Virongy Biosciences
Accelerate Discovery

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AAV SEC Purification Kit

1. INTRODUCTION & DESCRIPTION:

Virongy's **AAV SEC Purification Kit** provides a fast and reliable method for purifying adeno-associated virus (AAV) following harvest with **Virongy's AAV Extraction and Maturation Kit**. The kit includes 10 **Size-Exclusion Chromatography (SEC)** columns optimized for efficient separation of AAV particles from cellular debris and small-molecule contaminants, along with a ready-to-use wash and elution buffer for streamlined processing. Designed for simplicity and reproducibility, the kit enables researchers to obtain high-quality AAV preparations suitable for downstream applications.

Highlights:

1. Increase AAV Purity
2. Removal of protein/DNA contaminants
3. Removal of Excess Salt from Extraction
4. Simplifies Workflow

Add-on *Virongy's* **Low-Speed Viral Concentration Kit** *to efficiently concentrate and further purify your harvested virus, maximizing yield and quality for downstream applications.*

2. INTENDED USE:

This product has been manufactured for research use only and is not intended for diagnostic or therapeutic applications in humans or animals. THE PRODUCT IS INTENDED FOR USE ONLY BY PROFESSIONALS TRAINED IN MOLECULAR BIOLOGY AND VIROLOGY TECHNIQUES. This kit is designed for the purification of AAV particles using a specialized separation column and buffer system. While optimized for high-titer recovery, performance may vary depending on specific AAV serotypes and production conditions.

3. KIT COMPONENTS:

VBI AAV SEC Purification Kit	Mini	Midi
Number of AAV SEC Columns*	10	50
Cat. No.	AAVSEC-Mini	AAVSEC-Midi
Column Wash Buffer	10 mL	5 x 10 mL
Elution Buffer	10 mL	5 x 10 mL

AAV SEC Purification Kits should be stored at 4°C

* Each AAV preparation is for one DNA-transfected well of a 6 well plate.

MATERIALS (Not Included)	Recommended
Microcentrifuge Tubes	Fisher Scientific, Catalog No.05-408-129 or similar
EQUIPMENT: Liquid pipette	

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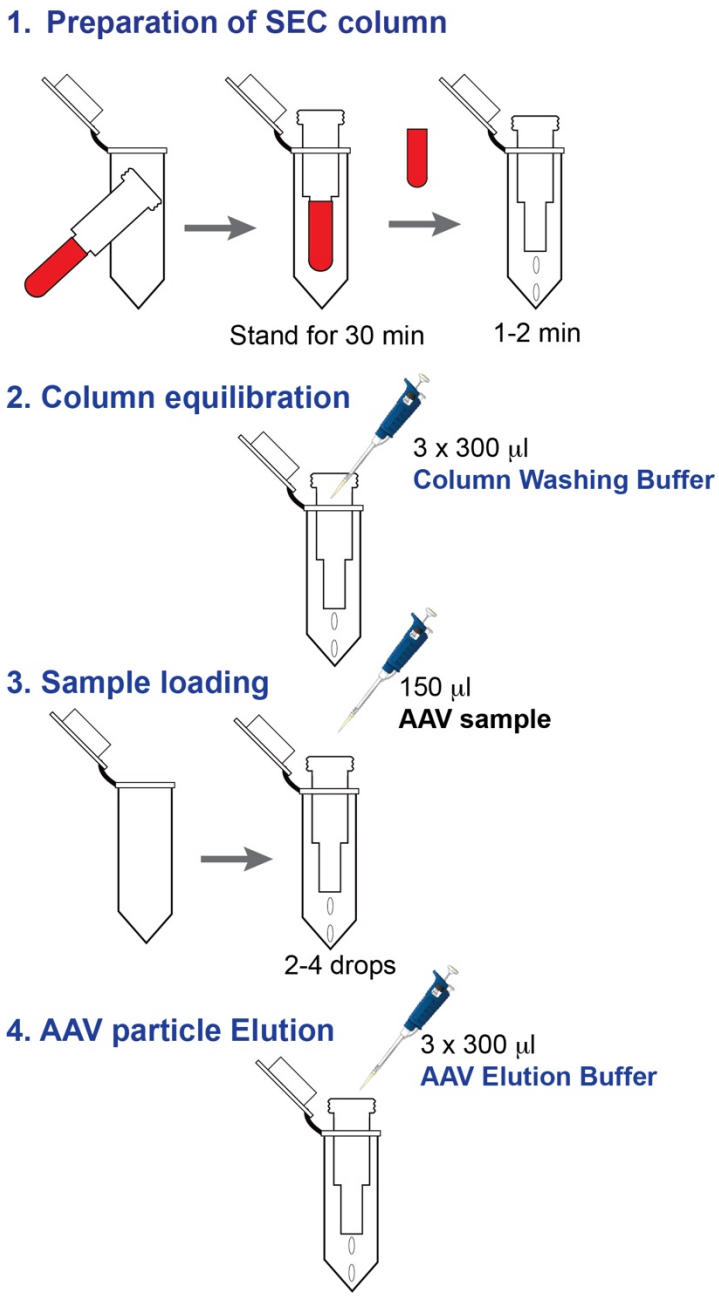
4. KIT PROCEDUES



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AAV SEC Purification Kit Protocol:

Before starting:
Complete AAV extraction using the **Extraction and Maturation Kit Protocol** found in **VBI-HBK-020**

SEC Purification Procedure Using Extracted Lysate from VBI-HBK-020:

1. **Preparation of SEC column**: with the screw cap and red rubber stopper still in place, vigorously shake the AAV SEC column to resuspend the resin. Place the column upright into a microcentrifuge tube and allow it to stand for **30 minutes** before continuing.
2. Remove the screw cap and red rubber stopper from bottom of the column. Place the column back into a microcentrifuge tube to collect the flow-through. Allow the residual storage buffer to drain for approximately **1–2 minutes**.

   Important: *Do **not** allow the column to dry out. Proceed to the next step immediately once dripping stops.*
3. **Column equilibration**: equilibrate the AAV SEC column by washing it **three times** with **300 µL ice-cold AAV Column Wash Buffer**.
   - Allow each wash to fully drain (~1 minute per wash).
   - Discard the flow-through after each wash.
   - Do **not** allow the column to completely dry between washes.
4. **Sample loading**: for one column, load **150 µL** of crude AAV particle sample (prepared using the **AAV Extraction and Maturation Kit, Cat No. AAV-EMK-mini**). Allow the sample to settle into the column resin. Approximately **2 - 4 drops** of flow-through should pass into the collection tube.
5. Transfer the column into a **clean, pre-labeled microcentrifuge tube** for AAV particle collection.
6. **AAV particle Elution**: Add **300 µL AAV Elution Buffer** to the column, and then collect the flow-through.
7. Repeat **Step 6 two additional times**, collecting each elution fraction in separate tubes if desired.

   Note: Additional AAV particles may remain in the column and can be recovered with further elution steps; however, these later fractions will typically have lower concentration.
8. *(Optional)* Pool the eluted fractions into a single tube and mix gently before use. It is recommended to perform an infection using a **serial dilution** of the purified AAV preparation to determine the optimal working dilution for your target cells.

Final Notes:

- The **eluted AAV** can be used **directly** for infection of cultured cells. It is recommended to **dilute 1:10-1:100 with PBS or cell culture media**. Dilute more as needed to avoid cytotoxicity.
- Alternatively, the eluted particle can be further concentrated using Virongy's **Low-Speed Viral Concentration Kit**

Storage Instructions:

- The eluted AAV particles can be stored at 4°C for up to 1 week.
- For long-term storage, store at -80°C