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RESEARCH NOTES

亲和反胶团选择性萃取分离酵母乙醇脱氢酶

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摘要 The reversed micelles were formed with cationic cetyltrimethylammonium bromide (CTAB) as surfac tant and n-hexanol as cosolvent in the CTAB (50mmol.L-1)/hexanol (15% by volume)/hexane system. Cibacron Blue 3GA (CB) as an affinity ligand in the aqueous phase was directly introduced to the reversed micelles with electrostatic interaction between anionic CB and cationic surfactant. High molecular weight (Mr) protein, yeast alcohol dehydrogenase (YADH, Mr = 141000) from baker's yeast, has been purified using the affinity reversed micelles by the phase transfer method. Various parameters, such as CB concentration, pH and ionic strength, on YADH forward and backward transfer were studied. YADH can be transferred into and out from the reversed mi celles under mild conditions (only by regulation of solution pH and salt concentration) with the successful recovery of most YADH activity. Both forward and backward extractions occurred when the aqueous phase pH＞pI with electrostatic attraction between YADH and CTAB. The recovery of YADH activity and purification factor have been improved with addition of a small amount of affinity CB. The recovery of YADH activity obtained was ～99% and the purification factor was about 4.0 -fold after one cycle of full forward and backward extraction. The low ionic strength in the initial aqueous phase might be responsible for the YADH transfer into the reversed micellar phase.

关键词 reversed micelles yeast alcohol dehydrogenase protein purification affinity technology cetyltrimethy lammonium bromide

分类号

Selective Affinity Separation of Yeast Alcohol Dehydrogenase by Reverse Micelles with Unbound Triazine Dye

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Abstract

The reversed micelles were formed with cationic cetyltrimethylammonium bromide (CTAB) as surfac tant and n-hexanol as cosolvent in the CTAB (50mmol.L-1)/hexanol (15% by volume)/hexane system. Cibacron Blue 3GA (CB) as an affinity ligand in the aqueous phase was directly introduced to the reversed micelles with electrostatic interaction between anionic CB and cationic surfactant. High molecular weight (Mr) protein, yeast alcohol dehydrogenase (YADH, Mr = 141000) from baker's yeast, has been purified using the affinity reversed micelles by the phase transfer method. Various parameters, such as CB concentration, pH and ionic strength, on YADH forward and backward transfer were studied. YADH can be transferred into and out from the reversed mi celles under mild conditions (only by regulation of solution pH and salt concentration) with the successful recovery of most YADH activity. Both forward and backward extractions occurred when the aqueous phase pH＞pI with electrostatic attraction between YADH and CTAB. The recovery of YADH activity and purification factor have been improved with addition of a small amount of affinity CB. The recovery of YADH activity obtained was ～99% and the purification factor was about 4.0 -fold after one cycle of full forward and backward extraction. The low ionic strength in the initial aqueous phase might be responsible for the YADH transfer into the reversed micellar phase.

Key words reversed micelles yeast alcohol dehydrogenase protein purification affinity technology

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