Fast purification/concentration of a bacteriophage T4 by using short monolithic columns

Separation mode: Anion-exchange chromatography
Instrumentation: Knauer high pressure gradient HPLC system (Knauer, Berlin, Germany)
Separation Device: CIM® QA (strong anion-exchange) Disk Monolithic Column; 3 × 12 mm I. D., V = 0.34 mL
Sample: Phage T4 (DSM No. 4505) incubated in the E. coli bacterial suspension. After the complete lysis, the sample was centrifuged for 20 minutes at 6000 × G (Sigma, Osterode am Harz, Germany). Clarified liquid solution was filtered through a 0.22 μm pore size filter (Millipore, Billerica, MA, USA).
Loading Volume: 20 ml
Mobile Phase: Buffer A: 125 mM Na-phosphate buffer, pH = 7.0
Buffer B: 125 mM Na-phosphate buffer with 1.5 M NaCl, pH = 7.0
Conditions: Gradient: stepwise gradients (at 0.5 M, 1.0 M and 1.5 M NaCl in buffer A); Flow rate: 2 mL/min (6 column volumes/min); Detection: UV at 280 nm.

Results:
Infective phages can efficiently be purified in a single chromatographic step with the use of a strong anion-exchange CIM® QA monolithic column.
Efficient separation of phages from DNA and proteins is obtained and the phage recoveries are in the order of at least 70%.

More details: Smrekar et al., J. Chromatography B, in press.