Empty/Partial/Full Capsid Assay Using Sedimentation Velocity Analytical Ultracentrifugation (SV-AUC)

SV-AUC – The Gold Standard for Empty/Partial/Full Capsid Assay

At BioAnalysis, we understand that precise product characterization is critical to your gene therapy program success. Our team partners with you to deliver the industry's most trusted empty/partial/full capsid analysis using SV-AUC technology. Sedimentation Velocity Analytical Ultracentrifugation (SV-AUC) has emerged as the gold standard technique for the empty/partial/full capsid assay in gene therapy product development. With unmatched resolution, accurate quantitation, and regulatory compatibility, SV-AUC is the preferred method for characterizing viral vectors such as AAV (Adeno-Associated Virus), lentivirus, and other delivery systems.

SV-AUC For Gene Therapy

Sedimentation Velocity Analytical Ultracentrifugation (SV-AUC) is a high-precision biophysical technique used to analyze the size, mass, and shape of

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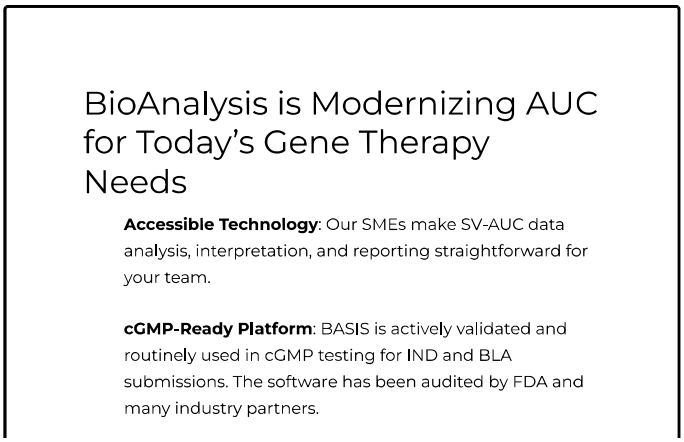
particles in solution.

Using high-speed centrifugation, SV-AUC separates these species based on their molecular and solution properties, producing detailed c(s) distribution plots that offer absolute quantification—no reference standards or calibration curves required, providing you with clear, actionable data that drives confident decision-making.

What Can SV-AUC Measure?

For gene therapy applications, SV-AUC excels in measuring and distinguishing between:

- **Empty Capsids**: Viral particles that do not contain genetic material
- **Partial Capsids**: Capsids with incomplete or truncated genomes
- **Full Capsids**: Properly loaded vectors carrying the therapeutic payload
- **Impurities**: residual DNA and protein, broken capsids
- **Aggregates**: High molecular weight assemblies or aggregates of viral capsids that can impact safety and efficacy



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Flexible Sample Requirements: Our team's experience with thousands of viral projects (across all titers and serotypes) allow for the lowest sample volumes required (100 µL for higher titer to 400 µL for low titers)

Rapid Results: BioAnalysis is believed to have the largest fleet of AUC instruments at a CRO which allows for streamlined workflows that can deliver comprehensive results in less than 14 days (or faster for expedited samples) without compromising quality.

Importance of Empty/Full Capsid in Gene Therapy

The **ratio of full to partial to empty capsids is a critical quality attribute (CQA)** that may impact safety, potency, and efficacy of viral gene therapy products. High levels of empty or partial capsids could:

- Reduce therapeutic efficacy by diluting the active dose
- Increase immunogenicity and risk of immune response
- Complicate dosing strategies across patient populations

Regulatory agencies require robust and validated methods for **empty/full capsid measurement** for:

- **IND (Investigational New Drug) submissions**
- **Lot release testing**
- **Commercial manufacturing quality control**

SV-AUC enables confident decision-making by offering consistent and reproducible quantitation of AAV and other viral vectors under **cGMP conditions**.

1. **Absolute Quantification**

SV-AUC is a first principle technique that directly measures capsid species without relying on external calibration or reference standards, allowing for direct and accurate measurement.

2. **Native-State Measurements**

SV-AUC operates in the product **formulation buffer** without labels, dyes, or matrix changes. The native structure and integrity of viral vectors is preserved.

3. **High Sensitivity & Resolution**

Metric	Value
LOD of capsids	< 0.5% to 5%
LOQ of capsids	< 1.5% to 15%
%CV for S-values	< 0.5%
%CV for Quantitation	< 10%

This precision allows for reliable detection and quantification of **even low-abundance species**, such as trace levels of empty capsids and aggregates.

4. cGMP and 21 CFR Part 11 Compliance

- With unique proprietary platforms like **BASIS™ (BioAnalysis SEDFIT Integrated Software)**, BioAnalysis has made SV-AUC uniquely suited as a **regulatory-grade assay. BASIS™** is fully compliant with FDA requirements:
- Electronic signatures
- Secure audit trails
- Data integrity

*Details about **BASIS™**, BioAnalysis' cGMP compliant AUC software can be found in the publication listed below and here

SV-AUC methods are easily developed, qualified, and validated for gene therapy products across **Phase I–III drug development. BASIS™** is routinely evaluated in internal reviews,

external client audits, and regulatory inspections.

SV-AUC vs. Other Empty/Full Capsid Methods

While multiple methods exist for capsid analysis, here's why SV-AUC remains the gold standard:

Technique	Limitations	SV-AUC Advantages
Transmission Electron Microscopy	Expensive, time-consuming, not QC-friendly	SV-AUC is faster, scalable, and QC-compatible
UV/Vis Spectroscopy	Lacks specificity; cannot resolve partial vs. full	SV-AUC separates and quantifies individual species accurately
Charge Detection Mass Spectrometry	Gas-phase, relative quantitation only, high cost	SV-AUC offers in-solution, absolute quantification
Mass Photometry	Limited dynamic range, semi-quantitative, requires dilution	SV-AUC offers better resolution, quantitation, and robustness
Size Exclusion Chromatography with Multi-Angle Light Scattering	Cannot distinguish empty from full due to similar hydrodynamic size	SV-AUC resolves capsids by sedimentation coefficient, enabling clear separation

Why Industry Leaders Choose SV-AUC

SV-AUC offers an **unmatched combination** of:

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- **Quantitative precision**
- **Regulatory readiness**
- **Native-state analysis**
- **High sensitivity and reproducibility**

It remains the **most reliable platform for AAV capsid analysis**, playing a critical role in **quality control, IND submissions, and commercial release testing**.

Complementary Technologies

While SV-AUC is the most robust method for full/empty capsid quantification, it can be complemented with:

- **Density Gradient Equilibrium (DGE)-AUC** for adding more detail to % E/P/F characterization via buoyancy **measurements**
- **Dynamic Light Scattering (DLS)** for measuring hydrodynamic size and polydispersity (aggregation)
- **Size Exclusion Chromatography with Multi-Angle Light Scattering** (**SEC-MALS)** for size exclusion profiling
- **Liquid Chromatography-Mass Spectrometry** (**LC-MS)** for capsid protein characterization
- **Charge Detection Mass Spectrometry (CDMS)** for orthogonal confirmation of mass
- **FRET (Förster Resonance Energy Transfer) or SPR (Surface Plasmon Resonance)** for analyzing functional binding or kinetics

For sponsors developing gene therapy programs, investing in SV-AUC testing with BioAnalysis ensures alignment with **FDA expectations** and provides high-confidence data that supports **product safety, consistency, and efficacy.**

Need help with your empty/partial/full capsid assay? Contact our SV-AUC experts to design a validated, compliant solution tailored to your gene therapy program.

Start Testing