

Human Respiratory Syncytial Virus, A2000/3-4

Catalog No. NR-28530

Product Description:

Human respiratory syncytial virus (RSV), A2000/3-4 was isolated from a nasal wash from an infant with RSV bronchiolitis in Nashville, Tennessee, USA, on March 4, 2000. NR-28530 lot 70039169 was produced by infecting *Homo sapiens* carcinoma cells (Hep-2; ATCC® CCL-23™) with the seed material and incubating in Eagle's Minimum Essential Medium (ATCC® 30-2003™) supplemented with 2% fetal bovine serum (ATCC® 30-2020™) for 5 days at 37°C with 5% CO₂.

Passage History:

HEp-2(11)/HEp-2(10) (Prior to deposit at BEI Resources/BEI Resources); HEp-2 = *Homo sapiens* carcinoma cells

Lot: 70039169

Manufacturing Date: 06OCT2020

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in HEp-2 Cells	Cell rounding, syncytia formation and detachment	Cell rounding, syncytia formation and detachment
Identification by Direct Fluorescent Antibody (DFA) Assay ¹	Fluorescence observed	Fluorescence observed
Sequencing of Species-Specific Region (~ 810 nucleotides)	≥ 98% identity with RSV, A2000/3-4 (GenBank: JX069803.1)	99.6% identity with RSV, A2000/3-4 (GenBank: JX069803.1)
Titer by TCID ₅₀ Assay in HEp-2 Cells by Cytopathic Effect and DFA ^{1,2} (8 days at 37°C with 5% CO ₂)	Report results	1.6 × 10 ⁴ TCID ₅₀ per mL
Amplification of RSV Sequence by RT-PCR	~ 900 base pair amplicon	~ 900 base pair amplicon
Sterility (21-day incubation) Harpo's HTYE broth, 37°C and 26°C, aerobic ³ 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, aerobic	No growth No growth No growth No growth No growth No growth No growth	No growth No growth No growth No growth No growth No growth No growth
Mycoplasma Contamination Agar and broth culture (14-day incubation at 37°C) DNA detection by PCR of extracted Test Article nucleic acid	None detected None detected	None detected None detected

¹Using Light Diagnostics™ Anti-Respiratory Syncytial Virus FITC Reagent (Millipore® 5022)

²The 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.

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

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