Biochemistry 463, Summer II **Your Name:** _____

University of Maryland, College Park **Your SID #:** _____

Biochemistry and Physiology **Profs. Doug Julin and Jason Kahn**

Exam II (100 points total) **August 10, 2007**

You have 80 minutes for this exam.

Exams written in pencil or erasable ink will not be re-graded under any circumstances.

Explanations should be concise and clear. I have given you more space than you should need. There is extra space on the last page if you need it.

You will need a calculator for this exam. No other study aids or materials are permitted.

Generous partial credit will be given, *i.e.*, if you don't know, guess.

Useful Equations:

$v_0 = V_{max}[S]/(K_M+[S])$ $\quad pH = -\log([H^+])$ $\quad \Delta G° = -RT\ln K_{eq}$

$K_a = [H^+][A^-]/[HA]$ $\quad \Delta G = \Delta H - T\Delta S$ $\quad pH = pK_a + \log([A^-]/[HA])$

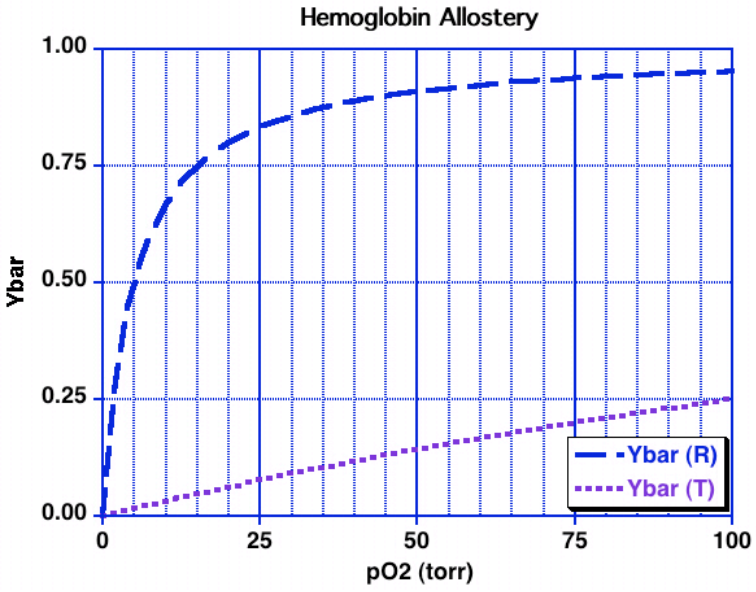
Honor Pledge: At the end of the examination time , please write out the following sentence and sign it, or talk to me about it:

"I pledge on my honor that I have not given or received any unauthorized assistance on this examination."

1. (30 pts) Hemoglobin and oxygen transport.

(a; 3 pts) Why have myoglobin and hemoglobin evolved to bind gaseous ligands in a bent conformation relative to the gas-iron bond?

(b; 6 pts)The graph at the right shows hypothetical "pure R" and "pure T" state binding curves. Sketch in the binding curve for Hb at pH 7.6, with a p50 of 26 torr. Sketch in the binding curve at pH 7.2. You do not need to know the p50: I am just asking for which direction the curve will shift.



Score for the page_____

(c; 16 pts) Blood picks up oxygen in the lungs. What happens to each of the following molecules during this process? Include in your answers how each species affects the R <-> T interconversion of hemoglobin.

Oxygen:

Carbon dioxide/bicarbonate:

Protons:

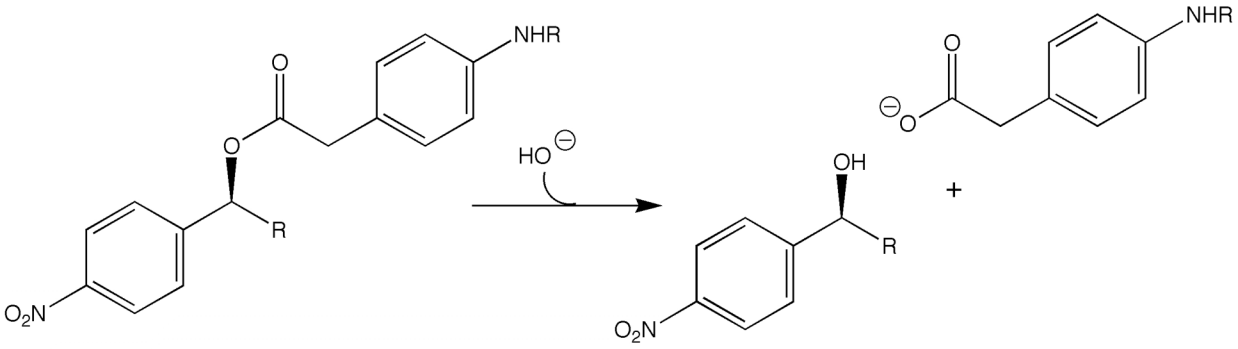
Chloride ion:

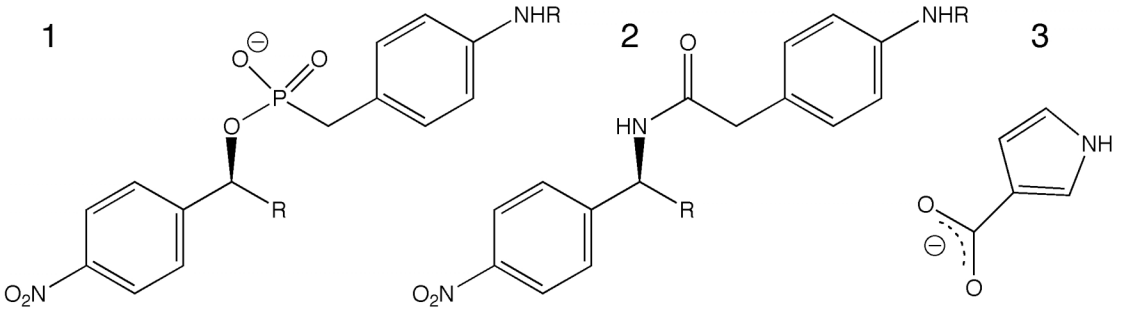
(d; 5 pts) The Bohr Effect refers to the observation that Hb is a stronger acid when bound to oxygen than when it is not. Why does the Bohr effect make sense in terms of the physiology of oxygen transport?

Score for the page_____

2. (15 pts) Enzyme kinetics

(a; 8 pts) Why are transition state analogs generally better (tighter-binding) enzyme inhibitors than substrate analogs? Which one of the inhibitors shown at the bottom below looks like it might be a transition state analog for the hydrolysis reaction at the top, and why?



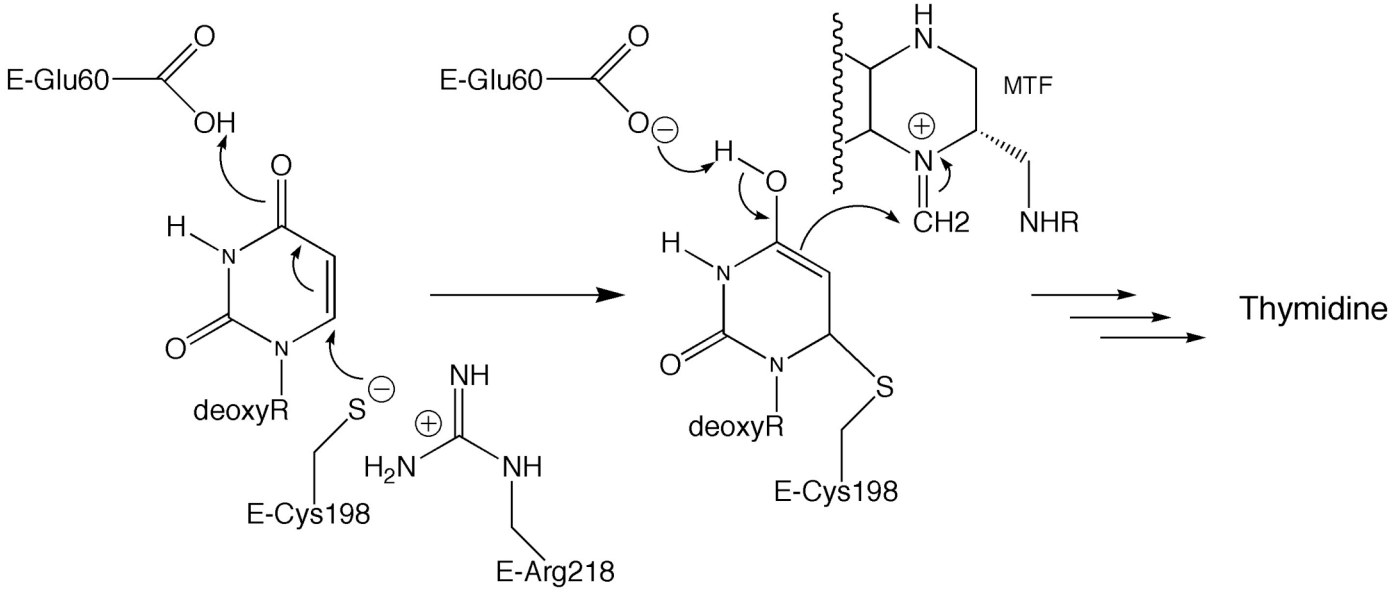


Score for the page_____

(b; 7 pts) Write down the Michaelis-Menten equation. We will not derive it here, but you should remember the ideas that made the derivation possible. To which critical intermediate did we apply the Steady State Approximation? What is the Steady State Approximation?

3. (25 pts) Enzymatic catalysis and inhibition

(a; 11 pts) A simplified version of the early steps of the reaction catalyzed by thymidylate synthase (TS) is shown below. Three of the active site residues are Cys 198, Arg 218, and Glu 60. Which of the common modes of enzymatic catalysis that we discussed in class is exemplified by each residue (one answer each)? Which one the following mutations do you think is most likely to give an active enzyme (circle one, no explanation needed): Cys 198 -> Val, Arg 218 -> Lys, Glu 60 -> Thr?



Score for the page_____

(b; 7 pts) The scheme below (page 722 of Berg 6e) shows inhibition of TS by FdUMP, which is an irreversible inhibitor used in chemotherapy. What is the difference between reversible and irreversible inhibition? How can one differentiate experimentally between a reversible inhibitor and an irreversible inhibitor?

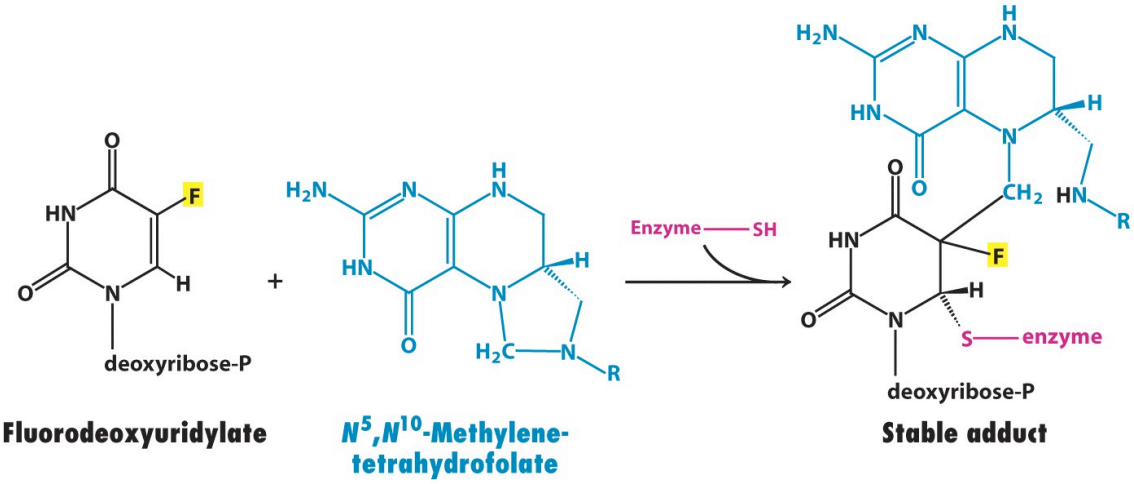


Figure 25-14
Biochemistry, Sixth Edition

Score for the page_____

(c; 7 pts) Sketch the Lineweaver-Burke plots corresponding to competitive and noncompetitive inhibition (two plots). Why does irreversible inhibition have the same kinetic profile as noncompetitive inhibition?

Score for the page_____

4. (30 pts) Metabolism and Glycolysis

(a; 10 pts) Draw out the entire pathway for glycolysis, including all structures.

Score for the page_____

(b; 10 pts) Discuss the experimental evidence that TIM is a kinetically perfect enzyme.

(c; 10 pts) Discuss the negative feedback loops that control flux through glycolysis, and explain why the thermodynamically downhill steps are the regulated ones.

Page	Score
1	
2	
3	
4	
5	
6	
7	
Total	

Score for the page_____