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生物化学工程、制药、食品和天然产物加工

基于λ噬菌体裂解基因表达的 PHB分离新工艺

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摘要 将S基因琥珀突变的λ噬菌体裂解基因(*S-RRz*)引入产聚β-羟基丁酸酯（PHB）的重组大肠杆菌VG1(pTU14)中以实现细胞的可控裂解破壁. 采用EDTA/Tris(pH值8.0)缓冲液处理结果表明，S-RRz在VG1(pTU14)中能够成功表达，且EDTA对细胞裂解的决定性作用是由于它模拟了S基因产物的功能. 当细胞内PHB含量为85%～90%时，大量积累的PHB颗粒可以改变细胞膜的通透性，实现重组细胞的内控自裂解. 对PHB与细胞进行直接分离的后处理工艺研究表明，在S-RRz成功表达的基础上，采用升温处理模拟S基因产物的功能诱导细胞自裂解，PHB产品纯度可以达到95%以上.

关键词 λ噬菌体裂解基因 聚β-羟基丁酸酯 细胞可控裂解

分类号

NEW TECHNIQUE FOR RECOVERY OF PHB FROM RECOMBINANT *Escherichia coli* BASED ON EXPRESSION OF LYTIC GENES OF PHAGE λ WITH S AMBER MUTATION

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Abstract

The lytic genes of phage λ with S amber mutation(*S-RRz*) were introduced into the recombinant *E. coli* VG1(pTU14) producing poly-β-hydroxybutyrate(PHB)to attain controllable lysis of cells. The results of EDTA/Tris(pH8.0)buffer treatment showed that *S-RRz* were successfully expressed in VG1(pTU14), and cell lysis was realized due to the action of EDTA on cytoplasm membrane.Here the function of EDTA was similar to that of *S* gene product.When PHB content was 85%—90%, membrane permeability would be increased by the abundantly accumulated in-cell PHB granules, and then the autolysis of recombinant cells occurred. After studies on different projects for direct separation of PHB from fermentation broth, a new technique, in which temperature treatment was introduced to simulate the function of S gene product, was presented, and the autolysis of cells was then easily realized based on the successful expression of *S-RRz*.By this simple technique, the final purity of PHB product could be up to 95%.

Key words lytic genes of phage λ with *S* amber mutation(*S-RRz*) poly-β-hydroxybutyrate (PHB) controllable lysis of cells

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