b|e|i RESOURCES

SUPPORTING INFECTIOUS DISEASE RESEARCH

Certificate of Analysis for NR-49758

Encephalomyocarditis Virus, TX 1579 (Tapir)

Catalog No. NR-49758

Product Description: Cell lysate and supernatant from *Cercopithecus aethiops* kidney epithelial (Vero) cells[1] infected with tissue culture adapted encephalomyocarditis virus (EMCV), TX 1579 (Tapir).

Passage History: V2/V6 (Prior to deposit at BEI Resources/BEI Resources); V# = Number of passages in Vero cells[2]

Lot[3]: 63732640 **Manufacturing Date: 10NOV2016**

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in Vero Cells[2]	Report results	Cell rounding and detachment
Sequencing of EMC Virus-Specific Sequence (817 nucleotides)	Consistent with EMCV	Consistent with EMCV[4]
Titer by $TCID_{50}$ Assay in Vero Cells[5,6]	Report results	8.9×10^7 $TCID_{50}$ per mL
Sterility (21-day incubation)		
Harpo's HTYE broth[7], 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
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 Test Article nucleic acid	None detected	None detected

[1]Removal of contaminating mycoplasma required six passages at BEI Resources.
[2]Vero cells: ATCC® CCL-81™
[3]Grown in Eagle's Minimum Essential Medium containing Earle's Balanced Salt Solution, non-essential amino acids, 2 mM L-glutamine, 1 mM sodium pyruvate and 1.5 g/L of sodium bicarbonate (ATCC® 30-2003) supplemented with 2% fetal bovine serum (ATCC® 30-2020) for 2 days at 37°C with 5% CO_2.
[4]Sequence information for EMCV, TX 1579 (Tapir) is not available in the NCBI database at this time; nucleotide sequence obtained for NR-49758, Lot No. 63732640 is ~ 85% identical to six EMCV genome sequences; whole genome sequencing of the deposited material indicated that this strain is most closely related to the EMCV diabetogenic variant (GenBank: M37588), with ~ 83% identity over the full length of the sequence.
[5]The Tissue Culture Infectious Dose 50% ($TCID_{50}$) 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.
[6]7 days at 37°C with 5% CO_2
[7]Atlas, Ronald M. Handbook of Microbiological Media. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.

Date: 15 MAR 2017 **Signature:**

BEI Resources Authentication

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