

A new ω -transaminase for the kinetic resolution of β -amino acids

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요약문

Alcaligenes denitrificans Y2k-2 was screened via enrichment culture from soil, which showed ω -transaminase (ω -AptA) activity able to kinetically resolve β -amino acid. The structural gene encoding the transaminase was cloned. The corresponding gene encodes AptA of 440 amino acids with a calculated molecular mass of 46.843 kDa. The gene was functionally expressed in *E. coli* using a IPTG inducible pET expression system (9.6 U/mg). The AptA produced from *E. coli* BL21 was purified to homogeneity (77 U/mg). The purified AptA showed an apparent molecular weight of 180 kDa as determined by gel filtration chromatography, suggesting a tetrameric structure. The enzyme showed an isoelectric point of 6.2. Optimal pH and temperature for enzyme activity were 9.0 and 45 °C, respectively. Pyruvate and pyridoxal 5'-phosphate increased the enzyme stability. The enzyme convert various β -amino acids to the corresponding β -keto acids utilizing the pyruvate as amine acceptor. The enzyme also exhibits high stereoselectivity ($E > 80$) in the kinetic resolution of racemic β -amino-*n*-butyric acid, producing optically pure D- β -amino-*n*-butyric (99% *ee*) with 53% conversion.

참고문헌

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