b|e|i RESOURCES

SUPPORTING INFECTIOUS DISEASE RESEARCH

Certificate of Analysis for NR-49912

Una Virus, MAC 150

Catalog No. NR-49912

Product Description: Cell lysate and supernatant from *Cercopithecus aethiops* kidney epithelial cells (Vero)[1] infected with Una virus, MAC 150

Passage History: V1/V2 (Prior to deposit at BEI Resources/BEI Resources); X = unknown, V# = Number of passages in Vero cells[2]

Lot[3]: 63856768 **Manufacturing Date: 04MAR2016**

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in Vero Cells[1]	Cell rounding and detachment	Cell rounding and detachment
Whole Genome Sequencing (11901 nucleotides)	Consistent with Una virus	Consistent with Una virus[4]
Titer by $TCID_{50}$ Assay[5,6] in Vero Cells[1]	Report results	2.8×10^8 $TCID_{50}$ 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 extracted Test Article nucleic acid	None detected	None detected

[1]Vero cells: ATCC® CCL-81™

[2]The first virus passage at BEI Resources was performed by lipofectamine-mediated transfection of extracted viral nucleic acid in order to remove contaminating mycoplasma.

[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 1 day at 37°C with 5% CO_2.

[4]The complete genomic sequence obtained for NR-49912 at BEI Resources has >99% identity with the limited sequence available for Una virus, MAC 150 (GenBank: DQ487375), and is also highly similar to other Una virus sequences in the NCBI database.

[5]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.

[6]4 days at 37°C and 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: 22 NOV 2016 **Signature:** Michael R. Gyrla

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