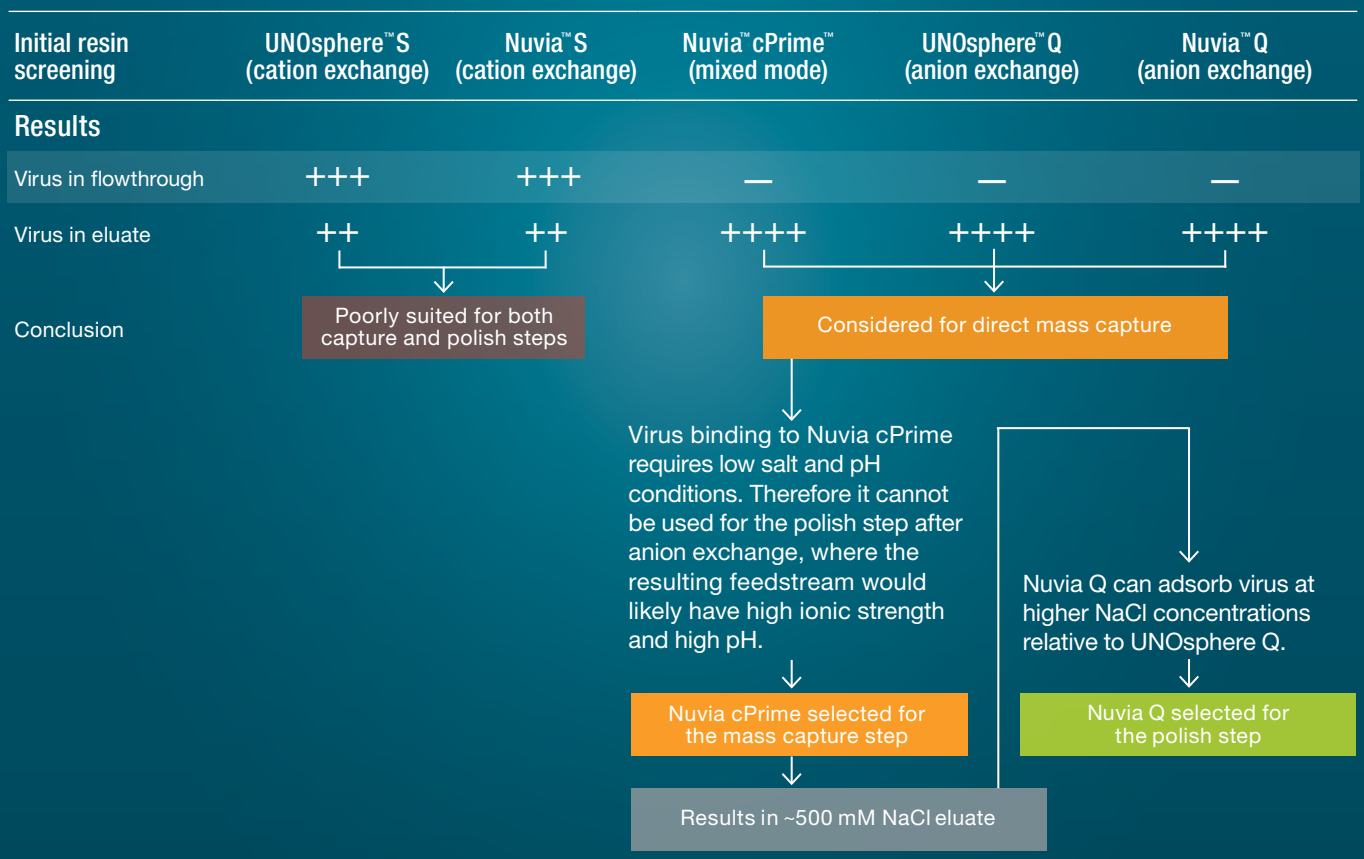
Quick Purification Strategy

Adenovirus Purification

Adenoviruses are commonly used as gene transfer vectors in gene therapy and clinical trials. However, their large size, complexity, and acid labile nature lead to challenges in their downstream purification. The following purification strategy provides an easy two-step protocol to purify adenoviruses.



Results

	Harvest	Nuclease-treated harvest	Nuvia cPrime	Nuvia Q
HCP, ng/10^{10} particles	ND	3,022	58	2
HC dsDNA, ng/10^{10} particles	3,144	30	ND	<0.02
Total virus (x10^{11} particles)	30.6	31.8	18.4	16.4

HCP, host cell protein. HC dsDNA, host cell dsDNA. ND, not determined.

Refer to experimental details on page 2.

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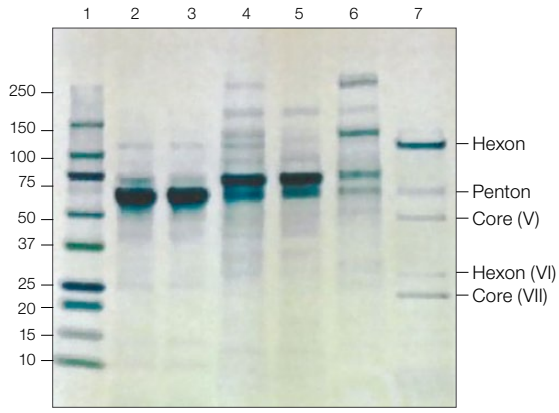
Adenovirus Purification Strategy Details

Capture Conditions

Step	Buffer	Column Volume	Flow Rate, cm/hr
Equilibration	25 mM histidine, pH 6.0 (buffer A)	10	120
Sample Loading	Culture supernatant diluted 1:3 with buffer A	48	120
Wash	Buffer A	5	120
Elution	75 mM Tris, 525 mM NaCl, pH 8.5	3	120

Polish Conditions

Step	Buffer	Column Volume	Flow Rate, cm/hr
Equilibration	75 mM, pH 8.0 (buffer A)	10	120
Sample Loading	Nuvia cPrime eluate diluted 1:1 with buffer A	2	120
Wash 1	75 mM, 250 mM NaCl, pH 8.0	2	120
Wash 2	15 mM Tris, 440 mM NaCl, pH 8.5	3	120
Elution	75 mM Tris, 1 M NaCl, pH 7.5	5	120



SDS-PAGE of intermediates and the final product. Lane 1, MW marker; lane 2, Nuvia cPrime load; lane 3, Nuvia cPrime flowthrough; lane 4, Nuvia cPrime elution/Nuvia Q load; lane 5, Nuvia Q flowthrough; lane 6, Nuvia Q pre-elution; lane 7, Nuvia Q product.

Resources

Download bulletin 6713 to get details about this workflow.

Visit **bio-rad.com/web/ProcessResins** to get technical details about the resins used in this workflow.

Visit **bio-rad.com/web/ProcessApplications** to see other applications in which these resins can be used.

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