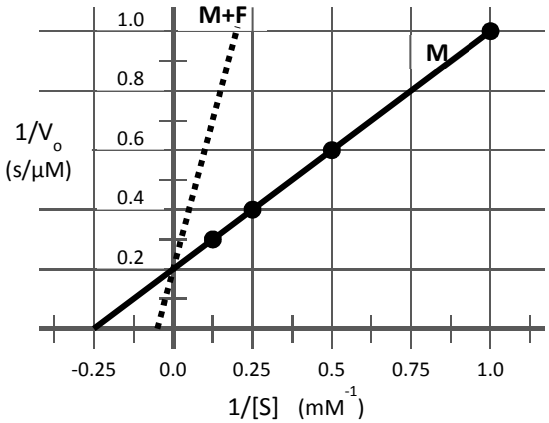
1. a. From each graph, you get one data point with values of V_o (the slope of the dotted line) and [S]. To create a Lineweaver-Burk plot, the reciprocals must be calculated:

[S] (mM)	**V_o** (μM/s)	**1/[S]** (1/mM)	**$1/V_o$** (s/μM)
1	1	1	1
2	5/3	0.5	0.6
4	5/2	0.25	0.4
8	10/3	0.125	0.3

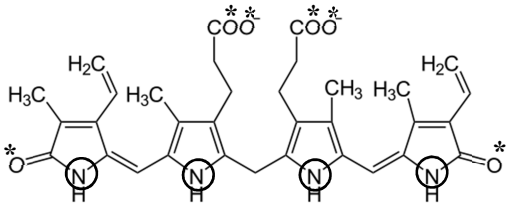


   b. x-intercept = $-1/K_m$
   K_m = -1/x-intercept = $-1/(-0.25\ mM^{-1})$ = <u>4 mM</u>
   c. y-intercept = $1/V_{max}$
   V_{max} = 1/y-intercept = 1/(0.2 s/μM) = <u>5 μM/s</u>
   d. $k_{cat} = V_{max}/[E_T]$ = (5 μM/s)/(0.0008 μM) = <u>6,250 s^{-1}</u>
   e. catalytic efficiency is measured by the specificity constant:
   k_{cat}/K_m = $(6{,}250\ s^{-1})/(4\ mM)$ = <u>1562.5 $s^{-1}\ mM^{-1}$</u>
   f. inhibitor line: x-intercept = $-1/K'_m = -1/(20\ mM) = -0.05\ mM^{-1}$
   y-intercept = $1/V'_{max}$ = 1/(5 μM/s) = 0.2 s/μM
   g. competitive
   h. $K'_m = \alpha K_m$ So, $\alpha = K'_m/K_m$ Also, $\alpha = 1 + [I]/K_I$
   Or, $K_I = [I]/(\alpha - 1) = [I]/\{(K'_m/K_m)-1\} = (5\ mM)/\{((20\ mM)/(4\ mM))-1\} = (5\ mM)/(5-1) =$ <u>1.25 mM</u>
   i. frigidol, since $K_I < K_m$

2. a. $k_{-1} >> k_2$ (k_2 is rate-limiting)
   b. A rate constant for a reaction with a single reactant (in $time^{-1}$ units).
   c. k_{-1} and k_2
   d. $K_d = k_{-1}/k_1$ and $K_d = [E][S]/[ES]$
   e. When rate of formation of ES = rate of breakdown of ES: $k_{-1}[ES] + k_2[ES] = k_1[E][S] + k_{-2}[E][P]$

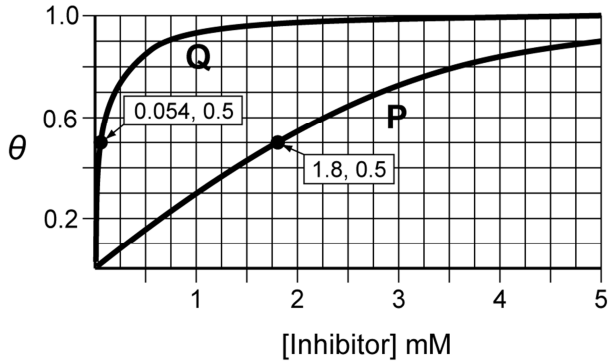
3. a. True
   b. False; for example, if the reaction is proceeding under steady-state conditions, the rate will be constant.

4. a. b.



   c. Higher. Bilirubin's carboxylates lack the nearby $NH3^+$ group of glycine, which through electron withdrawing (inductive effect) and through charge balance (electrostatic effect) promotes deprotonation of glycine's carboxyl group.
   d. double-reciprocal or Lineweaver-Burk plot
   e. substrate
   f. bilirubin is a competitive inhibitor
   g. A, D, E, F, H, I, J, K, L, (M)

5. a. 2, 6
   b. 1, 2, 3, 4
   c. 3, 4
   d. 1, 4
6. a. competitive
   b. mixed
   c. mixed, noncompetitive
   d. competitive
   e. Increasing enzyme concentration increases V_{max}, but doesn't change K_m. So:
      A. The slope would *decrease*
      B. The x-intercept would *not change*
      C. The y-intercept would *decrease*
   f. Increasing inhibitor concentration shows the same effect (but to a greater extent) as adding inhibitor to a solution that had none. So:
      A. The slope would *increase*
      B. The x-intercept would *decrease* (shift left)
      C. The y-intercept would *increase*
   g. Yes. P and A compete for binding, and Q and X compete for binding, but neither inhibitor interferes with the binding of the other substrate. It is therefore likely that both can bind together (in place of the two substrates).
   h. Q; its K_I is lower (indicating higher affinity)
   i.



7. a. $k_1[A][X] + k_4[C] = k_2[B][Y] + k_3[B]$
   b.

$$\Delta G'^{\circ} = -RT \ln \frac{[C]}{[B]}, \text{ or } \frac{[C]}{[B]} = e^{\Delta G'^{\circ}/-RT}$$

some of B will be coverted to C; call this concentration 'x'

$$\frac{[C]}{[B]} = \frac{x}{100\text{mM} - x} = e^{-7.2\text{kJ/mol}/-(8.31x10^{-3}\text{kJ/mol·K})(298\text{K})} = 18.31$$

$$x = 1831\,\text{mM} - 18.31x, \text{ or } 19.31x = 1831\,\text{mM}$$

$$x = [C] = 94.8\,\text{mM} \text{ and } [B] = 100 - 94.8 = 5.2\,\text{mM}$$

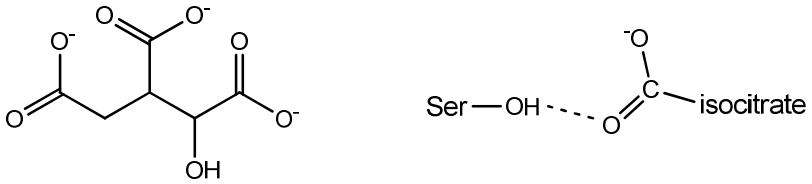
   c. It depends on the *exact* relationship between [B] and [C]. If [C]/[B] < 94.8/5.2 (or 18.2), the reaction will go B→C; if [C]/[B] > 18.2, C→B; if [C]/[B] = 18.2, the reaction will be at equilibrium.

8. $\Delta G = \Delta G'^{\circ} + RT\ln Q = 7.1\ \text{kJ/mol} + \left(0.008315\frac{\text{kJ}}{\text{mol·K}}\right)(310\ \text{K})\ln\left(\frac{[\text{Z}]^2}{[\text{X}]}\right)$
9. $\Delta G = \Delta G'^{\circ} + RT\ln Q$

   The reaction will be spontaneous when $\Delta G < 0$, so

   - $0 > \Delta G'^{\circ} + RT\ln\left(\frac{[\text{Y}]}{[\text{X}]}\right)$
   - $\frac{-\Delta G'^{\circ}}{RT} > \ln\left(\frac{[\text{Y}]}{[\text{X}]}\right)$
   - $e^{-\Delta G'^{\circ}/RT} > \frac{[\text{Y}]}{[\text{X}]}$
   - $[\text{X}] \cdot e^{-\Delta G'^{\circ}/RT} > [\text{Y}]$
   - $100\ \mu\text{M} \cdot e^{-4.9\ \text{kJ/mol}/(0.008315\ \text{kJ/mol·K})(310\ \text{K})} > [\text{Y}]$
   - $100\ \mu\text{M} \cdot 0.15 > [\text{Y}]$
   - $\mathbf{15\ \mu M > [Y]}$

10. a. Isocitrate dehydrogenase
    b. *Escherichia coli*
    c. 3D structural information; atomic positions
    d. Four
    e. Wild-type enzyme (with bound substrate), Ser113-phosphorylated enzyme (inactive), enzyme with Ser113→Asp mutation, enzyme with Ser113→Glu mutation
    f. Given the structure of isocitrate (left), and the description, the H-bond likely involves a carboxylate:



    g. The phosphoryl group has -2 charge, which repels the negatively charged carboxyl groups of isocitrate, preventing binding. (The bulkiness of the phosphoryl group could also interfere with binding.)
    h. No

11. e. Since [Y] > [X] at equilibrium, the system could be in any of the states described in choices a-d, and it could be far from equilibrium (if [Y] is much greater than [X]). Since no conclusion can be made, the answer is 'none of the above.'

12. Acetyl-CoA

13. a. True
    b. True
    c. True
    d. False – this is true of the sequential model
    e. False – the term 'zymogen' is specific to proteases; in general, the term is 'proprotein'
    f. True