

## **Certificate of Analysis for NR-48598**

## Purified Influenza A Virus, A/Brisbane/59/2007 (H1N1) (Tissue Culture Adapted)

## Catalog No. NR-48598

**Product Description:** Influenza A virus, A/Brisbane/59/2007 (H1N1) (tissue culture adapted) purified from infected Madin-Darby canine kidney (MDCK) cells<sup>1</sup> by high speed centrifugation of clarified supernatant

**Passage History:** E2/E1/E2/E1C10 (Submission laboratory/CDC/Baylor College of Medicine/BEI Resources; E# = Number passages in eggs; C# = Number passages in MDCK cells)

Lot<sup>2,3</sup>: 62449041 Manufacturing Date: 20APR2015

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in MDCK Cells <sup>1</sup>	Cell rounding and detachment	Cell rounding and detachment
Sequencing of Species- and Strain-Specific Regions Hemagglutinin gene (448 nucleotides)	Consistent with A/Brisbane/59/2007 (H1N1)	100% identity with A/Brisbane/59/2007 (H1N1) (GenBank: CY163640)
Matrix gene (951 nucleotides)	Consistent with A/Brisbane/59/2007 (H1N1)	99% identity with A/Brisbane/59/2007 (H1N1) (GenBank: CY163633)
Titer by TCID <sub>50</sub> Assay <sup>4,5</sup> in MDCK Cells <sup>1</sup> with Indirect Fluorescent Antibody (IFA) Readout <sup>6</sup>	Report results	1.6 × 10 <sup>8</sup> TCID <sub>50</sub> per mL
Sterility (21-day incubation)		
Harpo's HTYE broth <sup>7</sup> , 37°C and 26°C, aerobic	No growth	No growth
Trypticase soy broth, 37°C and 26°C, aerobic	No growth	No growth
Sabouraud broth, 37°C and 26°C, aerobic	No growth	No growth
Sheep blood agar, 37°C, aerobic	No growth	No growth
Sheep blood agar, 37°C, anaerobic	No growth	No growth
Thioglycollate broth, 37°C, anaerobic	No growth	No growth
DMEM with 10% FBS, 37°C and 5% CO <sub>2</sub>	No growth	No growth
Mycoplasma Contamination		
Agar and broth culture (14-day incubation at 37°C)	None detected	None detected
DNA detection by PCR of extracted Test Article nucleic acid	None detected	None detected

<sup>&</sup>lt;sup>1</sup>MDCK cells: ATCC<sup>®</sup> CCL-34™

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<sup>&</sup>lt;sup>2</sup>Prepared by growth of influenza A virus, A/Brisbane/59/2007 (H1N1) (tissue culture adapted) (NR-31657, lot 62263122) in MDCK<sup>1</sup> cells followed by virus purification by high speed centrifugation of clarified supernatant

<sup>&</sup>lt;sup>3</sup>Grown in Minimum Essential Medium containing Earle's salts (ATCC 30-2003) supplemented with 2 μg/mL TPCK-treated trypsin and 0.125% Bovine Serum Albumin (Gibco 15260-037) for 2 days at 35°C with 5% CO<sub>2</sub>.

<sup>&</sup>lt;sup>4</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 titer (or infectivity) of a virus preparation.

<sup>&</sup>lt;sup>5</sup>6 days at 35°C with 5% CO<sub>2</sub>

<sup>&</sup>lt;sup>6</sup>Using influenza A antibody FITC reagent (Millipore 5017)

<sup>&</sup>lt;sup>7</sup>Atlas, 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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**Date:** 21 JUL 2015

Signature: Milhard

**BEI Resources Authentication** 

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