Optimized AEX Buffer Formulations for AAV Full Capsid Enrichment

teknova:

Shira Karni, David Austin, Erik Lustre, Zoe Bowman, Bohdan Andreiuk, Bella Neufeld, Tony Tavares

Chromatography • AAV • Polishing

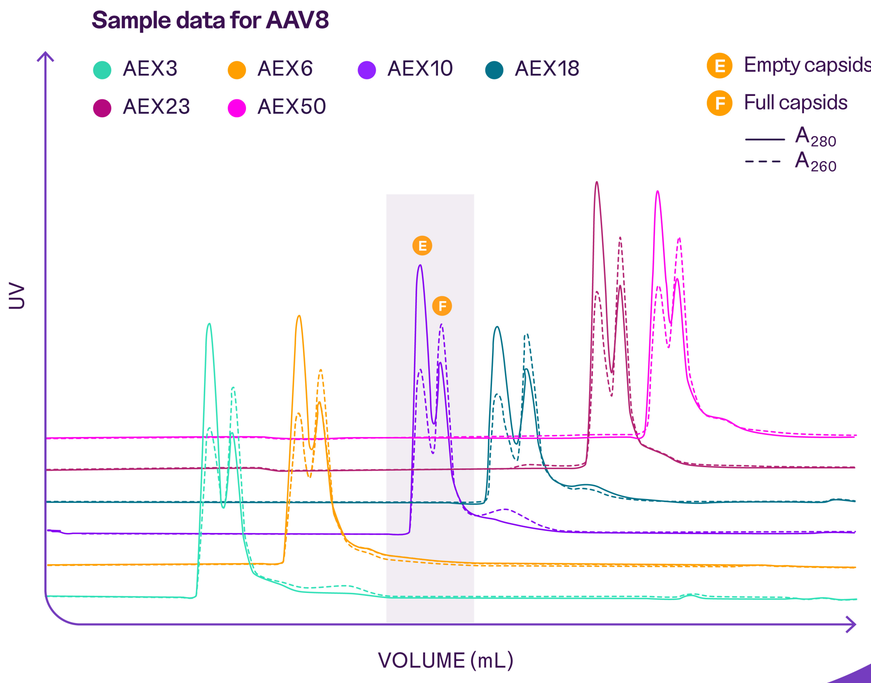
Reduce the time needed to determine the best polishing buffer to optimize the separation of empty and full capsids with our proprietary off-the-shelf buffer kit.

Approach: AEX design of experiments

A number of factors may affect successful polishing of a given AAV construct (including acidity, elution salt, stabilizer, excipient, surfactant, and base buffer). Therefore, a design of experiments (DOE) was executed to elucidate the extent of each contribution and to determine robust recipes.

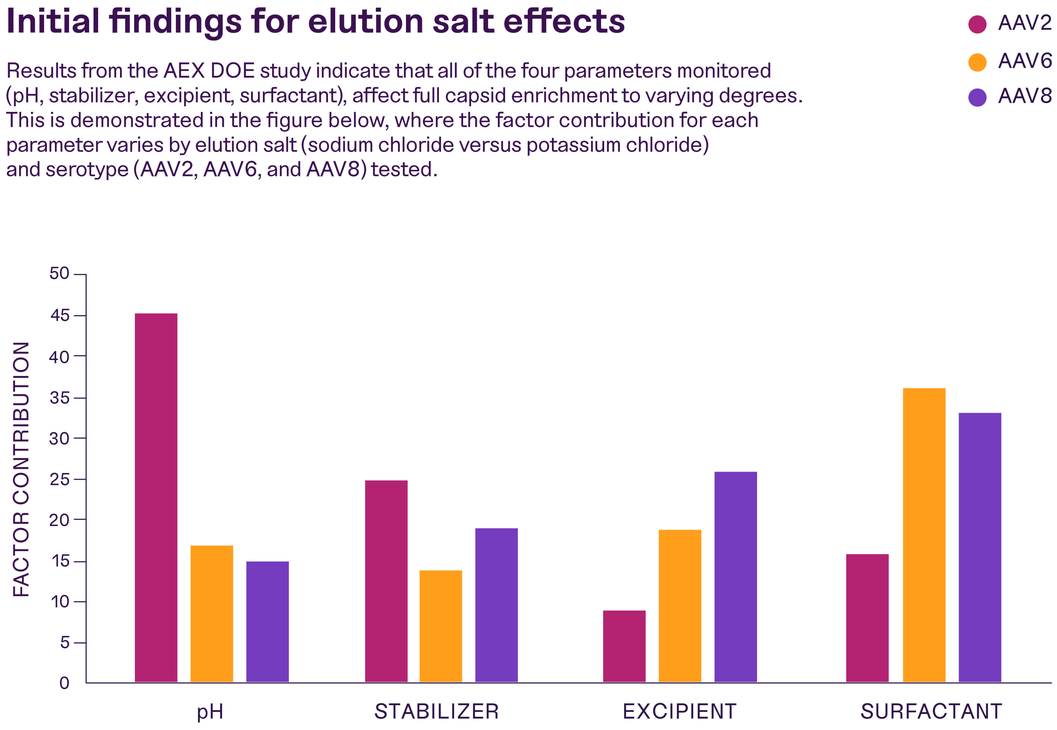
HUNDREDS OF DOE PARAMETERS → 6 EVALUATION METHODS → OPTIMIZED FORMULATIONS → EASY SCALE-UP

- Base buffer and concentration
- Acidity/alkalinity
- Excipient concentration
- Detergent concentration
- Up to 10 elution salts



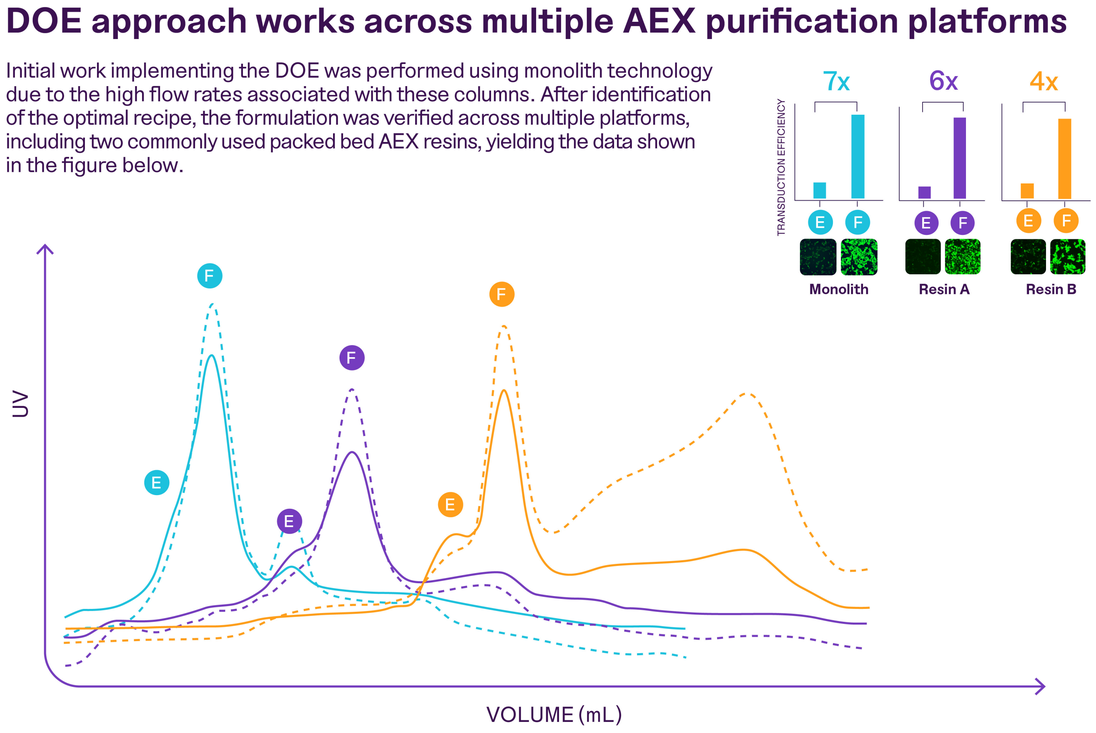
Initial findings for elution salt effects

Results from the AEX DOE study indicate that all of the four parameters monitored (pH, stabilizer, excipient, surfactant), affect full capsid enrichment to varying degrees. This is demonstrated in the figure below, where the factor contribution for each parameter varies by elution salt (sodium chloride versus potassium chloride) and serotype (AAV2, AAV6, and AAV8) tested.



DOE approach works across multiple AEX purification platforms

Initial work implementing the DOE was performed using monolith technology due to the high flow rates associated with these columns. After identification of the optimal recipe, the formulation was verified across multiple platforms, including two commonly used packed bed AEX resins, yielding the data shown in the figure below.



The DOE approach works across AEX upstream platforms

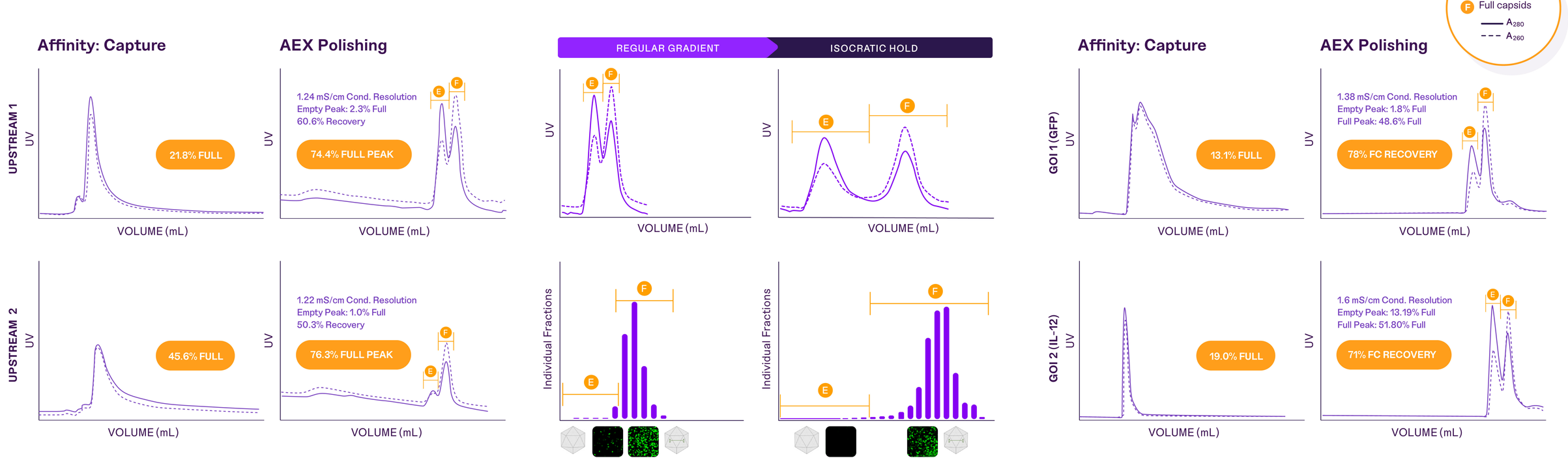
To ensure the kit is agnostic of the upstream process used (for instance, cell line, media choice, transfection and harvest conditions) and does not affect the ability to successfully utilize the buffer set, multiple processes were implemented and tested using the optimal buffer formulations.

Determine the optimal buffer for scale-up

The figure below demonstrates the utility of the kit for various chromatographic modalities. The buffer screening is shown using a linear gradient (left) and then finalized using an isocratic hold (right) to facilitate scale-up requirements.

The DOE approach works across multiple transgene sequences

Different DNA sequences packaged within viral capsids can impart differences in the pI of the overall construct. The optimized buffer formulations were verified across multiple sequence carrying constructs to ensure broad applicability.



Interested in trying our AAV•Tek™ AEX Buffer Screening Kit?

Now available for AAV2, AAV6, and AAV8, this first-of-its-kind kit can save you months in your process development at the polishing step by helping you find the ideal buffer formulation faster for the separation of empty and full capsids. Screening kits for AAV9 are also available for early-access testing.

SCAN QR CODE TO ORDER YOUR AEX KIT

www.teknova.com/AEX-Kit

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Our collaboration with Sartorius BIA Separations

We saw the successful achievement of over 85% full AAV capsid enrichment with 80-90% recoveries when pairing our AEX kit with Sartorius BIA Separations' monolith technology at the polishing step, given a specific starting material of AAV8 that contained 46% full capsids.

Have a question?

Get in touch with our team at **research@teknova.com**