研究论文

极端嗜盐古生菌启动子序列缺失突变的微量热研究

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摘要 用微量热方法和DNA缺失突变技术研究了来源于极端嗜盐古生菌R1上的一个推测的启动子片段(RM10) 在大肠杆菌中的启动子功能. 启动子片段融合到质粒pKK232-8上无启动子的氯霉素乙酰转移酶(CAT) 基因前来检测它驱动基因表达的能力, 缺失分析RM10启动子片段定位具有启动活性的重要功能区. 实验结果从热动力学角度揭示, 这个启动子片段上含有一35区和一10区特征的1382~1517 bp(碱基对) 区段是它在大肠杆菌中具有启动子功能的关键部分; 在1~1382 bp区段或1571~1848 bp区段上还存在它的负调控区. 该研究为基因启动子功能研究提供了一种新的、更加灵敏便捷的、化学与生物学相结合的方法.

关键词 微量热 嗜盐古菌 缺失突变 启动子

分类号

Microcalorimetric Study on Deletion Mutagenesis of the Gene Promoter Sequences from the Extremely Halophilic Archaea

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Abstract Microcalorimetric method and DNA deletion mutagenesis technique were combined to study a putative gene promoter fragment (RM10) from the extremely halophilic archaea, *Halobacteria halbium* R1, for its promoter function toward *Escherichia coli*. The promoter fragments were fused to the promoter-less chloramphenicol acetyltransferase (CAT) gene on plasmid pKK232-8 to evaluate its ability of driving CAT gene expression. Deletion analysis for RM10 was performed to identify important functional region responsible for promoter activity toward *Escherichia coli*. From the view of thermokinetics, the experimental results revealed that the 1382 to 1517 bp (base pair) with the typical —35 and —10 box sequences of bacterial promoters was very critical region for promoter function to *Escherichia coli*, and there was a negative control region from 1 to 1382 bp or from 1571 to 1848 bp. Our research work also provided a very sensitive and easily-performed novel method, combining the chemical and biological technique, for studying gene promoter function.

Key words <u>microcalorimetry</u> <u>halophilic archaea</u> <u>deletion mutagenesis</u> <u>gene promoter</u>

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