Purification and Characterization of an Intracellular Catechol 1,2-dioxygenase from Pseudomonas aeruginosa TKU002

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ABSTRACT

Pseudomonas aeruginosa TKU002 is the production bacillus for catechol 1,2-dioxygenase (abridged as C12O) screened by this laboratory, capable of accumulating intermediate product catechol, with decomposition into cis,cis-mucomic acid by the C12O produced by the bacillus. The preferable culture medium for TKU002 to yield C12O is 0.05% urea, 0.3% glycerin, 0.6% Sodium Benzoate. Cultured at 30℃ for 3 days, the bacilli are retrieved for ultrasound homogenization. The supernatant from centrifugation is purified into a singular C12O enzyme through separation of nucleic acid with streptomycin sulfer and purification steps by DEAE-Sepharose CL-6B and Sephacryl S-100, with molecular weight about 22 kDa, iso-electric point less than pH 5. In the substrate specificity respect, C12O shows a higher activity to pyrogallol; as for the influence of metallic ions on the enzyme, Zn2+ inhibits its activity, while Mn2+ promotes it

Keywords : 鄰苯二酚 ; 苯甲酸鈉 ; 兒茶酚

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