

The relative rates of reaction of various substrates can often be explained in terms of different affinities of the enzyme for the various substrate molecules.

K_M values can be determined from a Lineweaver-Burk plot that is a double reciprocal plot as follows

The reciprocal of the M-M equation is an equation of a straight line

A plot of $1/v$ vs. $1/[S]$ will yield a straight line with a slope of K_M/V_{max} .

Extrapolation of the straight line thru the axes yields a $1/[S]$ intercept of $-1/K_M$ and a $1/v$ intercept of $1/V_{max}$ as follows

So K_M and V_{max} can be determined from the intercepts.

Table 8-5 shows K_M values for various substrates of a number of enzymes.

If the K_M values reflect the relative affinities of an enzyme for the different substrates, differences in structures of the substrates may indicate the manner in which the enzyme binds substrate.

Consider an explanation for the differences in K_M values for synthetic substrates of chymotrypsin shown in Table 8-5.

The K_M values suggest that N-benzoyltyrosinamide binds to chymotrypsin more tightly than N-formyltyrosinamide.

Compare the structures of these substrates

Differences in the structures suggest that hydrophobic interactions involving the phenyl group may contribute to the binding.

The K_M value of glutamate is smaller than that for α -ketoglutarate suggesting that glutamate binds more tightly to glutamate dehydrogenase.

Compare the structures of glutamate and α -ketoglutarate

Differences in structures suggest that electrostatic interactions involving the $-\text{NH}_3^+$ group may contribute to the binding.

Additional information about the manner in which an enzyme binds a substrate molecule can be obtained from the inhibitory effect of small molecules other than the substrate on the reaction.

Malonate, for example, has been shown to be a strong inhibitor of succinate oxidation catalyzed by succinic dehydrogenase as below

Malonate decreases the rate of the succinate dehydrogenase reaction in the following way

Observe that V_{\max} can be approached at “high” substrate concentrations.