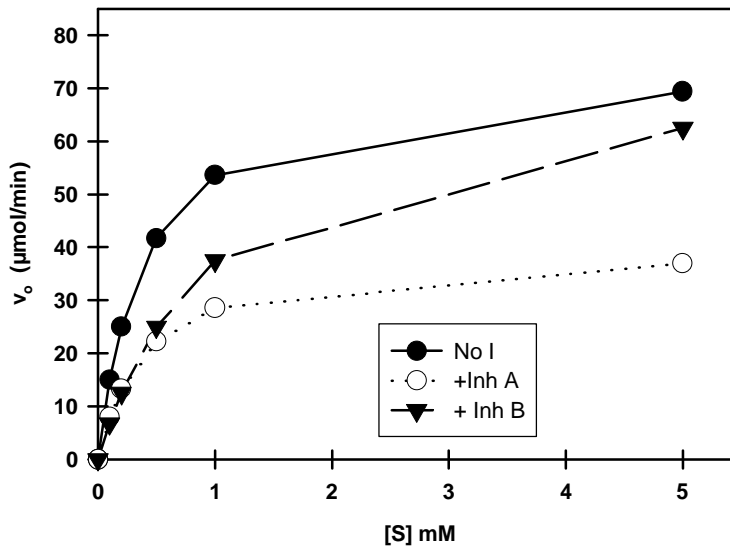


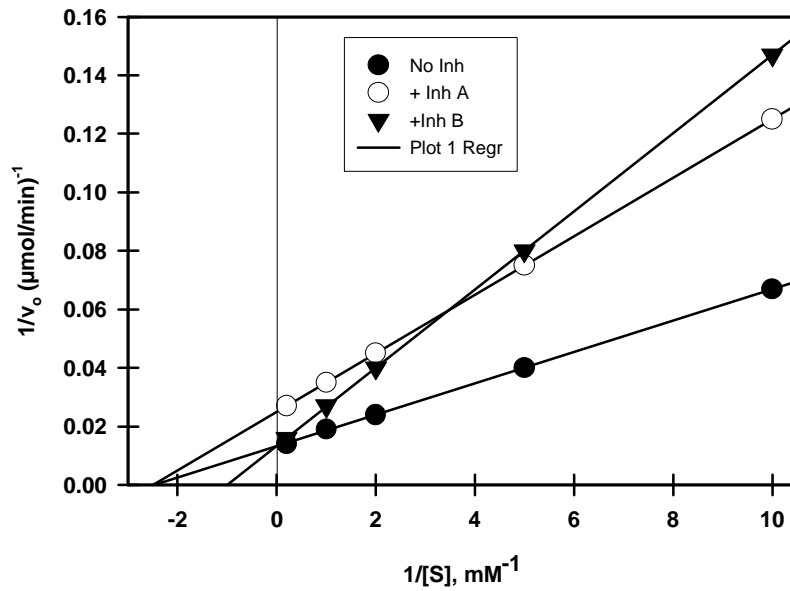
## 1. 50 Pts. Enzyme Kinetics Problem

- Draw plots of  $V_o$  vs  $[S]$  and  $1/V_o$  vs  $1/[S]$ , using the data given below - 25 Pts
- Determine  $K_m$  and  $V_{max}$  for uninhibited reaction (include units) - 10 Pts
- Determine what type of Inhibitor A and B are using the  $K_m'$  and  $V_{max}'$  for the inhibitors (show your reasoning by comparing  $V_{max}$  and  $V_{max}'$  and  $K_m$  and  $K_m'$ ) - 10 Pts
- Calculate the  $K_i$  for binding of Inhibitor A and B to the enzyme (include units) - 5 Pts

| [S] | $V_o$ (No I)               | $V_o$ (I = 0.1 mM A)       | $V_o$ (I = 2.0 mM B)       |
|-----|----------------------------|----------------------------|----------------------------|
| mM  | $\mu\text{mol}/\text{min}$ | $\mu\text{mol}/\text{min}$ | $\mu\text{mol}/\text{min}$ |
| 0.0 | 0.0                        | 0.0                        | 0.0                        |
| 0.1 | 15.0                       | 8.0                        | 6.8                        |
| 0.2 | 25.0                       | 13.3                       | 12.5                       |
| 0.5 | 41.7                       | 22.2                       | 25.0                       |
| 1.0 | 53.6                       | 28.6                       | 37.5                       |
| 5.0 | 69.4                       | 37.0                       | 62.5                       |

Exam 2 1997 -- Kinetics Problem  $v_o$  vs  $[S]$  Plot

Exam 2 1997 Kinetics Problem Double Reciprocal Plot



Summary Table of Kinetic Constants and Type of Inhibitor for A and B

| Parameter    | V <sub>max</sub> or V <sub>max</sub> ' | K <sub>m</sub> or K <sub>m</sub> ' | Type of Inhibitor   |
|--------------|--|------------------------------------|---|
| Units        | μmol/min                               | mM                                 |   |
| No Inhibitor | 75                                     | 0.4                                |   |
| Inhibitor A  | 40                                     | 0.4                                | Non Competitive<br>V <sub>max</sub> ' ≠ V <sub>max</sub><br>K <sub>m</sub> ' = K <sub>m</sub> |
| Inhibitor B  | 75                                     | 1.0                                | Competitive<br>V <sub>max</sub> ' = V <sub>max</sub><br>K <sub>m</sub> ' ≠ K <sub>m</sub>     |

**Calculation of K<sub>i</sub> values for Inhibitor A and B:**

Inhibitor A at I = 0.1 mM A

$$K_i = I / (V_{max} / V_{max}' - 1) = 0.1 \text{ mM} / ((75/40) - 1) = 0.11 \text{ mM A}$$

Inhibitor B at I = 2.0 mM B

$$K_i = [I] / (K_m' / K_m - 1) = 2.0 \text{ mM} / ((1.0/0.4) - 1) = 1.3 \text{ mM B}$$

2. 40 Pts. Protein Structure Problem (Use Words and Diagrams)

**Describe the four levels of protein structure and bonding stabilizing these structures.**

Be sure to illustrate each level of the structure of a protein with a drawing of the bonds or interactions involved among the polypeptide backbone and the amino acid side chains.

**SEE Lectures 8 and 9**

3. 10 Pts. Thought Question

Name the specific amino acid side chains you expect to find in the following regions of an enzyme and explain (briefly) why you expect to find them there:

| Enzyme Region   | Amino Acid  | Reason   |
|---|---|--|
| Interior  | Hydrophobic<br>(Ala, Val, Leu, Ile, Pro, Phe, Trp, Met and maybe Tyr)   | Hydrophobic amino acid side chains fold to the interior of proteins and enzymes, since this is a lower energy state in aqueous solution. Hydrophobics structure water and so the system has greater entropy (lower free energy or more stable) when they are inside and away from water.   |
| Surface   | Hydrophilic<br>(Ser, Thr, Cys, Asp, Glu, Asn, Gln, Lys, Arg, His and maybe Tyr)   | Hydrophilic or polar amino acid side chains tend to stay on the surface of the proteins and enzymes during folding since their interaction with water is energetically favorable.  |
| Active Site<br>1. General<br>2. Binding<br><br>3. Catalysis | Any amino acid side chain<br>(For binding substrate, any one may be required.<br>For catalytic function, it is usually more limited to hydrophilics.) | The amino acid side chains found in the Active site of an enzyme are governed by the nature of the substrate and the reaction being catalyzed. Since substrates often have both hydrophilic and hydrophobic character, at least in some cases, both general types of amino acid side chains are found in the active site. Amino acid side chains specifically assisting in catalysis in the active site are usually hydrophilic. |