

Discussion Exercise 5: Analyzing Graphical Data

Key

Problem 1: Use the graphs below to describe relationships between phenomena and a variable or answer questions concerning the relationships.

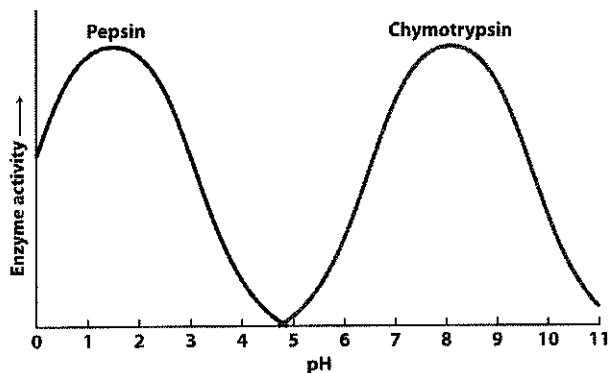


Figure 5.4
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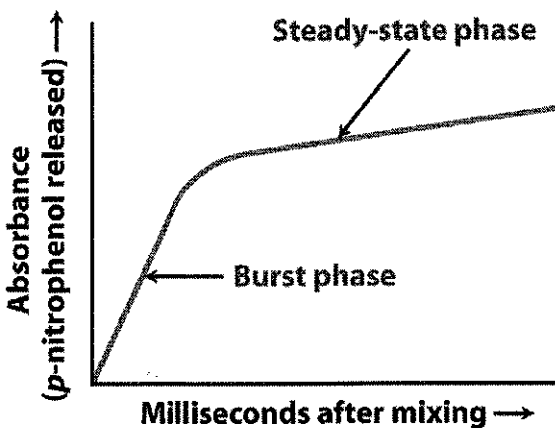


Figure 5.22
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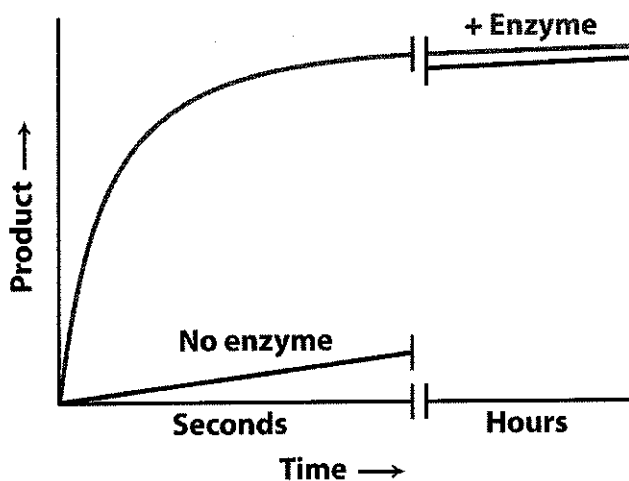
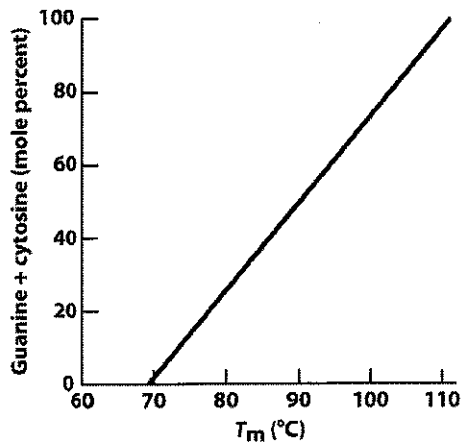


Figure 6.2
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Problem 11.20
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A. What is the relationship between the pH of a solution and the catalytic activity of pepsin?

It is a bell-shaped curve with maximum activity between a pH of 1 and 2.

B. How does doubling the mole % of G-C base pairs affect the melting point of the DNA double helix?

Because it is NOT directly linear, doubling the % G-C content has different effects. Doubling from 20% to 40% raises the T_m by 9°C, from 77 to 86° but raising the G-C content from 40% to 80% raises the temperature from 86 to 104°C. Even though it is not directly proportional, it is linear, with an increase of about 4.5° per 10% increase in GC content.

C. For an enzyme catalyzed reaction, when is the concentration of product formation approximately linear as a function of time?

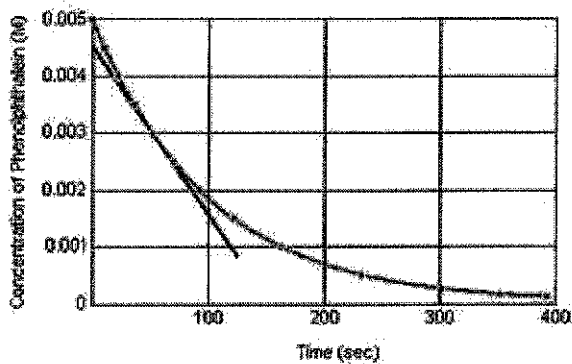
It is approximately linear at low concentrations of substrate, where it is first order (rate doubles as [S] doubles), and it is approximately linear at high concentration, where [S] has no effect on rate (zero order.)

D. What is the relationship of absorbance of product released to time during the burst phase of kinetics? How does the relationship change in the steady-state phase?

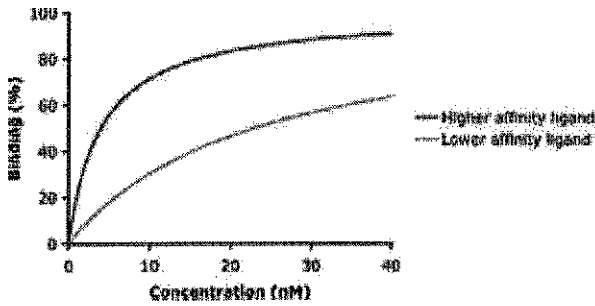
In the burst phase, it is linear with a fast rate (steep slope), but at steady state, it is linear with a more shallow slope (lower rate.)

Problem 2: Draw a graph showing each of these relationships.

A. During the initial part of a reaction reaction, the concentration of substrate is inversely proportional to the time of the reaction, then slowly approaches zero after a long time.



B. At low concentrations of a hormone, receptor/hormone binding increases linearly with concentration of hormone present, but at high concentrations of hormone, binding of receptor and hormone is unaffected by hormone concentration. (Plot % bound versus [hormone]).



C. An enzyme catalyzed reaction rate increases with an increase in temperature until it reaches 41 °C, at which point, the reaction abruptly stops.

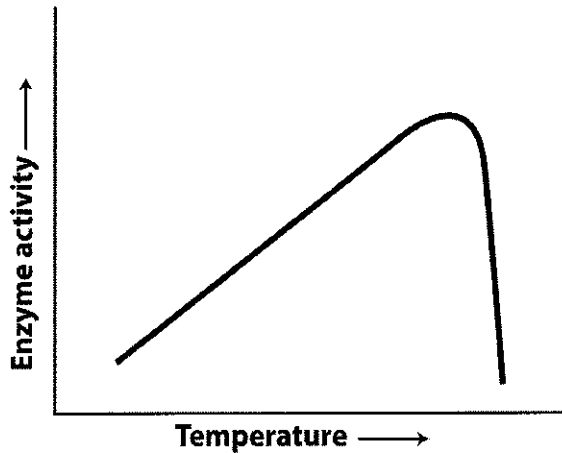
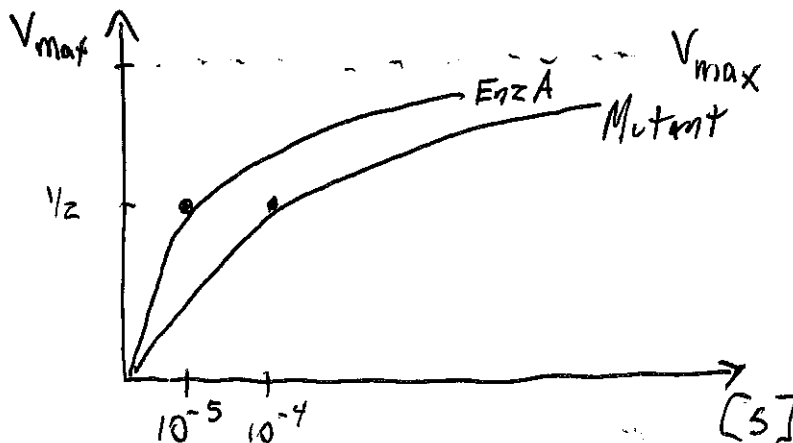


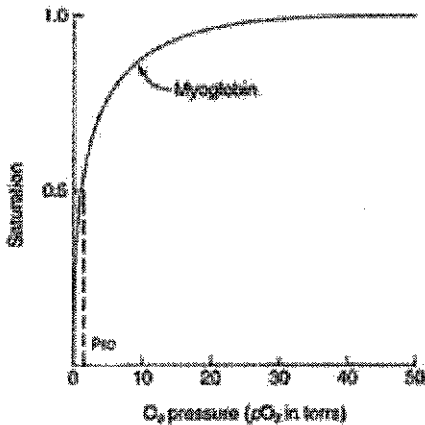
Figure 8.1a
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Problem 3: A mutation in Enzyme A has no effect on V_{max} , but raises K_M from 1×10^{-5} to 1×10^{-4} . Draw a Michaelis Menton curve for Enzyme A and its mutant. Did the mutation cause the enzyme to become a better or worse catalyst?

It is a worse enzyme at lower [S] but is unaffected at higher [S].



Problem 4: Myoglobin is an oxygen binding protein. Its binding curve is shown below. Using this curve, what is the value of “a” in the graph? What is its physical meaning? What is the value of “b”? What is its physical meaning?



“a” is the maximum asymptote. Its value is 1.0 because this is a % saturation, with the maximum saturation being 100%. “b” is approximately 2 torr. Physically, this means that if myoglobin is put into a system that has 2 torr oxygen pressure, that is enough oxygen so that half of the protein will have oxygen bound to it.

Problem 5: Answer the questions below based on the graphs.

A. Which of the graphs suggests cooperative binding?

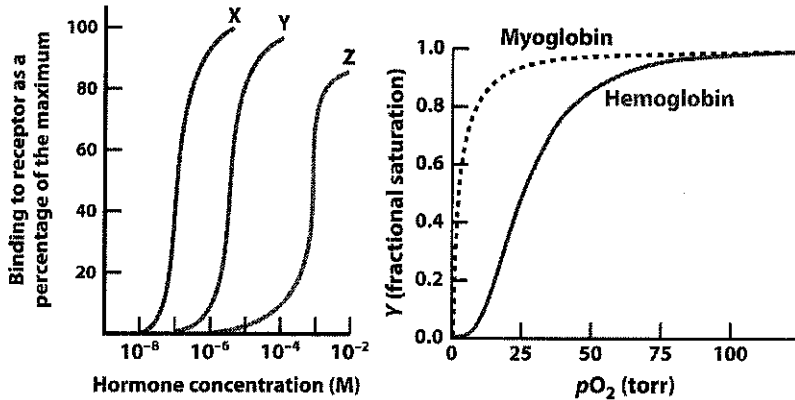
The hemoglobin curve suggests cooperative binding because it is sigmoidal on a linear x-axis. The hormone binding isn't cooperative, because it is sigmoidal on a semi-log scale.

B. Which hormone, X, Y, or Z, binds tightest to its receptor?

Hormone X binds tightest. It has a K_D of below 1×10^{-7} , so it is half bound when this hormone is at very low concentrations. Hormone Z binds about 4 orders of magnitude (10,000 times) worse.

C. Both hemoglobin and myoglobin bind oxygen well at high concentrations of oxygen, but which binds oxygen better at low concentrations of oxygen? Most tissues in the body have a pressure of oxygen in the 20-40 torr region. Which protein, hemoglobin or myoglobin, will have greater effects to its oxygen binding when a tissue oxygen level drops from 30 torr to 25 torr?

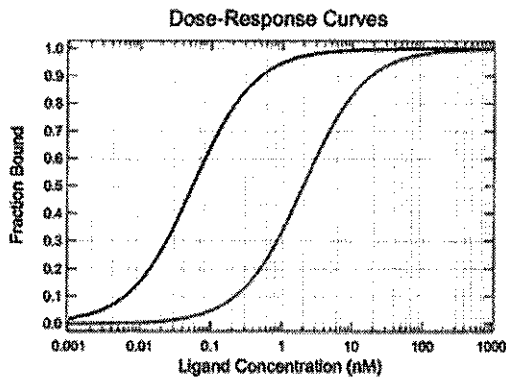
Myoglobin binds oxygen much better at low $[O_2]$ —for example it is 90% bound at 5 torr, but hemoglobin is only 2% bound. Hemoglobin has a steeper binding curve in the tissue region of oxygen pressure, so it is more affected. If the tissue levels of oxygen drop from 30% to 25%, hemoglobin will go from 65% bound to 45% bound, releasing oxygen to the tissue. Myoglobin, on the other hand, will only go from about 93% bound to 92% bound in that region.



Problem 13.22a
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Figure 5.1
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Problem 6: A dose-response curve is drawn to show the relationship between the amount of a drug administered and its effect. In the chart below, two different drugs are tested and placed on the same graph. Drug A gives the left curve and drug B gives the right curve.



A. Do these data suggest that drugs A and B exhibit cooperative binding?

No, this is a binding curve on a semi-log scale.

B. What is the dose of drug B that gives half maximal response?

The dose is 2 nM.

C. Which drug is most effective at lower doses? About how many times more effective is it?

Drug A is more effective. It is half bound at about 0.05 nM, making it about 40 times more effective.

