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# 人肿瘤坏死因子基因在变铅青链霉菌中的克隆与表达[1)]

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摘要 以质粒PIJ486为载体，将来源于质粒pHT1的肿瘤坏死因子(TNF-α) cDNA 克隆至变铅青 链霉菌(Streptomyces lividans TK54). 以新霉素(30μg/ml)为选择标记，获得了数百转化 子，实验表明No.7 转化子S.lividans TK54-HT 所含重组质粒 PIJT7 已克隆有TNFcDNA。L 929细胞毒实验结果表明该转化子TNF表达量可达108活性单位/升以上，中和实验确证其表达 产物为TNF-α。SDS-PAGE表明克隆菌株裂解上清液于17000道尔顿外有明显蛋白带，与TNF- α分子量相当，这表明TNF基因在变铅青霉菌中获得了成功的克隆与表达。

关键词 人肿瘤坏死因子-α,基因克隆和表达,变铅青链霉菌 1993

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# Gene Cloning and Expression of TNF in Streptomyces lividans TK54

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**Abstract**

With the plasmid pIJ486 as a vector, TNF cDNA originated from plasmid pHT1 h as been cloned into Streptomyces lividans TK54. After transformation, hundreds o f transformants, resistant to neomycin(30μg/ml) were selected. Result showed th at TNF cDNA was inserted into the plasmid pIJT7 harboured in No.7 transformant, namely Streptomyces lividans TK54-HT. Cytoxic activity experiment of L929 cell d emonstrated that the maximal TNF activity in Stre ptomyces lividans TK54-HT is m ore than 108 unit/L culture. Th neutrolization experiment showed that the produc t producing by Streptomyces lividans TK54-HT is human TNF-α. SDS-PAGE confirmed that there is a 17 000 dolton protein band which has the same molecular weight as that of TNF in the clear lysate supernatant of Streptomyces lividans TK54-HT cell. All of these results mentioned above showed that the TNF gene has been succe ssfully cloned and expressed in Streptomyces lividans TK54.

**Key words** Human TNF-α Gene cloning and expression Streptomyces lividans TK54

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