

## **Certificate of Analysis for NR-3510**

Kilbourne F77: A/Beijing/32/1992 (HA) x A/Beijing/353/1989 (NA) x A/Puerto Rico/8/1934 (H3N2), Reassortant X-115

Catalog No. NR-3510

**Product Description:** Pooled allantoic fluid from specific pathogen free (SPF) embryonated chicken eggs<sup>1</sup> infected with reassortant influenza A virus, A/Beijing/32/1992 (HA) x A/Beijing/353/1989 (NA) x A/Puerto Rico/8/1934 (H3N2)

Lot<sup>2,3</sup>: 60341178 Manufacturing Date: 03NOV2011

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity Using Embryonated Chicken Eggs <sup>1</sup> 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 (628 nucleotides)  Matrix (914 nucleotides)	Consistent with A/Beijing/32/1992 (H3N2)  Consistent with A/Puerto Rico/8/1934 (H1N1)	99% identity with A/Beijing/32/1992 (H3N2) (GenBank: CY033606) 100% identity with A/Puerto Rico/8/1934 (H1N1)
Neuraminidase (379 nucleotides)	Consistent with A/Beijing/353/1989 (H3N2)	(GenBank: CY033578) 99% identity with A/Beijing/353/1989 (H3N2) (GenBank: DQ508835)
Titer by CEID <sub>50</sub> Assay <sup>4,5</sup> in Embryonated Chicken Eggs <sup>1</sup>	Report results	1.6 × 10 <sup>9</sup> CEID <sub>50</sub> per mL
Sterility (21-day incubation)  Harpo's HTYE broth <sup>6</sup> , 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 <sub>2</sub>	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

<sup>&</sup>lt;sup>1</sup>10-day-old SPF Embryonated Chicken Eggs acquired from B&E Eggs, York Springs, Pennsylvania

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<sup>&</sup>lt;sup>2</sup>Derived from NIAID Catalog No. V-331-0E4982

<sup>&</sup>lt;sup>3</sup>Grown in the allantoic cavity of embryonated chicken eggs<sup>1</sup> for 2 days at 35°C in a humidified chamber

<sup>&</sup>lt;sup>4</sup>The 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.

<sup>&</sup>lt;sup>5</sup>2 days at 35°C in a humidified chamber

<sup>&</sup>lt;sup>6</sup>Atlas, 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: 01 JUN 2012

Signature: Dorothy C. Young

Title:

Technical Manager, BEI Authentication or designee

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