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Cloning of the Adenosine Deaminase Gene from Pseudomonas iodinum IFO 3558

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A cosmid library of Pseudomonas iodinum IFO 3558 genomic DNA was constructed and used to transform the adenosine deaminase deficient mutant, E. coli S43834. 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 S43834, 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 smallest 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

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Protoplast fusion of Lactobacillus acidophilus 88 and 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 2,300μF, 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 1mM 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 L. bulgaricus was better than that of L. acidophilus. Fusant No. 7 revealed excellent protease activity. Lipase activity of L. bulgaricus was better than that of L. acidophilus but fusant No. 11 exhibited the highest lipase activity.