

Kilbourne F25: A/turkey/Kansas/4880/80 (H1N1) Mutant, Low (L) Yield (Animal Isolate)

Catalog No. NR-3469

Product Description: Pooled allantoic fluid from specific-pathogen free (SPF) embryonated chicken eggs¹ infected with a low (L) yield mutant (Kilbourne F25) of influenza A virus, A/turkey/Kansas/4880/80 (H1N1).

Lot^{2,3}: 58411917

Manufacturing Date: 21NOV2008

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity Using Embryonated Chicken Eggs¹ Hemagglutination activity using allantoic fluid from infected eggs and 0.5% chicken red blood cells	Positive	Positive
Sequencing of Species-Specific Region (~ 450 nucleotides)	Influenza A virus	Influenza A virus
Titer by CEID₅₀ Assay^{4,5} in Embryonated Chicken Eggs¹	Report results	2.8 X 10 ⁸ CEID ₅₀ /mL
RT-PCR Assay of Extracted RNA⁶ Hemagglutinin gene Matrix gene	~ 470 bp amplicon ~ 1030 bp amplicon	~ 470 bp amplicon ~ 1030 bp amplicon
Sterility (21-day incubation) Harpo's HTYE broth ⁷ , 37°C and 26°C, aerobic Trypticase soy broth, 37°C and 26°C, aerobic Sabouraud broth, 37°C and 26°C, aerobic Sheep blood agar, 37°C, aerobic Sheep blood agar, 37°C, anaerobic Thioglycollate broth, 37°C, anaerobic DMEM with 10% FBS, 37°C and 5% CO ₂	No growth No growth No growth No growth No growth No growth No growth	No growth No growth No growth No growth No growth 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

²Derived from NIAID Catalog No. V-331-0E4149

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

⁶The H1 gene primers are described in Lee, M.-S., et al. "Identification and Subtyping of Avian Influenza Viruses by Reverse Transcription-PCR." *J. Virol. Methods* 97 (2001): 13-22. PubMed: 11483213; the matrix gene primers are described in Hoffmann, E., et al. "Universal Primer Set for the Full-Length Amplification of All Influenza A Viruses." *Arch. Virol.* 146 (2001): 2275-2289. PubMed: 11811679.

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

Date: 13 FEB 2009

Signature: Signature on File

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.

ATCC® is a trademark of the American Type Culture Collection.

You are authorized to use this product for research use only. It is not intended for human use.



Biodefense and Emerging Infections Research Resources Repository

P.O. Box 4137

Manassas, VA 20108-4137 USA

www.beiresources.org

800-359-7370

Fax: 703-365-2898

E-mail: contact@beiresources.org