

Product Information

Ad5 WT

Research use only

Catalog No: AD104-S

Lot No: 1011

Contents:

Wild-type Adenovirus, serotype 5 (Ad5, Genbank AY339865)

Volume: 100 μ L

Physical Titer: 5.0×10^{12} VP/mL

Infectious Titer: 5.2×10^{11} IU/mL

Ratio VP/IU: 10

Ratio A260/A280: 1.35

Ratio A320/A260: 0.22

Buffer:

20 mM Tris, 25 mM NaCl, 2.5 % glycerol, pH 8.0, sterile (GTS buffer, Hoganson *et al*)

Storage: -70°C, in a sealed container appropriately labeled

Virus Amplification:

HEK 293 cells cultured in DMEM supplemented with 10% FBS

Purification Method:

double CsCl banding, dialysis, filtration

Physical Titer: The concentration of virus particles (VP) in the virus stock was calculated given the extinction coefficient of 1.1×10^{12} VP per Abs_{260nm} unit in presence of SDS (Maizel *et al*)

Infectious Titer: The concentration of infectious units (IU) in the virus stock was determined by end-point dilution assay (TCID₅₀), using virus growth as indicator. The TCID₅₀ concentration was calculated using the Reed-Muench method.

The **A260/A280 ratio** is the ratio between the absorbance at 260 nm and the absorbance at 280 nm in presence of SDS. That ratio reflects the relationship between nucleic acid and protein in a purified virus suspension. For CsCl-purified adenovirus, this ratio falls typically between 1.2 and 1.4. Values outside that range indicate contamination (Vellekamp *et al*).

The **A320/A260 ratio** is the ratio between the absorbance at 320 nm and the absorbance at 260 nm in absence of SDS. It is the scattering ratio. It reflects the amount of any aggregation in the purified virus stock. The typical range for the scattering ratio for purified virus is approximately 0.22-0.27. It can rise rapidly from 0.3 to 0.5-0.7 as virus aggregation is initiated (Vellekamp *et al*).

Authentication: Molecular authentication was performed by sequencing viral DNA extracted from purified virus particles, using 3 primers

specific for AY339865. No discrepancy with found between the experimental quality-trimmed sequences and the published sequence of AY339865.

HANDLING INFORMATION

Biosafety level 2 practices and containment facilities are required for all activities involving the virus.

Protective clothing: laboratory coat, gloves, mask, and goggles.

Special care must be taken to avoid spread of infectious material by aerosol, direct contact or accidental injection.

Disposal: decontaminate all wastes with 10% bleach for 30 minutes.

Accidental Release Procedures: pour 1 volume of bleach over the spill and wait for 15 minutes. Wipe up carefully. Hold for autoclave waste disposal and

decontaminate work surfaces with 10% bleach.

First Aid/Treatment:

Contact: Immediately flush eyes and skin with plenty of water for at least 15 minutes. Call a physician.

Inhalation: N/A

Ingestion: Wash out mouth with water. Call a physician

Accidental injection: wash area with soap and water. Call a physician.

References:

Hoganson, D., et al., Development of a Stable Adenoviral Vector Formulation. *BioProcessing*, 2002. 1(1): p. 43-48.

Maizel, J.J., D. White, and M. Scharff, The polypeptides of Adenovirus 1: Evidence for multiple protein components in the virion and a comparison of types 2, 7A and 12. *Virology*, 1968. 36: p. 115-125.

Reed, L.J., & Muench, H. (1938). A simple method of estimating fifty percent endpoints. *Am. J. Hygiene*, 27, 493-497

Vellekamp, G., et al., Empty capsids in column-purified recombinant adenovirus preparations. *Hum. Gene Ther.*, 2001. 12(15): p. 1923-1936.

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