

Biochemistry 401 Lecture 40

Today we're going to wrap it up with our last lecture, metabolism in a nutshell.

Now one of the things that we discussed at great length was ATP. We saw ATP as a reactant. We saw ATP as a product. We saw how ATP was made. ATP is used as a universal energy source. It's used for such things as muscle contraction, in which we're going to transduce chemical energy to mechanical energy. It's used for active transport. It takes energy to transport molecules against their concentration gradient, and this active transport is necessary to allow cells to do their jobs properly. By having ions that be more concentrated on one side of the membrane than the other, we can have nerve impulses; we can have muscle contraction. ATP is also necessary for signal transduction and amplification in second messenger system signaling cascades. ATP is important for biosynthesis. We rely on ATP to support energetically unfavorable reactions so that our pathway can go forward. A thermodynamically unfavorable reaction can be made favorable by coupling the reaction to ATP hydrolysis.

ATP production relies on the oxidation of fuel molecules. We oxidize fuel to gain electrons with high electron transfer potential. These electrons are transferred from one electron carrier to the next in the electron transport chain, at each step, releasing energy that's used to pump protons against their concentration gradient to establish a chemiosmotic gradient. The ultimate electron acceptor is molecular oxygen, and this produces H_2O . The energy from electron transfer sets up the chemiosmotic gradient that is then released as protons flow into the matrix. It is the flow of protons back into the matrix by ATP synthase that powers the release of ATP, and allows ATP synthesis to occur. Now the fuels that we use include glucose, and the oxidation of glucose during glycolysis yields ATP, however, the complete oxidation of glucose yields 30 to 32 ATP molecules. The oxidation of palmitate, a 16-carbon fatty acid chain yields even more, and this is because palmitate is more reduced than glucose, and because there are more carbons of course. You can yield 106 ATP molecules.

Fuels are catabolized in an oxidative process and we use the electrons for energy to yield ATP. However, biosynthetic reactions are reductive, and they require electron carriers as well, in this case, NADPH. These synthetic reactions also require ATP. The electrons with high electron transfer potential that are needed for these reactions are supplied by NADPH not NADH, and this is

because of the relative concentrations of NADPH and NADP⁺ and NADH and NAD⁺. Now another fact is that many of the products that we need to make macromolecules in ourselves, and to make many of the small molecules that we need to carry out our daily affairs come from just a small number of starting molecules, and those that are in the TCA cycle and acetyl CoA and glycolysis are key players. So the major metabolic pathways that are catabolic also have anabolic purposes as well.

Anabolic and catabolic pathways are kept separate in many ways we don't just use the same pathways, running them backwards and forwards to get the products that we desire. We use different pathways. We use different enzymes in these pathways. Not all of them can go backwards. There are some very high-energy intermediates that just can't be made when you're trying to run the reaction backwards. This is a good thing. This makes sure that regulation plays a large role in ~~what pathways~~ may in which pathway is being undertaken. If we left it simply to the level of the substrates and reactants, we might find ourselves in major trouble, because it would be harder for the body to respond to global signals, if it were simply responding to the local signals in the cells. Anabolic and catabolic pathways are also compartmentalized in eukaryotes. One pathway occurs in the cytosol, another one occurs in the mitochondrion, or another one occurs in the ER, or another one occurs in the nucleus or in the lysosome, or in the peroxisome. There are many ways to keep pathways separate. There are many ways to regulate which pathways are going on, by regulating access to metabolites, access to enzymes. This separation of pathways allows for more fine control of these pathways, and we can have two enzymes that work in opposite directions respond in opposite ways to the same signal, for example, glycolysis and gluconeogenesis, glycogenolysis, and glycogenesis. These pathways are reciprocally regulated. Each of these pairs of pathways are reciprocally regulated, and they respond in opposite ways to the very same signals.

Chemical reactions in our bodies take advantage of enzymes. We aren't dependent solely on the concentrations of reactants for regulation and this allows for more fine control of the pathways. Now generally speaking, irreversible steps, especially the first irreversible reaction of the pathway, the first committed step, are usually regulated. The first committed step is a major decision point that decides which way that particular metabolite is going to go, what it's going to be used for, and so it's an excellent idea to make sure that you're going to use that

particular metabolite in the best way possible, in the most efficient way. Good use of resources depends on reacting to both local and global cues in the right way. When our bodies react in the wrong way that's when we have problems. That's when pathologies occur. Now control of enzymes occurs through allosteric interactions and covalent modifications to name a few. The allosteric interactions can include product inhibition, feed-forward regulation, and using an intermediate to make a potent activator. So something that is made just upstream of an enzyme can be used in a different way to synthesize a very potent activator. We saw this in glycolysis, where fructose 6-phosphate was phosphorylated to yield fructose 2,6-bisphosphate, which is a potent activator of phosphofructokinase 1 by using enzymes that are multimers. These enzymes can be more finely tuned by having them respond to many different allosteric regulators and have each subunit be turned up or be turned down and have effects on the neighboring subunits. Covalent modifications include phosphorylation, acetylation, methylation, ubiquitination. Such things as phosphorylation can have reciprocal effects on two separate pathways. So a phosphorylation cascade can activate one pathway and deactivate another. We can also have the same global signal effect many pathways in many tissues. For instance, glucagon stimulates lipoprotein lipase, stimulates glycogen phosphorylase, both of these signals mobilize stored energy for the needs of the body.

And so here we see a breakdown of where all the different pathways that we've studied occur. In the cytoplasm, glycolysis and the pentose phosphate pathway, and fatty acid synthesis occur. In the inner mitochondrial membrane we have oxidative phosphorylation, and in the mitochondrial matrix we have the citric acid cycle, the beta oxidation of fatty acids, and ketone body formation, and there's some pathways that are split, that have some reactions occurring in the cytoplasm and some that happen in the mitochondrion, and these include gluconeogenesis and the urea cycle.

So now we're going to go through some committed steps of pathways and it's a good idea for you to look at these and to be able to distinguish what pathway you're looking at. So here we see fructose 6-phosphate. The enzyme that plays a role is phosphofructokinase. This enzyme is activated by fructose 2,6-bisphosphate and AMP, and it's inhibited by ATP and citrate, and this reaction yields fructose 1,6-bisphosphate and ADP. So fructose 6-phosphate and ATP yield fructose 1,6-bisphosphate and ADP. Now just by looking at this you can tell