

**IMPROVEMENT OF THE THERMAL STABILITY OF IMMOBILIZED  
ALPHA AMYLASE BY COUPLING WITH PROLINE**

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$\alpha$ - Amylase was immobilized to Sepharose-4B activated by electrophilic method using cyanogen bromide. The  $\alpha$ -amylase coupled was 77% of the total protein added. Further L-proline was covalently linked to the immobilized  $\alpha$ -amylase by carbodiimide. Optimum carbodiimide concentration for the coupling of proline to the immobilized  $\alpha$ -amylase and the suitable proline concentration for the coupling were determined. Activity of immobilized  $\alpha$ -amylase was not altered after coupling to proline. The thermal stability of soluble  $\alpha$ -amylase, immobilized  $\alpha$ -amylase and immobilized  $\alpha$ -amylase-proline conjugates (samples coupled to two different proline concentrations) were studied at 45°C and 60°C. Soluble  $\alpha$ -amylase lost its total activity on the 30th and 16th days at 45°C and 60°C respectively. Immobilized  $\alpha$ -amylase-proline conjugate (< 85.35  $\mu$ g proline/g gel) lost only 78% activity at 45°C on the 30th day while the same preparation took 20 days at 60°C to lose the total activity. On the other hand the immobilized  $\alpha$ -amylase-proline conjugate (785.32  $\mu$ g proline/g gel) lost only 30% of its original activity at 45°C on the 30th day and took 30 days at 60°C to lose its total activity. These results show that the coupling of proline to immobilized enzymes increases their thermal stability.

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