Kinetics of carbohydrate uptake in *Saccharomyces fragilis*

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Uptake of sugars by *Saccharomyces fragilis* can proceed via energy independent facilitated diffusion or via energy coupled transport. Energy for this active transport can come from a proton gradient or from a high energy phosphate group (polyphosphate). Sorbose transport in this yeast is found to proceed via a combination of facilitated diffusion and proton symport. Also for other organisms such a mixed transport has been found (1,2). It was suggested, that only one carrier was operative, where the proton symport took place only after protonation of the carrier. However alternative explanations are possible. Kinetic analysis of these transports can give more exact information. Therefore the initial uptake kinetics have been described. Six sets of equations were derived: three in which active and passive transport share the same translocator, and three in which they use seperate carriers. Further it is considered that the binding of proton and sugar to the symporter can have three possible sequences: random binding or obligately ordered binding, with either proton binding first or otherwise sugar binding first.
Measurement of the initial uptake kinetics showed that sorbose enters the cell via facilitated diffusion and proton symport, using seperate carriers. Computer analysis of the uptake data also showed, that facilitated diffusion is pH independent from pH 4-8, while for proton symport a strong decrease in the maximal uptake velocity could be seen on raising pH, while the binding constant remained unchanged. These data indicate that the symport carrier has no obligatory binding sequence with respect to proton and sugar.
Further it is concluded that a group with pK=6.4 is responsible for the binding of protons. This seems to indicate that the imidazole ring from histidine is the proton binding group.
Kinetic analysis of 2-deoxyglucose uptake shows that for this sugar there is only one carrier operative. This transport is also an active process, however probably not coupled to a proton gradient but rather to the hydrolysis of polyphosphate. It seems that this sugar is taken up as sugar-phosphate. Transport of 2-deoxyglucose is also accompanied by influx of a small amount of protons, which is probably necessery for charge-compensation (3). Uptake kinetics showed that 2-deoxyglucose transport is only slightly pH dependent. Since the data do not fit into one of the symport models, this again shows that this sugar is not taken up via proton symport.

1. Höfer, M. and Misra, P.C. (1978) Biochem.J. 172, 15-22
2. Komor,E. and Tanner, W. (1974) J.Gen.Physiol. 54, 568-581
3. Jaspers, H.T.A. and van Steveninck, J. (1977) Biochim. Biophys. Acta 469, 292-300