SUPPORTING INFECTIOUS DISEASE RESEARCH

Kilbourne F36: A/swine/Cambridge/1947 (HA) x A/equine/Prague/1/1956 (NA) x A/Puerto Rico/8/1934 (H1N7)

Catalog No. NR-3521

Product Description: Pooled allantoic fluid from specific-pathogen free (SPF) embryonated chicken eggs¹ infected with (Kilbourne F36) influenza A virus, A/swine/Cambridge/1947 (HA) x A/equine/Prague/1/1956 (NA) x A/Puerto Rico/8/1934 (H1N7).

Lot^{2,3}: 59735813

Manufacturing Date: 04FEB2011

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 gene (~ 438 nucleotides)	Consistent with H1N1 influenza A virus ⁴	Consistent with H1N1 influenza A virus ⁴
Matrix gene (~ 932 nucleotides)	Consistent with A/Puerto Rico/8/1934 (H1N1) (GenBank: CY045765)	99% identity with A/Puerto Rico/8/1934 (H1N1) (GenBank: CY045765)
Titer by CEID ₅₀ Assay ^{5,6} in Embryonated Chicken Eggs ¹	Report results	2.8 X 10 ⁸ CEID ₅₀ /mL
Sterility (21-day incubation)		
Harpo's HTYE broth ⁷ , 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 to 11-day-old SPF Embryonated Chicken Eggs acquired from B&E Eggs, York Springs, Pennsylvania

²Derived from NIAID Catalog No. V-331-0E5033

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

⁴The H1 HA sequence of influenza A/swine/Cambridge/1947 is not in the NCBI database; the HA sequence obtained for NR-3521 is consistent with those of H1N1 influenza viruses isolated in Europe and North America in the late 1940s and early 1950s.

⁵The 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.
⁶2 days at 35°C in a humidified chamber

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

Date: 08 JUN 2011

) crothy C. Young Signature:

Title:

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