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Subcloning of the Adenosine Deaminase Gene from Pseudomonas iodinum IFO 3558

조영배^{*}, 백영석 , 전홍기 부산대학교 미생물학과

A cosmid library of <u>Pseudomonas iodinum</u> IFO 3558 genomic DNA was constructed and used to transform the adenosine deaminase deficient mutant, <u>E. coli</u> Sø3834. Cosmid clones containing the adenosine deaminase were isolated. One of these clones, named padd20, has an insert of approximately 20kb of genomic DNA. The add gene was further delineated by preparing a secondary plasmid library from padd20. The resulting recombinant plasmids were introduced into Sø3834, and transformants were selected on minimal medium plus 2,6-diamino purine. A number of transformants were obtained and one transformant, padd1.4, was identified as having the smallist insert of approximately 1.4kb. The presence of the add gene in padd1.4 was proved by testing activity of adenosine deaminase in the cell transformed with padd1.4.

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Studies on the Protoplast Fusion of Lactic Acid Bacteria via Electric Field or Chemical Induction

김혜정*, 조영배, 백형석, 전홍기 부산대학교 미생물학과

Protoplast fusion of <u>Lactobacillus acidophilus</u> 88 and <u>Lactobacillus bulgaricus IFO 13953 via electric field or chemical induction was attempted to obtain the improved strains. The highest numbers of electrofusants were obtained at a capacitance value of $2,300\,\mu\text{F}$, a field strength of 500V/cm and at a pulse controller setting of 49 Ohms. When various bivalent cations were added to the electroporation buffer, the electrofusion yield was slightly increased by the addition of lmM Mg++. Fusion yield of electroporation was better than that of chemical method. Electrofusants were very efficiently obtained by addition of polyethylene glycol in electroporation buffer. Acid production and protease activity of <u>L. bulgaricus</u> was better than that of <u>L. acidophilus</u>. Fusant No. 7 revealed excellent protease activity. Lipase activity of <u>L. bulgaricus</u> was better than that of <u>L. acidophilus</u> but fusant No. 11 exhibited the higest lipase activity.</u>