

Adenovirus purification kits

Adeno-X Maxi Purification Kit

- 2- or 6-prep format
- Easy-to-use, streamlined protocol
- Purifies up to 10¹² viral particles
- No CsCl needed

Introduction

Our Adeno-X Maxi Purification Kit is a chromatography-based system that allows you to purify recombinant adenovirus in 1.5 hours or less. The process is quick and easy, and can be scaled up or down without difficulty.

Our Adeno-X Maxi Purification Kit is easy and economical

The Adeno-X Maxi Purification Kit includes an improved protocol that allows you to purify a maximal amount of adenovirus directly from the cell pellet. You simply pellet the cells when the cytopathic effect is complete, and purify the virus. In addition, the kit is available in 2- and 6-prep formats, making it even more economical.

Protocol overview

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A simple chromatographic procedure

Adenovirus is purified chromatographically, without any ultracentrifugation steps, using a unique membrane that selectively binds adenoviral particles. The membrane is housed in a small, single-use cartridge (containing a one-way valve) that is supplied pre-attached to a standard-size BD Luer-Lok syringe. Adenovirus-containing cell lysate is brought into the syringe through the one-way valve and pushed out through the purification filter cartridge, eliminating the need for syringe removal when disposing of the filtered adenoviral medium and washes.

This simple procedure takes just 1–1.5 hours (Figure 1). After the cytopathic effect is complete, harvested cells are lysed and centrifuged. The supernatant is treated with Benzonase Nuclease to digest nucleic acids, clarified through a syringe-tip prefilter, and filtered through the purification cartridge, where the adenoviral particles bind to the filter and are eluted in a small volume of elution buffer.

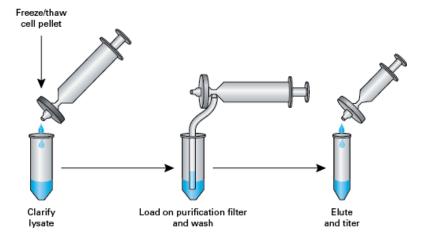


Figure 1. The Adeno-X Maxi Purification Kit protocol.







Results

Achieve high titers of high-quality virus

In a streamlined protocol, the virus is purified directly from the cell pellet. Our studies have shown that starting from the second day after infection, the vast majority of virus is found inside the cells, not in the supernatant (Figure 2). Therefore, combining the supernatant with the cell pellet unnecessarily complicates the protocol without significantly increasing the amount of virus purified.

The high titer and purity achieved by our Adeno-X purification kits is comparable to that achieved by CsCl density gradient centrifugation, with viral particle-to-infectious unit ratios as low as 20:1. Unlike techniques involving CsCl, our method is safe, and requires no advanced training or expensive equipment; it requires only simple manipulations to consistently produce high yields of purified adenovirus. The Adeno-X Maxi Purification Kit can yield up to 10¹² adenoviral particles, depending on the recombinant adenoviral backbone used and the toxicity of the expressed protein of interest. All of our Adeno-X purification kits, tested with recombinant adenovirus, are characterized by highly efficient recovery (Figure 3), producing high-purity, high-quality adenovirus with low viral particle/infectious unit (IFU) ratios.

The Adeno-X Maxi Purification Kit simplifies adenoviral purification by allowing you to purify virus from your cell pellets after the cytopathic effect is complete. Our kit provides ultra-convenient viral purification at a great price.

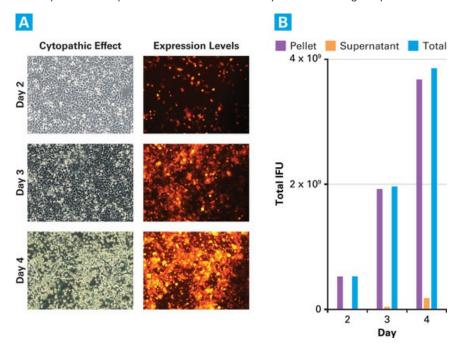


Figure 2. Time course of adenoviral yields from cell pellets and supernatants vs. the cytopathic effect. HEK-293 cells were transduced with an Adeno-X construct expressing DsRed-Express and incubated for 2–4 days. Panel A. The cytopathic effect was observed using phase contrast microscopy and expression was monitored by DsRed-Express fluorescence. By Day 4, the cytopathic effect was nearly complete. Panel B. On Days 2–4, cells were harvested, lysed, and centrifuged—and each pellet and supernatant was used to infect fresh HEK-293 cells to measure titer. Most of the virus was found in the cell pellets, especially by Day 4, with only 2% in the supernatant.







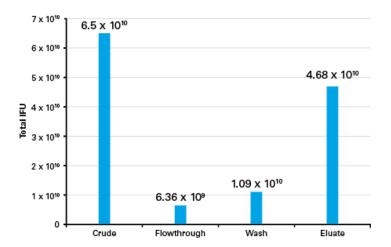


Figure 3. Mass balance analysis of the Adeno-X Maxi Purification Kit protocol. HEK-293 cells were seeded and infected with an Adeno-X construct expressing DsRed-Express as described in Figure 2, and harvested when the cytopathic effect was nearly complete. The resulting adenovirus was then purified according to the Adeno-X Maxi Purification Kit protocol. Total IFU was determined (as described in Figure 2) for each fraction (crude, flowthrough, wash, and eluate), and plotted. 72% of the starting material was recovered in the eluate as purified virus. The process was efficient and fast, allowing us to purify 4.68 x 10¹⁰ adenoviral particles in one hour.

Conclusions

The Adeno-X Maxi Purification Kit allows you to purify recombinant adenovirus directly from cell pellets harvested after the cytopathic effect is complete. The kit yields high titers of high-quality virus with low viral particle/infectious unit (IFU) ratios in 1.5 hours or less, using a simple and safe chromatographic procedure that requires no CsCl and is easily scalable. The Adeno-X Maxi Purification Kit is available in economical 2- or 6-prep formats, and can yield up to 10¹² adenoviral particles, depending on the recombinant adenoviral backbone used and the toxicity of the expressed protein of interest.



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A complete filtration-based purification system for concentrating and purifying infectious adenovirus. The Adeno-X Maxi Purification Kit is an alternative to CsCl density gradient ultracentrifugation. Each kit provides the buffers and filters needed to purify adenovirus from cell lysate.							
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