Bioreactors
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Lecture – 08
Solution to PP 2.2

Welcome to lecture number 8, the NPTEL online certification course on Bioreactors. When we finished up the last lecture, we had looked at a problem on in enzyme inhibition. Before that in the lecture itself we had looked at three types of inhibitions, the competitive inhibition, the non-competitive inhibition and the uncompetitive inhibition which were different in the way the inhibitor effects the reaction. In the first case, the inhibitor bound to the enzyme, in the second case the inhibitor bound to the enzyme substrate complex and in the third case it bound to both, I think I mixed up the last two but you get the idea, where the inhibitor binds made the difference. Then we looked at a problem which could help us understand this a little better. Let us work out that problem in this lecture.

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Practice problem 2.2

An enzyme normally displays Michaelis-Menten (M-M) behaviour in converting a substrate, S, to a product, P. It gave the following data when its M-M parameters for the above conversion were estimated in the presence of another substance I at different concentrations. Determine the type of inhibition, and the inhibition constant.

M-M parameters/I, mM→	0	0.5	1	5	10
v_{max}, mM min^{-1}	0.33	0.33	0.33	0.33	0.33
K_m, mM	0.120	0.126	0.132	0.180	0.240

Practice problem 2.2, An enzyme normally displays Michaelis-Menten behavior in converting a substrate, S, to a product, P. It gave the following data when its Michaelis-

Menten parameters for the above conversion were estimated in the presence of another substance I at different concentrations. Determine the type of inhibition and the inhibition constant, the Michaelis-Menten parameters V m and K m at different inhibitor concentrations were given in this table. Now, how do we go about it? Let us ask our regular questions.

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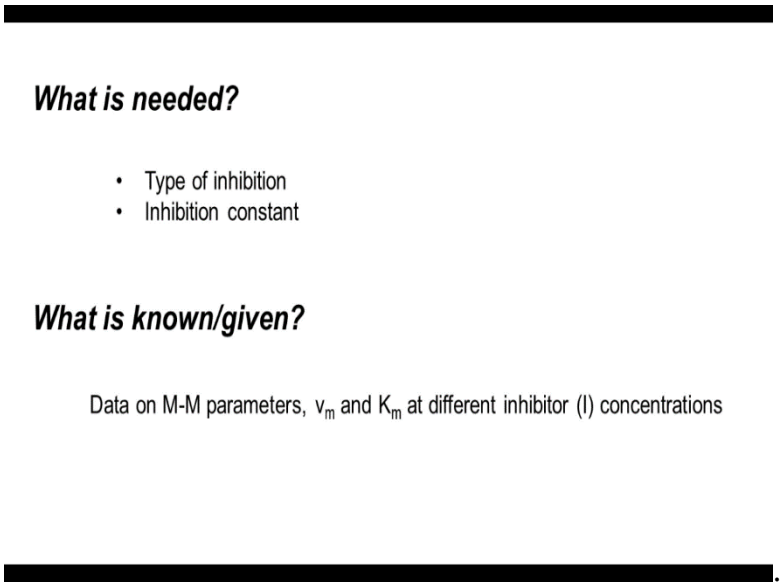

What is needed? The type of inhibition and the inhibition constant; k I. What is known or given, the data on the Michaelis-Menten parameters V m and K m at different inhibitor concentrations are given and How to connect what is needed to what is given?

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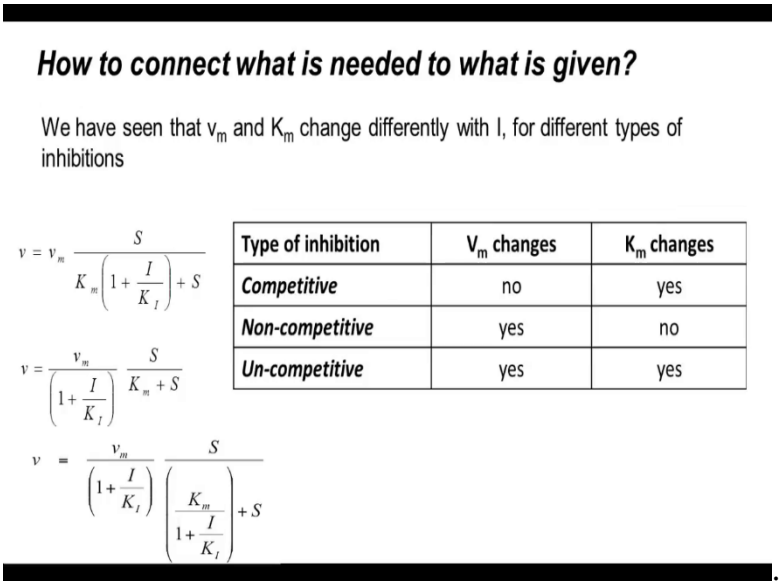

We have seen that V m and K m change differently with I for different types of inhibitions. For the competitive inhibition, the K m gets modified.

$$v = \frac{dP}{dt} = v_m \frac{S}{K_m\left(1+\frac{I}{K_I}\right)+S}$$

For the noncompetitive inhibition, the v m gets modified and for the uncompetitive inhibition both V m and K m got modified in this fashion, this is the summary. The type of inhibition is competitive, noncompetitive, uncompetitive. In the case of competitive inhibition, V m remains the same there is no change whereas K m changes. In the case of noncompetitive v m changes, but K m does not change and in uncompetitive both V m and K m change. So, we need to find out what changes here.

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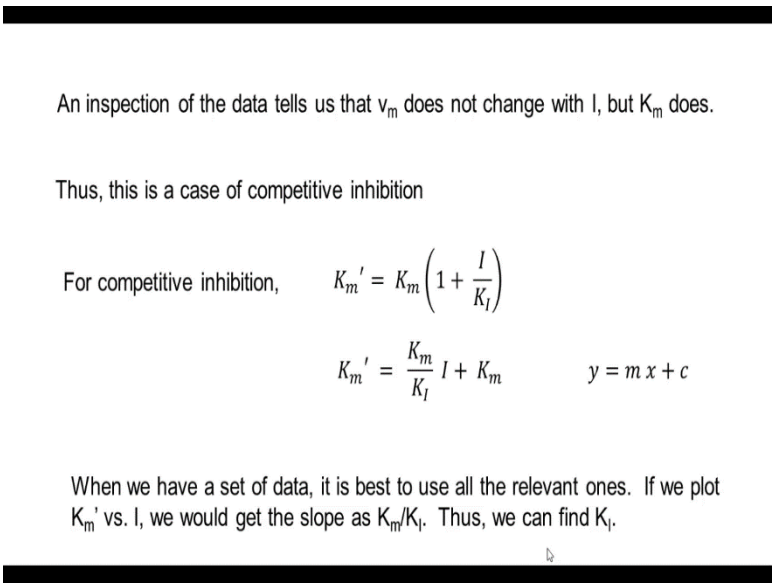


Now, if we look at the data, if we inspect the data, it tells us that V m does not change with I, but K m changes. Let me show this to you here. The data was this, as the inhibitor concentration changed, 0, 0.5, 1, 5 and 10, the V max remain the same at 0.33, whereas K m changed. This is what we saw, what we said earlier and this is what the data shows us.

This is clearly a case of competitive inhibition so that is straight forward. That is the first question, what kind of a competition it is? So, for a competitive inhibition, this Km', the new K m is

$$Km' = Km(1 + \frac{I}{K_I})$$

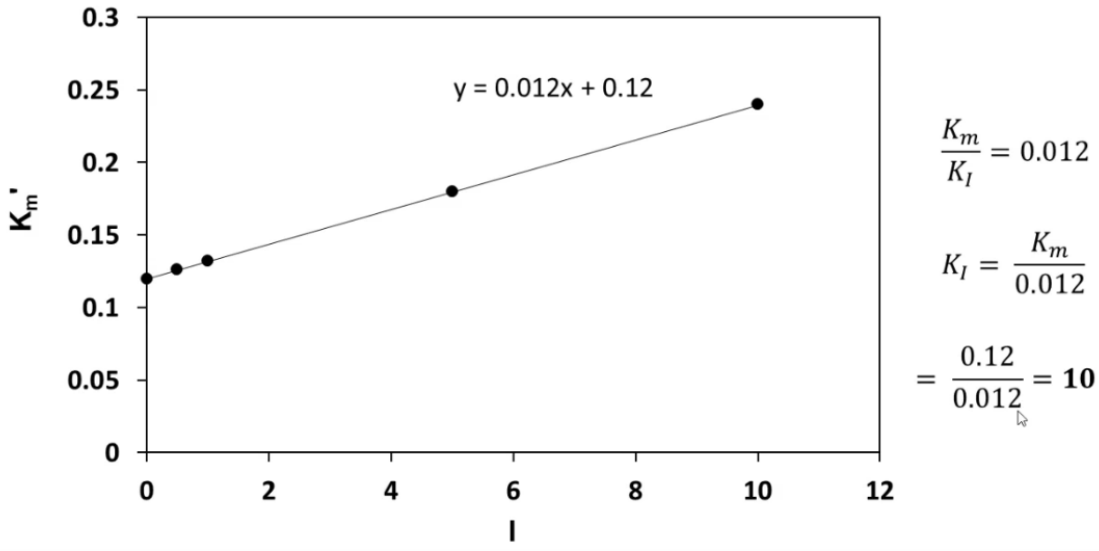
$$Km' = \frac{Km}{K_I} I + Km$$

I have written it of a form of y = mx + c. Remember we are dealing with a set of data here. Therefore, whenever we deal with data, we need to use the entire set of data. If we just pick points from there, the errors in the data collection would directly get transferred and probably get magnified. If you are using the data point, which happen to be incorrect

then the errors would be large in the estimates.

That is the reason why we need to use the entire set of data whenever we have data to work with and we are trying to use the entire set of data to find, whatever is necessary here which is the inhibition constant. To do that, I am looking at this in a y equals m x plus c form. Km' is y, I is x and the slope in that case is Km/K_I and the intercept is Km. We have data of Vm, Km with varying I's, so I is the independent x coordinate, Km'- the modified Km is the y coordinate. If we plot it that way, then we could get Km, K_I from here and K m from the intercept.

This is what we just mentioned, it is good to repeat this. When we have a set of data, it is best to use all the relevant ones. If we plot Km' versus I, you would get the slope as Km/K_I and thus we could find K_I.



$$\frac{K_m}{K_I} = 0.012$$

$$K_I = \frac{K_m}{0.012}$$

$$= \frac{0.12}{0.012} = \mathbf{10}$$

From a plot of Km' versus I as this, the equation was y = 0.012x + 0.12, y in this case is Km', x is I, so

Km' = 0.012 I + 0.12.

The intercept Km = 0.12 from the equation

$\therefore slope\ \frac{K_m}{K_I} = 0.012$

$\frac{K_m}{0.012} = K_I = \frac{0.12}{0.012} = 10mM$ = the inhibitor constant