

生物化学工程与技术

## 渗透剂对苹果酸脱氢酶包含体的复性作用

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摘要

将甜菜碱和海藻糖两种渗透剂作为添加剂, 考察了其促进苹果酸脱氢酶包含体的复性作用。结果显示, 甜菜碱和海藻糖均可有效促进大肠杆菌苹果酸脱氢酶包含体的复性。在本文实验条件下, 随添加甜菜碱浓度的升高, 复性后苹果酸脱氢酶的比活增大; 而海藻糖则出现最佳添加浓度使复性后苹果酸脱氢酶的比活最大; 同时在盐酸胍浓度较高、不添加渗透剂时复性效果较差的复性体系中, 若添加合适浓度的甜菜碱或海藻糖即可有效促进苹果酸脱氢酶的复性, 显示出渗透剂作为添加剂在实际包含体蛋白质复性过程中的应用前景。另外, 通过外源荧光分析发现, 添加甜菜碱和海藻糖均可降低苹果酸脱氢酶表面疏水性区域的暴露程度; 圆二色光谱分析则表明, 甜菜碱和海藻糖可以促进苹果酸脱氢酶 $\alpha$ -螺旋结构的生成。

关键词 [渗透剂](#) [大肠杆菌苹果酸脱氢酶](#) [复性](#) [1-苯胺基萘-8-磺酸](#) [圆二色光谱](#)

分类号

## Effect of osmolytes on refolding of *E.coli* malate dehydrogenase

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### Abstract

Recombinant *E.coli* malate dehydrogenase (eMDH) was expressed as inclusion bodies (IBs), and the eMDH IBs was used as a real oligomeric protein to study protein refolding from IBs assisted by two major osmolytes, betaine and trehalose. It was observed that the specific activity of eMDH was enhanced by increasing betaine concentration, while there was an optimal trehalose concentration at which the protein was favorably renatured. Furthermore, the renaturation could be improved even at high guanidinium chloride concentrations (up to  $0.4 \text{ mol}\cdot\text{L}^{-1}$ ) in the presence of betaine or trehalose. It was shown by binding of 1-anilino-8-naphthalene sulfonic acid (ANS) to eMDH as a function of osmolyte concentration at different guanidinium chloride concentrations that the hydrophobic residue exposure on eMDH surface was reduced in the presence of osmolyte. Circular dichroism (CD) spectra indicated that osmolyte could promote the formation of  $\alpha$ -helical in eMDH.

### Key words

[osmolyte](#) [E.coli malate dehydrogenase](#) [refolding](#) [1-anilino-8-naphthalene sulfonic acid](#) [circular dichroism](#)

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