

**Chikungunya Virus, PH/HU/86/NH/002**

**Catalog No. NR-49904**

**Product Description:** Cell lysate and supernatant from *Cercopithecus aethiops* kidney epithelial cells<sup>1</sup> infected with chikungunya virus (CHIKV), PH/HU/86/NH/002

**Passage History:** V2/V3 (Prior to deposit at BEI Resources/BEI Resources); V# = Number of passages in Vero cells<sup>2</sup>

**Lot<sup>3</sup>: 64108457**

**Manufacturing Date: 20MAY2016**

TEST	SPECIFICATIONS	RESULTS
<b>Infectivity in Vero E6 Cells</b>	Report results	Cell rounding and detachment
<b>Sequencing of Species-Specific Region (937 nucleotides)</b>	Consistent with CHIKV	Consistent with CHIKV <sup>4</sup>
<b>Titer by TCID<sub>50</sub> Assay<sup>5,6</sup> in Vero E6 Cells<sup>1</sup></b>	Report results	8.9 × 10 <sup>6</sup> TCID <sub>50</sub> per mL
<b>Amplification of CHIKV Sequence by RT-PCR</b>	~ 1070 bp amplicon	~ 1070 bp amplicon
<b>Sterility (21-day incubation)</b> Harpo's HTYE broth <sup>7</sup> , 37°C and 26°C, aerobic 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 and 5% CO <sub>2</sub>	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
<b>Mycoplasma Contamination</b> 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

<sup>1</sup>Vero 76, clone E6; ATCC® CRL-1586™

<sup>2</sup>The second virus passage at BEI Resources was performed by lipofectamine transfection of extracted viral nucleic acid in order to remove contaminating mycoplasma.

<sup>3</sup>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 3 days at 37°C with 5% CO<sub>2</sub>

<sup>4</sup>Sequence information for CHIKV, PH/HU/86/NH/002 is not available in the NCBI database; nucleotide sequence obtained for NR-49904, Lot No. 64108457 is ~ 99% identical to numerous CHIKV strains isolated in Southeast Asia.

<sup>5</sup>The Tissue Culture Infectious Dose 50% (TCID<sub>50</sub>) endpoint is the 50% infectious endpoint in cell culture. The TCID<sub>50</sub> 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<sub>50</sub>) is expected to kill half of the animals exposed. A reciprocal of the dilution required to yield the TCID<sub>50</sub> provides a measure of the titer (or infectivity) of a virus preparation.

<sup>6</sup>7 days at 37°C and 5% CO<sub>2</sub>

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

**Date:** 28 SEP 2016

**Signature:** 

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