**Enzyme Kinetics**

Enzymes are protein catalysts that, like all catalysts, speed up the rate of a chemical reaction without being used up in the process.

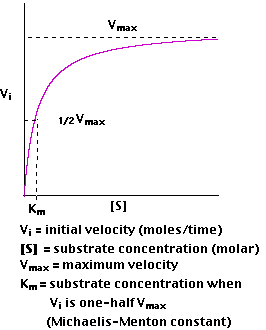
They achieve their effect by temporarily binding to the **substrate** and, in doing so, lowering the [activation energy](http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/E/Enzymes.html) needed to convert it to a product.

The rate at which an enzyme works is influenced by several factors, e.g.,

* the concentration of substrate molecules (the more of them available, the quicker the enzyme molecules collide and bind with them). The concentration of substrate is designated **[S]** and is expressed in unit of molarity.
* the [temperature](http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/E/Enzymes.html#pHandTemp). As the temperature rises, molecular motion - and hence collisions between enzyme and substrate - speed up. But as enzymes are proteins, there is an upper limit beyond which the enzyme becomes [denatured](http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/D/DenaturingProtein.html) and ineffective.
* the presence of inhibitors.
  + competitive inhibitors are molecules that bind to the same site as the substrate - preventing the substrate from binding as they do so - but are not changed by the enzyme.
  + noncompetitive inhibitors are molecules that bind to some other site on the enzyme reducing its catalytic power.
* [pH](http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/E/Enzymes.html#pHandTemp). The conformation of a protein is influenced by pH and as enzyme activity is crucially dependent on its conformation, its activity is likewise affected.

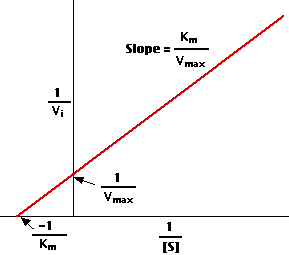
The study of the rate at which an enzyme works is called **enzyme kinetics**. Let us examine enzyme kinetics as a function of the **concentration of substrate** available to the enzyme.

* We set up a series of tubes containing graded concentrations of substrate, **[S]**.
* At time zero, we add a fixed amount of the enzyme preparation.
* Over the next few minutes, we measure the concentration of product formed. If the product absorbs light, we can easily do this in a spectrophotometer.
* Early in the run, when the amount of substrate is in substantial excess to the amount of enzyme, the rate we observe is the initial velocity of **Vi**.

Plotting **Vi** as a function of **[S]**, we find that

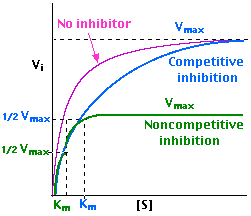
* At low values of **[S]**, the initial velocity,**Vi**, rises almost linearly with increasing **[S]**.
* But as **[S]** increases, the gains in **Vi** level off (forming a rectangular hyperbola).
* The asymptote represents the maximum velocity of the reaction, designated **Vmax**
* The substrate concentration that produces a **Vi** that is one-half of **Vmax** is designated the Michaelis-Menten constant, **Km**(named after the scientists who developed the study of enzyme kinetics).

**Km** is (roughly) an inverse measure of the affinity or strength of binding between the enzyme and its substrate. The lower the **Km**, the greater the affinity (so the lower the concentration of substrate needed to achieve a given rate).

Plotting the reciprocals of the **same data points** yields a "double-reciprocal" or Lineweaver-Burk plot. This provides a more precise way to determine **Vmax** and **Km**.

* **Vmax** is determined by the point where the line crosses the 1/**Vi** = 0 axis (so the **[S]** is infinite).
* Note that the magnitude represented by the data points in this plot **decrease** from lower left to upper right.
* **Km** equals **Vmax** times the slope of line. This is easily determined from the intercept on the X axis.

**The Effects of Enzyme Inhibitors**

Enzymes can be inhibited

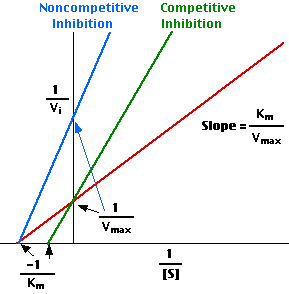
* **competitively**, when the substrate and inhibitor compete for binding to the same active site or
* **noncompetitively**, when the inhibitor binds somewhere else on the enzyme molecule reducing its efficiency.

The distinction can be determined by plotting enzyme activity with and without the inhibitor present.

**Competitive Inhibition**

In the presence of a competitive inhibitor, it takes a higher substrate concentration to achieve the same velocities that were reached in its absence. So while **Vmax** can still be reached if sufficient substrate is available, one-half **Vmax** requires a higher **[S]** than before and thus **Km** is larger.

**Noncompetitive Inhibition**

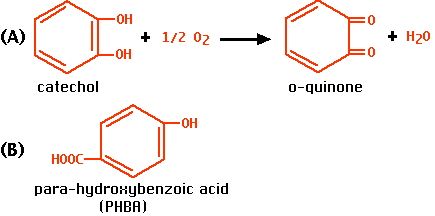
With noncompetitive inhibition, enzyme molecules that have been bound by the inhibitor are taken out of the game so

* enzyme rate (velocity) is reduced for all values of **[S]**, including
* **Vmax** and one-half **Vmax** but
* **Km** remains unchanged because the active site of those enzyme molecules that have not been inhibited is unchanged.

This Lineweaver-Burk plot displays these results.

**An Example**

When a slice of apple is exposed to air, it quickly turns brown. This is because the enzyme **o-diphenol oxidase** catalyzes the oxidation of phenols in the apple to dark-colored products. (A similar enzyme, tyrosinase, converts [tyrosine](http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/T/Tyr_phe.gif) to [melanin](http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/M/MSH.html).)

Let us determine: 

* the maximum rate at which the enzyme can perform (**Vmax**) and
* the Michaelis-Menten constant (**Km**) for this enzyme
  1. when it acts **alone**. We shall use **catechol** as the substrate. The enzyme converts it into **o-quinone** **(A)**, which is then further oxidized to dark products.
  2. when it acts in the presence of a **competitive inhibitor**. We shall use para-hydroxybenzoic acid (**PHBA**) **(B)**, which binds the same site as catechol but is not acted upon.
  3. when it acts in the presence of a **noncompetitive inhibitor**. We shall use phenylthiourea which binds to a copper atom in the enzyme which is essential for its activity.

Preparing for the Assay:

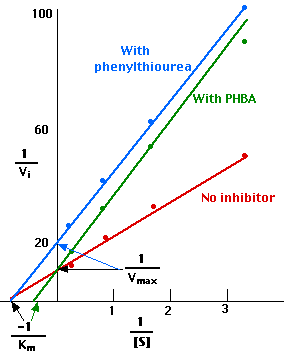
* Grind up pieces of apple and centrifuge the resulting soup.
* The supernatant is your enzyme preparation.
* Because of the speed with which colored products are formed, we can use the intensity of the color as a measure of product formation.
* We measure this in a spectrophotometer, an instrument that measures the absorbance of monochromatic light passed through the sample. Because the products are yellow-brown, they absorb green light (540 nm) best.

**First Experiment: No Inhibitor**

* Set up four tubes with different concentrations of catechol (the substrate). (Catechol is a relative of the active ingredients in the poison ivy plant and, like them, can cause serious contact sensitivity if it gets on one's skin.)
  + A = 4.8 mM; B = 1.2 mM; C = 0.6 mM; D = 0.3 mM
* Add a fixed amount of enzyme preparation to Tube A and measure the change in absorbance (**O**ptical **D**ensity) at 540 nm) at 1 minute intervals for several minutes.
* Record the average **change** in OD540 per minute (Δ OD540).
* Because the OD is directly proportional to the concentration of the products, we can use it as a measure of the rate or velocity of the reaction (**Vi**).
* Repeat with the other three tubes.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | Tube A | Tube B | Tube C | Tube D |
| **[S]** | 4.8 mM | 1.2 mM | 0.6 mM | 0.3 mM |
| **1/[S]** | 0.21 | 0.83 | 1.67 | 3.33 |
| Δ OD540 (**Vi**) | 0.081 | 0.048 | 0.035 | 0.020 |
| **1/Vi** | 12.3 | 20.8 | 31.7 | 50.0 |

The table above summarizes the results.

Making a Lineweaver-Burk plot of these results shows (**red**) that

* 1/**Vmax** = 10, so **Vmax** = 0.10
* −1/**Km** = − 0.8, so **Km** = 1.25 mM
* (In other words, when **[S]** is 1.25 mM, 1/**Vi** = 20, and **Vi** = 0.05 or one-half of **Vmax**.)

**Second Experiment: Effect of para-hydroxybenzoic acid (PHBA)**

As before, but this time add a fixed amount of a solution of PHBA to each of the four tubes.

The table below summarizes the results.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | Tube A | Tube B | Tube C | Tube D |
| **[S]** | 4.8 mM | 1.2 mM | 0.6 mM | 0.3 mM |
| **1/[S]** | 0.21 | 0.83 | 1.67 | 3.33 |
| ΔOD540 (**Vi**) | 0.060 | 0.032 | 0.019 | 0.011 |
| **1/Vi** | 16.7 | 31.3 | 52.6 | 90.9 |

The Lineweaver-Burk plot of these results is shown above in **green**.

* 1/**Vmax** = 10, so **Vmax** remains 0.10.
* Now, however, −1/**Km** = − 0.4, so **Km** = 2.50 mM
* (In other words, it now takes a substrate concentration **[S]** of 2.50 mM, to achieve one-half of **Vmax**.)

**Third Experiment: Effect of phenylthiourea**

As before, but this time add a fixed amount of a solution of phenylthiourea in each of the four tubes.

The table below summarizes the results.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | Tube A | Tube B | Tube C | Tube D |
| **[S]** | 4.8 mM | 1.2 mM | 0.6 mM | 0.3 mM |
| **1/[S]** | 0.21 | 0.83 | 1.67 | 3.33 |
| ΔOD540 (**Vi**) | 0.040 | 0.024 | 0.016 | 0.010 |
| **1/Vi** | 25 | 41 | 62 | 102 |

The Lineweaver-Burk plot of these results is shown above in **blue**.

* Now 1/**Vmax** = 20, so **Vmax** = 0.05.
* But −1/**Km** = − 0.8, so **Km** = 1.25 mM as it was in the first experiment.
* So once again it only takes a substrate concentration,**[S]**, of 1.25 mM to achieve one-half of **Vmax**.

**Summary**

Here, then, is a method by which catalytic power of different enzymes can be compared.

The table gives **Km** values (mM) for several enzymes - some of which you can encounter with links to other pages on this site.

|  |  |  |
| --- | --- | --- |
| **Enzyme** | **Substrate** | **Km** (**mM**) |
| Catalase | H2O2 | 1,100 |
| [Chymotrypsin](http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/G/GITract.html#pancreas) | Gly-Tyr-Gly | 108 |
| [Carbonic anhydrase](http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/B/Blood.html#CO2) | CO2 | 12 |
| [beta-galactosidase](http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/L/LacOperon.html) | [D-lactose](http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/C/Carbohydrates.html#disaccharides) | 4 |
| [Acetylcholinesterase](http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/M/Muscles.html#NeuromuscularJunction) | acetylcholine (ACh) | 0.09 |
| [beta-lactamase](http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/A/Antibiotics.html#BetaLactams) | benzylpenicillin | 0.02 |

|  |
| --- |
| [Link to a general discussion of other aspects of enzyme structure and function.](http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/E/Enzymes.html) |

|  |
| --- |
| [Welcome&Next Search](http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/W/Welcome.html) |

Published by [ur guide](http://www.triond.com/users/ur+guide)   
February 5, 2008, Category: [Microbiology](http://scienceray.com/category/biology/microbiology/)

A very easy quiz to develop confidence. An average person should score around 80%.

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Let’s see how much knowledge have you got. On an average, a person must score around 80% in this quiz.

1. Why a [bag](http://scienceray.com/biology/microbiology/quiz-on-enzymes/) of sugar is not converted to [carbon dioxide](http://scienceray.com/biology/microbiology/quiz-on-enzymes/) and water?
   1. rate of reaction is very slow
   2. sugar is not reactive
   3. equilibrium constant is very low
2. Which of the following is not the property of enzyme:
   1. to alter rate of equilibrium
   2. specificity to substrate
   3. alter rate of reaction
3. When biological catalysis was first recognized and described?
   1. 1600’s
   2. 1800’s
   3. 1700’s
4. On which food product, the biological catalysis was first recognized and described?
   1. rice
   2. wheat
   3. meat
5. In 1850’s, which scientist concluded that fermentation of sugar to alcohol is catalyzed?
   1. Edward Buchner
   2. Louis Pasteur
   3. Alfred Joseph
6. What was the name given to enzymes by Louis Pasteur?
   1. enzyme
   2. ferment
   3. catalyst
7. In which year Edward Buchner discovered that yeast extract can cause the fermentation of sugar to alcohol?
   1. 1888
   2. 1896
   3. 1897
8. Who gave the word “enzymes” to catalytic molecules?
   1. Louis Pasteur
   2. Frederick W. Kuhne
   3. Edward Buchner
9. In which year James Sumner isolated and crystallized urease?
   1. 1926
   2. 1928
   3. 1924
10. All enzymes are made up of which biomolecules (or, biomolecules)?
    1. proteins
    2. RNA
    3. Both a and b

Read more in [Microbiology](http://scienceray.com/category/biology/microbiology/)

[« Bacteria Are Talking to Each Other](http://scienceray.com/biology/microbiology/bacteria-are-talking-to-each-other/)

[Chromatography »](http://scienceray.com/biology/microbiology/chromatography/)

Answers:

1. 1
2. 1
3. 3
4. 3
5. 2
6. 2
7. 3
8. 2
9. 1
10. 3

More enzyme quiz related articles, refer:

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Quiz on Enzymes Three

Published by [ur guide](http://www.triond.com/users/ur+guide) in [Quizzes](http://purpleslinky.com/category/trivia/quizzes/)  
April 26, 2008

The great quiz is back with a bang, with more questions.

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Quiz on enzyme is back with more questions and more clues. On average your score should be around 75%.

1. What is covalent catalysis?
   1. Formation of hydrogen bond between enzyme and substrate
   2. Formation of covalent bond between enzyme and substrate
   3. Formation of transient covalent bond between enzyme and substrate
   4. None of the above
2. How metal ions participate in catalysis?
   1. By causing reduction and oxidation reactions between enzyme and substrate
   2. By causing ionic interactions between enzyme and substrate
   3. All of the above
   4. None of the above
3. What is the common enhancing rate of enzymes?
   1. 105 to 1017
   2. 1012 to 1020
   3. 10-17 to 10-9
   4. None of the above
4. Enzyme chymotrypsin carries out which type of catalysis?
   1. Covalent catalysis
   2. General acid base catalysis
   3. None of the above
   4. Both a and b
5. What is enzyme kinetics?
   1. Studying the mechanism of rate of reaction of enzyme
   2. Factors affecting enzyme activity
   3. Both a and b
   4. None of the above
6. What is Vmax?
   1. Maximum rate of reaction
   2. Rate of reaction increase with increase in enzyme concentration
   3. Both a and b
   4. None of the above
7. In which year, Victor Henri proposed that the combination of an enzyme with its substrate molecule to form an ES complex is necessary?
   1. 1904
   2. 1903
   3. 1902
   4. 1901
8. Who gave the general theory of enzyme action in 1913?
   1. Leonor Michaelis and Maud Menten
   2. Alfred Michaelis
   3. Maud Michaelis
   4. Leonor Menten and Maud Michaelis
9. In an enzyme catalyzed reaction, enzyme exists in which form?
   1. Free form
   2. Combined form
   3. ES form
   4. All of the above
10. The initial period when ES concentration builds up in an enzyme catalyzed reaction is called?
    1. Steady state
    2. Initial period
    3. Pre steady state
    4. Ephemeral state
11. The state which occurs after pre-steady state is called?
    1. Pro-steady state
    2. Late pre steady state
    3. Post steady state
    4. Steady state
12. Who introduced the concept of steady state kinetics in 1925?
    1. G.E. Briggs and Haldane
    2. Michaelis and Menten
    3. Briggs
    4. Menten
13. What is Km in Michaelis-Menten equation?
    1. Michaelis-Menten constant
    2. Substrate
    3. Enzyme
    4. All of the above
14. The initial reaction rate reflects a steady state in which [ES] is constant. This is called?
    1. Michaelis and Menten state
    2. Steady state assumption
    3. Initial state assumption
    4. None of the above
15. Which enzymes are said to follow Michaelis-Menten kinetics?
    1. Enzymes which show parabolic dependence of rate of reaction and substrate
    2. Enzymes which show circular dependence of rate of reaction and substrate
    3. Enzymes which show hyperbolic dependence of rate of reaction and substrate
    4. None of the above
16. Double-reciprocal plot is also called?
    1. Michaelis plot
    2. Line plot
    3. Lineweaver-Burk plot
    4. Inverse plot
17. What is Kd?
    1. Dissociation constant
    2. Michaelis constant
    3. Lineweaver constant
    4. None of the above
18. What is the unit of Kcat?
    1. Reciprocal of concentration
    2. Reciprocal of time
    3. Both a and b
    4. None of the above
19. Km and Kcat of an enzyme reflects?
    1. Cellular environment
    2. Concentration of substrate normally encountered in vivo
    3. Reaction chemistry
    4. None of the above
20. What is the Km of substrate whose concentration in cell is high?
    1. Low
    2. High
    3. Depends on reaction
    4. Depends on parameters

Read more in [Quizzes](http://purpleslinky.com/category/trivia/quizzes/)

[« Disney Dames: Which One are You?](http://purpleslinky.com/trivia/quizzes/disney-dames-which-one-are-you/)

[Quiz on Helmenthiasis Chemotherapy »](http://purpleslinky.com/trivia/quizzes/quiz-on-helmenthiasis-chemotherapy/)

ANSWERS:

1. 3
2. 3
3. 1
4. 3
5. 3
6. 1
7. 2
8. 1
9. 4
10. 3
11. 4
12. 1
13. 1
14. 2
15. 3
16. 3
17. 1
18. 2
19. 1, 2,3
20. 2

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Published by [ur guide](http://www.triond.com/users/ur+guide) in [Quizzes](http://purpleslinky.com/category/trivia/quizzes/)  
November 27, 2008

A new and better quiz on enzymes.

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A good score is around 80% in this quiz.

1. What is the order of reaction, if rate of reaction depends upon the concentration?

1. zero order
2. no order
3. more than zero order
4. first order

2. What does “k” denotes in the equation:

V= k[S]

And what is the unit of “k”?

1. Equilibrium constant and its unit is second2
2. Equilibrium constant and its unit is second-1
3. First order and its unit is second2
4. First order and its unit is second-1

3. What is the order of reaction, if it depends on two different molecules?

1. zero order
2. first order
3. second order
4. third order

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[« Is Earth Smart?](http://purpleslinky.com/trivia/quizzes/is-earth-smart/)

[Gk Quiz »](http://purpleslinky.com/trivia/quizzes/gk-quiz/)

4. What is the order of reaction, if it depends on two molecules of same substrate?

1. zero order
2. first order
3. second order
4. third order

5. What is the value of rate constant in second order reaction?

1. Ms
2. M
3. s
4. M-1 s-1

6. What is the [relationship](http://purpleslinky.com/trivia/quizzes/quiz-on-enzymes-four/) between constant and activation energy?

1. no relationship
2. directly proportional
3. inversely proportional
4. can’t be determined

7. How enzymes decrease activation energy?

1. restricting the movement of substrate
2. reducing entropy
3. making weak interactions
4. all of the above

8. What is binding energy, DGB?

1. Energy derived from enzyme-substrate interaction
2. Energy derived from enzyme-product interaction
3. Energy derived from product-substrate interaction
4. None of the above

9. Which energy is mainly responsible for lowering activation energy?

1. activation energy
2. transition energy
3. binding energy
4. none of the above

10. In which state weak interactions are optimized?

1. initial state
2. final state
3. intermediate state
4. transition state

11. In which state, substrate specifically to the enzyme active site?

1. initial state
2. final state
3. intermediate state
4. transition state

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12. Which scientist proposed lock and key model in 1894?

1. Daniel Koshland
2. Emil Fischer
3. Einstein
4. Michael

13. Who were the first scientists to propose that enzyme must me complementary to the reaction transition state?

1. Michael Polanyi and Haldane
2. Linus Pauling and Haldane
3. Haldane
4. Emil Fischer

14. Who elaborated the work of Michael Polanyi and Haldane in year1946?

1. Daniel Koshland
2. Emil Fischer
3. Linus Pauling
4. Einstein

15. What are the physical and thermodynamic factors which are responsible for lowering of activation energy?

1. reduction in entropy
2. increase in entropy
3. lowers binding energy
4. none of these

16. What is induced fit?

1. when enzyme change shape due to absence of substrate
2. when enzyme do not change shape due to absence of substrate
3. when enzyme change shape due to presence of substrate
4. when enzyme do not change shape due to presence of substrate

17. Who postulated induced fit in year1958?

1. Daniel Koshland
2. Emil Fischer
3. Linus Pauling
4. Einstein

18. Name specific catalytic group which contribute to catalysis?

1. alumina
2. glucose
3. acetic acid
4. none of the above

19. Specific acid-base catalysis uses which ions?

1. H+
2. OH-
3. Both a and b
4. None of the above

20. What is general acid-base catalysis?

1. use proton donors
2. use of proton acceptors
3. no use [water](http://purpleslinky.com/trivia/quizzes/quiz-on-enzymes-four/) as proton acceptor or donor
4. all of the above

ANSWERS:

1. 1
2. 2
3. 3
4. 3
5. 4
6. 2
7. 4
8. 1
9. 3
10. 4
11. 4
12. 2
13. 1
14. 3
15. 1
16. 3
17. 1
18. 4
19. 3
20. 4

|  |
| --- |
| [**Quiz Instructions**](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) in a pop-up window. |

http://www.bio.cmu.edu/courses/03231/MCQF04/nr/1pixel.gif

|  |  |
| --- | --- |
| **Quiz on Lecture 15: Enzyme Kinetics - Steady State** | |
| 1. Which of the statements regarding enzymes is false? | |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | Enzymes are proteins that function as catalysts. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | Enzymes are specific. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | Enzymes provide activation energy for reactions. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | Enzyme activity can be regulated. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | Enzymes may be used many times for a specific reaction. |
| 2. The relationship between an enzyme and a reactant molecule can best be described as: | |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | a temporary association. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | an association stabilized by a covalent bond. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | one in which the enzyme is changed permanently. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | a permanent mutual alteration of structure. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | noncomplementary binding. |
| 3. When [S] = KM, the velocity of an enzyme catalyzed reaction is about: | |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | 0.1\*Vmax. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | 0.2\*Vmax. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | 0.3\*Vmax. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | 0.5\*Vmax. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | 0.9\*Vmax. |
| 4. The active site of an enzyme | |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | remains rigid and does not change shape. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | is found at the center of globular enzymes. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | is complementary to the rest of the molecule. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | contains amino acids without sidechains. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | None of the above choices are correct. |
| 5. The active site of an enzyme differs from an antibody-antigen binding site in that the enzyme active site | |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | contains modified amino acids. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | catalyzes a chemical reaction. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | is complementary to a specific ligand. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | contains amino acids without sidechains. |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/rbd.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQLec14.htm) | None of the above are correct. |
|  | |
| [http://www.bio.cmu.edu/courses/03231/MCQF04/nr/cfc0.gif](http://www.bio.cmu.edu/courses/03231/MCQF04/MCQTEMP.htm)  http://www.bio.cmu.edu/courses/03231/MCQF04/nr/10.gifhttp://www.bio.cmu.edu/courses/03231/MCQF04/nr/10.gifhttp://www.bio.cmu.edu/courses/03231/MCQF04/nr/10.gifhttp://www.bio.cmu.edu/courses/03231/MCQF04/nr/10.gif  **Lecture 14 (Handout): Substrate Saturation Kinetics**  This hand-out is available as a [PDF](http://www.bio.cmu.edu/courses/03231/LecF04/Lec14/MMCurves.pdf) image.  **Examples of Data Analysis**  Experimental data, *e.g.* from initial velocity measurements (vo) can be analyzed to yield values for KM and Vmax in several ways. Computer programs are available that provide automated and objective estimates of these parameters. The processed data are then published in one or more of the following graphical formats:   1. **Saturation Curve**: vo *vs.* [A]. http://www.bio.cmu.edu/courses/03231/LecF04/Lec14/SubSatn.gif The above "data", with small errors added, were generated on the [Enzyme Kinetics Calculations](http://www.bio.cmu.edu/courses/03231/DryLab/MM/MMCalc.htm) page. The line corresponds to the calculated vo values for Vmax = 0.5 mM/min and KM = 2 mM. 2. **Double Reciprocal Plot**: 1/vo *vs.* 1/[A]. http://www.bio.cmu.edu/courses/03231/LecF04/Lec14/DblRecip.gif The data from the Substrate Saturation Curve are plotted in a "Lineweaver-Burke Plot". The Y-intercept of this plot is 1/Vmax. The X-intercept is -1/KM. The slope is KM/Vmax. Thus, KM = slope/intercept. 3. **Eadie-Hofstee Plot**: vo *vs.* vo/[A]. http://www.bio.cmu.edu/courses/03231/LecF04/Lec14/Eadie.gif The data from the Substrate Saturation Curve are plotted in an "Eadie-Hofstee Plot". The form of this graph should remind you of the Scatchard Plot for ligand binding. It is shown here only for that comparison. No details on the equation used for this plot or the meaning of the slope and intercepts are provided here. (See Problem Set #6.)   To obtain an accurate value of KM and Vmax from these data, it is necessary to obtain velocity measurements at high and low substrate concentrations, as shown in the above figures.  Given the total enzyme concentration, it is possible to determine k2 from Vmax. Since KM is a function of three rate constants it is not possible to obtain values of the substrate on (k1) and off (k-1) rates from steady state analysis.  [[http://www.bio.cmu.edu/courses/03231/LecF04/BCITop2.gif](http://www.bio.cmu.edu/courses/03231/LecF04/Lec14/MMCurves.html#top)](http://www.bio.cmu.edu/courses/03231/LecF04/Lec14/MMCurves.html#top)  [[http://www.bio.cmu.edu/courses/03231/LecF04/bluBack.gif](http://www.bio.cmu.edu/courses/03231/LecF04/Lec14/lec14.html#MMplots)Back to **Lecture 14**](http://www.bio.cmu.edu/courses/03231/LecF04/Lec14/lec14.html#MMplots) notes.  [[smBack](http://www.bio.cmu.edu/courses/03231/biochemF04.htm)Return to **Home Page**](http://www.bio.cmu.edu/courses/03231/biochemF04.htm).  Diabetes is the common term for several metabolic disorders in which the body no longer produces insulin or uses the insulin it produces ineffectively.  It is a common condition and is characterised by abnormally high blood sugar levels.  Diabetes is known as *"diabetes mellitus"* - where diabetes comes from the Greek word for siphon, which describes the excessive thirst and urination of this condition, and mellitus is the Latin word for honey, because diabetic urine is filled with sugar and is sweet.  **Diabetes essentially changes the way your body uses food**  The key to the problem is insulin - as insulin's role in the body is to help glucose get into the body cells where it is used to make energy.  Diabetes is characterized by a partial or complete lack of insulin production by the body. The most common forms of diabetes are type 1 diabetes and type 2 diabetes. In both types of diabetes, people have little or no ability to move sugar out of the blood stream and into the cells, where it is used as the body's primary fuel.  **Symptoms and complications**  Symptoms of diabetes include:   * Frequent urination * Extreme thirst and/or hunger * Weight loss * Fatigue * Numbness * Sores that are slow to heal, and * Increased infections   Learning how to best manage your diabetes is key to your treatment.  Poor control of diabetes can lead to an increased risk of:   * Heart disease * High blood pressure * Stroke * Kidney and bladder failure * Gum disease * Blindness * Foot and leg infections   **How common is diabetes?**  According to the World Health Organization, over 175 million people throughout the world have diabetes.  Of these, 90% have type 2 diabetes, and 10% have type 1 diabetes.  **There are three main types of diabetes:**  **Type 1 diabetes**  Type 1 diabetes is a lifelong condition that is treated with injections of insulin. Injections must be given each day and some people require multiple injections a day to help maintain blood glucose control.  Type 1 diabetes develops when an "autoimmune reaction" destroys beta cells in the pancreas. Autoimmune reaction means that the body creates antibodies against its own cells. As a result, the pancreas stops producing insulin or cannot produce enough insulin on its own. Treatment involves daily insulin injections, in conjunction with healthy eating and regular exercise.  Symptoms of type 1 diabetes are usually:   * Extreme thirst * Frequent urination * Sugar in the urine * An acetone-like smell around the body * Fatigue, weakness, drowsiness * Excessive weight loss over a short period of time, for no apparent reason   Although the cause of diabetes is unknown, there are certain risk factors that can increase the risk of developing type 1 diabetes.  Risk factors for type 1 diabetes include:   * Ethnic background or race (more common in people of Caucasian descent) * Having a parent with type 1 diabetes   Type 1 diabetes most often affects people under 20 years of age. It was previously called juvenile-onset diabetes or Insulin-Dependent Diabetes Mellitus (IDDM).  **Type 2 diabetes**  Type 2 diabetes is a term for several disorders with different causes and degrees of severity. It is the most common type of diabetes.  Often, people with type 2 diabetes can still make their own insulin in the pancreas, but the insulin that is produced is not used as effectively by the body.  Many people manage type 2 diabetes simply by following a healthy diet and regular exercise. In overweight individuals, type 2 diabetes often improves as a result of weight loss, a healthy diet and exercise.  With the progression of the disease, some people may have to take oral medication(s) or insulin injections.  Type 2 diabetes is much more common than type 1 diabetes.  Although the cause of type 2 diabetes is unknown, there are some risk factors that can predispose some people to this condition.  Risk factors of type 2 diabetes include:   * Age (being over 45 years old) * Being overweight or obese * Having a family history of diabetes * Ethnic background or race (Native/Indigenous, African, Hispanic or Asian descent) * Having given birth to a large baby (over 4 kg or 9 lbs) * Impaired glucose intolerance   The symptoms of type 2 diabetes are the same as type 1 diabetes. Some people may also experience slow healing cuts and bruises, recurring gum or bladder infections, or tingling in their hands or feet.  Other terms previously used for type 2 diabetes are adult-onset diabetes and Non-Insulin Dependent Diabetes Mellitus (NIDDM).  **Gestational diabetes**  Gestational diabetes is another common type of diabetes. It is a temporary condition that occurs during pregnancy.  Extra demands on the pancreas cause some women to develop diabetes during pregnancy. Often, it goes away after delivery. But, later in life, diabetes may return.  Gestational diabetes affects 2% to 4% of all pregnancies, with an increased risk of developing diabetes for both the mother and the child.  The risk of type 2 diabetes returning is greater if the mother has given birth to a baby that weighed over 4 kg (9 lbs) at birth.  Treatment will involve following a healthy diet, physical activity, and in some cases, insulin therapy. | |

**Types of Diabetes and Prevention Measures**

by Athira



A girl with diabetes taking insulin injection

My last article on [tips to prevent diabetes](http://healthruns.com/prevent-diabetes/) was a small but concise list of steps you could take to prevent diabetes, But readers began asking questions related to diabetes so much that I decided to write a full article on types of diabetes and more preventive measures. If you are a diabetic patient, do not skip the introduction part as it explains in detail what causes diabetes which you need to know. Diabetes is the most common non-communicable disease. Everyone knows that diabetes is related with insulin and pancreas. So it won’t be appropriate to tell about diabetes without mentioning about pancreas and insulin. So let’s see from where insulin is secreted.

**Pancreas and Islets Of Langerhans**

Pancreas is the endocrine organ in our body. This organ consists of one million microscopic clusters of cell which is called the *islets of Langerhans* weighing not more than one to one and half grams. These cells are of four types. They are ***Beta*** *(68%),* ***alpha*** *(20%),* ***delta*** *(10%) and* ***PP cells*** *or pancreatic polypeptide cells (2%). The percentage composition is representing the islets cell clusters of an adult.*

The most important trigger for releasing insulin is nothing but blood glucose level.

Diabetes mellitus is characterized by “a defective or deficient insulin secretary response”. It results in impaired carbohydrate /glucose synthesis. Thus ultimately results in hyperglycemia. Diabetes mellitus do consists of many disorders having hyperglycemia as the most common feature.

In short Diabetes mellitus is a chronic disorder of the nutrient metabolism. Thus it is characterized by a deranged carbohydrate, protein and fat metabolism.

**Types of diabetes**

There are three types.

1. With just 5% of incidence Maturity Onset Diabetes of the Young [MODY] is due to the genetic defects of beta cells.

2. Type 1 diabetes: it was once called juvenile onset diabetes. Type one is otherwise called as IDDM, insulin dependent diabetes mellitus. It has 10%of incidence.

3. Type 2 diabetes: this one was once called as the non insulin dependent diabetes mellitus. Type 2 is otherwise called as NIDDM, non insulin dependent diabetes mellitus. Life style is the major culprit here.

**Diagnosis for diabetic patients**

If you experience excessive thirst, hunger, maturation tendencies, sudden loss of weight, fatigue, bad breath after consulting a dentist and when he couldn’t see any issues with your teeth; then please do check your blood glucose level and urinary glucose level. Even if you don’t feel any of these symptoms after the age of 25 it’s recommended to go for a routine health check which includes fasting blood sugar, random blood sugar, HbA1C, haemoglobin level, triglycerides, cholesterol, LDL, VLDL, HDL…

A non diabetic person’s *fasting blood sugar* should be less than **120**. *Random blood sugar* level should be less than **180**.

Apart from the blood glucose level there is yet another tool to diagnose diabetes. It’s a test which The American Diabetic Association recommends. HbA1C should be tested every half yearly. The [HbA1C] *glycosylated haemoglobin* which is a better indicator of diabetes than blood glucose level should be ***less than 7***. The glycosylated haemoglobin is that part of the hemoglobin on RBC that is attached to the glucose. The level is an indicator of average blood glucose level of last six months. If you are detected diabetic then please don’t delay to get yourself a self check meter, by which you can monitor your blood glucose and get the information to take timely action if you have low blood sugar or high blood sugar.

**Do You Think You Are Not In The Risk Group?**

It’s said that if one of your parents is diabetic you have 50%chance of developing diabetes. If both your parents are diabetic there is 75%chance of getting diabetes.

Diabetes is emerging as a common chronic disease even in developing countries like India. By 2025 one out of four diabetic patients in world will be an Indian. It’s a fact that this high ratio is due to Indians having a genetic predisposition to diabetes.

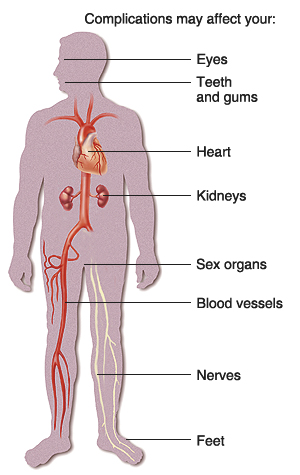
Which ever be the nation as far as obesity rate increases, side by side diabetes rate also increases.

Diabetes is highly related to stressful life style. So ***in short each and every individual in today’s world can’t escape from the clutches of diabetes*** so easily unless you follow a healthy life style. This can be clearly demonstrated by the fact that rural areas diabetes is 4 fold less than in urban areas. So modernization is a reason for diabetes.

But how is urbanization which means better living gives you this chronic disease?

For better time management in the busy world people have made kitchen to be just for cooking frozen food stuffs. People are depending on fast food, French fries, potato chips….Not having a check on dietary needs consuming excess quantities. By Using lots of sugar, sweetened colas, using lots of fat in diet for improving tastes, not doing exercises, not having food in time, not having good sleep, stressed up for making more and more money people are welcoming this chronic disease to their lives.

Smoking has got a direct link with diabetes. I won’t tell you to try hard to reduce smoking. But I can [tell you to quit smoking](http://healthruns.com/how-to-stop-smoking-for-good/).



Complications caused by diabetes

**Why diabetes is deadly?**

* Study conducted by Duke University, North Carolina says that diabetic patients have seen losing their antioxidants stored in the body. But this complication can be overcome by including [fruits in your diet](http://healthruns.com/25-grape-fruit-diet-tips/) and/or taking supplements.
* It increases the risk of cardiovascular diseases.
* It lowers your resistance levels.
* You experience more fatigue than people of your age.
* Wounds take longer to heal.

**Complications of diabetes**

To admit the truth I have considered diabetes like any other non communicable disease until my father was tested positive for diabetes 5 years back. I admit it’s a chronic disease. But from then you start putting a check on what you consume. You start workout. A diabetic patient is unable either to produce sufficient insulin or use insulin effectively.  After years of being diabetic patient without taking care of the disease condition, one will be risking his own heart, blood vessels, nerves [diabetic neuropathy], kidneys [diabetic nephrology], and eyes [diabetic retinopathy] they will feel numbness in the hands and feet or tingling in the extremities. When diabetes starts affecting your kidneys albumin will be excreted in the urine. It will be detected if its quantity is above 300mg/liter of urine. Studies show that more than 70%of diabetes’s patients have nerve damage or some form of neuropathy. Of these symptoms some are considered positive as this makes a person to seek medical advice. They are tingling sensation, numbness, and pain. The symptoms in the second category, negative list are for example loss of sensation/ feeling … is often neglected.

**Managing Diabetes**

Diabetes is a chronic disease. It leads to many complications. But it’s manageable too. Diabetes will lead to failure of other organs. ***It’s not a deadly disease if taken care*. *You can lead a normal life.***

***Eat small meals at regular intervals*.** Don’t over burden with heavy food. All you got to do is supply body with nutrients regularly. Eat every 2 hours.

***Try not to include any***

***1***. ***Sweets***

***2. Sweetened colas***

***3. Fried items***

***4. Red meat***

***5. Chips*** in your diet.

If you don’t lie to have special diabetes recipes including ragi, then don’t. Instead have ***moderate quantities*** of a variety of foods. Keep in mind that like obese patients you too have to stick on to ***regular meal time***. Diabetes patients according to the Columbia University do show a deficiency to magnesium levels. Other than people with kidney complaints can go for magnesium supplements. ‘Bilberry’, a herb that prevents diabetic retinopathy has also found helpful in preventing abnormal blood clotting. ***Gymnema silvestre***, an Indian herb helps to control blood sugar. It may help to reduce the need for insulin. Lots of research has been carried out in these areas.

***EXERCISE REGULARLY.***

*D*o [exercise daily](http://healthruns.com/3-exercise-swiss-ball/) by which you are not keeping diabetes under control, but also it’s a shield against cardiovascular diseases. It’s said that those burning 13000 to 15000 kilojoules a week through exercise are having least chances to prevent type 2 diabetes. Control your body weight. Proper taking care of diabetes and blood pressure will keep your kidneys healthy.

You can monitor your own glucose level. ***Self monitoring of blood glucose*** will warn you and you can be your own health adviser.

***[Foot Care](http://healthruns.com/12-foot-care-tips/) is of great significance***. Always wear slippers/shoes. Using a mirror closely inspect even the bottom of feet for any wounds, blisters, redness. If it’s a new shoe, for the initial day do wear that for not more than an hour. Do a pedicure every week. You can very much moisturize, but it would be better to leave the in between skin of your toes.