

Certificate of Analysis for NR-21659

Influenza A Virus, A/turkey/Wisconsin/1/1966 (H9N2)

Catalog No. NR-21659

Product Description: Pooled allantoic fluid from specific-pathogen free (SPF) embryonated chicken eggs¹ infected with influenza A virus, A/turkey/Wisconsin/1/1966 (H9N2).

Lot²: 59772075 Manufacturing Date: 17FEB2011

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity Using Embryonated Chicken Eggs ¹ Hemagglutination activity using allantoic fluid from infected eggs and 0.5% chicken red blood cells	Positive	Positive
Sequencing of Species- and Strain-Specific Regions Hemagglutinin (~ 460 nucleotides) Matrix gene (~ 915 nucleotides)	Consistent with A/turkey/Wisconsin/1/1966 (H9N2) (GenBank: CY087824) Consistent with A/turkey/Wisconsin/1/1966 (H9N2) (GenBank: CY087825)	99% identity with A/turkey/Wisconsin/1/1966 (H9N2) (GenBank: CY087824) 99% identity with A/turkey/Wisconsin/1/1966 (H9N2) (GenBank: CY087825)
Titer by CEID₅₀ Assay³,⁴ in Embryonated Chicken Eggs¹	Report results	1.6 X 10 ⁹ CEID ₅₀ /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 Blood 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 ²Grown in the allantoic cavity of embryonated chicken eggs ¹ for 2 days at 35°C in a humidified chamber

Date: 27 JUL 2011 Signature: Doothy C. Young

Title: Technical Manager, BEI Authentication or designee

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

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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.