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SUPPORTING INFECTIOUS DISEASE RESEARCH

Certificate of Analysis for NR-55

Vaccinia Virus, Western Reserve (NIAID, Tissue Culture Adapted)

Catalog No. NR-55

(Derived from ATCC® VR-1354™)

Product Description:

Vaccinia virus (VACV), Western Reserve (WR) (NIAID, tissue culture adapted) was derived from the original New York City Board of Health (NYCBH) strain by intracerebral passages in mice followed by tissue culture adaptation. NR-55 lot 70055086 was produced by infecting *Cercopithecus aethiops* kidney cells (Vero; ATCC® CCL-81™) with seed material (BEI Resources lot 3580751) and incubating in Eagle's Minimum Essential Medium (ATCC® 30-2003™) supplemented with 2% fetal bovine serum (ATCC® 30-2020™) for 3 days at 37°C with 5% CO_2.

Passage History:

Unk(rabbits?)(unk), M(24), HeLa(unk), BSC-1(4, including plaque purification), HeLa(1)/V(3) (Prior to deposit at BEI Resources/ BEI Resources); Unk = Unknown; M = Mouse brain(i.c.); HeLa = *Homo sapiens* adenocarcinoma epithelial cells; BSC-1 = *Cercopithecus aethiops* kidney cells; V = *Cercopithecus aethiops* kidney cells (Vero) cells

Lot: 70055086 **Manufacturing Date: 16SEP2022**

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in Vero Cells	Cell rounding and detachment	Cell rounding and detachment
Sequencing of Species-Specific Region (~ 1010 nucleotides)	≥ 98% identity with VACV, WR (GenBank: AY243312)	99.1% identity with VACV, WR (GenBank: AY243312)
Titer by $TCID_{50}$ Assay in Vero Cells by Cytopathic Effect[1] (4 days at 37°C with 5% CO_2)	Report results	1.6×10^7 $TCID_{50}$ per mL
Sterility (21-day incubation)		
Harpo's HTYE broth, 37°C and 26°C, aerobic[2]	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, aerobic	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]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.

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

/Sonia Bjorum Brower/
Sonia Bjorum Brower 05 DEC 2022
Technical Manager or designee, ATCC Federal Solutions

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

Support Provided by NIAID

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