

生物技术—研究报告

过表达海藻糖-6-磷酸合成酶拮抗酵母菌工程菌株的构建

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摘要:

本研究构建了带G418抗性的载体pFL61-G418, 从酿酒酵母WT303菌株中克隆了真菌抗逆境基因tps1(海藻糖-6-磷酸合成酶基因), 再将tps1基因插入到pFL61-G418载体的NotI位点, 构建pFL61-G418-tps1表达载体, 并将该表达载体转化进入野生型拮抗酵母膜醭毕赤酵母菌株中。构建过表达海藻糖-6-磷酸合成酶拮抗酵母菌工程菌株, 增强拮抗酵母菌的抵抗逆境的生存能力, 从而提高其生活力及与病原菌的竞争力。结果表明成功地构建了过表达海藻糖-6-磷酸合成酶拮抗酵母菌工程菌株, 说明pFL61穿梭质粒可以用来转化野生型拮抗酵母, G418抗性可以作为转化子筛选的阳性标记。该重组表达载体的成功构建, 为野生型拮抗酵母的遗传改良提供了重要的理论依据。

关键词: 过表达

Construction of Vector for High Trehalase by Overexpression of tps1 in Antagonistic Yeast *Pichia membranefaciens*

Abstract:

The G418 resistance expression vector pFL61-G418 was constructed and trehalose-6-P synthase gene tps1 from the strain WT-303 of *Saccharomyces cerevisiae* was cloned. pFL61-G418-tps1 expression vector was constructed, and it was transformed into *Pichia membranefaciens* by lithium acetate method. Overexpression of tps1 gene improved the stress tolerance of *P. membranefaciens*, enabled the cells to increase viability and biocontrol ability. We succeeded in construction of vector for high trehalase by overexpression of tps1 gene in antagonistic yeast. The results showed that yeast shuttle vector pFL61 could be used to transform wild type antagonistic yeast and G418 resistance can be used as a dominant selectable marker to select transformants. The plasmid pFL61-G418 provides a visible and effective method for studying the functions of exogenous genes expression in wild type antagonistic yeast.

Keywords: overexpression

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