Derivation of the Rate Equation for the Aspartate Aminotransferase Mechanism from the Michaelis-Menten Assumptions

The rate equation for the enzymatic mechanism exemplified by aspartate aminotransferase has been derived from steady-state theory (ALBERTY[1], VELICK and VAVRA[2]), but it has not been reported before that the derivation can be made using the Michaelis-Menten conditions.

Symbols are those recommended by the Enzyme Commission of the International Union of Biochemistry, modified in that the 2 half-reactions are treated separately, the symbols referring to the second enzyme form being distinguished where necessary by the prime ('), and in that certain additions are required, namely:

$$e_t = e + e' \text{ (total enzyme concentration)},$$

K_a, K_b are the 'true K_m' for the substrates A and B, as defined by VELICK and VAVRA[2].

The 2 half-reactions are:

$$E + A \rightleftharpoons E\,A \xrightarrow{k} E' + \text{product 1},$$

$$K_A \equiv (e - p)a/p\,,$$

$$E' + B \rightleftharpoons E'\,B \xrightarrow{k'} E + \text{product 2},$$

$$K_B \equiv (e' - p')b/p'\,.$$

For any constant overall reaction rate, the rates of the 2 half-reactions must be equal, for if they were not, the enzyme form produced by the faster would accumulate, and the enzyme form consumed by it would be depleted until the 2 rates became equal, i.e. $v = k\,p = k'\,p'$. Therefore $(p/p') = (k'/k)$ ($= R$, say). Thus the concentrations of the 2 enzyme-substrate complexes are in a fixed ratio during any period of constant reaction rate, irrespective of substrate concentrations. The value of R could be altered if k and k' themselves were altered but not otherwise.

Note that,

$$e = p\,(1 + K_A/a)\,, \qquad e' = p\,(1 + K_B/b)/R\,.$$

Therefore,

$$e = \frac{e_t\,R\,(1 + K_A/a)}{1 + K_B/b + R\,(1 + K_A/a)}\,,$$

$$v = k\,p = k'\,e/R\,(1 + K_A/a) = \frac{k'\,e_t}{1 + K_B/b + R\,(1 + K_A/a)}\,.$$

The rate equation derived from steady-state theory may be written,

$$V_{max}/v = 1 + K_a/a + K_b/b\,,$$

and the following correspondences are readily obtained:

$$V_{max} = k'\,e_t/(1 + R)\,, \qquad K_a = R\,K_A/(1 + R)\,,$$

$$K_b = K_B/(1 + R)\,.$$

The new equation predicts half-maximal velocity when half the total enzyme exists as total enzyme-substrate complex. This may be achieved in an infinite variety of combinations of substrate concentrations, but fixing the concentration of one, fixes the other.

The factors $1/(1 + R)$ and $R/(1 + R)$ express the concentration of $E'B$ and EA respectively as proportions of the total enzyme-substrate complex. It should be noted that R cannot be equated with the ratio of pyridoxal to pyridoxamine forms of aspartate aminotransferase. Indeed it has no simple physical interpretation, since there are, in reality, more than 2 forms of enzyme-substrate complex.

Résumé. On peut déduire l'équation de vitesse de la réaction catalysée par la transaminase glutamique – oxalacetique (EC 2.6. 1.1) en utilisant les postulats de Michaelis et Menten, ainsi que l'hypothèse de «Steady-State».

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Effect of Hormones on Acid Mucopolysaccharide Synthesis in Mouse Skin. An Enzyme Study

It has been found that uridine diphosphate-D-glucuronic acid (UDPGA) and glucosamine-6-phosphate (Gm-6-P) are important intermediates as monosaccharide units for the synthesis of acid mucopolysaccharides (AMPS)[1]. UDPGA can be converted enzymatically from uridine diphosphate-D-glucose (UDPG) by a dehydrogenase[2], and hexose-6-phosphate and L-glutamine (L-glut.) form Gm-6-P and glutamic acid by the L-glut.-D-hexose-6-phosphate transamidase[3]. The metabolism of AMPS is known to be influenced by various hormones[4]. However, some of the data are still contradictory[5,6].

In this investigation, UDPG dehydrogenase (UDPG-DH), L-glut.-D-fructose-6-phosphate(F6P)-transamidase and L-glut.-D-glucose-6-phosphate(G6P)-transamidase activities were studied in the normal skin of mice, as were the effects of various hormones on these enzymes comparatively.

Materials and methods. Thirty-six female and 12 male Swiss albino mice, weighing 16–23 g, were kept on an optimal laboratory diet and were given water ad libitum. They were divided into 7 groups, i.e. normal and treated with 4 different hormones. In the sex hormone-treated

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