INHIBITION OF KIDNEY AND INTESTINAL MUTAROTASE BY ACTIVELY ABSORBED SUGARS[1]

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It has been proposed (Keston, 1954) that the enzyme mutarotase is involved in reabsorption of sugars by the kidney. According to the postulated mechanism, mutarotase accelerates conversion of sugars toward a preferentially absorbed form which then, due to a concentration gradient, can diffuse back into the blood stream. The hypothesis fell into disrepute when it was shown that 1-deoxyglucose (Crane and Krane, 1956) and α-methyl glucoside (Wilson and Landau, 1960) were actively absorbed and could inhibit transport of other actively absorbed sugars (Jorgensen, Landau and Wilson, 1961). Since these sugars are non-mutarotable, it was not possible to reconcile the results with Keston's hypothesis.

Recent developments in the field of active transport have popularized the concept of a membrane acceptor site to which transported substances become transiently bound. Such binding might be expected to influence rate of mutarotation of a sugar as a coincidental effect. The possibility arose therefore that "mutarotase" activity detected in extracts of kidney (Keston, 1954) and intestine (Bailey and Pentchev, 1963) may represent coincidental activity of such an acceptor protein. This hypothesis would require that the transported sugars 1-deoxyglucose

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and α-methylglucoside should be capable of interacting with the intestinal "acceptor-mutarotase" protein. This possibility has now been confirmed by the experiments described below.

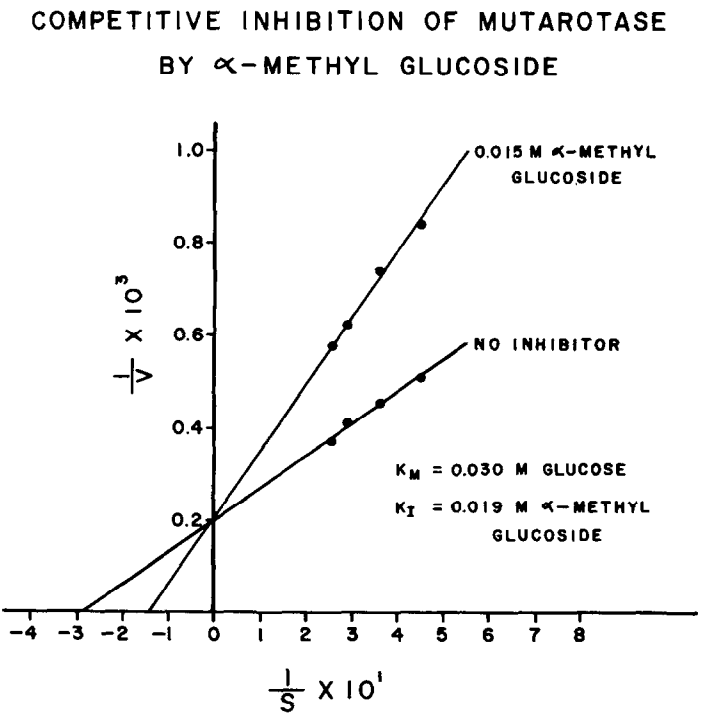

Figure 1. - α-D-glucose in concentrations of 2.2, 2.7, 3.3 and 3.9 x 10^{-2} M was incubated with 10 ml of centrifuged and diluted 1% rat kidney homogenate in .005 M EDTA pH 7.4 at 27°C in presence and absence of α-methyl-D-glucoside (0.3%). Optical rotation was measured at successive time intervals in Keston polarimeter attachment to Beckman Model DU Spectrophotometer. First order rate constants (K) were derived for each catalyzed reaction. Rate (V) is expressed as product of K catalyzed - K spontaneous and substrate concentration (S).

Interaction with the mutarotase active site was adjudged from ability of the particular sugar to function as inhibitor of the enzyme catalyzed mutarotation of α-D-glucose. Based upon this criterion, a number of sugars which themselves are incapable of mutarotation (1-deoxy-D-glucose, α-methyl-D-glucoside and β-methyl-D-xyloside) interacted with the catalytic site of the enzyme. Conversely, a number of

sugars (D-mannose, 2-deoxy-D-glucose and 2-deoxy-D-galactose) inherently capable of mutarotation, failed to interact. A third group (D-galactose, 3-O-methyl-D-glucose, D-xylose, 6-deoxy-D-galactose and L-arabinose) containing a mutarotable hydroxyl, were inhibitors of enzyme activity. Results can be seen in Figure 1 and Figure 2.

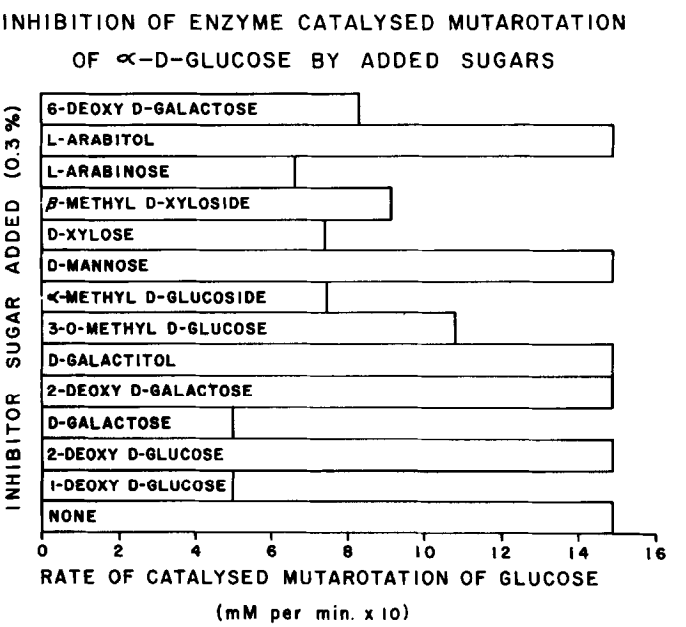

Figure 2. - First order rate constants (K) for enzyme catalyzed mutarotation of α-D-glucose were measured at 27° in presence of each sugar. Individual sugars were added in equilibrium form at same concentrations as substrate (0.3%). Enzyme solution was 10 ml centrifuged 1% homogenate of rat mucosa in .005 M EDTA at pH 7.4. Rate (V) is expressed as product of (K catalyzed - K spontaneous) and substrate (glucose) concentration.

Ability of sugars to inhibit intestinal enzyme correlates well with literature data for active transport of carbohydrates in small intestine. The comparison is given in Table 1. From the results obtained, it appears possible that the protein "mutarotase" plays a role in sugar transport, although in a different manner to that originally proposed.

Table 1

COMPARISON OF ABILITY OF SUGARS TO INHIBIT INTESTINAL "MUTAROTASE" AND LITERATURE DATA ON ACTIVE TRANSPORT

Sugar	Inhibitor of Mutarotase	Literature data on active transport
D-galactose	Yes	Yes (Fisher and Parsons, 1953)
D-mannose	No	No (Wilson and Vincent, 1955)
3-O-methyl-D-glucose	Yes	Yes (Wilson and Vincent, 1955)
1-deoxy-D-glucose	Yes	Yes (Crane and Krane, 1956)
2-deoxy-D-glucose	No	No (Crane and Krane, 1958)
α-methyl-D-glucoside	Yes	Yes (Wilson and Landau, 1960)
2-deoxy-D-galactose	No	No (Wilson and Landau, 1960)
D-xylose	Yes	No (Wilson and Landau, 1960); but shares same carrier as glucose (Salomon, Allums and Smith, 1961)
6-deoxy-D-galactose	Yes	Yes (Wilson and Crane, 1958)
L-arabinose	Yes	No (Wilson and Vincent, 1955); no data on carrier system.
L-arabitol	No	No data
D-galactitol	No	No data
β-methyl-D-xyloside	Yes	No data

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