MICHAELIS MENTON RELATIONSHIP

In 1913, Michaelis and Menton formulated a relationship based on the assumption that an enzyme combines with a substrate to form an enzyme-substrate complex prior to decomposition of the substrate.

Assumptions:

1. Enzyme and substrate combine reversibly to form the enzyme-substrate complex (ES complex).
2. The ES complex decomposes to yield enzyme and products in the reaction.
3. The initial velocity of this reaction is proportional to the concentration of the ES complex.
4. The ES complex remains in a steady state for the period of the reaction, e.g. the rate of breakdown of the ES complex is equal to the rate of its formation.

The overall reaction can be described by:

\[ E + S \xrightarrow{k_1} ES \xrightarrow{k_2} E + P \] (1)

According to the law of mass action, the rate of formation of the ES complex is proportional to the concentration of free enzyme and the concentration of free substrate.

\[ \text{rate} = k_1[E]_f[S]_f \] (2)

Since initially the amount of substrate combined with enzyme is only a minute fraction of the total substrate present, we can let the concentration of free substrate be represented by the total concentration of substrate.

\[ [S]_f = [S]_t \] (3)

A considerable fraction of the total enzyme concentration is combined with substrate as ES complex so the concentration of free enzyme must be represented by the difference between the total enzyme concentration and the concentration of the ES complex.

\[ [E]_f = [E]_t - [ES] \] (4)

Substituting equations 3 and 4 in equation 2, the rate of formation of the ES complex is:

\[ \text{rate} = k_1([E]_t - [ES]) ([S]_t) \] (5)

The ES complex may break down in two ways. Thus the rate of breakdown of the ES complex can be expressed as the sum of these two reactions. From equation 1:

\[ \text{rate} = k_2[ES] + k_3[ES] \] (6)
In accordance with assumption \( k \), we can set equations 5 and 6 equal to each other

\[
k_1([E]_t - [ES]) ([S]_t) = k_2[ES] + k_3[ES]
\]  
(7)

and solving for the concentration of the ES complex we have

\[
[ES] = \frac{[E]_t [S]_t}{\frac{k_2 + k_3}{k_1} + [S]_t}
\]  
(8)

Having solved for the concentration of the ES complex, we can go to assumption 3 which states that:

\[
v = k_3[ES]
\]  
(9)

Substituting equation 8 in equation 9 we have

\[
v = \frac{k_3[E]_t[S]_t}{\frac{k_2 + k_3}{k_1} + [S]_t}
\]  
(10)

The term involving the 3 rate constants can be replaced by a new constant \((K_m)\) called the Michaelis constant.

\[
v = \frac{k_3[E]_t[S]_t}{K_m + [S]_t}
\]  
(11)

Assumption 3 tells us that the initial rate of the reaction will depend on the concentration of the ES complex. Once an equilibrium has been reached between E, S and ES complex, the maximum rate has been reached, the rate of decomposition of the ES complex \((k_3)\) becomes the rate limiting step for the entire reaction. At this point, all the active sites on the enzyme protein are combined with substrate, and the reaction rate cannot be increased unless additional enzyme is added. In other words, velocity is now dependent on the amount of enzyme present.

\[
V_{max} = k_3[E]_t
\]  
(12)

Substituting equation 12 into equation 11, we arrive at the final Michaelis-Menton equation.

\[
v_{max} \frac{[S]}{K_m + [S]}
\]

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