

Certificate of Analysis for NR-28530

Human Respiratory Syncytial Virus, A2000/3-4

Catalog No. NR-28530

Product Description: Cell lysate and supernatant from HEp-2 cells¹ infected with human respiratory syncytial virus, A2000/3-4

Lot²: 63339791 Manufacturing Date: 22MAY2015

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in HEp-2 Cells ¹	Cell rounding and sloughing	Cell rounding and sloughing
Identification by Direct Fluorescent Antibody (DFA) Assay ³	Fluorescence observed	Fluorescence observed
Sequencing of Species-Specific Region (851 nucleotides)	Consistent with human respiratory syncytial virus, A2000/3-4	99% identity with human respiratory syncytial virus, A2000/3-4 (GenBank: JX069803)
Titer by TCID ₅₀ Assay ^{4,5} in HEp-2 Cells ¹ by Cytopathic Effect	Report results	2.8 × 10 ⁶ TCID ₅₀ per mL
Sterility (21-day incubation) Harpo's HTYE broth ⁶ , 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: 19 OCT 2015

BEI Resources Authentication

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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BEI Resources www.beiresources.org E-mail: contact@beiresources.org
Tel: 800-359-7370

Fax: 703-365-2898

²Grown in Eagle's Minimum Essential Medium (ATCC[®] 30-2003[™]) supplemented with 2% fetal bovine serum (ATCC[®] 30-2020[™]) for 4 days at 37°C and 5% CO₂

³Using Light Diagnostics[™] Respiratory Syncytial Virus FITC Reagent (Millipore 5022)

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.

⁵8 days at 37°C and 5% CO₂

⁶Atlas, Ronald M. <u>Handbook of Microbiological Media</u>. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.