Financial support by Deutsche Forschungsgemeinschaft (grant Do 33/9) is gratefully acknowledged.


Isolation of an Electrophoretically Homogeneous Transketolase from the Yeast Candida utilis

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Commercial dried Candida utilis was found to be a good source for the preparation of all the enzymes of the pentose phosphate cycle. 6-Phosphogluconate dehydrogenase from Candida was crystallized by an Italian group (Rippa et al., 1970); crystallized glucose 6-phosphate dehydrogenase (Chilla et al., 1973) and a highly purified ribose 5-phosphate isomerase have been obtained from Candida by our group (Domagk et al., 1973). In collaboration between the two laboratories, a purification scheme has been worked out in which all the three activities plus transketolase are obtained in different fractions. Transketolase from liver and spinach was partially purified (Horecker et al., 1953). Purification of the enzyme from baker's yeast has yielded a crystalline preparation (de la Haba et al., 1955). C. utilis was used as the starting material for transketolase by Canadian workers (Kiely et al., 1969), but their procedure gives only a 12-fold purification. The procedure proposed here includes autolysis of the dried yeast, filtration through DEAE-cellulose, chromatography on phospho- and DEAE-cellulose, and gel filtration on Ultrogel AcA-44 and AcA-34 (LKB). The purified transketolase has a specific activity of 14 units/mg of protein. Some properties of the enzyme are reported.

This work was supported by Fonds der Chemischen Industrie, Frankfurt.

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Steady-State Kinetics Studies: Mechanism of Action of Alcohol Dehydrogenases from Yeast, Horse Liver and Rat Liver

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The alcohol dehydrogenases (EC 1.1.1.1) from yeast and horse liver have already been subjected to kinetic investigation of the initial velocity in the steady state, whereas this