NOTES TO SACCHARIDE METABOLISM

Prof. MUDr. Stanislav Štípek, DrSc.
Institute of Medical Biochemistry
First Faculty of Medicine, Charles University in Prague

Purposes of metabolism

- The major role is to oxidize food to provide energy in the form of ATP
- Food molecules are converted to new cellular material and essential components
- Waste products are processed to facilitate their excretion in the urine
- Some specialized cells in human babies oxidize food to generate heat. (Heat is also by-product of metabolism)

Storage of food in the body

- Animals take in food at periodic intervals
- Blood glucose increases from 5 to 7.8 mM in an hour after a meal
- Foods are stored in various cells (organs) to be used later (during starvation)
- During starvation the fuel is mobilized and moved in the body

Glycogen synthesis

- Energy injected from ATP by ***hexokinase*** and ***glucokinase***
- ***UDP-glucose*** as activated glucose donates the glucosyl group to non-reducing end of glycogen
- ***Branching enzyme*** transfers block of about 7 glucosyl units to C6-OH of glucose in the chain
- ***Glycogen synthase*** is under hormonal control

Glycogen degradation

- Glycogen breakdown occurs at the non-reducing ends by ***phosphorolysis***, not by ***hydrolysis***
- It is catalyzed by ***glycogen phosphorylase, which is under hormonal control***
- ***Debranching enzyme*** transfers three of the glycoside units of the branch to the 4-OH of another chain and it also clips (hydrolyses) off the 1 $\rightarrow$ 6 linked glucose.
- Glucose is released from resulting G-6-P by ***glucose-6-phosphatase***

Glycogen metabolism

- The synthesis and degradation pathways are different and therefore independently controllable.
- Glucose-6-phosphatase is found in liver but not in muscle or adipose tissues, which therefore cannot release glucose into the blood. Only the kidney, among other tissues, has the enzyme.
- In extrahepatic cells (as well as in liver) glucose-6-phosphate is on the path to glucose oxidation.
- The effect of insulin on cells is to activate glycogen synthesis and inactivate glycogen breakdown. The effect of glucagon on cells is the reverse one.

Energy production from foodstuffs
(a preliminary overview)

- **Energy production from glucose (glucose oxidation)**

* Glycolysis ... splitting of glucose into two C3 fragments (pyruvates) ***in cytoplasm***
* Tricarboxylic acid cycle ... conversion of carbon atoms two and three of pyruvate to CO_2 ***in mitochondria***
* Electron transport system..... transfer of electrons from the glucose metabolites via electron carriers to oxygen ***in inner membrane of mitochondria***

Why does liver have glucokinase and the other tissues hexokinase ? ? ?

- In starvation, when glucose supply in the blood is the all-important problem, the entry of glucose into muscle and other tissues is restricted because of the lack of insulin, while entry of glucose into brain, liver and blood cells is not insulin dependent
- Salvation of the following illogical situation: The liver synthesizes glucose from amino acids supplied by the muscle so that it can keep the blood glucose level up to permit normal brain function and it ***would not make sense for the liver to take up glucose in competition with the brain***

Stoichiometry of the citric acid cycle

Acetyl-S-CoA + $2H_2O$ + $3NAD^+$ + FAD + GDP + Pi →

$2CO_2$ + 3NADH + $3H^+$ + $FADH_2$ + CoA-SH + GTP

$\Delta G^{o'} = -40$ kJ mol^{-1}

Cycle operates in a **unidirectional way** because of three irreversible reactions:

- synthesis of citrate $\Delta G^{o'} = -32.2$ kJ mol^{-1}
- decarboxylation of isocitrate $\Delta G^{o'} = -20.9$ kJ mol^{-1}
- decarboxylation of α-ketoglutarate $\Delta G^{o'} = -33.5$kJ mol^{-1}

Inner membrane of mitochondria

* **Transfer of electrons** via electron carriers (*respiratory chain*) to oxygen in the membrane
* **Proton pumping** from the matrix to the cytosolic site of the inner mitochondrial membrane (proton gradient). *Protonmotive force* = pH gradient + membrane potential
* Protons flow through *ATP synthase* and power the **synthesis of ATP.**

The balance sheet of ATP production by electron transport

- The F_0 complex has 12 c subunits in mammals: one complete turn means flow of 12 protons.
- The F_1 part has three ATP-producing sites: one complete turn of the shaft means synthesis of three ATP from ADP + Pi
- **...4 H^+ have to be pumped out of the matrix for production of 1 ATP**
- Transport of 2 electrons from NADH to oxygen (complexes I, III, IV) pumps 10 protons, from $FADH_2$ to oxygen (complexes III, IV) 6 protons .
- **...Oxidation of 1 NADH produces 2.5 ATP ... P/O ratio**
- **...Oxidation of 1 $FADH_2$ produces 1.5 ATP ... P/O ratio**
- Cytoplasmic NADH efficiency is dependent on the type of shuttle !! It produces 1.5 - 2.5 molecules of ATP.

Yield of ATP from oxidation of a molecule of glucose to CO_2 and H_2O

- Glycolysis of free glucose 2 ATP from substrate level
- Glycolysis of free glucose 5 or 3 ATP from 2 NADH (shuttles)

- Pyruvate dehydrogenase 5 ATP from 2 NADH
- Citric cycle 2 ATP from succinyl-S-CoA
- Citric cycle 15 ATP from 6 NADH
- Citric cycle 3 ATP from 2 $FADH_2$

- **Total 30 - 32 ATP from 1 free glucose**

Synthesis of glucose (gluconeogenesis)

- Starting point is **pyruvate**. Acetyl-CoA cannot be converted to pyruvate and therefore fatty acids cannot be converted into glucose
- **Three reactions of glycolysis are irreversible** hexokinase, phosphofructokinase and pyruvate kinase reaction. They cannot be used for gluconeogenesis.
- Synthesis of phosphoenolpyruvate requires 2 high energy -P in **pyruvate carboxylase** and **PEP carboxykinase** reactions

Sources of pyruvate used by the liver gluconeogenesis

- Muscle amino acids (**alanine**)
- Muscle **lactate**
- **Glycerol** from triacylglycerol hydrolysis

Pentose phosphate pathway
Hexose monophosphate shunt
Direct oxidation pathway of glucose

- It supplies **ribose-5-phosphate** for nucleotide and nucleic acid synthesis
- It supplies **NADPH** for fat synthesis (liver, adipose cells) and for defense antioxidant reactions (red blood cells, reduction of glutathione, reduction of methemoglobin)
- It provides a route for excess pentose sugars in diet to be brought into the mainstream of glucose metabolism
- The pathway has two main parts, the oxidative step (conversion of hexose to pentose and reduction of $NADP^+$ to NADPH) and non-oxidative section (interconversions of C3, C4, C5, C6 and C7 monosaccharides (***transaldolase and transketolase***).

Xu5P + R5P = GAP + S7P

C5 + C5 = C3 + C7

S7P + GAP = E4P + F6P

C7 + C3 = C4 + C6

Xu5P + E4P = GAP + F6P

C5 + C4 = C3 + C6

2Xu5P + R5P = GAP + 2F6P

3C5 = C3 + 2C6

3R5P = GAP + 2F6P

Control of carbohydrate and fat metabolism

•The energy needs of organism vary in various situations

•Metabolic pathways need to work in different directions after a meal when metabolites are being stored as compared with intervals between meals when storage metabolites are being utilized

How are enzyme activities controlled ?

•to change the **amount** of enzyme (synthesis of the new enzyme molecules) .... slow

•to change the **rate of catalysis** of the enzyme (***allosteric control*** or covalent modification - ***phosphorylation***) ... rapid

Allosteric control

•Ligand (allosteric effector or moderator with a different structure than substrate) binds to binding site other than for substrate (“allo” = other) and modifies the activity of the enzyme.

•Therefore: The allosteric effector need not have any relationship whatsoever to the substrate of the enzyme regulated (and usually does not). This means that any metabolic pathway can be connected in a regulatory manner to any other metabolic area.

Control of enzyme activity by phosphorylation

Covalent modification of the enzyme : phosphorylation on -OH group of serine or threonine (ATP, protein kinase). In consequence of that the enzyme undergoes a conformational change such that its activity is modified (increased or decreased).

Two classes of controls of metabolic pathways

Intrinsic control - internal (in the cell) - largely allosteric. These are the automatic controls in which metabolites signal to other pathways and parts of their own pathway, so that a smoothly running chemical machine results (without any metabolic pile-ups and/or shortages).

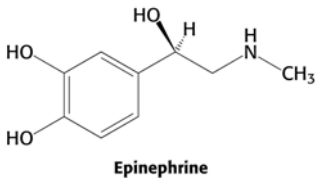
Extrinsic control (external to the cell, internal to the organism). The signals (hormones, neurotransmitters) instruct cells on what their major metabolic direction should be - such as whether to store fuel or release it.

Extrinsic (hormonal) control of carbohydrate and fat metabolism in the body

Insulin - small protein secreted by B-cells of the pancreatic islets of Langerhans when blood glucose concentration rises (after meal)

Glucagon (hunger hormone, small protein) is produced by A-cells of the pancreatic islets of Langerhans when blood glucose is low.

Epinephrine (derivative of tyrosine) is liberated by the adrenal medulla following a stress



Epinephrine

Norepinephrine (derivative of tyrosine) is liberated by sympathetic nerve endings (preparation for fight or defense)

Glucagon (hunger hormone, small protein) supports breakdown of food storage and releasing of the energy

Insulin does the opposite - the storage of the glucose and fat.

Summary of the glycogen phosphorylase control.

•In the absence of hormone stimulation, phosphorylase is in the unphosphorylated 'b' form, which is inactive unless allosterically (partially) activated by the presence of AMP (not cAMP). This activation does not involve phosphorylation of the protein.

•In normal muscle contraction, Ca^{2+} ions are released into the cell by the motor neuron signal; they allosterically partially activate phosphorylase b kinase; and this results in a partial activation of the phosphorylase. Unlike the cAMP-induced activation of the phosphorylase b kinase, the Ca^{2+} activation of this enzyme does not involve phosphorylation and occurs only as long as the muscle is contracting because the Ca^{2+} is immediately removed on cessation of the neuronal signal.

Summary of the glycogen phosphorylase control (continuation)

•Epinephrine in muscle increases the level of cAMP.

•cAMP allosterically activates PKA, which, in turn, activates phosphorylase b kinase. The latter phosphorylates the 'b', form of phosphorylase, producing the active 'a' form, which does not require AMP for activation. The process is an amplifying cascade.

•Phosphoprotein phosphatase I is capable of converting the 'a' form back to the 'b' form but as long as cAMP is present to activate PKA, the latter activates a phosphatase inhibitor protein so that inactivation of phosphorylase occurs only after removal of the hormonal signal.

Control of glycolysis in muscle

In muscle PFK_1 must not be inhibited when epinephrine is released, since maximum glycolysis is needed in emergency (in contrary to glycolysis in liver). It is reported that, in the presence of epinephrine, the level of the fructose-2:6-bisphosphate increases.

Speculation: It may be that the increase is due to the increased level of substrate for PFK_2 (fructose-6-phosphate) resulting from cAMP-induced glycogen breakdown. Fructose-6-phosphate is known to activate PFK_2.

Intrinsic control of citric cycle and electron transport area

Major internal controls are the availability of NAD^+ and ADP as substrates. Allosteric control also exists.

High NADH/ NAD^+ ratio inhibits the dehydrogenases in the cycle.

When ADP/ATP ratio is low, electron transport is inhibited because oxidation and phosphorylation are tightly coupled. This tight coupling is called **respiratory control**.

In addition to these controls ATP inhibits citrate synthase; and ATP inhibits and ADP stimulates isocitrate dehydrogenase. Succinyl-CoA and NADH inhibit α-ketoglutarate dehydrogenase.

According to
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