

技术及应用

N⁺离子注入技术在中性蛋白酶高产

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摘要 为获得中性蛋白酶高产菌株, 选用低能 (30 keV) N⁺离子束以不同剂量注入中性蛋白酶, 产生菌枯草芽孢杆菌 (Bacillus subtilis) AS1.398, 研究其诱变效应, 菌株存活率曲线为典型的“马鞍型”剂量-效应曲线, 且在马鞍形区域内具有较高的正突变率。经多次筛选, 获得一株稳定高产突变株ZC-12, 突变株产中性蛋白酶活力为初始菌株的1.84倍。通过对PCR扩增诱变前后两菌株编码中性蛋白酶基因的DNA序列的测序和比对, 在所报道的该酶催化区域内, 有3个氨基酸位点发生突变, 表明N⁺离子束注入细胞的辐射诱变方法具有独特的诱变效果, 可用于菌种选育。

关键词 [枯草芽孢杆菌](#); [中性蛋白酶](#); [离子注入](#); [诱变选育](#); [基因克隆](#)

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Application of Low Energy N⁺ Ions Implantation Technology in Breeding of Neutral Protease Producing Bacteria

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Abstract This study aimed to investigate the biological effects induced by low energy ions implantation on neutral protease producing bacteria. A bacterium, Bacillus subtilis AS1.398, which could produce neutral protease was mutagenized by implanting 30 keV N⁺ ions beam and a high yield mutant strain ZC-12 was obtained. And the activity of the mutant was 0.84 times more than the original strain. Mutagenic effects were investigated, and the results indicate that the survival rate curve takes a “saddle” shape and high positive mutation rate is occurred in this region, these show that unique mutation effects of ion implantation are presented. The DNA sequences of the neutral protease genes in the original strain and the mutant were cloned and analyzed. Through blasting this two [CM(38)]sequences, five amino acid sites of mature peptide were changed showing that unique mutation effects of ion implantation were presented. As a result, low energy ions implantation was successful in neutral protease producing bacteria breeding.

Key words [Bacillus subtilis](#); [neutral protease](#); [ion implantation](#); [mutation breeding](#); [gene clone](#)

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