

Purification of Adeno-Associated Viruses with Macro-Porous IEX Resin

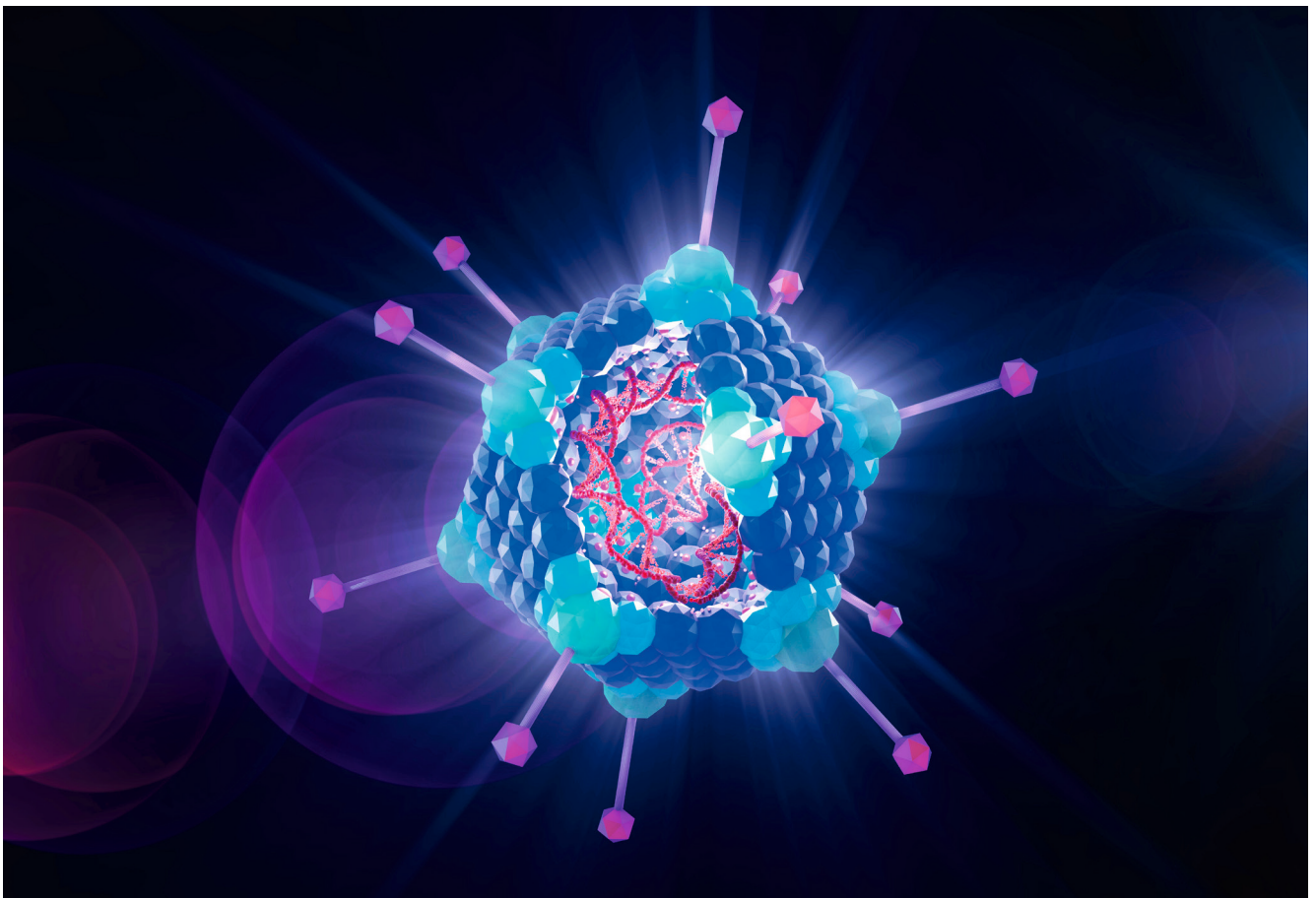
Abstract

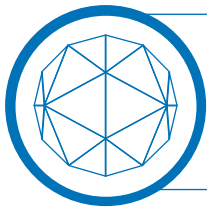
Oligonucleotides are highly important biopharmaceuticals with strongly increasing relevance. Due to their physico-chemical properties, biomolecules have been a focus of research and industry for a long time, and their importance continues to grow. As therapeutic targets evolve, so do the requirements for chromatographic purification strategies. Gene therapy underscores the need for innovative methods to purify large biomolecules. Among these, adeno-associated viruses (AAVs) have emerged as essential tools in the production of gene therapeutics. However, their size and complexity require purification techniques specifically tailored to their unique properties. YMC addresses this challenge with MacroSep IEX Q, an advanced ion exchange resin developed for the purification of large biotherapeutics. Unlike conventional IEX resins, MacroSep IEX Q features a macro-porous structure that enables efficient binding and separation of large particles like AAVs.

Key Benefits of MacroSep IEX Q:

- **High dynamic binding capacity (DBC)** for large biomolecules, ensuring high loadability.
- **Superior separation efficiency** even at elevated flow rates, driving enhanced productivity.

With its advanced properties, MacroSep IEX Q is an ideal solution for downstream processing (DSP) platforms, enabling successful and productive purification of large biotherapeutics.



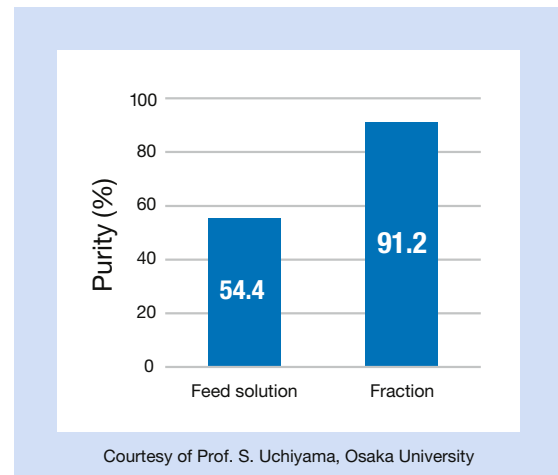
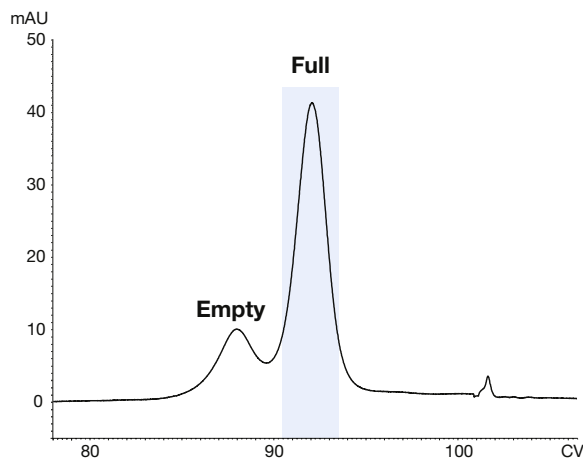


Case Study: Separation of Full and Empty Capsids

Efficient and safe purification of biomolecules is critical but often presents significant complexity. One key step in the purification of AAVs is the separation of empty capsids from fully loaded capsids. These separations are essential to ensure therapeutic efficacy and safety. In this study, AAV serotype 2 (AAV2) was chosen as the model system. AAV2 is one of the earliest identified and characterised serotypes, making it one of the most extensively studied and well-understood AAV types.

The example below demonstrates real-life data from an AAV2 purification process using MacroSep IEX Q. This innovative ion exchange resin achieved excellent separation of full and empty capsids. Purity levels of the full AAV2 species were analysed before and after the chromatographic step using analytical ultracentrifugation. The AEX purification step with MacroSep IEX Q has increased the purity level of the target immensely. Starting from a purity of 54.4 % in the feed solution, the final purity after purification was 91.2 %.

Good Resolution: Efficient Separation of Full and Empty AAVs!



Courtesy of Prof. S. Uchiyama, Osaka University

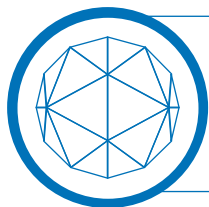
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Column:	MacroSep IEX Q 50x5.0 mm ID
Eluent:	A) 20 mM Bis-tris propane-HCl (pH 9.0), 1% sucrose, 0.1% poloxamer 188 B) 20 mM Bis-tris propane-HCl containing 0.5 M choline chloride (pH 9.0), 1% sucrose, 0.1% poloxamer 188
Flow rate:	1 mL/min
Gradient:	10 CV equilibration 0%B, 48 CV load of sample, 20 CV wash 0%B, 28 CV 0-50%B, 10 CV strip 100%B, 10 CV CIP 1 M NaOH, 10 CV regeneration 1 M NaCl, 10 CV equilibration 0%B
Temperature:	ambient (25 °C)
Detection:	UV at 280 nm
Injection:	48 mL
Sample:	AAV2 (0.72x10 ¹¹ vg/mL)

Excellent Dynamic Binding Capacity for Large Biomolecules

Efficient purification processes with increased productivity rely heavily on high loadability. The DBC of a chromatography resin is a critical parameter. A higher DBC allows for greater loadability in packed columns, directly enhancing the overall process productivity. Large biomolecules, such as

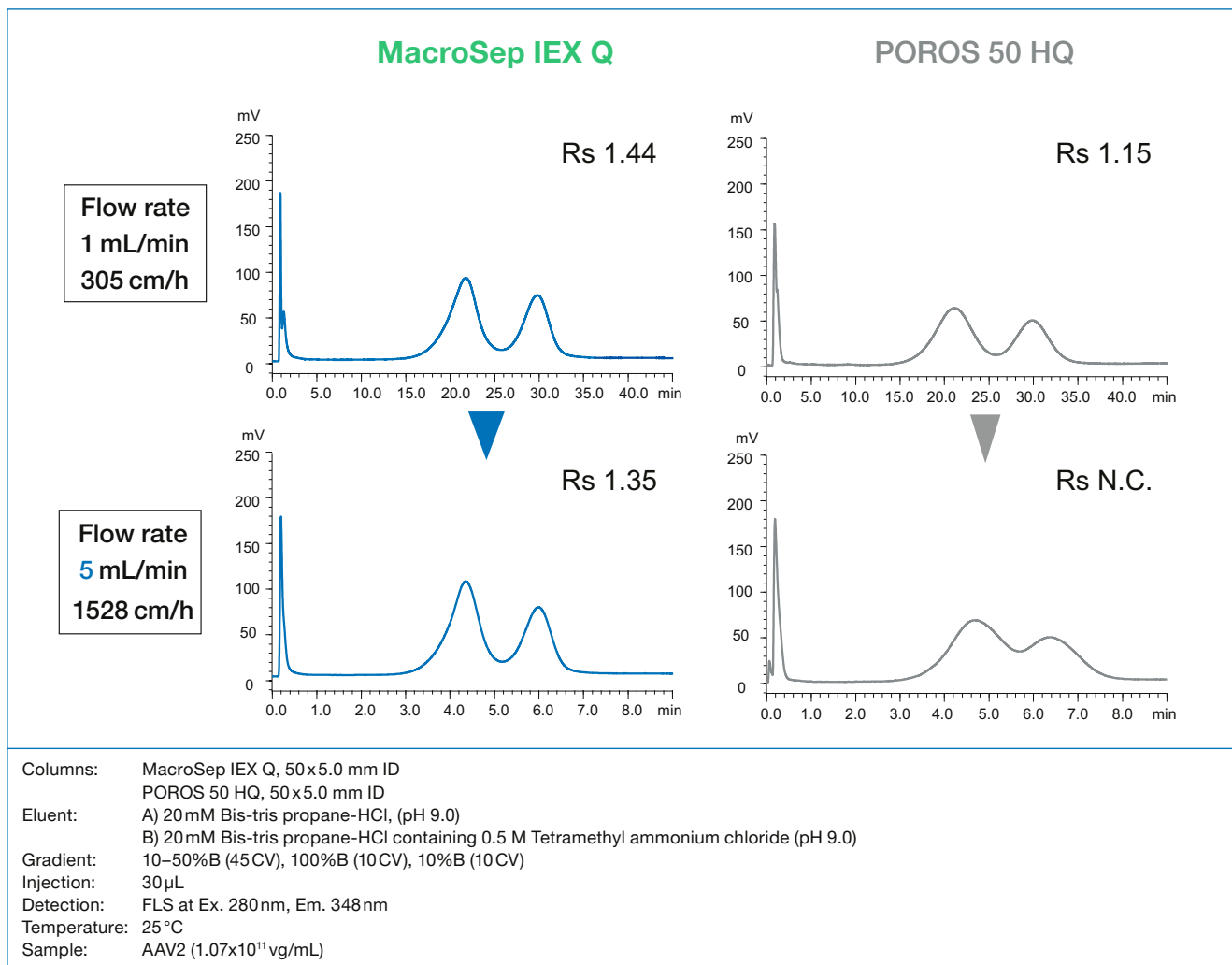
viral vectors, require resins with optimised, macro-porous structures for efficient binding. MacroSep IEX Q was specifically developed to meet these requirements, making it an effective solution for purifying large biomolecules.



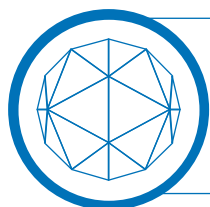
High Productivity with Improved Pressure-Flow Characteristics

In addition to achieving high purity levels, the productivity of a purification process is strongly influenced by the pressure-flow properties of the resin. MacroSep IEX Q is based on hydrophilic polymer beads that exhibit exceptional pressure-flow characteristics, allowing its use at high flow rates without significantly compromising resolution. A direct com-

parison with a conventional IEX resin demonstrates that MacroSep IEX Q provides superior resolution, even under demanding conditions. Its macro-porous structure ensures reliable performance across both standard and elevated flow rates. This ability to maintain resolution at higher flow rates can substantially improve the throughput of DSP workflows.



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Supporting Your DSP Optimisation






With its innovative design and demonstrated performance, MacroSep IEX Q represents a robust tool for the purification of large biomolecules. By addressing key priorities such as loadability, resolution, and process productivity, this resin offers a practical solution for optimising downstream

processes. For further insights or technical support, we invite you to explore how MacroSep IEX Q can enhance your purification processes. Contact our team for detailed data or to discuss potential applications in your workflows.

Specifications

Specifications	MacroSep IEX Q
Matrix	methacrylate-based hydrophilic porous polymer
Charged Group	-R-N ⁺ (CH ₃) ₃
Particle Size	30 µm
Pore Size	900 nm
pH Range	2–12
Pressure Resistance	regular use: 2 MPa max: 3 MPa
Ion Exchange Capacity	min 0.08 meq/mL-resin
Compression Factor	1.05
Temperature	4–60 °C

Screening Options

				
Bulk Packing Material	MiniChrom™ Columns	Screening Kit	RoboColumns®	Analytical Columns
Chromatography resins for self-packing	Prepacked columns with different dimensions and volumes	Prepacked columns (1 mL and 5 mL)	Miniaturized prepacked columns (e.g. 50 µL, 200 µL and 600 µL) High-throughput process development	Prepacked bioinert columns in analytical dimensions