Chem 162/262 (2005) Assignment #3:

Enzymes: Mechanisms & Inhibition

1. In their famous 1971 PNAS paper Michael Page and William Jencks proposed that enzymes may work as "entropy traps". Based on simple thermodynamic arguments they arrived to the conclusion that enzymes can accelerate bimolecular reactions by as much as 10^8.

   a) Explain the "entropy trap" concept of Page and Jencks in your own words.
   b) Many scientists now believe that while the basic idea behind the "entropy trap" concept remains valid, the 10^8 value largely overestimates rate acceleration that could be achieved by simply binding two reactants of the bimolecular reaction to the enzyme. Discuss reasons why the 10^8 estimate might be wrong.
   c) Is the concept of "entropy trap" relevant to designing enzyme inhibitors as potential drugs? Discuss why.

2. Transition state analogs are potent inhibitors of enzymes. Consider each of the following three hypothetical enzymatic reactions and draw (using IsisDraw or a similar program) a structure of the reactant, the transition state, and the reaction product. Design a transition state analog that might inhibit each of these hypothetical enzymes and provide the structure of the inhibitor. You are encouraged to solve this problem without much help from the literature; if you use literature sources for your answer please provide full citation for your source.
   a) Hydrolysis of D-arginine into D-ornithine and urea
   b) Hydrolysis of 5-methylcytidine into 5-methyluridine with loss of ammonia
   c) Phosphorolysis of 6-thioinosine into 6-thiohypoxanthine and α-D-ribose 1-phosphate

3. In order for cells replicate, sufficient quantities of nucleotides, such as deoxythymidylate (dTTP), are needed. dTTP is made from deoxyuridylate (dUTP) by thymidylate synthase. Enzymes responsible for synthesis of dUTP from uridine are ribose-1-phosphate uridine phosphorylase, uridine phosphorylase, UMP kinase, ribonucleotide reductase, orotate phosphoribosyltransferase, thymidine kinase, and dUTP diphosphohydrolase. Significant decrease in thymidylate synthase activity is seen after administration of drug 5-fluorouracil. However, this compound does not significantly reduce the activity of the enzymes that are involved in conversion of uracil to dUTP.

   a) Explain why 5-fluorouracil does not inhibit enzymes that convert uracil to dUTP.
   b) Explain why administration of 5-fluorouracil leads to reduced thymidylate synthase activity.

Project development.
You will be submitting a drug design proposal at the end of this course. As a second part of this project, you are expected to validate one (or few) targets pertaining to the disease that you want to work on. As part of your second assignment, present and critically discuss the evidence that validates your target(s). Make sure to read and reference the original research that provided validating data, and discuss limitations of such studies.

Some students may wish to work on a project where the target is unknown. For example, you may have heard or read about a surprisingly valuable side-effect of a currently existing drug and want to design a combinatorial library around this structure to find compounds that show stronger "side-effect". In this case, please describe what approaches you would take to identify the target of your drug.

Please see "Drug Design project tips" document for more information on how to approach writing the final project.