SUPPORTING INFECTIOUS DISEASE RESEARCH

Influenza A Virus, A/Netherlands/22/2003 (H3N2)

Catalog No. NR-49236

Product Description: Cell lysate and supernatant from Madin-Darby Canine Kidney (MDCK) cells¹ infected with influenza A virus, A/Netherlands/22/2003 (H3N2)

Passage History: C1/C2 (Contributor/BEI Resources; C# = Number passages in MDCK cells)

Lot²: 70010073

Manufacturing Date: 19JAN2018

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in MDCK Cells ¹	Cell rounding and detachment	Cell rounding and detachment
Sequencing of Hemagglutinin and Matrix Coding Regions Hemagglutinin (690 nucleotides) Matrix (930 nucleotides)	Consistent with A/Netherlands/ 22/2003 (H3N2) Consistent with A/Netherlands/ 22/2003 (H3N2)	100% identity with A/Netherlands/22/2003 (H3N2) (GenBank: CY112957.2) 100% identity with A/Netherlands/22/2003 (H3N2)
Titer by TCID ₅₀ Assay ^{3,4} in MDCK cells ¹ With Direct Fluorescence Assay (DFA) Readout ⁵	Report results	(GenBank: CY112958.2) 1.6 × 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 Blood agar, 37°C, aerobic Blood agar, 37°C, anaerobic Thioglycollate broth, 37°C, anaerobic DMEM with 10% FBS, 37°C and 5% CO ₂	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

¹MDCK; ATCC[®] CCL-34[™]

²Grown in Eagle's Minimum Essential Medium (ATCC[®] 30-2003) supplemented with 0.125% bovine serum albumin (Invitrogen[™] 15260-037) and 1 µg per mL L-1-tosylamido-2-phenylethyl chloromethyl ketone (TPCK)-treated trypsin for 2 days at 37°C and 5% CO₂

³The 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 infectious titer (or infectivity) of a virus preparation.

⁴8 days at 37°C and 5% CO₂

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⁵Using Light Diagnostics™ Influenza A Antibody FITC Reagent (Millipore 5017)

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

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E-mail: contact@beiresources.org Tel: 800-359-7370 Fax: 703-365-2898