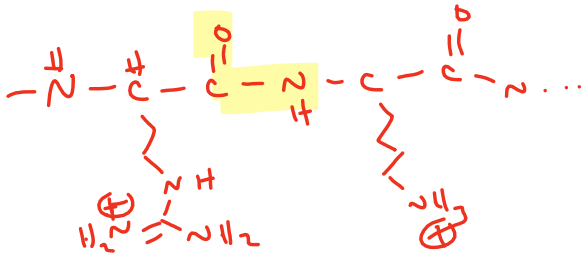
PS6key

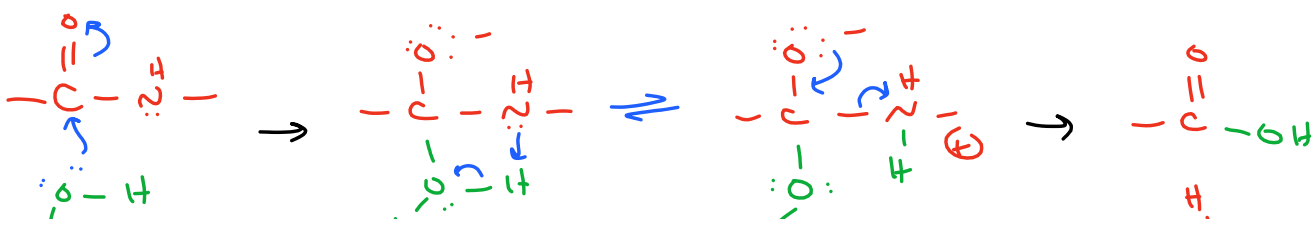
Thursday, April 13, 2017 5:05 AM

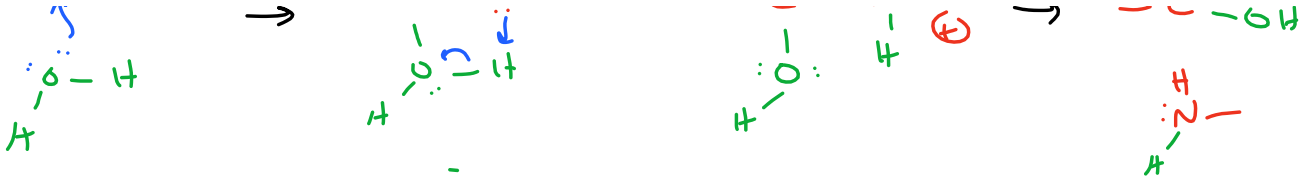
Problem Set 6 (**Due: April 10th 5:00 PM)**

1. Insulin is a protein hormone that is secreted by the pancreas. The different parts of this question will explore some aspects of the production, structure, and function of insulin. Refer to this image to answer these questions: https://upload.wikimedia.org/wikipedia/commons/1/18/Insulin_path.svg. You can also use this table of amino acids to help you figure out what the single letter codes mean. http://www.sigmaaldrich.com/life-science/metabolomics/learning-center/amino-acid-reference-chart.html
   a. Insulin is synthesized by on the ribosome as a protein that contains and extra sequence at the N-terminus that allows it to be sent to the endoplasmic reticulum (or the ER – this is a part of the cell). Examine the sequence of this signal peptide (in red in the image).
      i. What do you notice about the chemical properties of most of the amino acids? They are hydrophobic
      ii. What is the name of the transporter that allows insulin to move into the ER? Sec61 translocon
   b. In the image, you should note that once insulin gets into the ER, it is folded and oxidized. This occurs as it is anchored to the lipid bilayer.
      i. What part of insulin favorably inserts itself into the bilayer? The signal sequence – it is primarily hydrophobic
      ii. You may notice that the membrane embedded region contains an arginine (r) and a glutamic acid (d). Under normal conditions, what is the charge on each of these amino acids? Glu = negative, Arg = positive Based on this, how do you think that these amino acids are able to be part of a transmembrane peptide? Two possibilities – they could interact with each other, forming a salt bridge (ion-ion interaction) that effectively neutralizes the charge. OR The pKa of the side chains could be modified so that they are actually neutral instead of charged.
      iii. The "oxidation" part of this step refers to cysteine side chains getting oxidized. What happens when cysteine gets oxidized? A covalent bond is formed with another cysteine side chain (Cys – S-S-Cys) How do you think this affects the stability of the insulin structure? Locks the structure in place. Normally, secondary and tertiary structure is held in place by intermolecular forces – these are weaker than covalent bonds. So the presence of the covalent crosslinks helps make the structure more stable.
      iv. Once inside the secretory vesicle, the protein chain is broken in two places by enzymes that catalyze hydrolysis reactions.
         1. These enzymes recognize specific amino acid sequence (show in dark blue in the image). What do these recognition sequences have in common? They are both two basic amino acids linked together (Arg-Arg and Lys-Arg).
         2. Draw the peptide bond that is formed between arginine and glycine (the target of the PC2 enzyme).



         3. Show the mechanism for this hydrolysis reaction (this is the 3 step process that we learned earlier in the term).



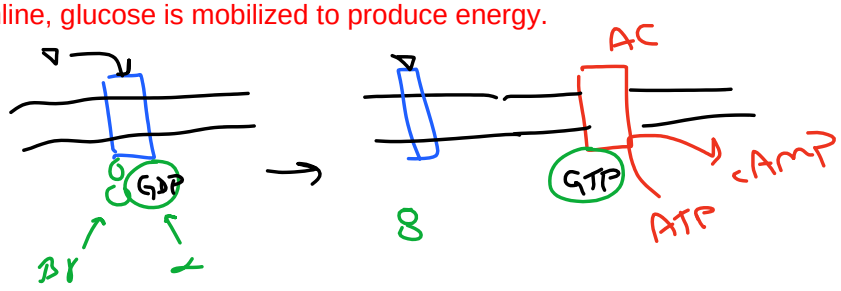


2. Once insulin is secreted into the blood, it seeks out insulin receptors.
   a. Describe how the insulin receptor works. Insulin binding causes tyrosine residues on the inside of the cell to get phosphorylated. The phosphate comes from ATP (gets converted to ADP). Once the intracellular tyrosines are modified, this initiates an intracellular signal.

   b. Clearly explain how an enzyme inside a cell might be able to "sense" insulin binding to the outside of the cell. If an enzyme recognizes a phosphorylated tyrosine a not a tyrosine, insulin binding would trigger this new interaction. The image below shows how this might be possible.

   c. Type 2 diabetes is marked by the development of insulin resistance. What does this mean and why is it important? Cells no longer respond to insulin. This means that the sugar in the blood does not get taken up by cells. Consequently, the blood sugar remains elevated and more insulin is secreted by the pancreas. So now there is elevated sugar and insulin in the blood. This can lead to hypertension, obesity, dyslipidemia (elevated lipid level in the blood), and glucose intolerance (collectively, this is metabolic syndrome = bad).

3. Epinephrine (aka adrenalin) is a hormone that is secreted by adrenal glands. It functions in a very similar way as glucagon and has a very similar physiological effect.
   a. Using any combination of words and images, describe how epinephrine can stimulate energy (ATP) production in cells. Epinephrine activates a GPCR. The alpha subunit dissociates from the GPCR and activates adenylate cyclase – this catalyzes the conversion of ATP to cAMP. This secondary messenger can then lead to the activation or inhibition of many enzymes involved in metabolism: punchline, glucose is mobilized to produce energy.



   b. Look up the structure of epinephrine.
      i. Which amino acid does it most closely resemble? Tyrosine
      ii. Predict how the epinephrine receptor recognizes and binds to epinephrine.