

Zika Virus, DAK AR 41524

Catalog No. NR-51813

Product Description:

Zika virus (ZIKV), DAK AR 41524 was isolated from a mosquito (*Aedes africanus*) in Kédougou, Senegal, on November 17, 1984.

Passage History:

A1C1V2C1/C2 (Prior to deposit at BEI Resources/BEI Resources); A = *Aedes pseudoscutellaris* (AP61) cells; V = Vero cells; C = *Aedes albopictus* (C6/36) cells¹

Lot: 70029217²

Manufacturing Date: 10OCT2019

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in C6/36 cells ¹	Report results	Cell rounding and detachment
Identification by Indirect Fluorescent Antibody (IFA) Assay ³	Fluorescence observed	Fluorescence observed
Sequencing of Species-Specific Region (~ 850 nucleotides)	≥ 98% identity with ZIKV, DAK AR 41524 (GenBank: KX601166.2)	100% identity with ZIKV, DAK AR 41524 (GenBank: KX601166.2)
Titer by TCID ₅₀ Assay in C6/36 cells with IFA Readout ^{1,3,4,5}	Report results	2.8 × 10 ⁸ TCID ₅₀ per mL
Genome Copy Number by ddPCR ⁶	Report results	7.3 × 10 ⁷ genome copies per mL
Amplification of ZIKV Sequence by RT-PCR	~ 1080 base pair amplicon	~ 1080 base pair amplicon
Endotoxin Content (<i>Limulus</i> Amoebocyte Lysate Assay)	Report results	2.5 EU per mL
Sterility (21-day incubation) Harpo's HTYE broth, 37°C and 26°C, aerobic ⁷ Trypticase Soy broth, 37°C and 26°C, aerobic Sabouraud broth, 37°C and 26°C, aerobic Blood agar, 37°C, aerobic Blood agar, 37°C, anaerobic Thioglycollate broth, 37°C, anaerobic DMEM with 10% FBS, 37°C and 5% CO ₂	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

¹*Aedes albopictus* clone C6/36 cells (ATCC® CRL-1660™)

²Grown 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 9 days at 28°C with 5% CO₂.

³Using Anti-Zika NS1 Monoclonal Antibody (Aalto Bio Reagents AZ 1225)

⁴The 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.

⁵Assay plates were incubated 9 days at 28°C and 5% CO₂

⁶ddPCR data was obtained post-vial from 9 replicates on the BioRad QX200 Droplet Digital PCR (ddPCR™) System.

⁷Atlas, Ronald M. *Handbook of Microbiological Media*. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.

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04 MAR 2020

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