

## *Rickettsia asiatica*, Strain IO-1

### Catalog No. NR-10405

(Derived from ATCC® VR-1593™)

We have been unsuccessful in our attempts to purify NR-10405 from contaminating *Mycoplasma orale*. Please determine whether or not this product is acceptable for your intended use.

**Product Description:** Cell lysate and supernatant from African green monkey kidney (Vero) cells<sup>1</sup> infected with *Rickettsia asiatica*, strain IO-1.

**Lot<sup>2</sup>: 58365931**

**Manufacturing Date: 20AUG2008**

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in Vero Cells <sup>1</sup>	Report results	Cell rounding and sloughing
Identification by Sequencing of Citrate Synthase Gene (~ 1020 bp)	Identical to GenBank AF394901 <i>Rickettsia asiatica</i>	Identical to GenBank AF394901 <i>Rickettsia asiatica</i>
Titer by TCID <sub>50</sub> Assay <sup>3,4</sup> in Vero Cells <sup>1</sup>	Report results	1.6 X 10 <sup>6</sup> TCID <sub>50</sub> /mL
PCR Amplification of Extracted DNA	~ 1154 bp amplicon	~ 1154 bp amplicon
<b>Sterility (21-day incubation)</b> Harpo's HTYE broth <sup>5</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 Brucella 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 (30-day incubation at 37°C) DNA detection by PCR of extracted Test Article nucleic acid	Report results Report results	<b>Growth</b> Contaminated with <i>Mycoplasma orale</i>

<sup>1</sup>Vero cells: ATCC® CCL-81™

<sup>2</sup>Grown in Minimum Essential Medium with Earle's salts (Invitrogen™ 10370-021) supplemented with 10% irradiated fetal bovine serum (Lonza 14-471), 2 mM L-glutamine (Invitrogen™ 25030-081) and 1 mM sodium pyruvate (Invitrogen™ 11360-070) for 13 days at 32°C and 5% CO<sub>2</sub>.

<sup>3</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>4</sup>12 days at 32°C and 5% CO<sub>2</sub> with media overlay

<sup>5</sup>Atlas, Ronald M. *Handbook of Microbiological Media*. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798

**Date:** 20 MAY 2009

**Signature:** Signature on File

**Title:** Technical Manager, BEI Authentication or designee

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