Dr. Mallery | Biology 150 – Workshop | Fall Semester

ENERGY and METABOLISM **ANSWERS**

IN THE GRAND SCHEME

The purpose of this workshop is to get the Learning Communities to begin thinking about why certain chemical reactions occur within cells and why there seems to be a directionality to biochemical pathways. It is not so important that the students know the names of the molecules in glycolysis, Krebs cycle, or the electron transfer chain, as it is that they understand that energy must be expended to perform all the processes that keeps a cell alive. The students need to consider that the real purpose of a biochemical pathways is to conservatively capture small amounts of energy into more useful molecules that can be selected for by a cell later to accomplish some other bit of work. Metabolic pathways have evolved as they have because they provide a steady source of replenish able energy cells can use.

FOR REVIEW

A. Seven (7) different forms of energy include : ① Mechanical, ② Heat, ③ Sound, ④ Electrical, ⑤ light, ⑥ radioactive radiation, and ⑦ magnetic.

B. a) *nature of chemical bond* - pairs of electrons being shared between atoms, which represent stored energy, potential energy. When a covalent bond is broken in a chemical reaction, energy is released. If the bond being broken is within an organism, the organism may be able to use the released energy to do work and carry out its life processes.

b) *nucleotides* - the building blocks of the nucleic acids consisting of a 5-carbon sugar, a phosphate group, and a nitrogenous base (adenine, thymine, cytidine, guanine, and uracil). Nucleotides are also components of the biologically important coenzymes FAD, NAD^+, and ATP.

c) *proton pump* - a proton pump establishes a hydrogen ion gradient across a cell membrane and uses that gradient to produce ATP. Protons are actively pumped across the membrane, and their diffusion back through special protein channels in the enzyme ATP synthase is coupled to the phosphorylation of ADP with P producing ATP.

d) *entropy* - is defined as a measure of the randomness or disorder of a system. Entropy is the tendency of energy to spontaneously convert to a less organized pattern and therefore provides a directionality to all energetic processes, i.e., disorder is constantly increasing in the universe.

e) *potential vs. kinetic energy* - Potential energy is "Stored Energy", while kinetic energy is the "energy of motion".

f) *free energy* - A thermodynamic quantity that is the difference between the enthalpy (heat content) and the product of the absolute temperature and entropy of a system; also called Gibb's free energy. Best defined as the energy content in a system available to do work.

g) *exergonic/endergonic reaction* - If the products of a reaction contain more free energy after the reaction is complete than the reactants, then the reaction is endergonic, and another energy source had to contribute in the reaction in order for it to go to completion. If the products contain less free energy, then the reaction is exergonic, and will proceed spontaneously. Cells function by coupling exergonic and endergonic reaction together in living systems with the energy released by the exergonic reaction driving the endergonic reaction to completion.

h) *phospho-anhydride bond* - Because of the triplet phosphates linked to adenosine the covalent bond linkages are referred to as phosphoric acid anhydrides [-P-O-P] and are considered high energy bonds. The term "high energy bond" is incorrect and misleading, as it wrongly suggests that the bond itself contains the energy. In fact, the breaking of chemical bonds requires an input of energy. The free energy released by the hydrolysis of the phospho-anhydride linkages does not come from the breaking of the bond but results from the products of the reaction [ADP and P] having smaller free energy contents than the reactants. The hydrolysis of ATP to ADP + P has a $\Delta G^{0'}$ difference between reactants and products of -7.3Kcal/mol.

i) *aerobic* - involving oxygen; referring to an organism, environment, or cellular process that requires oxygen

j) *oxidation/reduction* - Oxidation is the loss of an electron by an atom or molecule, while reduction is the gaining of an electron by an atom or molecule. The e^- is frequently accompanied by a hydrogen proton. Oxidation involves the release of energy from the molecule, while reduction signifies the addition of energy to a molecule. By definition, whenever one substance is oxidized, another substance must be reduced; therefore oxidation-reduction reactions (redox Rx) transfer energy between molecule within cells.

k) *chemiosmosis* - the major pathway by which aerobic cells produce ATP. A proton pump in the inner mitochondrial membrane forces protons out of the mitoplasm producing a concentration gradient between the mitoplasm and peri-mitochondrial space. Following their diffusive concentration gradient, the protons diffuse back into the mitoplasm through an integral membrane protein, ATP synthase, which facilitates the phosphorylation of ADP with P.

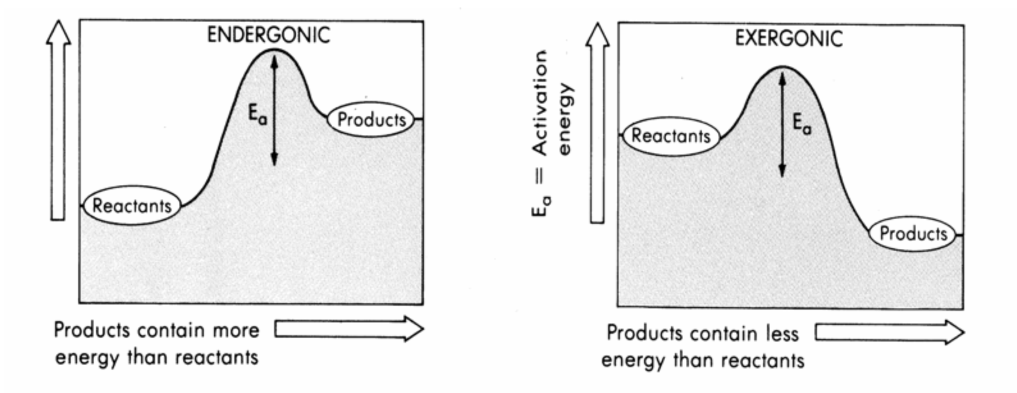
l) *anabolic* - The phase of metabolism in which simple substances are synthesized into the complex materials of living tissue; smaller inorganic [CO_2 and H_2O] are made into carbohydrates; the biosynthetic pathways of cellular metabolism are anabolic. Catabolic -the metabolic breakdown of complex molecules into simpler ones, often resulting in a release of energy; larger organic are degraded into smaller inorganics [glucose to CO_2 and H_2O]. The oxidative pathways of cellular respiration are catabolic.

m) *phosphorylation* - To add a phosphate group to (an organic molecule); commonly involves adding P to ADP to make ATP; reaction is often catalyzed by enzymes called kinases; is the primary mechanism by which cells make and transform energy poor compounds into energy rich molecules.

n) *dehydrogenase* - one of the six main classes or types of enzymes that catalyzes the removal and transfer of hydride ion from a donor substrate in an oxidation-reduction reaction.

o) *cytochrome* - any of a class of iron-containing proteins that are important in cell respiration as catalysts of oxidation-reduction reactions; found in the inner cristae membranes of mitochondria.

ENERGY GRAPHS



ENERGY FLOW

The five terms are : (A) ATP, (B) ADP, (C) P, (D) Energy Added, and (E) Energy released. The significance of this energy flow diagram is that it indicates that all cellular form energy are derived from the light energy of the sun and are converted into more useful intracellular form (ATP, ADP, P) allowing work to be done as the energy is eventually transformed and lost to the cells.

CELL RESPIRATION

STAGE	SUBSTANCE(S) PRODUCED
1. Glycolysis	ATP & NADH
2. Fermentation	NAD^+
3. Oxidation of Pyruvate	NADH
4. Citric Acid Cycle	ATP, $FADH_2$, NADH
5. Electron Transfer Chain	ATP, FAD, NAD^+

ELECTRON TRANSFER CHAIN - Paragraph Fill In.

A. NADH
B. $FADH_2$
C. NAD+
D. FAD
E. cytochrome proteins
F. oxygen
G. protons
H. water
I. proton pumps
J. ATP
K. chemiosmosis

GLYCOLYSIS - Two phases:

a) an ***energy investment phase*** - is so defined because the cell uses ATP to phosphorylate glucose and then fructose, raising those glycolytic intermediate to higher free energy states [more chemically reactive], thereby allowing them to be acted upon and converted into other intermediates, providing a favorable directionality for the oxidation of glucose to pyruvate to proceed. Glucose is phosphorylated to Glu-6-P as ATP is hydrolyzed by the enzyme hexokinase and fructose-6-P is phosphorylated to Fruc-1,6-diP by ATP hydrolysis.

and b) an ***energy-yielding phase*** occurs when the favorable intermediates produced above now can undergo oxidation producing 2 molecules of NADH, where some energy is conserved and can be used to make ATP by Chemiosmosis later in the mitochondria. 2 molecules of glyceraldehyde-P are oxidized and the electrons used to reduce 2 molecules of NAD^+. Additionally, 4 molecules of ADP are phosphorylated to make 4 ATP's by substrate level phosphorylation, producing a net yield of 2 ATP, thus energy yielding.

ELECTRON TRANSFER CHAIN

Electron Protein Carrier	Prosthetic Group (coenzyme)
cytochromes	iron heme groups four organic rings binding an iron atom
iron-sulfur proteins	iron and sulfur complexes
flavoproteins	FMN and FAD; Flavin mononucleotide & Flavin adenine dinucleotide
ubiquinone	an organic phenolic ring capable of being reduced to a semiquinone or hydroquinone

MAXIMUM ENERGY YIELDS (in ATP Equivalents) for EACH GLUCOSE OXIDIZED IN CELL RESPIRATION

Process	Substrate Level ATP's	Reduced Coenzymes Produced	ATP Produced by Oxidative Phosphorylation	Totals
Glycolysis	4 ATP (2 ATP net)	2 NADH	4 to 6 ATP	6-8
Oxidation of pyruvate	none	2 NADH	6 ATP	6
Krebs Cycle	2 GTP (=2 ATP)	6 NADH 2 $FADH_2$	18 ATP 4 ATP	24

A DIFFERENT KIND OF CONCEPT MAP

Process	Main Function (concept)	Inputs	Outputs
Glycolysis	oxidation of glucose to 2 pyruvate with net synthesis of 2 ATP, occurs anaerobically in all cells and as component part of aerobic respiration	glucose 2 ATP 2 NAD^+ 4 ADP + P	2 pyruvate 4 ATP (2 net) 2 NADH
Pyruvate to Acetyl-coA	oxidation of pyruvate to acetyl coA; CO_2 is released as part of oxidation of glucose; key link of anaerobic to aerobic metabolism; multi-enzyme complex involves 5 different coenzymes; key regulatory point.	2 pyruvate 2 CoASH 2 NAD^+	2 acetyl coA 2 CO_2 2 NADH
Krebs Cycle	series of redox reactions that produce NADH & $FADH_2$, GTP [an ATP equivalent] by substrate level phosphorylation, and decarboxylation of glucose (-CO_2)	2 acetylCoA 2 OAA 2 GTP + P 6 NAD^+ 2 FAD	2 CoASH 4 CO_2 2 ATP 6 NADH 2 $FADH_2$
Electron Transfer chain & oxidative phosphorylation	NADH (from glycolysis & Krebs cycle) and $FADH_2$ (from Krebs) transfer electrons to carrier protein molecules embedded in the cristae membranes of mitochondria. In series of redox reactions, H^+ are pumped into the intermembrane space, and electrons are passed to ½O_2. By Chemiosmosis, a proton motive force drives H^+ back into mitoplasm through ATP synthase making ATP	NADH $FADH_2$ H^+ ½O_2 ADP + P	NAD^+ FAD H_2O ATP
Fermentation	Regenerates cytoplasmic NAD+ so that glycolysis may continue. Pyruvate is reduced to ethyl alcohol and CO_2 in what may be most important *human* biochemical Rx.	pyruvate NADH	NAD^+ ethanol CO_2
Anaerobic Respiration (lactic acid production)	Regenerates cytoplasmic NAD+ so that glycolysis may continue. Pyruvate is reduced to lactic acid in anaerobic process and tissues as muscle.	pyruvate NADH	NAD^+ lactate

SOME IDENTIFICATIONS

1. (F) is the peri-mitochondrial (inner membrane) space and is the site of the higher [H^+]. The space depicted by the label (D = cytoplasm, might also be chosen, but is not as correct as the label (F)
2. The outer mitochondrial membrane is more freely permeable (label B).
3. Label (C) is cristae membranes.
4. Label (D) is the cytoplasm of the cell.
5. The area with the highest [H^+] has the lowest pH values and that is label (F).
6. The site of glycolysis is the cytoplasm of the mitochondria, which is label (D).
7. The molecule that is extruded by the mitochondria is H^+ 's.
8. The system depicted by the box labeled (A) is the electron transfer chain.
9. Label (C) is identifying the ATP synthase enzyme responsible for synthesizing ATP.
10. The cellular process depicted in this figure by labels A, B, and C is Chemiosmosis or oxidative phosphorylation.

SOME THOUGHT QUESTIONS

A. What is the net yield (of everything produced and/or consumed) for a every molecules of Glucose that goes through glycolysis?

    1 glucose is destroyed yielding 2 molecules of pyruvate
    4 molecules of ADP are phosphorylated by substrate level phosphorylation making 4 ATP's
    2 molecules of NAD^+ are reduced to 2 molecules of NADH.

B. Wine is an alcoholic beverage made by FERMENTATION of the juice of the grape. The four chemical products made in the process of the fermentation of wines are: a. NAD^+, b. CO_2, c. ethanol, and d. lactic acid (but also, acetic acid = vinegar, butyric acid- of butter, and propionic acid, a mold inhibitor in breads). The alcoholic content of natural wine comes from fermentation. Wine contains alcohol because of the yeast that live in the grape juices. The yeast carry out fermentation because conditions are anaerobic in a wine vat or bottle where the wine is fermenting. Fermentation starts when yeast on the skins of ripe grapes comes into contact with the grape juice (called must). Run off into casks, the new wine then undergoes a series of chemical processes, including oxidation, precipitation of proteins, and fermentation of chemical compounds, that create characteristic bouquet. After periodic clarification and aging in casks, the wine is ready to be bottled. Natural wines do not have alcohol levels higher than about 12%, because at higher levels the alcohol becomes toxic to the yeast cells and kills them, thus preventing further fermentation or further production of alcohol.

C. Oxygen is the terminal (final) electron acceptor of the electron transfer chain in oxidative respiration. When air, containing as little as 0.1% carbon monoxide by volume, is inhaled, the oxygen carried by hemoglobin is replaced by the carbon monoxide, resulting in fatal oxygen starvation throughout the body. When no oxygen is present, the preceding electron transfer chain proteins acceptors (the cytochromes) can not pass along their electrons and/or protons. The electron transfer chain essentially becomes backlogged or jammed and stops passing electrons. Without continued electron transport the chemiosmotic synthesis of ATP stops. The ATP synthase, which is responsible for making 32 of the total 36 ATP per glucose molecule, can not work as efficiently as the proton gradient production slows when the lack of oxygen backs things up. As the amounts of ATP drop there is not enough energy to do the work of life, e.g., pumping the heart, carrying out vital biochemical reactions, and continued muscle contraction.

D. Metabolic water is the water formed at the end of the electron transport chain. Oxygen is the terminal electron acceptor. It combines with the electrons and protons to form a molecule of water. This *metabolic water* (formed by the processes of oxidative metabolism within the organism) can be sued to help meet the water needs of cells and organisms.

E. Fermentation may be inefficient when compared with oxidative respiration, but it is still better than nothing at all. The ATP yield from glucose may be small in anaerobic conditions, but there is still some yield. ("*An ugly date is better than no date at all*" - well maybe?). Fermentation also allows organisms to live in otherwise uninhabitable anaerobic environments, and additionally allows normally aerobic cells to keep functioning (albeit, at a reduced level) under temporarily anaerobic conditions (e.g., muscles cells during short, sprint/strenuous exercises).

F. Jogging is considered aerobic because the cardiovascular system (heart and lungs) can adjust its activity to continually supply the oxygen needing to keep oxidative respiration (glycolysis, Krebs cycle, and electron transfer) functioning harmoniously. A continued supply of oxygen is needed to keep these metabolic pathways from "backing up" (see answer C. above). Sprinting is considered anaerobic because a continued supply of oxygen is not needed nor is continued ATP production by Chemiosmosis. What is required is the glycolytic production of ATP, albeit small, by the anaerobic oxidation of glucose. Anything that requires only 2 minute or less of complete exertion is considered to be a "sprint/anaerobic" exercise. There is sufficient ATP stores in muscle cells and in the form of creatine-phosphate, which can convert ADP into ATP in the short term (100 meters). Golf is considered non-aerobic, I suspect, for the maximum exertion is only during the swinging of the golf club (so where does baseball fit in this - remember the words of Roberto Clemente on his induction into the Baseball Hall of Fame ....*"Baseball has been berry, berry good to me.")* Such short periods of exertion [swinging a bat or a golf club, lifting a weight, throwing a punch, etc...] probably only use up stored muscle ATP and/or creatine phosphate and do not call upon glycolysis to produce its 2 ATP net per glucose.