Preparation and Properties of Urease Covalently Bound to Polyaminostyrol

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Immobilized enzymes gain increasing significance within many fields of natural science. The application of immobilized urease reaches from analytical chemistry (enzyme electrodes) to the medical area (lowering blood urea) [1].
The quality of carrier bound enzymes depends on the preparation conditions as well as on the physical properties of the carrier's surface. At the fixation of urease to polyaminostyrol (PAS) by an azo linkage as an example, it is shown, how the reaction conditions can be systematically optimized. Therefrom rules are deducable, that are commonly applicable to such syntheses. In the case in hand, the optimum conditions are a) a pH-value of the reaction mixture between 4.5 and 5.5, b) a reaction time of 25 min., and c) an enzyme supply of about 30 mg per 100 mg PAS. Furthermore, the number of attachment points between enzyme and carrier is of great importance for the activity of the immobilized enzyme. This parameter can be simply controlled by the rate of diazotization. The influence of the surface properties of the carrier is demonstrated by results obtained from PAS preparations with different surface size and porous structure. The experimental findings are supported by scanning electron micrographs.
Parallel to these experiments the adsorption of urease to PAS has been studied in order to compare these results with those obtained for the covalently bound urease.
The reaction conditions strongly affect the storage stability of the immobilized enzyme. The activity of urease adsorbed to PAS decreases within 50 days by 45 % whereas urease covalently bound to PAS under optimum reaction conditions looses 5 % of its activity at the most.

The K_m-value for immobilized urease is drifted from $2.75 \cdot 10^{-3}$ mole/l to $6.3 \cdot 10^{-3}$ mole/l, whereas the activation energy E_a for the urease catalyzed urea hydrolysis is reduced by about 10 % to 31.5 kJ/mole.

1. Mosbach, K. (1976) FEBS Lett. 62 (Suppl.) E80 - E95.