

Certificate of Analysis for NR-13659

Influenza A Virus, A/California/04/2009 (H1N1)pdm09, Cell Isolate (Produced in Eggs)

Catalog No. NR-13659

Product Description: Pooled allantoic fluid from specific pathogen free (SPF) embryonated chicken eggs¹ infected with influenza A virus, A/California/04/2009 (H1N1)pdm09.

Passage History: C2/E1 (CDC/BEI); C# = Number passages in MDCK cells, E# = Number of passages in embryonated chicken eggs

Lot²: 58632566 Manufacturing Date: 02MAY2009

TEST	SPECIFICATIONS	RESULTS
Identification by Hemagglutinin Gene Sequencing (389 nt)	Homologous to A/California/04/2009 (H1N1)pdm09 (GenBank: GQ117044)	Homologous to A/California/04/2009 (H1N1)pdm09 (GenBank: GQ117044)
Titer by CEID₅₀ Assay³,⁴ in Embryonated Chicken Eggs¹	Report results	8.9 × 10 ⁶ CEID ₅₀ per mL
Sterility (21-day incubation) Harpo's HTYE broth ⁵ , 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 Brucella agar, 37°C, anaerobic Thioglycollate broth, 37°C, anaerobic DMEM with 10% FBS, 37°C and 5% CO ₂	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

¹10- to 11-day-old SPF Embryonated Chicken Eggs acquired from B&E Eggs, York Springs, Pennsylvania

Date: 04 DEC 2014

Signature:

Title:

Technical Manager, BEI Authentication or designee

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²Grown in the allantoic cavity of embryonated chicken eggs¹ for 2 to 3 days at 33.5°C in a humidified chamber without CO₂

³The Chicken Embryo Infectious Dose 50% (CEID₅₀) 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% (LD₅₀) is expected to kill half of the animals exposed. A reciprocal of the dilution required to yield the CEID₅₀ provides a measure of the infectious titer (or infectivity) of a virus preparation.

⁴2 days at 35°C in a humidified chamber

⁵Atlas, Ronald M. <u>Handbook of Microbiological Media</u>. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.