

代謝疾患に関与するMBP 融合タンパク質 (MCAD : medium-chain acyl-CoA dehydrogenase) の精製

Column: MBPTrap HP 5 ml

Sample: N-terminal MBP-MCAD in E. coli lysate

Sample volume: 15 ml

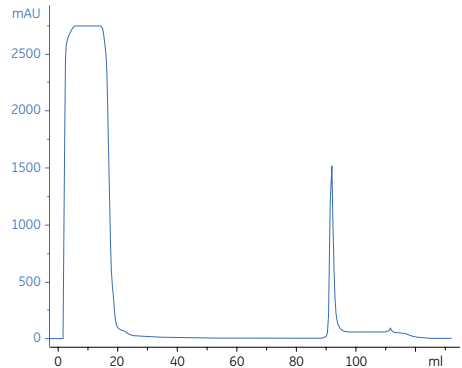
Flow rate: 5.0 ml/min

(0.5 ml/min during sample loading)

Binding buffer: 20 mM Tris-HCl, 200 mM NaCl, 1 mM EDTA, 1 mM DTT, pH 7.4

Elution buffer: 10 mM maltose in binding buffer

System: ÄKTAprime plus



Lane

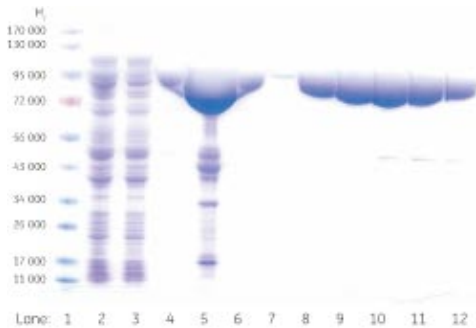
1 Molecular weight marker

2 Start material, N-terminal MBP-MCAD in E. coli lysate, dil. 6 ×

3 Flow through MBPTrap HP, dil. 6 ×

4-6 Eluted fractions from MBPTrap HP

7-12 Eluted fractions from gel filtration



Column: HiLoad 16/60 Superdex 200pg

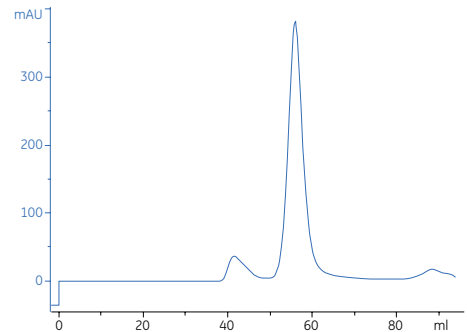
Sample: Eluted fraction from MBPTrap HP 5 ml

Sample volume: 2 ml

Flow rate: 0.4 ml/min

Buffer: 20 mM HEPES, 200 mM NaCl, pH 7.0

System: ÄKTAprime plus



MBPTrap HP で精製した後、続いてHiLoad 16/60 Superdex 200 pg で最終精製を行いました。選元条件下のSDS-PAGEにより、極めて純度高く目的タンパク質を精製できていることがわかります。