

Certificate of Analysis for NR-3575

Kilbourne F118: A/Port Chalmers/1/1973 (HA, NA) x A/Puerto Rico/8/1934 (H3N2), Reassortant X-41

Catalog No. NR-3575

Product Description: Pooled allantoic fluid from specific-pathogen free (SPF) embryonated chicken eggs¹ infected with reassortant influenza A virus, A/Port Chalmers/1/1973 (HA, NA) x A/Puerto Rico/8/1934 (H3N2) (Kilbourne F118; X-41).

Lot^{2,3}: 59597984 Manufacturing Date: 07JAN2011

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 Strain- and Species-Specific Regions Hemagglutinin (~ 684 nucleotides) Matrix gene (~ 919 nucleotides)	Consistent with A/Port Chalmers/1/1973 (GenBank: CY009348) Consistent with A/Puerto Rico/8/1934 (GenBank: CY033578)	100% identity with A/Port Chalmers/1/1973 (GenBank: CY009348) 99% identity with A/Puerto Rico/8/1934 (GenBank: CY033578)
Titer by CEID ₅₀ Assay ^{4,5} in Embryonated Chicken Eggs ¹	Report results	2.8 X 10 ⁸ CEID ₅₀ /mL
Sterility (21-day incubation) Harpo's HTYE broth ⁶ , 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 Fertile Chicken Eggs acquired from B&E Eggs, York Springs, Pennsylvania

Date: 31 MAR 2011

Signature:

Title: Technical Manager, BEI Authentication or designee

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www.beiresources.org

E-mail: contact@beiresources.org

Tel: 800-359-7370 Fax: 703-365-2898

²Derived from NIAID Catalog No. V-331-0E5382

³Grown in the allantoic cavity of embryonated chicken eggs¹ for 48 hours 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. Handbook of Microbiological Media. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.