

Certificate of Analysis for NR-44233

Human Respiratory Syncytial Virus, A1998/3-2, Purified from HEp-2 Cells

Catalog No. NR-44233

Product Description: Human respiratory syncytial virus (RSV), A1998/3-2 purified from clarified supernatant from infected HEp-2 cells¹ by high speed centrifugation

Lot^{2,3}: 62973124 Manufacturing Date: 17NOV2014

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in HEp-2 Cells ¹	Cell rounding and syncytia formation	Cell rounding and syncytia formation
Sequencing of Species-Specific Region (829 nucleotides)	Consistent with human RSV, A1998/3-2	99% identity with human RSV, A1998/3-2 (GenBank: JX069801)
Titer by TCID ₅₀ Assay ^{4,5} in HEp-2 Cells ¹ With Direct Fluorescence Assay (DFA) Readout ⁶	Report results	8.9 × 10 ³ TCID ₅₀ per mL
Sterility (21-day incubation) Harpo's HTYE broth ⁷ , 37°C and 26°C, aerobic Trypticase soy broth, 37°C and 26°C, aerobic Sabouraud broth, 37°C and 26°C, aerobic Sheep blood agar, 37°C, aerobic Sheep blood agar, 37°C, anaerobic Thioglycollate broth, 37°C, anaerobic DMEM with 10% FBS, 37°C and 5% CO ₂	No growth	No growth
Mycoplasma Contamination Agar and broth culture (14-day incubation at 37°C) DNA Detection by PCR of Test Article nucleic acid	None detected None detected	None detected None detected

¹HEp-2 cells: ATCC[®] CCL-23™

Date: 06 FEB 2015

Signature:

Title: Technical Manager, BEI Authentication or designee

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

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²Prepared by growth of RSV, A1998/3-2 (BEI Resources NR-28529, lot 59927457) for a single passage in HEp-2 cells followed by virus purification by high speed centrifugation of clarified supernatant

³Grown in Eagle's Minimum Essential Medium (ATCC[®] 30-2003[™]) supplemented with 2% fetal bovine serum (ATCC[®] 30-2020[™]) for 6 days at 37°C and 5% CO₂

⁴The Tissue Culture Infectious Dose 50% (TCID₅₀) endpoint is the 50% infectious endpoint in cell culture. The TCID₅₀ is the dilution of virus that under the conditions of the assay can be expected to infect 50% of the culture vessels inoculated, just as a Lethal Dose 50% (LD₅₀) is expected to kill half of the animals exposed. A reciprocal of the dilution required to yield the TCID₅₀ provides a measure of the titer (or infectivity) of a virus preparation.

⁵7 days at 37°C and 5% CO₂

⁶Using Light Diagnostics™ RSV FITC Reagent (Millipore 5022)

Atlas, Ronald M. Handbook of Microbiological Media. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.