Compartmentalization of Metabolic Pathways-Notes.15/06/2021-S.M

For Eukaryotic cell to properly function, they are divided into intracellular space in sub cellular compartments, each harboring specific metabolic activities. Compartmentalization of metabolic pathways is a prerequisite for certain cellular functions.

All reactions occurring in cells take place in certain space – **compartment**, which is separated from other compartments by means of **semi permeable membranes**. They help to separate even chemically quite heterogeneous environments and so to optimize the course of chemical reactions.

Enzymes catalyzing individual reactions often have different temperature and pH optimums and if there was only one cellular compartment a portion of enzymes would probably not function or them-catalyzed reactions would not be sufficiently efficient. By dividing the cellular space, **optimal conditions** for individual enzymatically catalyzed reactions are created.

At the same time, cell also protects itself against the activity of **lytic enzymes**. For example, sealing the cellular digestion in lysosomes prevents an unwanted auto-digestion of other organelles within cell. Common processes that accompany the disruption of some of the compartments (like spilling the content of lysosomes or mitochondria) are **necrosis** or activation of **apoptosis** (the process of programmed cell death).

Compartmentalization affects the **regulation of metabolic** pathways as well, making them more accurate and targeted and less interfering with each other. It is sometimes possible to regulate the course of the reaction at the point of **entry of** particular **substrate into the compartment** (transport across the membrane, often mediated by transport mechanisms).

Despite its advantages, compartmentalization at the same time puts greater demand on the energy consumption. It arises from a frequent need to use ATP-dependent transporters, transporting substances across membranes against the concentration gradient and thus creating different environments in different compartments.

Compartmentalization of metabolic pathways:

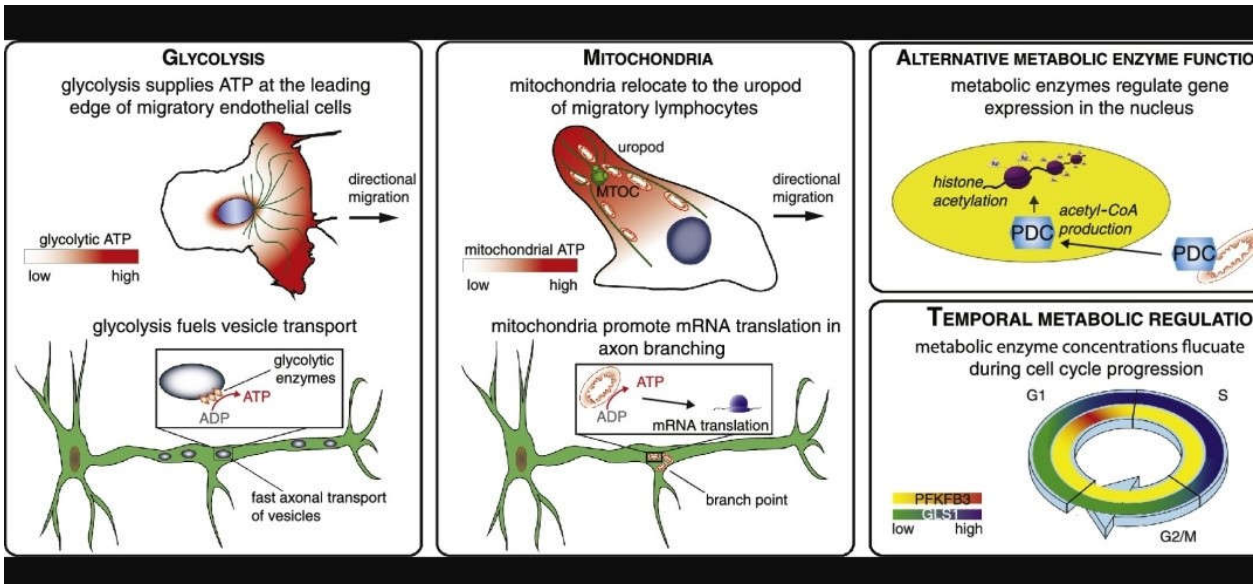
Cytosol (Cytoplasm without organelles):

1. Metabolism of saccharides: glycolysis, part of gluconeogenesis, glycogenolysis and synthesis of glycogen, phosphate pentose cycle.
2. Metabolism of fatty acids: FA synthesis.

3) Metabolism of amino acids: synthesis of nonessential AA, some of the transamination reactions

4) Other pathways: parts of heme and urea synthesis pathways, metabolism of purines and pyrimidines

Mitochondria:
1) Metabolism of saccharides: PDH, part of gluconeogenesis (conversion of pyruvate to OAA)
2) Metabolism of fatty acids: beta-oxidation of FA (Linen's spiral), synthesis (hepatocytes only) and degradation (extra hepatic tissues) of ketone bodies.



Glycolysis. This sequence of reactions in the cytosol converts one molecule of glucose into two molecules of pyruvate with the concomitant generation of two molecules each of ATP and NADH. The NAD^+ consumed in the reaction catalyzed by glyceraldehyde 3-phosphate dehydrogenase must be regenerated for glycolysis to proceed. Under anaerobic conditions, as in highly active skeletal muscle, this regeneration is accomplished by the reduction of pyruvate to lactate. Alternatively, under aerobic conditions, NAD^+ is regenerated by the transfer of electrons from NADH to O_2 through the electron-transport chain. Glycolysis serves two main purposes: it degrades glucose to generate ATP, and it provides carbon skeletons for biosyntheses.

> *Phosphofructokinase, which catalyzes the committed step in glycolysis, is the most important control site*. ATP is both a substrate in the phosphoryl transfer reaction and a regulatory molecule. A high level of ATP inhibits phosphofructokinase—the regulatory sites are distinct from the substratebinding sites and have a lower affinity for the nucleotide. This inhibitory effect is enhanced by citrate and reversed by AMP. Thus, the rate of glycolysis depends on the need for ATP, as signaled by the ATP/AMP ratio, and on the availability of building blocks, as signaled by the level of citrate. *In liver, the most important regulator of phosphofructokinase activity is fructose 2,6-bisphosphate* (F-2,6-BP). Recall that the level of F-2,6-BP is determined by the activity of the kinase that

forms it from fructose 6-phosphate and of the phosphatase that hydrolyzes the 2-phosphoryl group. When the blood-glucose level is low, a glucagon-triggered cascade leads to activation of the phosphatase and inhibition of the kinase in the liver. The level of F-2,6-BP declines and, consequently, so does phosphofructokinase activity. Hence, glycolysis is slowed, and the spared glucose is released into the blood for use by other tissues.

2.

Citric acid cycle and oxidative phosphorylation. The reactions of this common pathway for the oxidation of fuel molecules—carbohydrates, amino acids, and fatty acids—take place inside mitochondria. Most fuels enter the cycle as acetyl CoA. The complete oxidation of an acetyl unit by the citric acid cycle generates one molecule of GTP and four pairs of electrons in the form of three molecules of NADH and one molecule $FADH_2$. These electrons are transferred to O_2 through the electron-transport chain, which results in the formation of a proton gradient that drives the synthesis of nine molecules of ATP. The electron donors are oxidized and recycled back to the citric acid cycle only if ADP is simultaneously phosphorylated to ATP. *This tight coupling, called respiratory control, ensures that the rate of the citric acid cycle matches the need for ATP.* An abundance of ATP also diminishes the activities of two enzymes in the cycle—*isocitrate dehydrogenase* and *α-ketoglutarate dehydrogenase.* The citric acid cycle has an anabolic role as well. In concert with *pyruvate carboxylase,* the citric acid cycle provides intermediates for biosyntheses, such as succinyl CoA for the formation of porphyrins and citrate for the formation of fatty acids.

3.

Pentose phosphate pathway. This series of reactions, which takes place in the cytosol, consists of two stages. The first stage is the oxidative decarboxylation of glucose 6-phosphate. Its purpose is the production of NADPH for reductive biosyntheses and the formation of ribose 5-phosphate for the synthesis of nucleotides. Two molecules of NADPH are generated in the conversion of glucose 6-phosphate into ribose 5- phosphate. The dehydrogenation of glucose 6-phosphate is the committed step in this pathway. This reaction is controlled by the level of $NADP^+$, the electron acceptor.

The second stage of the pentose phosphate pathway is the nonoxidative, reversible metabolism of five-carbon phosphosugars into phosphorylated three-carbon and six-carbon glycolytic intermediates. Thus, the nonoxidative branch can either introduce riboses into glycolysis for catabolism or generate riboses from glycolytic intermediates for biosyntheses.

4.

Gluconeogenesis. Glucose can be synthesized by the liver and kidneys from noncarbohydrate precursors such as lactate, glycerol, and amino acids. The major entry point of this pathway is pyruvate, which is carboxylated to oxaloacetate in mitochondria. Oxaloacetate is then metabolized in the cytosol to form phosphoenolpyruvate. The other distinctive means of gluconeogenesis are two hydrolytic steps that bypass the irreversible reactions of glycolysis. *Gluconeogenesis and glycolysis are usually reciprocally regulated so that one pathway is minimally active while the other is highly active.* For example, AMP inhibits and citrate activates fructose 1,6-bisphosphatase, an essential

enzyme in gluconeogenesis, whereas these molecules have opposite effects on phosphofructokinase, the pacemaker of glycolysis. Fructose-2,6-bisphosphate also coordinates these processes by inhibiting fructose 1,6-bisphosphatase. Hence, when glucose is abundant, the high level of F-2,6-BP inhibits gluconeogenesis and activates glycolysis.

5.

Glycogen synthesis and degradation. Glycogen, a readily mobilizable fuel store, is a branched polymer of glucose residues.In glycogen degradation, a phosphorylase catalyzes the cleavage of glycogen by orthophosphate to yield glucose 1-phosphate, which is rapidly converted into glucose 6-phosphate for further metabolism. In glycogen synthesis, the activated intermediate is UDP-glucose, which is formed from glucose 1-phosphate and UTP. Glycogen synthase catalyzes the transfer of glucose from UDP-glucose to the terminal glucose residue of a growing strand. *Glycogen degradation and synthesis are coordinately controlled by a hormonetriggered amplifying cascade so that the phosphorylase is active when synthase is inactive and vice versa.* Phosphorylation and noncovalent allosteric interactions regulate these enzymes.

6.

Fatty acid synthesis and degradation. Fatty acids are synthesized in the cytosol by the addition of two-carbon units to a growing chain on an acyl carrier protein. Malonyl CoA, the activated intermediate, is formed by the carboxylation of acetyl CoA. Acetyl groups are carried from mitochondria to the cytosol as citrate by the citrate-malate shuttle. In the cytosol, citrate is cleaved to yield acetyl CoA. In addition to transporting acetyl CoA, *citrate in the cytosol stimulates acetyl CoA carboxylase, the enzyme catalyzing the committed step. When ATP and acetyl CoA are abundant, the level of citrate increases, which accelerates the rate of fatty acid synthesis.*

A different pathway in a different compartment degrades fatty acids. Carnitine transports fatty acids into mitochondria, where they are degraded to acetyl CoA in the mitochondrial matrix by β-oxidation. The acetyl CoA then enters the citric acid cycle if the supply of oxaloacetate is sufficient. Alternatively, acetyl CoA can give rise to ketone bodies. The $FADH_2$ and NADH formed in the β-oxidation pathway transfer their electrons to O_2 through the electron-transport chain. Like the citric acid cycle, β-oxidation can continue only if NAD^+ and FAD are regenerated. Hence, *the rate of fatty acid degradation also is coupled to the need for ATP*. Malonyl CoA, the precursor for fatty acid synthesis, inhibits fatty acid degradation by inhibiting the formation of acyl carnitine by carnitine acyl transferase 1, thus preventing the translocation of fatty acids into mitochondria.