

Certificate of Analysis for NR-3647

Kilbourne F50: A/Ulan Ude/01/2000 (HA) x A/Puerto Rico/8/1934 (NA) (H3N1)

Catalog No. NR-3647

Product Description: Pooled allantoic fluid from specific pathogen free (SPF) embryonated chicken eggs¹ infected with reassortant influenza A virus, A/Ulan Ude/01/2000 (HA) x A/Puerto Rico/8/1934 (NA) (H3N1)

Lot^{2,3}: 61982338 Manufacturing Date: 12SEP2013

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 Hemagglutinin, Matrix, and Neuraminidase Coding Regions		
Hemagglutinin (694 nucleotides)	Consistent with A/Ulan Ude/01/2000 (H3N2)	99% identity with A/Ulan Ude/01/2000 (H3N2) (GenBank: CY121408)
Matrix (927 nucleotides)	Consistent with A/Puerto Rico/8/1934 (H1N1)	100% identity with A/Puerto Rico/8/1934 (H1N1) (GenBank: CY033578)
Neuraminidase (619 nucleotides)	Consistent with A/Puerto Rico/8/1934 (H1N1)	99% identity with A/Puerto Rico/8/1934 (H1N1) (GenBank: CY033579)
Titer by CEID ₅₀ Assay ^{4,5} in Embryonated Chicken Eggs ¹	Report results	1.6 × 10 ⁸ CEID ₅₀ per mL
Sterility (21-day incubation)		
Harpo's HTYE broth ⁶ , 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
Blood agar, 37°C, aerobic	No growth	No growth
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 ₂	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

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

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²Derived from NIAID Catalog No. V-331-0E5535

³Grown in the allantoic cavity of embryonated chicken eggs¹ for 2 days at 35°C in a humidified chamber

⁴The 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.

⁵2 days at 35°C in a humidified chamber

⁶Atlas, 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: 15 NOV 2013 Signature: Minhael Q. Cymple

Title: Technical Manager, BEI Authentication or designee

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