Integrating continuous process steps by desalting with microporous chromatographic media

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CONCLUSION

• A simple and versatile desalting method using microporous ion exchange media was developed.
• which is easily scalable.
• and can be integrated into fully continuous processes.
• Due to low resin costs, it is also attractive to be used in disposable mode.

INTRODUCTION

• Desalting of protein solutions can be difficult in large scale and is often neglected in process development.
• Particularly in integrated continuous processes ion removal adds complexity.
• Membrane techniques face fouling problems.

GOAL:
Development of a desalting method based on chromatographic beads for different model applications.

EFFECT OF DESALTING

5-7 fold higher binding capacities on macroporous ion exchange media were achieved with desalted refolding solution.

COLUMN SWITCHING PROGRAM

Continuous operation on a Semba Octave 10 system.

Loading (1), rinsing (2), regeneration NaOH/HCl (3/4), re-equilibration (5/6).

CASE STUDY 3: Desalting of a scFv refolding solution

AEX:
Dowex Marathon A2
CEX:
Diaion Sepabead SK110

Model system:
scFv inclusion bodies expressed in E. coli were dissolved and refolded resulting in a solution with pH 10.4 and a conductivity of 10.0 mS/cm.

Continuous desalting yielded conductivities below 2 mS/cm at minimal protein loss but significant impurity depletion.

CASE STUDY 2: Desalting of a mAb after salting out

AEX:
Amberlite IRA-400
CEX:
Dowex Marathon MSC

Model system:
mAb in cell culture supernatant salted out with a saturated ammonium sulfate solution. The precipitate was re-dissolved resulting in a solution with pH 7.0 and a conductivity of 8 mS/cm.

Continuous desalting yielded conductivities of approx. 1 mS/cm at minimal protein loss but significant impurity depletion.

CASE STUDY 1: Desalting of a GFP homogenate

AEX:
Amberlite IRA-400
CEX:
Diaion WK40L

Model system:
Green fluorescent protein was expressed in E.coli in soluble form. The clarified homogenate had a pH of 7.3 and a conductivity of 10.0 mS/cm.

Continuous desalting yielded conductivities of approx. 2 mS/cm at minimal protein loss.

REFERENCE:

This work has been supported by the Austrian BMDW, BMVIT, SFG, Standortagentur Tirol, Government of Lower Austria and Business Agency Vienna through the Austrian FFG-COMET Funding Program.