

## CHEM205 Problem Set 2. Enzyme Kinetics

Due 11am Friday October 20

Question 1 (10 points). An enzyme is discovered to be implicated in a disease process. The enzyme is known to have  $K_m = 1.0$  mM, and  $V_{max} = 10.0$  M/min for its natural substrate. As a therapeutic strategy, a medicinal chemist tries to design a competitive inhibitor for the enzyme. On testing, the inhibitor is shown to be a pure non-competitive inhibitor with  $V_{max} = 1.0$  M/min at  $[I] = 1$  mM.

- Draw the double reciprocal plot for the enzyme in the presence and absence of the inhibitor. Clearly label which line on the plot corresponds to which case.
- What is the inhibition constant,  $K_i$  ?
- What would  $K_m$  have been if the inhibitor had been competitive, assuming the same  $K_i$  ?
- Draw a line on the same plot corresponding to the expected competitive inhibitor, assuming the same  $K_i$

Assume  $[E]_{total}$  is the same for all the experiments.

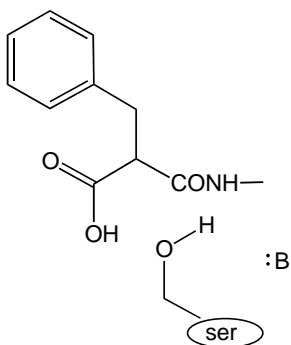
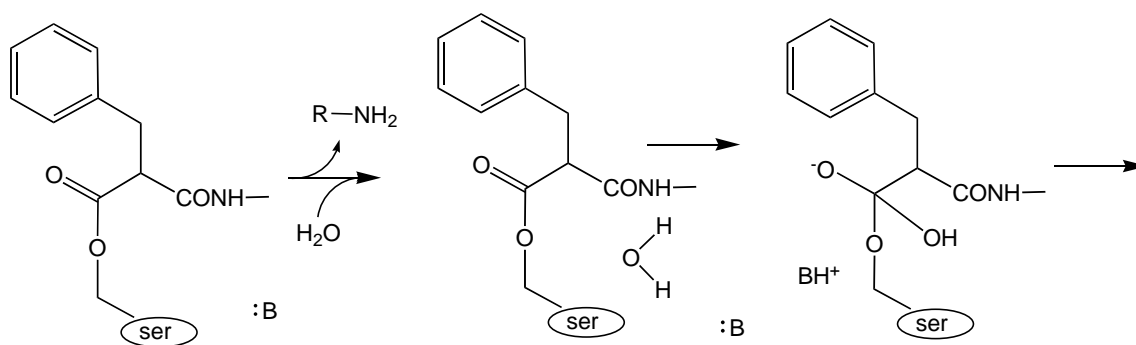
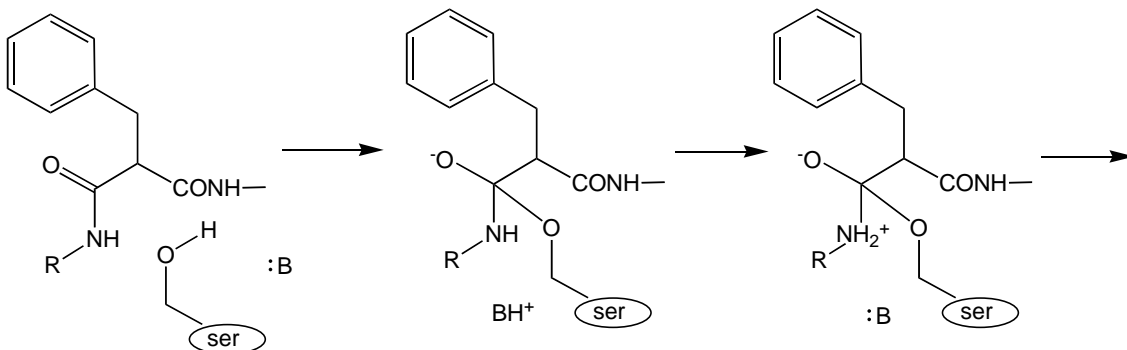
Question 2 (9 points). Explain how the lysozyme mechanism illustrates

- Transition state stabilization (and ES destabilization)
- General acid-base catalysis
- Electrostatic catalysis

Question 3 (12 points). Explain how the serine protease mechanism illustrates:

- Transition state stabilization
- Covalent catalysis
- General acid-base catalysis
- Electrostatic catalysis

**Question 4** (12 points). Shown below are the steps in amide hydrolysis catalyzed by chymotrypsin. Draw curved arrows to indicate the movement of electrons through the process. Indicate lone pairs of electrons by pairs of dots. Indicate the step at which C-N bond cleavage occurs.



Question 5 (10 points). For the hydrolysis of a dipeptide KX (X is any amino acid) catalyzed by trypsin:

- (a) What is the rate determining step?
- (b) What experimental evidence supports this?
- (c) Use the mechanism of trypsin-catalyzed peptide hydrolysis to explain which (if any) of the 20 common amino acids at X will cause the greatest increase in the reaction rate.

Question 6 (8 points). The low-barrier H-bond between the two aspartate residues in the aspartic proteases becomes two “normal” H-bonds in ES (Figure 14.29 in Garrett and Grisham). If the energy difference in H-bonding modes is used to deprotonate the water molecule in ES, by how much could the  $pK_a$  of the water molecule be changed?