Application of immobilized bovine enterokinase in repetitive fusion protein cleavage for the production of MUC1

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Keywords:	enterokinase, immobilization, fusion protein cleavage, protein purification, MUC1

Scheme of the batch binding chamber used for enterokinase purification with metal chelate affinity chromatography.
169x83mm (600 x 600 DPI)

Application of immobilized enterokinase in repetitive fusion protein cleavage. The reaction solution was removed after 24 h (increased process time of 27 h (#) and 33 h (#')) and new substrate solution was added to the enzyme-support preparation starting a new cleavage reaction. The set reaction conditions involved 1.16 mg*mL⁻¹ MUC1-IgG2a Fc in 50 mM Tris-HCl, pH 8.0 incubated with 10 U*gcarrier⁻¹ (determined with GD4K-2NA) immobilized enterokinase in a reaction volume of 1 mL at 25°C under shaking conditions.

87x63mm (600 x 600 DPI)

Repeated fusion protein cleavage by immobilized enterokinase in the absence and presence of magnesium. The reaction solution was removed after 24 h and new substrate solution was added to the enzyme-support preparation starting a new cleavage reaction. The set reaction conditions involved 0.625 mg*mL⁻¹ MUC1-IgG2a Fc in 50 mM Tris-HCl, pH 8.0 incubated with 10 U*mL⁻¹ 1*carrier-1 (determined with GD4K-2NA) immobilized enterokinase in a reaction volume of 1 mL at 25°C under shaking conditions. The reaction buffer was supplemented with 1 mM magnesium ions. A) Repetitive batches in the absence of magnesium; B) Repetitive batches in the presence of magnesium.

125x168mm (600 x 600 DPI)