

**Influenza A Virus, A/Macha/O1453/2021 (H1N1)**

**Catalog No. NR-59464**

**Product Description:**

Influenza A virus, A/Macha/O1453/2021 (H1N1) was isolated from a human in Zambia in 2021. NR-59464 lot 70062492 was produced by infecting Madin-Darby canine kidney-SIAT1 cells (MDCK-SIAT1; Sigma 05071502-1VL) with influenza A virus, A/Macha/O1453/2021 and incubating in Dulbecco's Modified Eagle Medium (ATCC® 30-2002™) supplemented with 0.3% bovine serum albumin and 5 µg/mL N-acetyl trypsin for 4 days at 33°C and 5% CO<sub>2</sub>. The cell lysate and supernatant were spin-clarified at 500 × g for 10 minutes at 4°C.

**Passage History:**

hNEC(1), MDCK-SIAT1(1) / MDCK-SIAT1(1) (Johns Hopkins University/BEI Resources); hNEC = human Nasal Epithelial Cells; MDCK-SIAT1 = Madin-Darby canine kidney-SIAT1 cells

**Lot: 70062492**

**Manufacturing Date: 31OCT2023**

TEST	SPECIFICATIONS	RESULTS
<b>Identification by Infectivity in MDCK-SIAT1 Cells</b>	Cell rounding and detachment	Cell rounding and detachment
<b>Sequencing of Hemagglutinin and Neuraminidase Coding Regions</b> Hemagglutinin (~ 1750 nucleotides)  Neuraminidase (~ 1430 nucleotides)	Consistent with hemagglutinin type 1 (H1) Consistent with neuraminidase type 1 (N1)	Consistent with H1  Consistent with N1
<b>Next-Generation Sequencing (NGS) of Complete Genome Using Illumina® MiSeq™ Platform</b>	Consistent with sequence of depositor's material	Consistent with sequence of depositor's material
<b>Titer by TCID<sub>50</sub> Assay in MDCK-SIAT1 Cells by Hemagglutination Assay<sup>1,2</sup></b> (7 days at 33°C and 5% CO <sub>2</sub> )	Report results	2.8 × 10 <sup>9</sup> TCID <sub>50</sub> /mL
<b>Sterility (21-day incubation)</b> Harpo's HTYE broth, 37°C and 26°C, aerobic <sup>3</sup> 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
<b>Mycoplasma Contamination</b> 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

<sup>1</sup>The Tissue Culture Infectious Dose 50% (TCID<sub>50</sub>) endpoint is the 50% infectious endpoint in cell culture. The TCID<sub>50</sub> 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% (LD<sub>50</sub>) is expected to kill half of the animals exposed. A reciprocal of the dilution required to yield the TCID<sub>50</sub> provides a measure of the infectious titer (or infectivity) of a virus preparation.

<sup>2</sup>Assay performed using 0.5% turkey red blood cells

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

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