

## Kilbourne F5: A/New Jersey/10/76 (H1N1) Mutant, Low (L) Yield

**Catalog No. NR-3462**

**Product Description:** Pooled allantoic fluid from specific-pathogen free (SPF) embryonated chicken eggs<sup>1</sup> infected with a low (L) yield mutant (Kilbourne F5) of influenza A virus, A/New Jersey/10/76 (H1N1).

**Lot<sup>2,3</sup>: 58405666**

**Manufacturing Date: 21NOV2008**

| TEST   | SPECIFICATIONS  | RESULTS   |
|--|---|---|
| <b>Identification by Infectivity Using Embryonated Chicken Eggs<sup>1</sup></b><br>Hemagglutination activity using allantoic fluid from infected eggs and 0.5% chicken red blood cells   | Positive  | Positive  |
| <b>Sequencing of Species-Specific Region (445 nucleotides)</b>   | Influenza A virus   | Influenza A virus   |
| <b>Titer by CEID<sub>50</sub> Assay<sup>4,5</sup> in Embryonated Chicken Eggs<sup>1</sup></b>  | Report results  | 2.8 X 10 <sup>7</sup> CEID <sub>50</sub> /mL  |
| <b>RT-PCR Assay of Extracted RNA<sup>6</sup></b>   | ~ 470 bp amplicon   | ~ 470 bp amplicon   |
| <b>Sterility (21-day incubation)</b><br>Harpo's HTYE broth <sup>7</sup> , 37°C and 26°C, aerobic<br>Trypticase soy broth, 37°C and 26°C, aerobic<br>Sabouraud broth, 37°C and 26°C, aerobic<br>Sheep blood agar, 37°C, aerobic<br>Sheep blood agar, 37°C, anaerobic<br>Thioglycollate broth, 37°C, anaerobic<br>DMEM with 10% FBS, 37°C and 5% CO <sub>2</sub> | No growth<br>No growth<br>No growth<br>No growth<br>No growth<br>No growth<br>No growth | No growth<br>No growth<br>No growth<br>No growth<br>No growth<br>No growth<br>No growth |
| <b>Mycoplasma Contamination</b><br>Agar and broth culture (14-day incubation at 37°C)<br>DNA detection by PCR of extracted Test Article nucleic acid   | None detected<br>None detected  | None detected<br>None detected  |

<sup>1</sup>10 to 11-day-old SPF Fertile Chicken Eggs acquired from B&E Eggs, York Springs, Pennsylvania

<sup>2</sup>Derived from NIAID Catalog No. V-331-0E4090

<sup>3</sup>Grown in the allantoic cavity of embryonated chicken eggs<sup>1</sup> for 2 days at 37°C in a humidified chamber

<sup>4</sup>The Chicken Embryo Infectious Dose 50% (CEID<sub>50</sub>) is the dilution of virus that under the conditions of the assay can be expected to infect 50% of the inoculated embryonated chicken eggs, 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 CEID<sub>50</sub> provides a measure of the infectious titer (or infectivity) of a virus preparation.

<sup>5</sup>2 days at 35°C in a humidified chamber

<sup>6</sup>The primers are described in Lee, M.-S., et al. "Identification and Subtyping of Avian Influenza Viruses by Reverse Transcription-PCR." *J. Virol. Methods* 97 (2001): 13-22. PubMed: 11483213.

<sup>7</sup>Atlas, Ronald M. *Handbook of Microbiological Media*. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.

**Date:** 05 JUN 2009

**Signature:** Signature on File

**Title:** Technical Manager, BEI Authentication or designee

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