分离工程

N-和C-末端组氨酸标记基因重组AxCeSD的柱层析分离特性

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摘要

关键词

组氨酸标记 固定化金属亲和层析 尺寸排阻层析 AxCeSD

分类号

Column chromatographic separation characteristics of gene recombinant protein AxCeSD with N- and C-terminal histidine-tags

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Abstract

Immobilization metal affinity chromatography (IMAC) and size-exclusive chromatography (SEC) have been widely used in the purification of recombinant protein. In order to apply the column chromatography to the separation and purification of the gene recombinant with histidine-tags, the column chromatographic separation characteristics of N-terminal histidine-tagged (N-AxCeSD) and C-terminal histidine-tagged (C-AxCeSD) gene recombinant protein AxCeSD, one of the subunit involved in the cellulose synthesis in Acetobacter xylinum were studied. In the ring-shaped three-dimensional structure of AxCeSD, N-terminal histidine-tags were located in the inner of ring, while C-terminal histidine-tags were located in the outer. A higher imidazole concentration was necessary for eluting the C-AxCeSD from the IMAC column due to the C-terminal histidine-tags had stronger chelating interaction with the Ni²⁺ on the IMAC media. Moreover, the retention time for eluting C-AxCeSD from the same SEC gel column was shorter than that for N-AxCeSD, because the larger protein homolog was formed in the C-AxCeSD solution through the inter-molecular hydrogen bonds between the C-terminal histidine-tags.

Kev words

histidine-tags immobilization metal affinity chromatography size-exclusive chromatography AxCeSD

DOI:

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