

CONTINUOUS DESALTING OF REFOLDING SOLUTION BY ION EXCHANGE CHROMATOGRAPHY

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Recombinant proteins expressed in *E. coli* as insoluble inclusion bodies need to be resolubilized and refolded to obtain its native structure. These steps require certain salts, which lead to buffers with elevated conductivity. When loading such a refolding solution on an ion exchange column for capturing only relatively low binding capacities can be achieved. In order to overcome this problem, an additional process step has to be introduced. The traditional approach is dilution, diafiltration or dialysis. Here we present a novel alternative process for salt removal of protein solutions. We applied anion and cation exchangers of a micro-pore type, where only salts can penetrate into the pores, but no proteins, in order to desalt the solution. The columns were connected together to run in a serial setup. In order to increase operation performance, a continuous process was developed comprising of four columns, two anion and two cation exchangers. Continuous mode was achieved by staggered cycling operation, where one set of columns was loaded while the other set was regenerated. Proof of concept using a scFv as model protein was performed. The refolding solution could be successfully deionized resulting in constantly low conductivity below 0.5 mS/cm. By running the process continuously process time could be reduced by 38.5% and at the same time productivity was increased to 163% compared to batch operation. Desalting of the protein solution resulted in 5-7 fold higher binding capacities in subsequent ion exchange capture step by conventional protein binding resins.