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Application Highlight

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High Resolution Cofactor and Plasmid Separation by Sartobind® IEX Lab

The phosphate groups on cofactors and the backbone of nucleic acids contribute negative charges to these molecules. They can therefore be easily bound on strong or weak anion exchangers. By use of the included Luer to UNF 10-32 adapters, Sartobind[®] Lab membrane adsorbers can be connected to most liquid chromatography systems for convenient application of complex elution gradients, enabling high resolution separations.

Although the bed height in the three-membrane-layer Sartobind[®] Lab units is only 0.8 mm, it is possible to effectively separate separate molecules based on their minimal charge differences, such as the redox cofactors NAD⁺, NADH, NADP⁺ and NADPH, at high resolution (Figure 1).

Sartobind[®] IEX Lab membrane adsorbers can also be used for the purification of plasmids following alkaline lysis (Figure 2).



Device	Sartobind® IEX Lab, 0.4 mL MV
System	FPLC
Sample	100 μL cofactor mixture (50 μΜ ΝΑΟ⁺, 50 μΜ ΝΑDΗ, 150 μΜ ΝΑDΡ⁺, 150 μΜ ΝΑDΡΗ)
Flow Rate	10 mL/min (~25 MV/min)
Buffer A	0.2 mM TEA, pH 7.7
Buffer B	Buffer A + 1 M KCl
Gradient	0% B for 0.8 min 0 – 20% B in 3.5 min 20 – 100% B in 1.2 min



Figure 1: Separation of Cofactors Using Sartobind[®] Q Lab (0.4 mL MV)

Device	Sartobind® Q Lab, 0.4 mL MV
System	FPLC
Sample	1 mL of alkaline lysate containing pBR322, prepared from 100 mL of HB101 culture according to Maniatis et al. (1982)
Flow Rate	0.3 mL/min (~0.75 MV/min)
Buffer A	20 mM Tris pH 8, 1 mM EDTA, 750 mM NaCl
Buffer B	20 mM Tris pH 8, 1 mM EDTA, 850 mM NaCl
Gradient	0% B for 10 min 0 - 100% B in 20 min



Figure 2: Plasmid Purification Following Alkaline Lysis Using Sartobind® Q Lab (0.4 mL MV)

Reference

Maniatis, T., Fritsch, E.F. and Sambrook, J. In Molecular Cloning: A Laboratory Manual (1982), pp 150–162, Cold Spring Harbor, New York.

Note

Literature published up to c.2022 may reference the use of Sartobind® MA, which is a name previously used for the Sartobind® Lab membrane adsorbers. When these products were renamed, there was no change made to fit, form or function. Therefore, results collected and methods established using Sartobind® MA remain valid also for Sartobind® Lab.

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