b|e|i RESOURCES

SUPPORTING INFECTIOUS DISEASE RESEARCH

Certificate of Analysis for NR-51182

Powassan Virus, M11665

Catalog No. NR-51182

Product Description:

Powassan virus (POWV), M11665 was isolated from a tick *(Ixodes cookei)* in Laurier Township, Ontario, Canada on June 2, 1965. NR-51182 lot 70015198 was produced by infecting *Cercopithecus aethiops* kidney epithelial cells (Vero; ATCC® CCL-81™) with the deposited material and incubating in Eagle's Minimum Essential Medium (ATCC® 30-2003) supplemented with 2% fetal bovine serum (ATCC® 30-2020) for 6 days at 37°C with 5% CO_2.

Passage History:

X(1)SM(1)/V(3) (Prior to deposit at BEI Resources/BEI Resources); X = Unknown; SM = Suckling mice; V = Vero cells

Lot: 70015198 **Manufacturing Date: 16MAY2019**

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in Vero Cells	Cell rounding and detachment	Cell rounding and detachment
Sequencing of Species-Specific Region (~ 840 nucleotides)	≥ 98% identity with POWV	≥ 98% identity with POWV[1]
Titer by $TCID_{50}$ Assay in Vero Cells by Cytopathic Effect[2] (8 days at 37°C and 5% CO_2)	Report results	2.8×10^8 $TCID_{50}$ per mL
Amplification of POWV Sequence by RT-PCR	~ 1480 base pair amplicon	~ 1480 base pair amplicon
Sterility (21-day incubation)		
Harpo's HTYE broth, 37°C and 26°C, aerobic[3]	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
Sheep blood agar, 37°C, aerobic	No growth	No growth
Sheep 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

[1]Sequence information for POWV, M11665 is not available in the NCBI database; nucleotide sequence obtained for NR-51182 lot 70015198 is ≥ 98% identical to numerous POWV strains.

[2]The Tissue Culture Infectious Dose 50% ($TCID_{50}$) endpoint is the 50% infectious endpoint in cell culture. The $TCID_{50}$ 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% (LD_{50}) is expected to kill half of the animals exposed. A reciprocal of the dilution required to yield the $TCID_{50}$ provides a measure of the titer (or infectivity) of a virus preparation.

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

/Heather Couch/
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Program Manager or designee, ATCC Federal Solutions

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