Seminar 1, Tuesday 8/3

Material and energy budget

1. In his studies of glycolysis in cancer, Otto Warburg noted that some tumors produce their own weight in lactate in as little as 8 hours. What is then the rate of glycolysis in molar amounts per hour? (Assume some typical wet weight of a mammalian cell.)

2. Energy content of nutrients is often measured by heat of combustion, which doesn't consider how efficient cells are at metabolizing the nutrient. One study estimates the actual yield of ATP to be about 10 mol ATP / MJ heat energy for sugar, and about 7 mol /MJ for protein. Given this data, how many ATP do you get from 1 glucose molecule, or one "average" amino acid?

Further reading: http://www.ncbi.nlm.nih.gov/pubmed/6689941

3. How long could a cell survive by oxidizing its own protein contents for energy?

4. How much ammonia would an average cell produce per hour if oxidizing only protein for energy? (Assume each amino acid catabolized releases 1 ammonium ion.) At this rate, how long does it take for ammonia to reach toxic levels in cell culture?

Ammonia in cell culture: http://www.ncbi.nlm.nih.gov/pubmed/8672289

5. How long could an adipocyte survive if it would oxidize all of its fat contents for energy? Assume that the fat is similar to palmitate, and yield of about 100 ATP for a palmitate molecule.

6. An average adult turns requires about 2500 kJ energy per day. How many ATP equivalents (Gibb's energy of hydrolysis) does this correspond to if translated directly? Do you think this is a realistic estimate of ATP turnover? Why / why not?

7. How much sugar would a person have to consume to generate 2500 kJ of energy per day from glycolysis? What is the average daily intake of sugar in adults?

8. ATP and other high-energy molecules are only intermediates in metabolism; ATP is ultimately used by cell to *do work*. In the end, where does all the energy consumed by cells (or people) go?

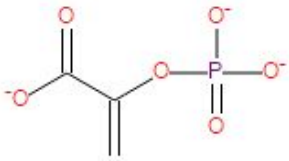
9. How long would you expect a cell to survive if ATP synthesis was completely blocked?

10.. Proliferating cells must renew all their components every cell cycle. Based on the biomass composition, what metabolites are in highest demand? What is their expected synthesis rate?

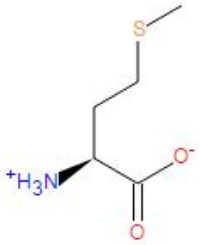
For your final report: What cell type(s) are you considering in your PhD work? What do you know / can you estimate about their mass, composition, biosynthesis, energy needs?

Oxidation and reduction

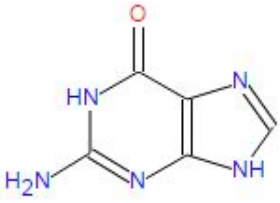
1. What is the definition of an oxidation number? What are the oxidation numbers for the various carbons in the metabolites below?



phosphoenolpyruvate

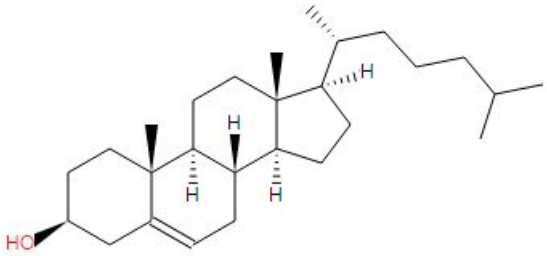


methionine



guanine

2. Cholesterol (below) is a rather complex, hydrophobic molecule which is synthesized by a complicated pathway, starting from acetyl-CoA. Without looking at this pathway, can you tell if it is oxidative or reductive? What electron carrier do you expect to be active in the pathway? Does the synthesis require energy?



3. Purine nucleotides like guanine (problem 1, above) are synthesized from amino acids and formyl (aldehyde) groups. Is this an oxidative or reductive pathway?

4. Is an aminotransferase a redox enzyme? (Are carbons being oxidized / reduced in aminotransferase reactions?) See for example alanine aminotransferase at humancyc.org. Does the reaction require energy?

5. Look up the pathway “serine and glycine biosynthesis” (SER-GLYSYN-PWY) in humancyc.org. Which reactions in this pathway are redox-reactions? What is being oxidized, reduced? Is energy required for serine synthesis?

6. What are the major electron carriers for oxidative and reductive metabolism, respectively? Why are the two processes separated? List the known enzymes using each electron carriers in humancyc.org. Do you recognize any pathways?

7. Look up the structure for NAD and identify the redox active site. Compare with the structure of NADP. What function does the extra phosphate group in NADP serve? What metabolites (moieties) does NAD(P) consist of?

8. How many electrons are transferred when NAD is reduced to NADH?

9. Flavin adenine dinucleotide (FAD) is another major electron carrier. How many electrons are transfered when it is reduced to $FADH_2$? What is the major difference in function between FAD and NAD(P) ?

10. The amino acid cysteine has a --SH group that is prone to conversion to cysteine dimers, --S-S--, in the presence of oxygen. Is this an oxidation or reduction? What atom is being oxidized / reduced ? What does this means for cells that need to keep --SH groups intact?

For your final report: In your favorite pathway, can you identify oxidation and reduction reactions? Are there electron carriers involved?

Carbohydrates

1. What is the yield of ATP and NADH when catabolizing glucose to lactate? Which are the ATP-yielding enzymes?

2. Consider the situation when pyruvate obtained in glycolysis is *not* converted to lactate, but is further oxidized in mitochondria. What happens with the cytosolic NADH/NAD?

3. Acid-base homeostasis is an important factor for glycolysis. Which reactions of glycolysis produce protons? What is the overall balance of H+ when glucose is converted to pyruvate, or to lactate?

4. Glycolysis really yields only a net of *one* ATP per glyceraldehyde (triose) molecule; the second ATP obtained in lower glycolysis is the reversal of an ATP-driven phosphorylation in upper glycolysis. In which reaction is energy (ATP) actually extracted from glyceraldehyde?

5. Some cells take up lactate from their surroundings as fuel, converting it to pyruvate which is then oxidized in the mitochondrion. What does this mean for NAD/NADH balance in the cytosol? How does it affect H+ balance?

6. Intermediates of the glycolytic pathway are also used for biosynthesis of other compounds. For example, dihydroxyacetone-phosphate can be converted to glycerol for synthesis of various lipids, and 3-phosphoglycerate can be converted to serine. What is the energetic cost when these compounds are “drained” from glycolysis?

7. What would (theoretically) happen to cellular pH if lactate dehydrogenase is blocked experimentally in a glycolytic cell? Consider all components of the reaction.

8. While glucose is the main blood sugar in humans, glyceraldehyde is the precursor of the energy-yielding steps in lower glycolysis. Would it not be more advantageous for a cell to take up and catabolize glyceraldehyde? Can you think of reasons why glyceraldehyde is *not* the main blood sugar?

9. Fructose is an important sugar in the human diet, on par with glucose in abundance. The initial steps in catabolism of fructose differs a bit from glucose. What are the key differences? Is the energy yield of fructose catabolism comparable to glucose?

10. Galactose is another dietary sugar. It is catabolized via the Leloir pathway. Lookup the pathway in humancyc. How does it compare to that of glucose or fructose? Are there corresponding enzymes? At what point do the pathways “connect” ? Compare with the bow-tie model of metabolism.

11. Glucose-6-phosphate pool can be obtained from glycogen instead of glucose. Lookup / draw the pathway leading from glycogen to glucose-6-phosphate. What is the ATP yield when catabolizing glycogen to lactate? Compare this with the yield of glycolysis from glucose. What is the difference?

12. There is one unique enzyme in glycogen metabolism that is able to phosphorylate sugar *without* using ATP. Which one? How is the reaction made possible?

13. The liver can contain 3--7% glycogen w/w in the fasted state. How much glucose does this represent? Compare with the blood glucose content.

See also: http://www.ncbi.nlm.nih.gov/pubmed/13331917

For your final report: Does your favorite pathway interact with carbohydrate metabolism? Does it involve cytosolic ATP or NADH, for example?