

Certificate of Analysis for NR-3685

Kilbourne F125: A/Victoria/3/1975 (HA) x A/equine/Prague/1/1956 (NA) x A/Puerto Rico/8/1934 (H3N7), Reassortant X-48R

Catalog No. NR-3685

Product Description: Cell lysate and supernatant from Madin-Darby Canine Kidney (MDCK) cells¹ infected with reassortant influenza A virus, A/Victoria/3/1975 (HA) x A/equine/Prague/1/1956 (NA) x A/Puerto Rico/8/1934 (H3N7)

Lot^{2,3}: 63511774 Manufacturing Date: 13JUL2015

| TEST | SPECIFICATIONS | RESULTS |
|--|---|---|
| Identification by Infectivity in MDCK cells 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 (692 nucleotides) | Consistent with A/Victoria/3/1975 (H3N2) | 100% identity with A/Victoria/3/1975 (H3N2) (GenBank: CY121197) |
| Matrix (864 nucleotides) | Consistent with A/equine/Prague/1/1956 (H7N7) | 99% identity with A/equine/Prague/1/1956 (H7N7) (GenBank: CY096908) |
| Neuraminidase (489 nucleotides) | Consistent with A/equine/Prague/1/1956 (H7N7) | 100% identity with A/equine/Prague/1/1956 (H7N7) (GenBank: CY096909) |
| Titer by TCID ₅₀ Assay ^{4,5} in MDCK Cells ¹ | Report results | 5.0 × 10 ⁵ TCID 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 |

¹MDCK; ATCC[®] CCL-34™

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²Derived from NIAID Catalog No. V-331-0TC445

³Grown in Eagle's Minimum Essential Medium (ATCC[®] 30-2003™) supplemented with 0.125% BSA (Invitrogen™ 15260-037) and 2 μg per mL L-1-tosylamido-2-phenylethyl chloromethyl ketone (TPCK)-treated trypsin (Sigma T1426) for 3 days at 37°C and 5% CO₂

⁴The Tissue Culture Infectious Dose 50% (TCID₅₀) is the dilution of virus that under the conditions of the assay can be expected to infect 50% of the infected cells, 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 TCID₅₀ provides a measure of the infectious titer (or infectivity) of a virus preparation.

⁵10 days at 37°C and 5% CO₂

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: 09 DEC 2015

Signature:

BEI Resources Authentication

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