

Spondweni Virus, SAAr 94

Catalog No. NR-51973

Product Description:

Spondweni virus (SPONV), SAAr 94 was isolated from *Mansonia uniformis* mosquitoes in Lake Simbu, Natal, South Africa in 1955. NR-51973 lot 70032791 was produced by infecting *Cercopithecus aethiops* kidney epithelial cells (Vero E6; ATCC® CRL-1586™) and incubating in Eagle's Minimum Essential Medium (ATCC® 30-2003™) supplemented with 2% fetal bovine serum (ATCC® 30-2020™) for 12 days at 37°C with 5% CO₂.

Passage History:

X(5)V(1)/VE6(2) (Prior to deposit at BEI Resources/BEI Resources); X = Unknown; V = Vero cells; VE6 = Vero E6 cells

Lot: 70032791

Manufacturing Date: 02MAR2020

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in Vero E6 Cells	Cell rounding and detachment	Cell rounding and detachment
Identification by Indirect Fluorescent Antibody (IFA) Assay ¹	Fluorescence observed	Fluorescence observed
Sequencing of Species-Specific Region (~ 870 nucleotides)	≥ 98% identity with SPONV, AR 94 polyprotein gene (GenBank: KX227370.1)	100% identity with SPONV, AR 94 polyprotein gene (GenBank: KX227370.1)
Titer by TCID ₅₀ Assay in Vero E6 Cells by Cytopathic Effect and IFA ^{1,2} (8 days at 37°C with 5% CO ₂)	Report results	2.8 × 10 ⁴ TCID ₅₀ per mL
Amplification of SPONV Sequence by RT-PCR	~ 910 base pair amplicon	~ 910 base pair amplicon
Sterility (21-day incubation) Harpo's HTYE broth, 37°C and 26°C, aerobic ³ 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, aerobic	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

¹Using Monoclonal Anti-Flavivirus Group Antigen, Clone D1-4G2-4-15 (BEI Resources NR-50327) and Light Diagnostics™ Goat Anti-Mouse IgG FITC Reagent (Millipore® 5008)

²The 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.

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

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