Contd. The Topic: Enzyme kinetics.

According to molecular kinetics. The frequency with which enzyme and substrate molecules react together is controlled by two main factors, The energy barrier to reaction, and the entropy of activation, which is the entropy change when the activated complex is formed from the reactants. Both of these factors provide valuable information about the molecular nature of the processes occurring. For example, a large positive entropy of activation is good evidence that charges are being neutralized when the process occurs, the entropy increase is due to the release of water molecules that had been bound by the ions. This is an example of a mechanistic detail that cannot easily be obtained from structural studies, since activated complexes do not exist long enough to be detected by any but the most sophisticated high speed techniques.

First of all to carry out kinetic studies with a pure enzyme, trypsin, was the British physical chemist John Alfred Valentine Butler (V Butler, 1941). Many studies of this have been made the same type with a variety of enzymes, and the results have contributed greatly to the understanding of enzyme action (Laidler and Bunting, 1973). When an enzyme and substrate are brought together, the steady state is usually established within a few milliseconds. For convenience, most investigations of enzyme kinetics have been concerned with the steady state. When steady state is being established — during what is called the transient phase of the reaction requires special high speed techniques. To overcome two problems. Two problems have to be overcome. The first is to make measurements within short periods of time. The second is to bring the enzyme and substrate together rapidly (as otherwise the reaction may be over before they are properly mixed).

The second problem may sometimes be overcome by the use of flow methods in which solutions are forced together very rapidly. An important variant of this method was the stopped flow method and then relaxation method. Sometimes the individual steps are too fast for their rates to be measured by flow methods and then relaxation methods have to be used. In recent years much

further work has been done using high speed techniques, and many of the kinetic details of enzyme have been worked out.

Finally Enzyme kinetics may be defined as the study of the chemical reactions that are catalyzed by enzymes.

Kinetic ~~studies~~ studies on enzymes that only bind one substrate, such as triosephosphate isomerase, aim to measure the affinity with which the enzyme binds the substrate and the turnover rate.

Representation of Michaelis-Menten kinetics by Michaelis-Menten Kinetics.

a graph called a Lineweaver-Burk Plot

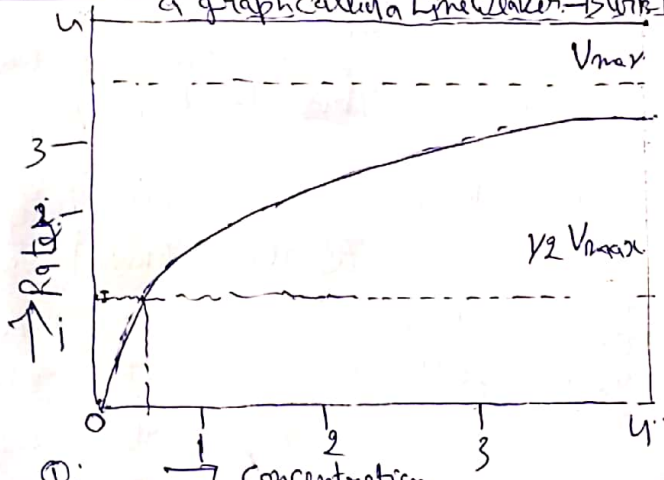

Image-1 an enzyme displaying Michaelis-Menten kinetics

This eqn. concerns the steady state of an enzymatic reaction with one substrate and is given by:

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

Lineweaver-Burk Plot: the eqn. This plot is given by:

[S] → substrate conch

Where Vmax - the maximum rate of reaction. When all enzyme active sites are saturated with substrate.

Km - The substrate concentration that gives half maximal velocity.

Km is a measure of the affinity an enzyme has for its substrate, as a lower Km means that less of the substrate is required to reach half of Vmax.

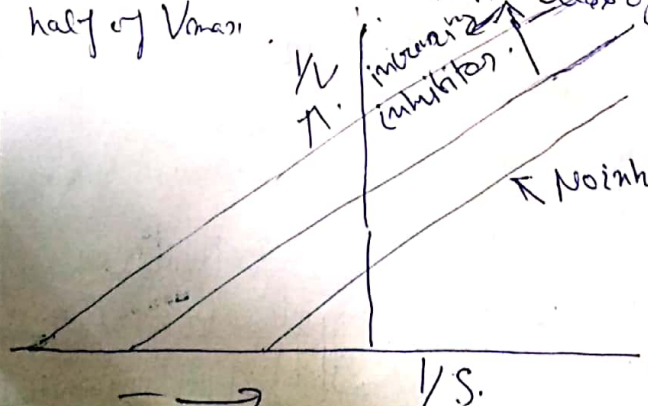

Image-2 Lineweaver-Burk Plot.

The eqn generated for the Plot is as:

$$\frac{1}{v} = \frac{K_m}{V_{max}} \frac{1}{S} + \frac{1}{V_{max}}$$

The Plot of rate against concentration (in image 1) has the shape of a rectangular hyperbola. However, a more useful representation of Michaelis-Menten kinetics is a graph called a Lineweaver-Burk Plot.