## Enzyme Kinetics



# Question - Answers & Quick Notes

**Useful for CSIR NET and other Life Science Examinations** 



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Q1. In a biochemical reaction catalyzed by enzyme following the Michaelis-Menten equation, what will be the substrate concentration, when the observed velocity of reaction is 90% of the maximum velocity?

(1) 18 Km (2) 9 Km

(3) 5 Km (4) 11 Km



Given, 
$$[s] = \frac{8}{5}$$
  
 $V_{\text{max}} = 100$   $V = 90\%$  of  $V = 90\%$  of  $V = 90\%$  Max.  
 $V_0 = \frac{V_{\text{max}} \cdot [s]}{V_{\text{max}} \cdot [s]}$   
 $V_0 = \frac{100 \cdot [s]}{V_0 \cdot [s]}$ 

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Q1. In a biochemical reaction catalyzed by enzyme following the Michaelis-Menten equation, what will be the substrate concentration, when the observed velocity of reaction is 90% of the maximum velocity?

(1) 18 Km

(2) 9 Km **\** 

(3) 5 Km

I Km



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Q2. For an enzyme catalyzed reactions exhibiting Michaelis Menten equation what would be increase in substrate concentration to increase the rate of reaction from 10% of  $V_{\text{max}}$  to 90% of  $V_{\text{max}}$  -

- (1) 81 fold (2) 8 fold
- (3) 4 fold (4) 2 fold



Given,
$$V_{1} = 10\% \text{ of } V_{\text{max}}$$

$$[S_{1}] = \chi$$

$$V_{0} = \frac{V_{\text{max}} \cdot [S]}{K_{\text{m}} + [S]}$$

$$10 = \frac{100 \cdot [S]}{K_{\text{m}} + [S]}$$

$$10 K_{\text{m}} + 10[S] = 100[S]$$

$$[S] = \frac{90}{30} K_{\text{m}}$$

$$S_{0} \cdot (\text{in this case.} \dots \text{we can say})$$

$$[S_{2}] = \frac{1}{3} K_{\text{m}}$$

$$S_{1} \cdot (\text{can we in fold})$$

$$[S_{2}] = \frac{3 K_{\text{m}}}{3 K_{\text{m}}} = 9 \times 9$$

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Vmax = 100 (assume)

 $V_0 = \frac{V_{\text{max}} \cdot [S]}{K_{\text{m}} + [S]}$ 

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- (1) 81 fold (2) 8 fold
- (3) 4 fold (4) 2 fold



Q3. The V<sub>max</sub> and K<sub>m</sub> from a Lineweaver-Burk plot of an enzyme reaction where  $\frac{1}{v} = 40 \mu \text{M}^{-1}$  min at  $\frac{1}{[s]} = 0$  and  $\frac{1}{[s]}$ 

= -1.5 × 10<sup>2</sup> mM<sup>-1</sup> at 
$$\frac{1}{v}$$
 = 0 are

(1) 
$$0.025 \,\mu\text{M min}^{-1}$$
 and  $0.67 \times 10^{-2}\text{mM}$   
(2)  $0.025 \,\mu\text{M}^{-1}$  min and  $0.67 \times 10^{-2}$ 

(3) 
$$0.025 \,\mu\text{M min}^{-1}$$
 and  $1.5 \times 10^2 \,\text{mM}^{-1}$ 

(4) 
$$0.038 \,\mu\text{M min}^{-1}$$
 and  $0.67 \times 10^{-2}\text{mM}$ 



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#### **Enzyme Kinetics**

The Michaelis-Menten equation

**Lineweaver - Burk Plot** 



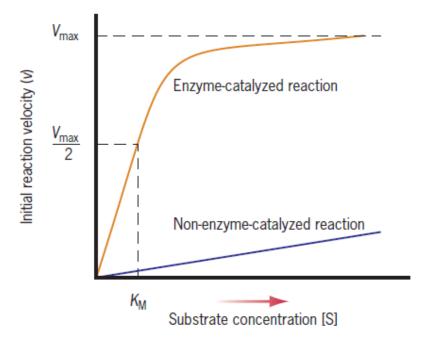


FIGURE 3.17 The relationship between the rate (velocity) of an enzyme-catalyzed reaction and the substrate concentration. Since each enzyme molecule is only able to catalyze a certain number of reactions in a given amount of time, the velocity of the reaction (typically expressed as moles of product formed per second) approaches a maximal rate as the substrate concentration increases. The substrate concentration at which the reaction is at half-maximal velocity  $(V_{max}/2)$  is called the Michaelis constant, or  $K_{\rm M}$ .



$$V = V_{\text{max}} \frac{[S]}{[S] + K_{\text{M}}}$$

According to the equation, when the substrate concentration [S] is set at a value equivalent to  $K_{\rm M}$ , then the velocity of the reaction (V) becomes equal to  $V_{\rm max}/2$ , or one-half the maximal velocity. Thus,  $K_{\rm M} = [S]$ , when  $V = V_{\rm max}/2$ .



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To generate a hyperbolic curve such as that in Figure 3.17 and make an accurate determination of the values for  $V_{\rm max}$  and  $K_{\rm M}$ , a considerable number of points must be plotted. An easier and more accurate description is gained by plotting the reciprocals of the velocity and substrate concentration against one another, as formulated by Hans Lineweaver and Dean Burk. When this is done, the hyperbola becomes a straight line (FIGURE 3.18) whose x intercept is equal to  $-1/K_{\rm M}$ , y intercept is equal to  $1/V_{\rm max}$ , and slope is equal to  $K_{\rm M}/V_{\rm max}$ . The values of  $K_{\rm M}$  and  $V_{\rm max}$  are, therefore, readily determined by extrapolation of the line drawn from a relatively few points.

In most cases, the value for  $K_{\rm M}$  provides a measure of the affinity of the enzyme for the substrate. The higher the  $K_{\rm M}$ , the greater the substrate concentration that is required to reach one-half  $V_{\rm max}$  and, thus, the lower the affinity of the enzyme for that substrate.

$$\frac{1}{V_0} = \frac{K_m}{V_{\text{max}}} \cdot \frac{1}{[S]} + \frac{1}{V_{\text{max}}}$$



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$$\frac{1}{V_0} = \frac{K_m}{V_{\text{max}}} \cdot \frac{1}{[S]} + \frac{1}{V_{\text{max}}}$$

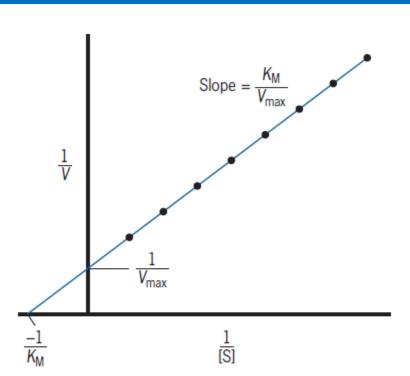


This is of the form y = mx + c, which is the equation of a **straight line graph**; a plot of y against x has a slope m and intercept c on the y-axis.

$$\frac{1}{V_0} = \frac{K_m}{V_{\text{max}}} \cdot \frac{1}{[S]} + \frac{1}{V_{\text{max}}}$$



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$$\frac{1}{V_0} = \frac{K_m}{V_{\text{max}}} \cdot \frac{1}{[S]} + \frac{1}{V_{\text{max}}}$$

**FIGURE 3.18 A Lineweaver-Burk plot** of the reciprocals of velocity and substrate concentration from which the values for the  $V_{\text{max}}$  and  $K_{\text{M}}$  are readily calculated.



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$$\frac{1}{V_0} = \frac{K_m}{V_{\text{max}}} \cdot \frac{1}{[S]} + \frac{1}{V_{\text{max}}}$$

It is clear that,

when 
$$\frac{1}{[S]} = 0$$

then  $\frac{1}{V} = \frac{1}{V_{max}}$ 

(by putting 
$$\frac{1}{[S]}$$
 = 0 in the equation)

when 
$$\frac{1}{[V]} = 0$$

$$0=rac{K_m}{V_{max}}rac{1}{[S]}+rac{1}{V_{max}}$$

Set  $\frac{1}{V} = 0$  and solve for  $\frac{1}{|S|}$ :

$$-\frac{1}{V_{max}} = \frac{K_m}{V_{max}} \frac{1}{[S]}$$

$$-1 = K_m \frac{1}{[S]}$$

$$-\frac{1}{K_m} = \frac{1}{[S]} = \text{x-intercept}$$



Given is, 
$$\frac{1}{V} = 40 \mu M^{-1} min$$
.

$$at \frac{1}{(S)} = 0$$

$$also Given \frac{1}{(S)} = -1.5 \times 10^{2} m M^{-1}$$

$$at \frac{1}{V} = 0$$

$$when, \frac{1}{V} = 0 \quad then.$$

$$\frac{1}{(S)} = -\frac{1}{Km} \quad So$$

$$-1.5 \times 10^{2} m M^{-1} = -\frac{1}{Km}$$

 $V_{\text{max}} = 3$ 

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$$= \frac{1}{1.5} \times 10^{2} \text{ mM}$$

$$= 0.67 \times 10^{2} \text{ mM}$$

$$= 0.66 \times 10^{2} \times 10^{2} \text{ mM}$$

$$= 0.66 \times 10^{2} \times 10^{2$$

Vmax = 1 = 0.025 4 Mmin

1.5 × 102 mM-1

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Q3. The V<sub>max</sub> and K<sub>m</sub> from a Lineweaver-Burk plot of an enzyme reaction where  $\frac{1}{v} = 40 \mu \text{M}^{-1}$  min at  $\frac{1}{[s]} = 0$  and  $\frac{1}{[s]} = 0$ 

= -1.5 × 10<sup>2</sup> mM<sup>-1</sup> at 
$$\frac{1}{v}$$
 = 0 are  
(1) 0.025  $\mu$ M min<sup>-1</sup> and 0.67 × 10<sup>-2</sup>mM  $\checkmark$ 

(2) 
$$0.025 \,\mu\text{M}^{-1}$$
 min and  $0.67 \times 10^{-2}$ 

(3) 
$$0.025 \,\mu\text{M min}^{-1} \text{ and } 1.5 \times 10^2 \,\text{mM}^{-1}$$

(4) 
$$0.038 \,\mu\text{M min}^{-1}$$
 and  $0.67 \times 10^{-2}\text{mM}$ 



### Thank you!



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