SUPPORTING INFECTIOUS DISEASE RESEARCH

West Nile Virus, 385-99

Catalog No. NR-158

(Derived from ATCC[®] VR-1507[™])

Product Description: Cell lysate and supernatant from *Cercopithecus aethiops* kidney epithelial cells (Vero)¹ infected with West Nile Virus (WNV), 385-99

Lot²: 4594475

Manufacturing Date: 26JUN2008

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity Using Vero Cells ¹	Report results	Cell rounding and sloughing
Sequencing of Species-Specific Region (819 nucleotides)	Consistent with WNV, 385-99	100% identity with WNV, 385-99 (GenBank: AY842931)
Titer by TCID ₅₀ Assay ^{3,4} in Vero Cells ¹	Report results	$1.6 \times 10^7 \text{ TCID}_{50} \text{ per mL}$
Sterility (21-day incubation) Harpo's HTYE broth ⁵ , 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

¹Vero cells: ATCC[®] CCL-81[™]

²NR-158 was grown from ATCC[®] VR-1507™ (Lot: 3398666) in Minimum Essential Medium containing Earle's salts and non-essential amino acids (Invitrogen™ 10370-021) supplemented with 2% irradiated fetal bovine serum (Lonza 14-471F), 2 mM L-glutamine (Invitrogen™ 25030-081), and 1 mM sodium pyruvate (Invitrogen™ 11360-070) for 6 days at 37°C and 5% CO₂.

³The Tissue Culture Infectious Dose 50% (TCID₅₀) endpoint is the 50% infectious endpoint in cell culture. The TCID₅₀ is the dilution of virus that under the conditions of the assay can be expected to infect 50% of the culture vessels inoculated, 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 titer (or infectivity) of a virus preparation.

⁴5 days at 37°C and 5% CO₂

⁵Atlas, Ronald M. <u>Handbook of Microbiological Media</u>. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.

Date: 02 APR 2015

Signature: Michael

Title:

Technical Manager, BEI Authentication or designee

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