

Name: Scooter Libby Date: Nov. 17, 2005

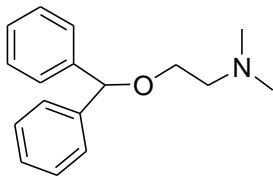
**Topics in Organic Chemistry: Modern Medicinal Chemistry  
(CHEM 515)  
Mid-term Examination**

Profs. Malachowski and White

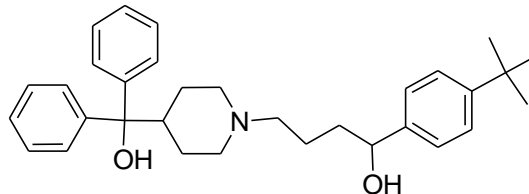
Due: November 8, 2005, 10 AM

*Honor Code:* You may take this examination while consulting your course lecture notes and the information posted on the course Blackboard site. You are not to consult any other electronic or written material during the exam. You have 3 consecutive hours to complete the exam. You may word process your answers. You may use molecular models to assist you in answering the exam questions. You should not discuss the exam with anyone until all students have handed in their exam. There are a total of twelve questions on 12 pages.

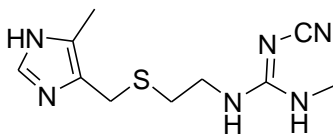
1. The sedative effects of antihistamines (histamine antagonists), e.g. diphenhydramine (the active ingredient in Benadryl), are believed to be the result of antagonism of H<sub>1</sub> receptors in the brain. Two other histamine antagonists, terfenadine (the active ingredient in Seldane) and cimetidine (the active ingredient in Tagamet) do not show similar sedative effects. Explain. (7 pts.)



*diphenhydramine*

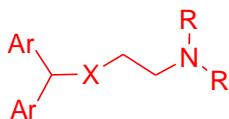


*terfenadine*



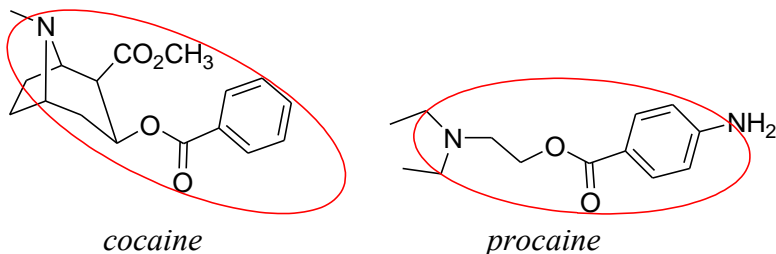
*cimetidine*

Cimetidine is an H<sub>2</sub> histamine antagonist and therefore does not interact with H<sub>1</sub> histamine receptors. Terfenadine is an H<sub>1</sub> histamine antagonist based on its structural match with the H<sub>1</sub> histamine antagonist pharmacophore structure (below) that was described in lecture at the start of our cimetidine discussion. However, terfenadine is a more polar molecule with two hydroxy groups and therefore can not easily penetrate the lipophilic blood-brain barrier. Cimetidine would also be more polar and unlikely to penetrate the blood-brain barrier. Consequently, neither one would be able to interact with the H<sub>1</sub> histamine receptors in the brain.



*H<sub>1</sub> histamine antagonist pharmacophore*

2. In the nineteenth century, cocaine was known to have local anesthetic properties and the medical community hoped to develop a drug that could replicate these local anesthetic properties without the adverse addictive properties of cocaine. Success was achieved in 1909 with the development of procaine (Novocaine).



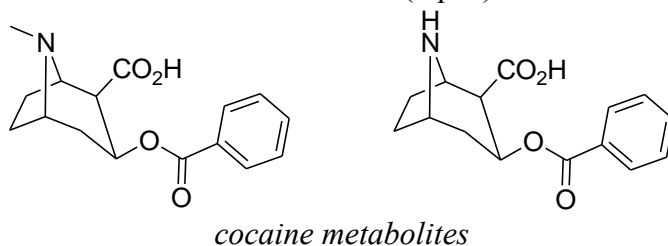
a) Determine the pharmacophore, the essential structural elements that lead to the desired bioactivity, for molecules with local anesthetic properties based on the structures of cocaine and procaine. Circle the pharmacophore core in the cocaine and procaine structures shown above and briefly describe the structure. (3 pts.)

The local anesthetic pharmacophore is a benzoate ester linked by two or three carbons to a tertiary amine.

b) What type of structural modification was made to eliminate the unwanted properties of cocaine? (2 pts.)

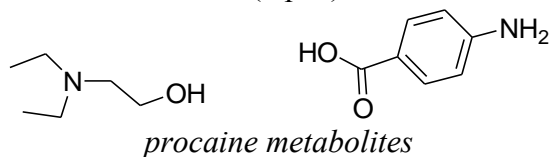
Primarily structural reduction occurred with the loss of the bicyclic ring and one of the ester groups, although a primary amine is added on the benzene ring.

c) Metabolism of cocaine affords two common metabolites (shown below). What enzymes are involved in cocaine metabolism? (2 pts.)



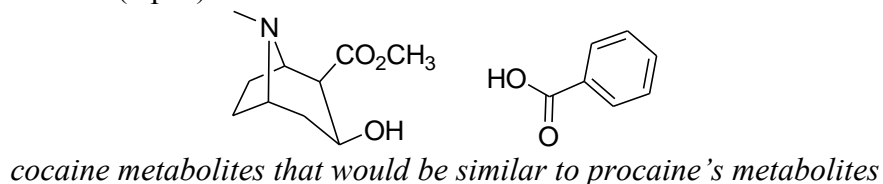
The methyl ester of cocaine is converted to a carboxylic acid which is a process catalyzed by hydrolase or esterase enzyme. The methyl group of the tertiary amine is dealkylated; that is an oxidative process catalyzed by a cytochrome P-450.

d) Procaine is metabolized to afford the metabolites shown below. Which enzyme is involved in its metabolism? (2 pts.)



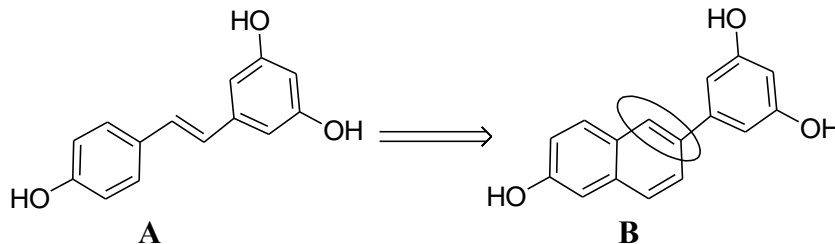
Since this is an ester cleavage, it would also be catalyzed by an esterase or hydrolase.

e) Suggest a reason why the analogous metabolic products (shown below) are not seen for cocaine. (2 pts.)



The methyl ester is less sterically hindered and therefore more easily cleaved than the secondary alcohol ester cleavage shown above. With procaine, the ester is a primary alcohol ester and, although less reactive than a methyl alcohol ester, it will be more reactive than a secondary alcohol ester.

3. A recent report of anti-cancer drug development showed the following structural modification of the known drug, **A**, into an analog **B**, that was tested for similar anti-cancer activity.



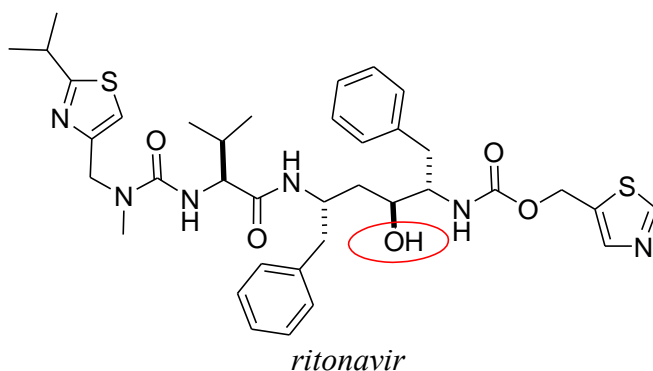
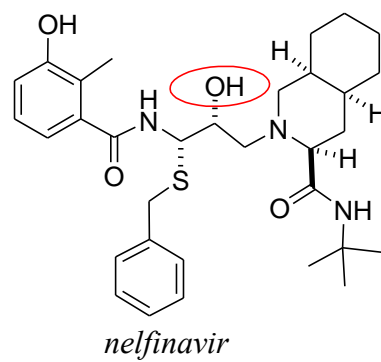
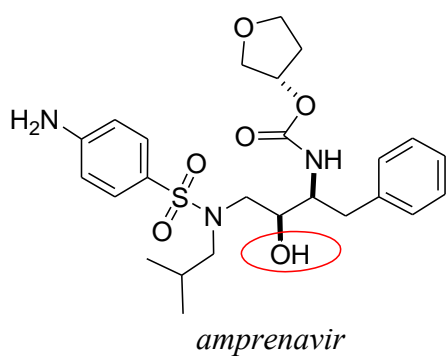
a) In their report, the researchers mentioned that one goal of the modification to compound **A** was to reduce the reactivity of the alkene that links the two benzene rings. Why is the analogous alkene in **B** (circled) more stable? (3 pts.)

As part of an aromatic benzene ring, the circled alkene would be more stable.

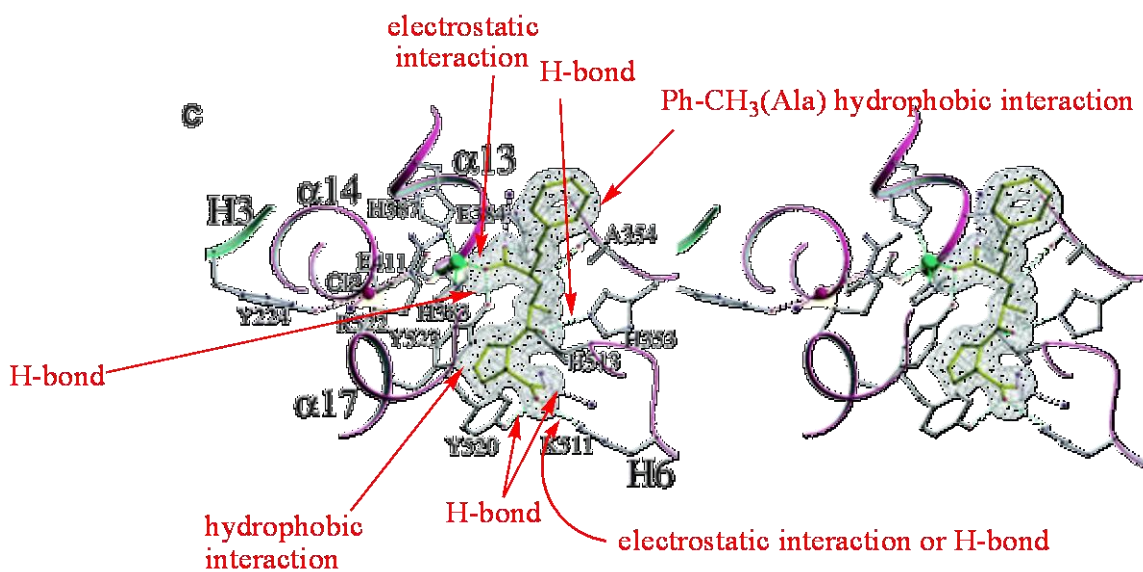
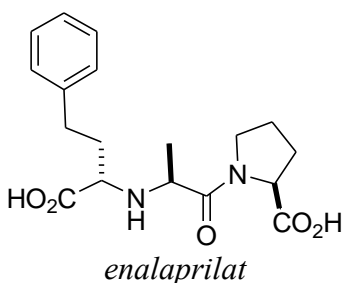
b) Besides stabilizing the alkene, name one other effect of the change in structure **A** seen in compound **B**. (4 pts.)

The incorporation of the alkene in a ring reduces conformational flexibility making the alkene co-planar with the benzene ring in **A**.

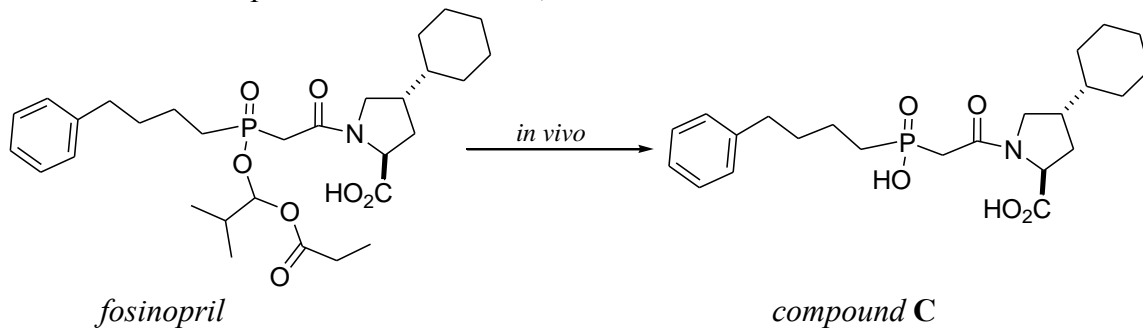
4. Three HIV protease inhibitors are shown below. Assuming they all interact with HIV protease in a manner similar to crivivan (indinavir), circle the atom(s) in the drugs shown below that form hydrogen bonds with the Asp25/Asp25' at the enzyme's active site. (6 pts.)



5. A recent paper followed the results of the Nature (2003, 421, 551-554) paper that we discussed in class with more crystal structures of ACE inhibitors in the active site of human testicular angiotensin converting enzyme. The crystal structure of enalaprilat with ACE is shown below. In the picture below, identify three different intermolecular forces (hydrogen bond, hydrophobic force, electrostatic interaction) that contribute to enalaprilat's binding to ACE. (6 pts.)



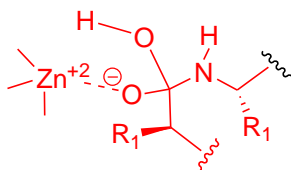
6. Another example of an ACE inhibitor is the drug called fosinopril. *In vivo* fosinopril is converted into compound C shown below, which is the actual inhibitor of ACE.



a) Based on this *in vivo* activation process, what is fosinopril's actual role in ACE inhibition? (2 pts.)

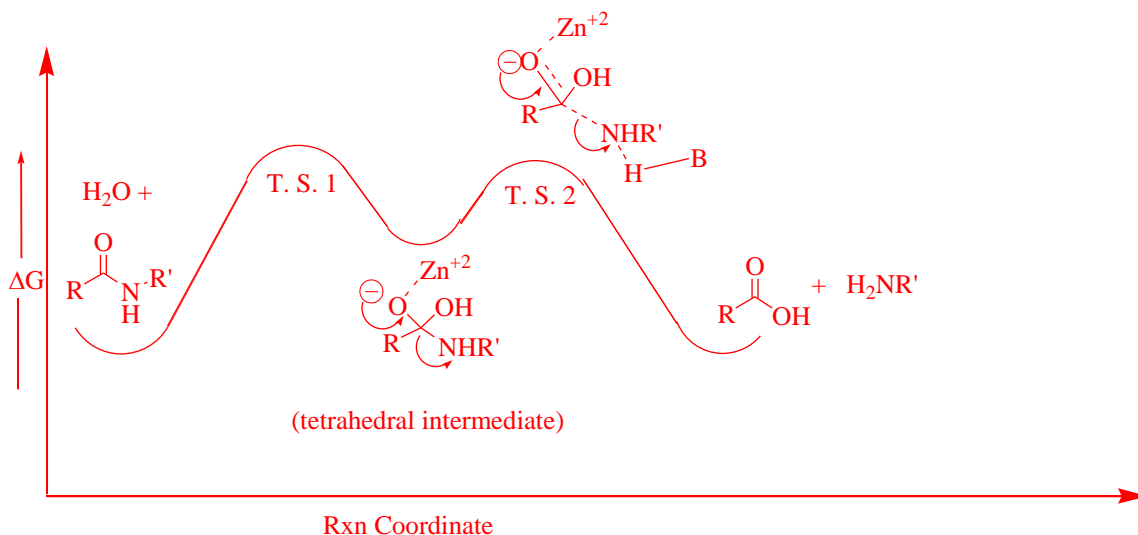
Fosinopril is a pro-drug.

b) Fosinopril inhibits ACE by a different mechanism than captopril. Fosinopril is a "transition state analog" inhibitor like crivivan. Draw the "transition state" structure that fosinopril mimics in inhibiting ACE. (You don't need to draw the entire "transition state" structure, just the important part.) (4 pts.)

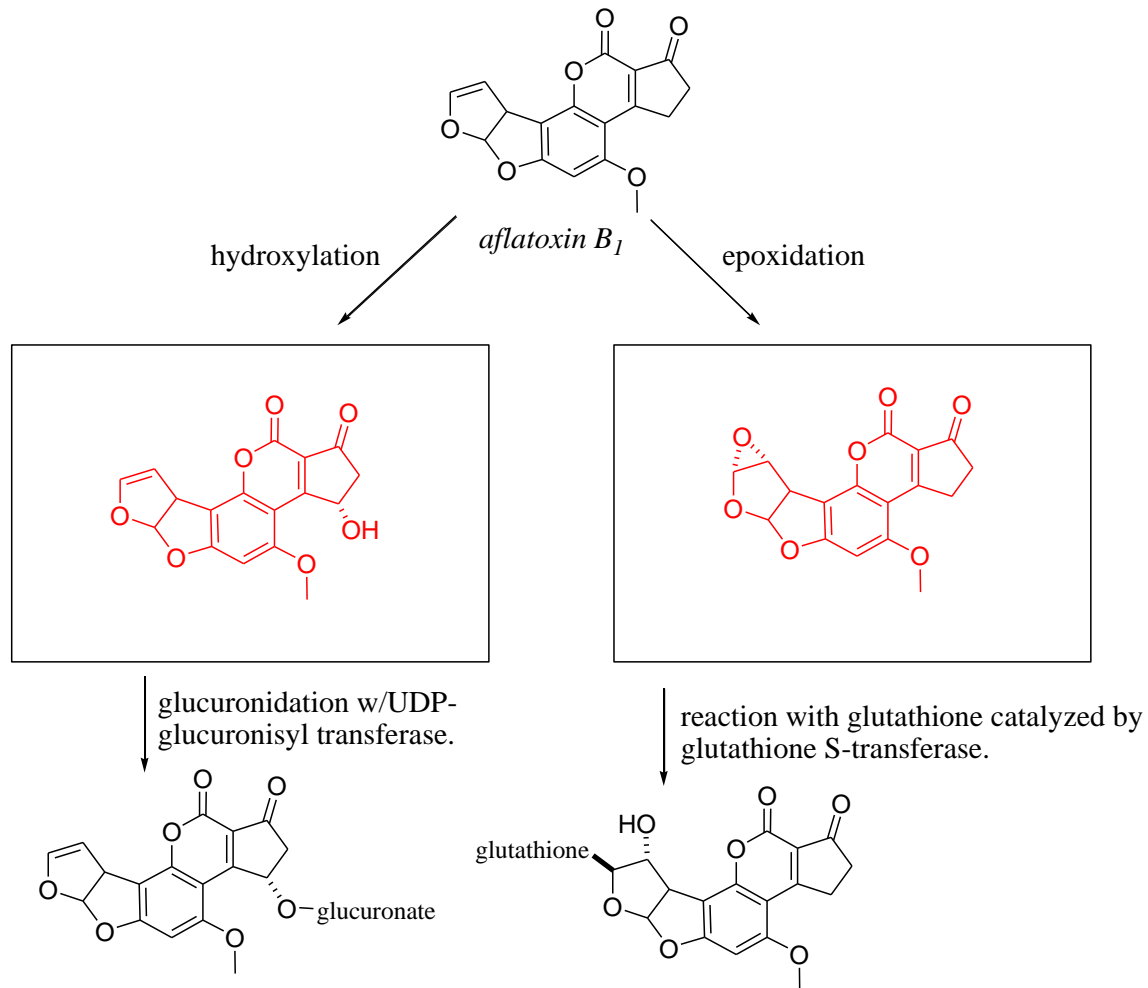


c) "Transition state analog" is the accepted term for these types of inhibitors, however in the protease reactions, the inhibitor really looks more like another point on the reaction pathway. What is the correct name for the structure that fosinopril and crivivan actually mimic? (Hint: An energy diagram might be useful in illustrating this point.) (3 pts.)

reaction intermediate:



7. Hydrophobic molecules that are foreign to the body are usually metabolized along two general pathways. Both of these are amongst the pathways we discussed in drug metabolism lectures. One of the most potent carcinogens, aflatoxin B1, a fungal toxin, is processed in these two different ways. Unfortunately, one pathway creates the lethal form of aflatoxin B1 that leads to its carcinogenic properties.



a) Draw the structure of the intermediate metabolic products in the two boxes shown. (2 pts.)

b) What type of enzyme probably catalyzes the first set of reactions (hydroxylation and epoxidation)? (2 pts.)

cytochrome P-450 oxidases

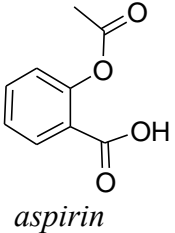
c) Which phase of metabolic reactions is represented in the second set of transformations (reaction with glucuronic acid and glutathione)? (2 pts.)

phase II, conjugation reactions

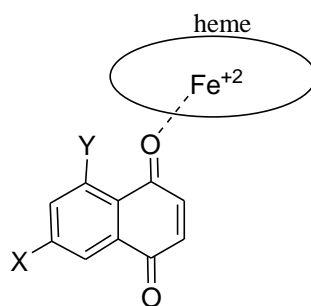
d) What is the body's primary goal in the series of metabolic reactions shown above? (2 pts.)

Primary goal is to polarize molecules to make them more water soluble and thereby excrete them.

8. Does Lipinski's "rule of five" predict good oral bioavailability for aspirin? (5 pts.)

	<p>Evaluation of aspirin by Lipinski's rule of five:</p> <ol style="list-style-type: none"><li>1. Aspirin has fewer than 5 H-bond donors (only 1).</li><li>2. Aspirin's molecular wt. is below 500 (180)</li><li>3. Aspirin's clogP is less than 5 (calcd: <math>1.00=1.96+(-0.32)+(-0.64)</math>)</li><li>4. Aspirin has fewer than 10 N's and O's.</li></ol> <p>Since none of these criteria are met, aspirin should be orally bioavailable.</p>
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9. The Malachowski group is currently working on developing a new anti-cancer treatment by developing inhibitors of an enzyme called indoleamine 2,3-dioxygenase (IDO). Currently, one lead compound has the naphthoquinone structure (X, Y=H) shown below. One proposed mode of IDO inhibition involves binding of the ketone oxygen in the naphthoquinone to the heme iron atom at the active site of IDO.



*naphthoquinone structure bound to  $Fe^{+2}$  at IDO active site.*

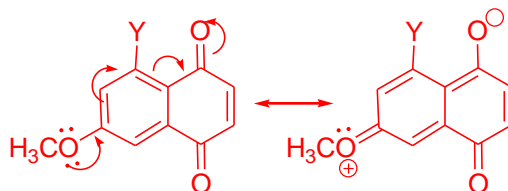
a) To test the proposed mode of binding, we could make naphthoquinone analogs that modify the electronic nature of the ketone oxygen. One way to do this would be to vary the groups at the X position. Suggest one group in the X position that would make the ketone oxygen more electron rich and one that would make it more electron poor. (4 pts.)

Make more electron rich with electron releasing groups at X:  $-OCH_3$ ,  $-OH$ ,  $-CH_3$ , etc. (look for  $\sigma_{para}$  values that are negative).

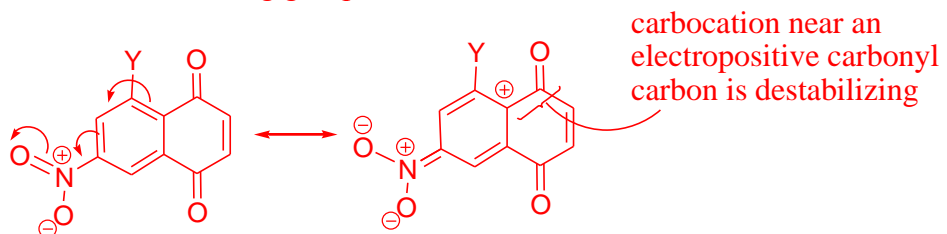
Make ketone less electron rich with electron withdrawing groups at X:  $-CN$ ,  $-NO_2$ ,  $-Cl$ , etc. (look for  $\sigma_{para}$  values that are positive).

b) Choose one of the groups that you selected in part a and draw a resonance structure that dramatizes the effect that the group has on the ketone oxygen. (3 pts.)

Electron releasing group effect:



Electron withdrawing group effect:



c) Changing substituents at position Y might afford similar electronic changes, however conclusions about the electronic effects of substituents at Y would be less certain. Why? (2 pts.)

Y's proximity to the ketone (ortho) means there is potential for inductive effects or steric effects to also play a role.

10. a) An X-ray beam of wavelength  $1.54 \text{ \AA}$  shines on a crystal. Two beams scattered from the crystal are exactly in phase at the detector. What are the possible path differences in  $\text{\AA}$  for the two scattered beams? One answer is zero. Give three other possible answers. (4 pts.)

The PD (or 2 PD if you were looking at your notes) must be an integer number of wavelengths— $1.54 \text{ \AA}$ ,  $3.08 \text{ \AA}$ ,  $4.62 \text{ \AA}$

b) The two beams in (a) are exactly out of phase at the detector. Give three possible values in  $\text{\AA}$  for their possible path differences. (4 pts.)

The PD (or 2 PD) must be  $1/2$ ,  $3/2$  or  $5/2$  times the wavelength  $0.77 \text{ \AA}$ ,  $2.31 \text{ \AA}$ ,  $3.85 \text{ \AA}$

11. Tough Week for W!

a) We've seen that "W" in crystal structure may stand for bridging waters that hydrogen bond between two parts of a protein or between a drug and the protein. Draw a geometrically correct bridging water that hydrogen bonds to a drug carbonyl oxygen and a protein lysine amino group. (5 pts.)

Lysine's amino group is not the amide group! At neutral pH it should be + charged and can only be an H bond donor. Geometrically correct H bonds have the H sandwiched between the carbonyl O and the water O with all 3 colinear. Or the lysine  $\text{N-H}\dots\text{O}=\text{C}$ , in a collinear arrangement. "Two centered" bonds are more complicated and weren't necessary.

b) Sometimes crystallographers do not know if their crystal contains protein or just salt so they use the highly un-technical "smush" test. Using any handy pointy object, they press down on a questionable crystal. If it is hard, or shatters cleanly, then it is salt. If, on the other hand, it "squishes" then it is protein. Explain the basis for this test. (5 pts.)

HINT: Part (a) may be useful.

Salt crystals have no water and are held together by strong ionic bonds. Conversely, protein have irregular shapes that don't take up the unit cell and contain lots of water and there are lots of weak interactions including waters H bonded to the protein.

12. According to the article “Crystal structure of the human angiotensin-converting enzyme-lisinopril complex” (*Nature*, 42, **2003**, 551-54), the structure of the enzyme alone is the same as / different from the enzyme bound to the drug. This is an example of lock and key / induced fit.

a) Circle the correct underlined term. (2 pts.)

b) Consider the article “Crystal Structure at 1.9 Å Resolution of Human Immunodeficiency Virus (HIV) II Protease Complexed with L-735,524, and Orally Bioavailable Inhibitor of the HIV Proteases” (*J. Biol. Chem.*, **269**, 1994, 26344-48). Do you find evidence for the “lock and key” or induced fit model? You must support your answer with structural information found in the article. (4 pts.)

Induced fit—the HIV protease flaps move upon substrate binding.

c) For which type of model might it be easier to design a drug? Explain your reasoning. (4 pts.)

For the lock and key you know where all the active site functional groups are and they don't move. So it's easy to figure out what type of inhibitor to design.

Resistance mutations in dihydropteroate synthase were studied in “Structure and Function of the Dihydropteroate Synthase from *Staphylococcus aureus*” (*JMB*, **268**, 1997, 21-30). These mutations were found to be all around the active site / in almost all regions of the protein.

d) Circle to correct underlined choice. Do you think this enzyme binds to its substrate by a lock-and-key or by an induced fit mechanism? Justify your answer in three sentences or less. (4 pts.)

Given that mutations that confer resistance are all over the protein, one can imagine that conformational changes are required during substrate binding or catalysis. This would argue for induced fit. BUT the one substrate used did not cause conformational changes except for one Asp that was mentioned.