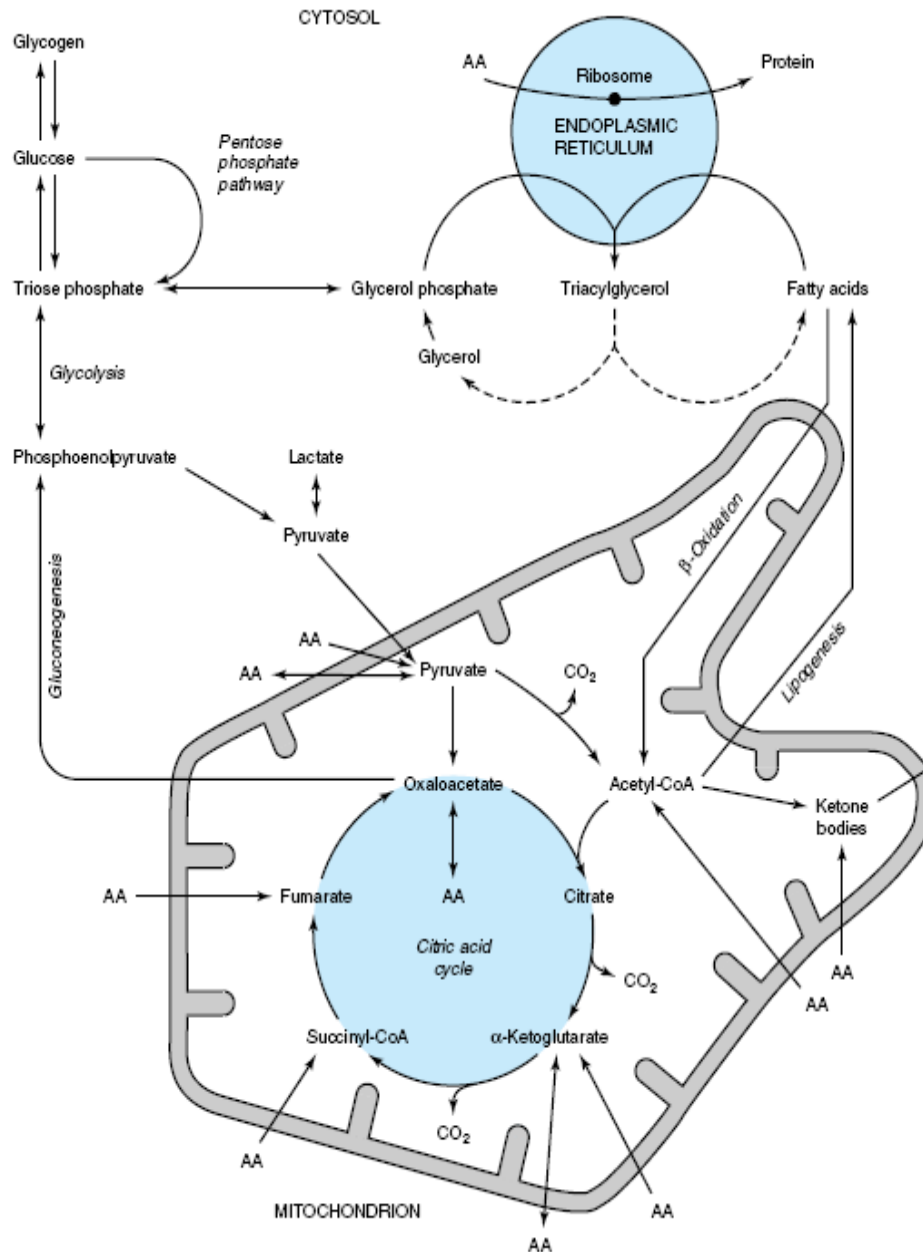


Fig. 4.5.1. Intracellular localization of different metabolic pathways in a hepatocyte (AA = amino acid). Note the separation of anabolic and catabolic pathways. (Source: Murray *et al*, 2003, p 127 fig 15-7)

Movement of Molecules across Mitochondrial Membranes

The restriction of movement of key molecules like ATP, NADH, and acetyl-CoA across the mitochondrial membrane helps to regulate the concentrations of these metabolites in the cytosolic and mitochondrial compartments. However, in spite of the spatial localization of reactions, many metabolic intermediates do need to be exchanged between the cytosol and the mitochondrion for integration or continuation of different metabolic pathways. Crossing the inner mitochondrial membrane (IMM) is not an easy task. The IMM is freely permeable to small, uncharged

molecules like O_2 , water, CO_2 and NH_3 but prevents the *unrestricted* passage of many larger, charged, biologically important molecules. In order to solve this problem and at the same time maintain the special characters of the milieu in the cytosol and mitochondrial matrix, the cell has ingeniously evolved *specific transporters* and *shuttle systems*.

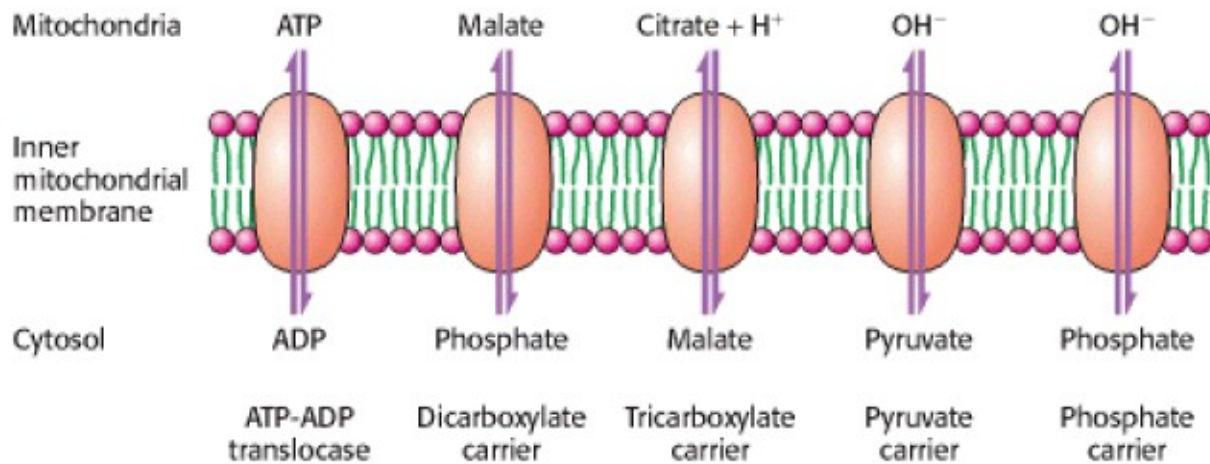


Fig. 4.5.2. Transport proteins (orange-colored ovals) in the inner mitochondrial membrane for exchange diffusion of charged ions and metabolites (Source: Berg *et al*, 2002, fig 18-40)

Transport proteins or **carriers** span the inner mitochondrial membrane and allow exchange of anions against OH^- and cations against H^+ ions. They enable selective uptake and output of ionized metabolites without altering the osmotic and electrical equilibrium on both sides of the membrane. Thus, the adenine nucleotide transporter allows ATP formed in the mitochondrial electron transport system to exit to the cytosol and supply energy there, while at the same time it ensures that ADP from the cytosol enters the mitochondrion for re-phosphorylation (Chapter 9). The pyruvate transporter brings in pyruvate, formed by glycolysis in the cytosol, for entry into the mitochondrial citric acid cycle (Chapter 5). However, if excess citrate accumulates inside the mitochondrion, the citrate transporter moves citrate out into the cytosol for lipogenesis (Chapter 6).

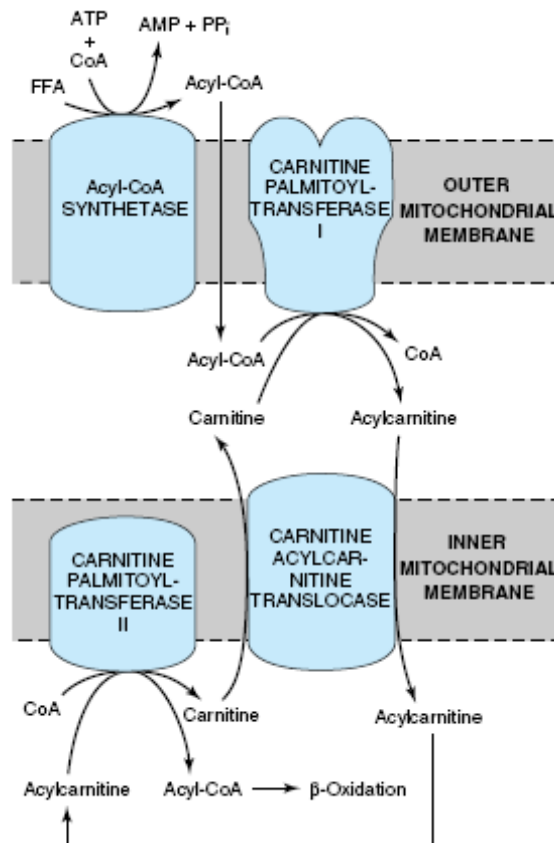


Fig.4.5.3. Carnitine-aided carrier system for transport of acyl-CoA from the cytosol into the mitochondrion.

(Source: Murray *et al*, 2003, p 181 fig 22-1)

Shuttle systems also help to transfer functional groups across the IMM. They operate through substrate pairs that are present on both sides of the membrane, linked by suitable enzymes and assisted often by carrier proteins. The malate-aspartate shuttle depicted below (fig 4.15), and the glycerophosphate shuttle for transfer of H⁺ and electrons across the IMM are discussed in detail in Chapter 5 Section 1.

In the same way other membrane-located receptors and transporters discriminate between the myriads of molecules that bombard the cell surface and the mitochondrial membranes, and allow specific binding and/or entry of only desirable molecules into the cell. You will come across many of them as you traverse the metabolic pathways.

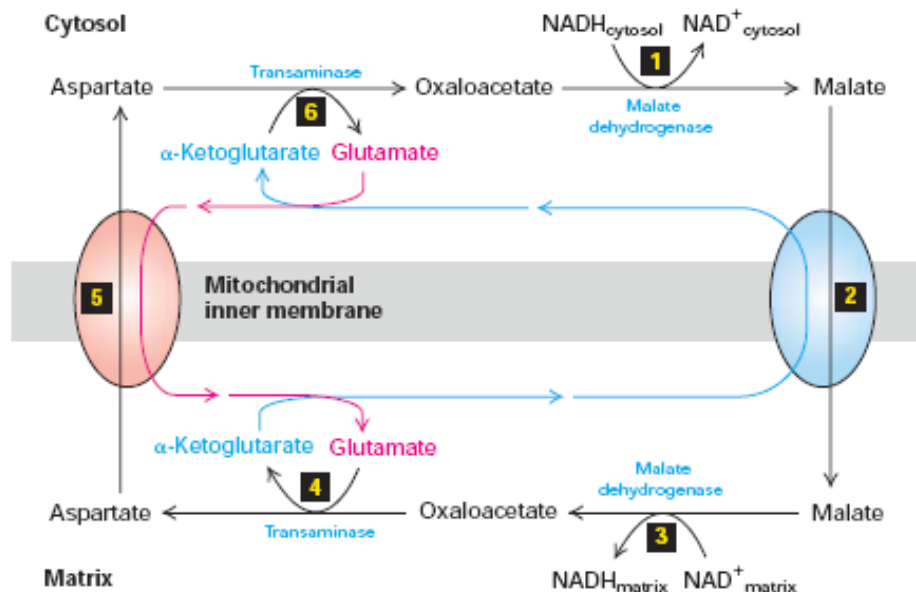


Fig. 4.5.4. The malate-aspartate shuttle: The numbers indicate successive reactions that enable transfer of reducing equivalents from the cytosol to the mitochondrial matrix. Blue and red ovals represent transporters in the mitochondrial inner mitochondrial membrane. (Source: Lodish *et al*, 2003, p 810 fig 3-11)

Chapter 4 Section 6

SPECIALIZED METABOLIC ROLES OF TISSUES

Different tissues and organs may have specialized metabolic roles. **Blood** is the vehicle for transport of water-soluble molecules like O_2 , CO_2 , nutrients, energy fuels, urea and regulatory hormones between different organs. The products of protein and carbohydrate digestion, namely amino acids and glucose, are absorbed by epithelial cells of the intestinal villi and carried by the hepatic portal vein from the gastrointestinal tract to the liver.

The **liver is central to the overall metabolism** in the body. It is the processing site for several crucial metabolic pathways that determine the blood concentrations of many metabolites. The demand of extrahepatic tissues for fuels and precursors, varies with the organ, nutritional state and level of activity. The liver shows remarkable flexibility in changing the activity and synthesis of its own enzymes to meet the requirements of the body. When blood glucose is high, the extrahepatic tissues slow down their uptake of glucose. The liver however, continues to take in and conserve this excess glucose as glycogen or fat by glycogenesis and lipogenesis respectively. When blood levels of glucose are low, the liver releases glucose for other tissues by carrying out glycogenolysis from its own stores, and gluconeogenesis (from non-carbohydrate sources), but it keeps its *own* rate of glucose utilization low. The Cori's cycle and the glucose-alanine cycle (Chapter 5) operating between the liver and muscle are excellent examples of such metabolic inter-relationship. When required, the liver can direct glucose into the pentose phosphate pathway to obtain $NADPH.H^+$ for synthesis of fatty acids and ribose 5-phosphate, and for detoxification of many drugs and xenobiotics.

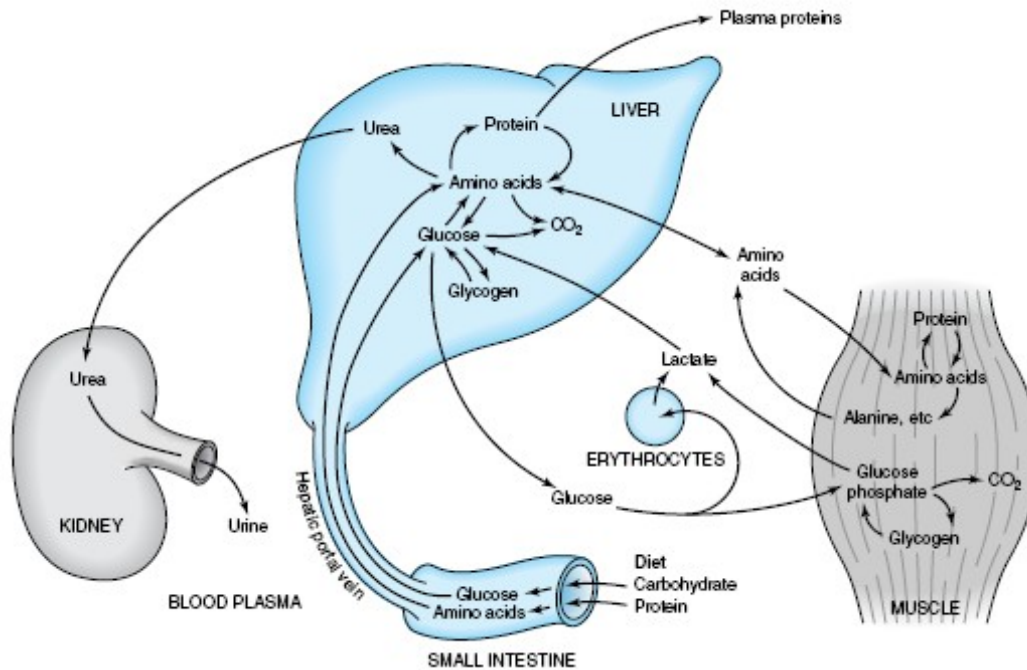


Fig. 4.6.1. Inter-relationship between different organs in carbohydrate and protein metabolism. The liver is central to many metabolic pathways (see text for details). (Source: Murray *et al*, 2003, p 125 fig 15-5)

The preferred energy fuel for the liver is fatty acids which it utilizes by β -oxidation like many other tissues. However, under conditions of starvation or a high-fat diet, the blood levels of glucose are low but fatty acids are in excess of the carrying capacity of blood. The liver then diverts the products of fatty acid oxidation in its cells into the formation of ketone bodies (ketogenesis) that can be used as fuel by other extrahepatic tissues but not by its own cells.

Toxic nitrogenous waste products like ammonia are produced by transamination and deamination in other tissues. The urea cycle in the liver converts two waste products viz. ammonia and CO_2 , to non-toxic urea which is then excreted by the kidneys. Most of the blood proteins like albumins and lipoprotein complexes (like VLDL), are also synthesized by the liver.

As can be expected under these circumstances, the liver has a very high turn-over rate of its own proteins.

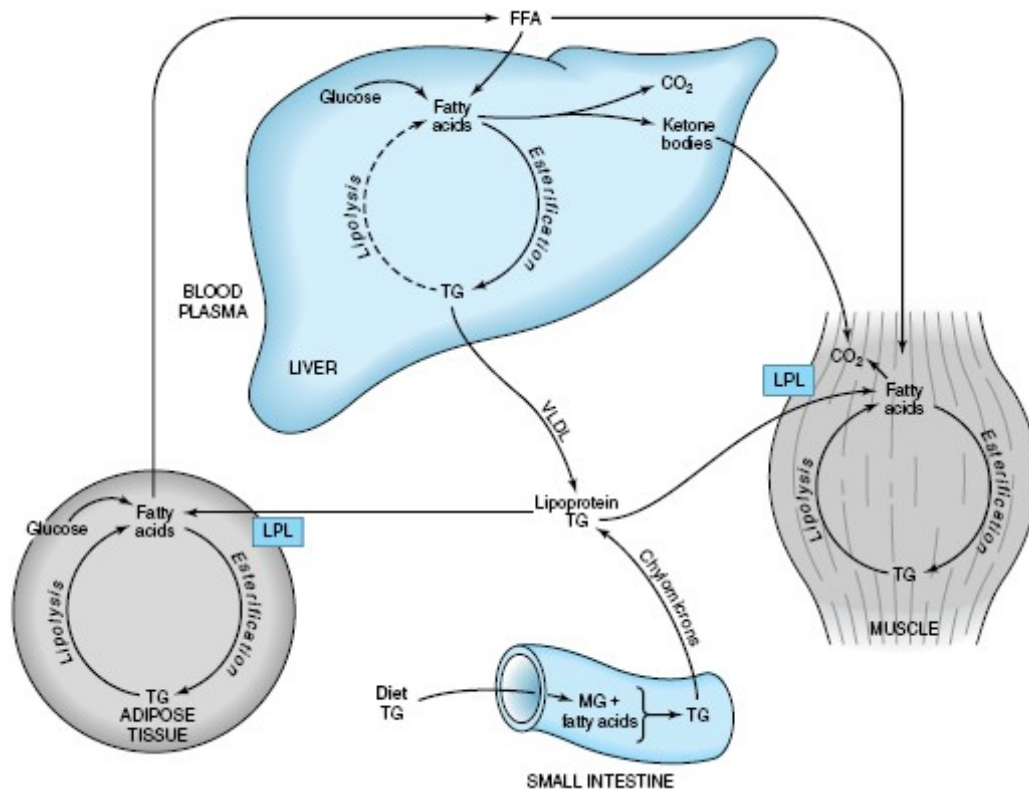


Fig 4.6.2. Differences in the metabolism of lipids in different organs (see text below).
(Source: Murray *et al*, 2003, p 126 fig 15-6)

The absorbed fatty acids and glycerol are processed differently in the gastrointestinal tract. The **intestinal cells** re-synthesize glycerides from them and package the products with proteins into lipoprotein complexes called chylomicrons. These are absorbed in the lacteals of the intestinal villi, transported by **lymph** and enter the blood circulation later. The blood carries the lipoprotein complexes to different tissues and fatty acids obtained from them undergo β -oxidation to generate energy. The **adipose tissue** synthesizes and stores triglycerides (lipogenesis), the main fuel reserve of the body. When required, it carries out lipolysis to release fatty acids for other tissues.

We take up the metabolism of glucose as an example of how the *same* compound may have *different* metabolic fates in different tissues. Most tissues of the body including **muscle** and **brain**, use glucose as energy fuel by aerobic glycolysis, releasing CO_2 into the blood. **Erythrocytes** and very **active skeletal muscle** obtain energy by anaerobic glycolysis, releasing lactate into the blood, which the liver converts to glucose by gluconeogenesis. Glucose is used for lipogenesis in **adipocytes** and for the pentose phosphate pathway in many tissues. The **liver** and **muscle** store excess glucose as glycogen, though unlike the liver, the muscle does so for its own utility. In times of need, amino acids from degraded muscle protein undergo gluconeogenesis to produce glucose in the **liver**.

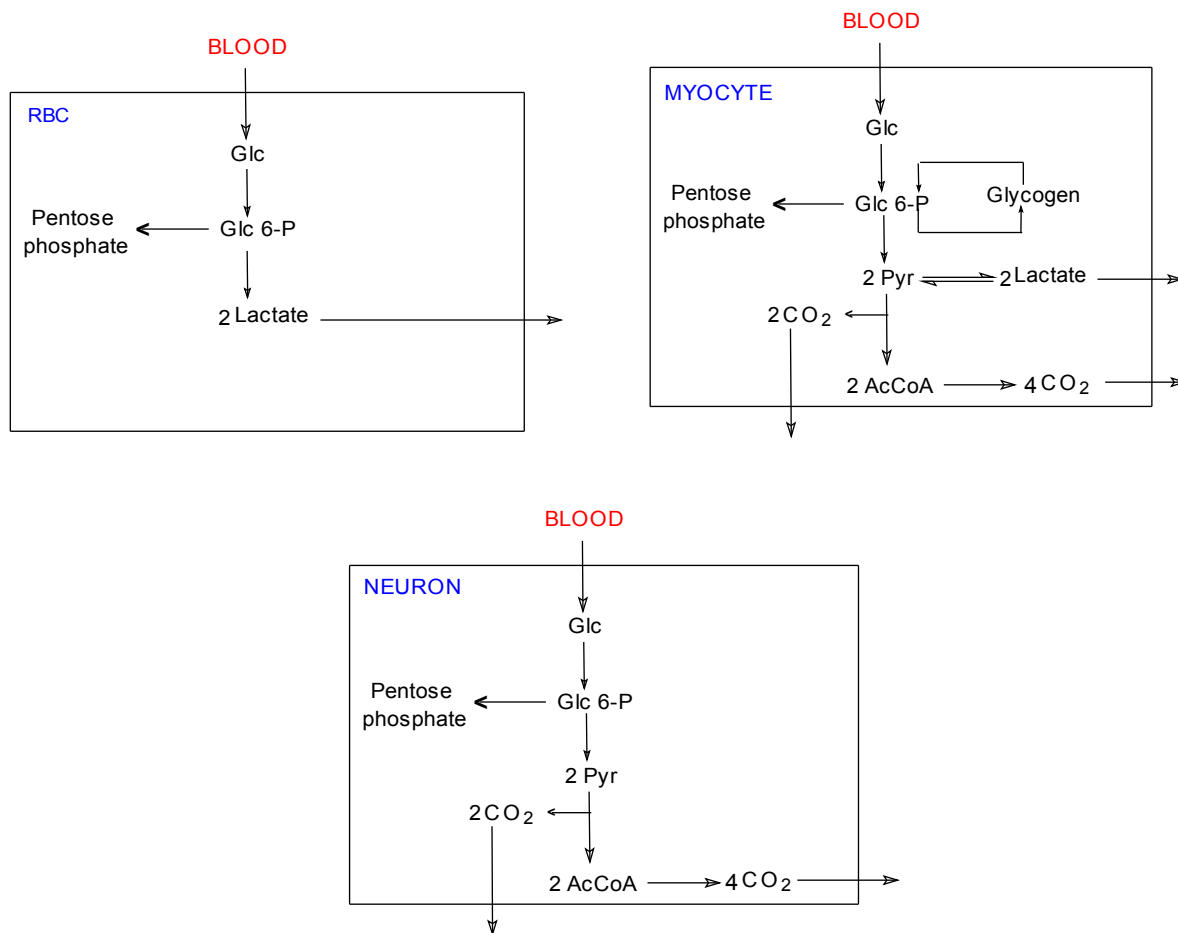


Fig 4.6.3. Differences in glucose metabolism in selected cell types of the body (Glc = glucose; Glc6P = glucose 6-phosphate; Pyr = pyruvate; AcCoA = acetyl-CoA)

Chapter 4 Section 7

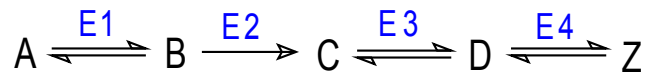
REGULATION OF METABOLIC PATHWAYS

Complex regulatory mechanisms have evolved at the molecular, cellular and organismic levels to ensure that the flux of intermediates in metabolic pathways is in the desired *direction* and at an appropriate *rate* at any one point in *time*.

We have seen above that even though the biosynthetic and degradative pathways of a molecule are generally separate, there are some common steps or cross-links between them, or with molecules of other pathways. The complex *network* of metabolic pathways requires **strict and co-ordinated regulation** both within the cell and in the whole body. At the same time, regulatory mechanisms must have **in-built flexibility** because a biological system must respond constantly to changes in its internal and external environment and thereby maintain its own homeostasis.

In a chain of biological reactions the potential control points are **non-equilibrium reactions**.

Consider a linear reaction sequence that starts with a compound A and terminates with the final product Z for which the different enzymes used are E1, E2, E3 and E4:



All the reactions, except B to C, are equilibrium reactions i.e. the forward and the backward reactions proceed at equal rates so that there is no *net* flux of metabolites in either direction. For the reaction B to C, catalyzed by the enzyme E2, the equilibrium lies far to the right and hence it is not readily reversible. Under the conditions of the cell if this reaction proceeds to completion, the entire sequence of reactions will proceed from left to right. The enzyme E2, catalyzing such a reaction, is the ideal point for regulating the entire sequence. This reaction is regarded as a **key reaction** and E2 is a **regulatory enzyme**.

Metabolic pathways are stimulated or inhibited by controlling the activity of **regulatory enzymes**. Such enzymes are usually located in the earlier part of a long metabolic sequence so that reactants are not unnecessarily wasted.

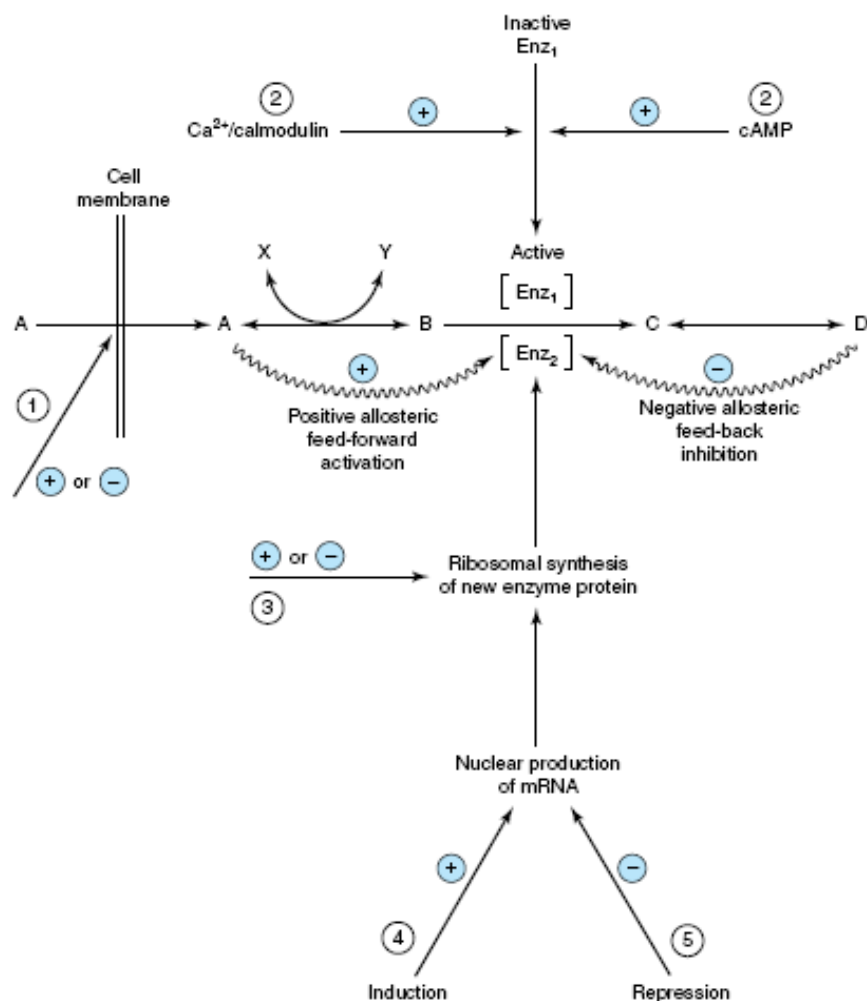


Fig 4.7.1. Mechanisms that control the activity of an enzyme, Enz_1 : The reaction sequence is from A to D in which B to C is the non-equilibrium reaction. Positive/negative symbols imply stimulation/inhibition respectively. The points of regulation are numbered from 1 to 5.
(Source: Murray *et al*, 2003, p 128 fig 15-8)

The **amount** of an enzyme, its **catalytic activity** and the **availability of its substrate(s)** are of paramount importance in determining the rate of a reaction. The *quantity* of enzyme available depends on the rate of transcription of its encoding genes and on the rate of its degradation. *Catalytic* activities of enzymes are regulated by **allosteric and covalent modulation/modifications** of their structures (Chapter 3). Such changes may depend on the feedback of reaction products, or may involve extracellular signals, often amplified by intracellular enzyme cascades that lead to phosphorylation/dephosphorylation of the target enzyme protein.

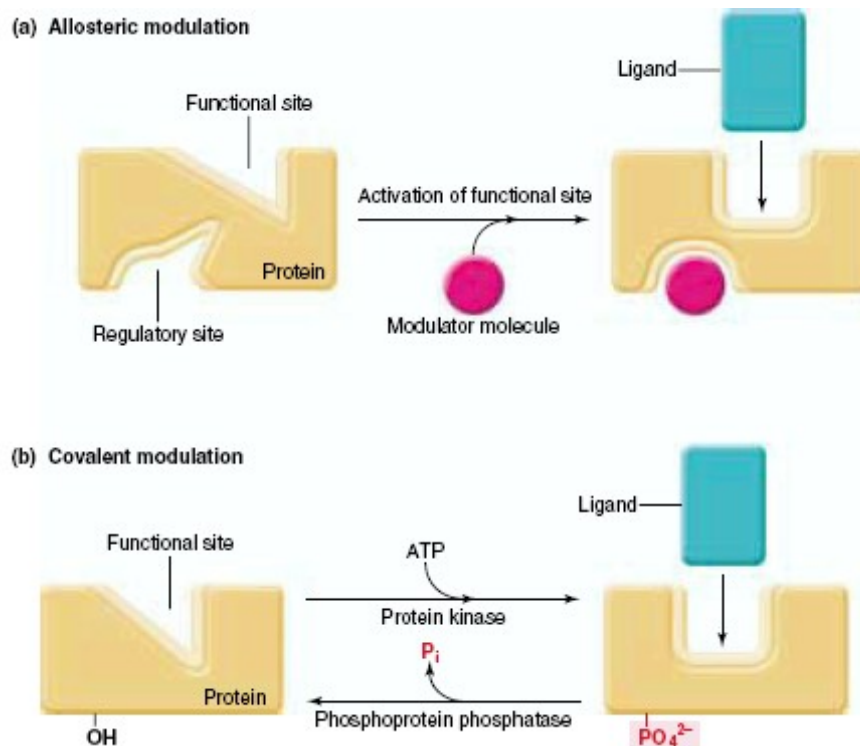


Fig. 4.7.2. Schematic representations of allosteric and covalent modulations of the active (functional) site of an enzyme protein. (Source: Vander *et al*, 2001, p 58 fig 4-7)

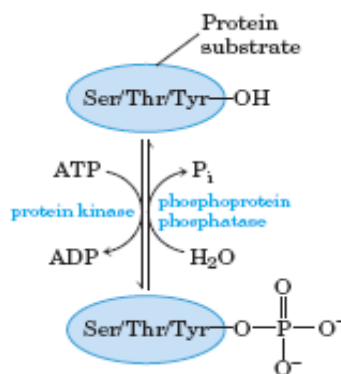


Fig 4.7.3. Covalent modification of an enzyme protein by reversible phosphorylation of a functional group in a constituent amino acid. (Source: Nelson and Cox, 2003, p 574 fig 15-14)

The *availability of substrate* may depend on transport mechanism in membranes or on the rate of its generation in a previous reaction. Reaction rates are also influenced by the routing of substrates into appropriate sub-cellular compartments. This is because different compartments have different concentrations of substrates, products and other effectors required by the various enzymes.

Time factors in such enzyme regulations may be different. Allosteric action is by local molecules (e.g. ATP, Ca²⁺) and is effective within milliseconds. Covalent modification often involves reversible phosphorylation. It is usually triggered by an extracellular factor like a hormone, and may require seconds or minutes. The induction or repression of the genes that enable enzyme synthesis, may take up to several hours or days.

Different metabolic pathways may intersect at the level of certain small metabolites like acetyl-CoA, keto-acids and glyceraldehyde 3-phosphate. The rate of reactions in one metabolic pathway will then be affected by the **generation/removal of its intermediates** by other pathways. The cell or the body makes regulatory adjustments depending on the immediate need.

When both anabolic and catabolic pathways are linked through a common product (e.g. pyruvate is the end-product of glycolysis and also a precursor for gluconeogenesis), they are likely to include reversible reactions catalyzed by common enzymes. However, there will always be at least one or a few non-equilibrium reactions for which the catabolic and anabolic routes require separate enzymes. The latter will be the targets for regulation and will decide the direction of the reaction sequence. You will come across **reciprocal regulation** of anabolic and catabolic pathways in glucose and fatty acid metabolism (Chapters 5 and 6 respectively).

When a pair of reactions functions in opposite directions, with one of them using a substrate and the other regenerating it, the phenomenon is known as **substrate cycling**. This was regarded as imperfect metabolism and so designated as “*futile*”. As stated above, such pairs of reactions are not simultaneously active in cells and are reciprocally controlled by allosteric or covalent factors. However, a limited degree of substrate cycling does occur in many pairs of irreversible reactions and may help in amplification of metabolic signals and in thermogenesis.

The **energy charge of a cell**, which indicates the relative amounts of intracellular ATP, ADP and AMP, plays a critical role in regulating many metabolic pathways. A cell in a high-energy state will not be conducive to catabolism but is likely to exhibit anabolism, and *vice versa*.

The concentration of ATP in a typical cell is approximately 5mM. The cell always endeavours to keep the concentration and supply of ATP constant. Accordingly, regulatory mechanisms respond to changes in [ATP], and are even more sensitive to changes in [AMP]. The ratios of the concentrations of co-factors like NADH/NAD^+ , NADPH/NAD^+ and **acetyl-CoA/CoA** also affect the regulatory enzymes.

The flux of substances in a metabolic pathway is finally linked to the *general state of the body*. Regulation depends on the **nutritional state**, and on the endocrine/paracrine/autocrine mechanisms of action of **hormones** and **growth factors**. Insulin, glucagon, cortisol, epinephrine, thyroxine and growth hormone are some of the hormones critical to successful metabolic balance in the body.

Summary 4: OVERVIEW OF METABOLISM

Metabolism is the sum total of all catabolic and anabolic reactions in the body.

Metabolic pathways are multi-step sequences of enzyme-catalyzed reactions in which the product of one reaction is the substrate for the next reaction in the sequence. Different pathways intersect to form an integrated and purposeful network which collectively constitutes metabolism.

Catabolism breaks complex biomolecules into smaller units. The overall pathways comprise three stages and are essentially oxidative, convergent and exergonic. Anabolism involves synthesis of complex biomolecules or performance of other cellular actions. The pathways are essentially reductive, divergent and endergonic. Amphibolic pathways like the citric acid cycle, link catabolic and anabolic pathways.

Metabolic pathways involve energy transactions and the ATP-ADP cycle plays a key role in energy transfers.

Other key molecules in metabolism are the nicotinamide and flavin co-enzymes, which can undergo reversible oxidation-reductions. Coenzyme A is a carrier of the acetyl group which is a crucial intermediate in many pathways.

Metabolic reactions are conducted in specific cellular compartments/organelles.

The liver plays a major role in metabolism. Other organs like the muscle, intestine, kidney, adipose tissue, brain and blood have specialized metabolic functions.

Metabolic pathways are regulated by control of their key enzymes through allosteric modulation, covalent modification and induction/repression of the genes controlling their synthesis.

Credits for illustrations in Chapter 4

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