

# **Certificate of Analysis for NR-46445**

### Ehrlichia chaffeensis, Strain St. Vincent

### Catalog No. NR-46445

This reagent is the property of the U.S. Government.

### **Product Description:**

Ehrlichia chaffeensis (E. chaffeensis), strain St. Vincent was isolated in 1996 from the blood of a male patient in Georgia, USA, who was bitten by a tick and subsequently developed fatal human monocytic ehrlichiosis (HME). NR-46445 lot 70018415 was produced by infecting DH82 cells (ATCC® CRL-10389™) and incubating in Dulbecco's Modified Eagle's Medium containing 5% fetal bovine serum (ATCC® 30-2020™) and 2 mM L-glutamine for 12 days at 37°C with 5% CO₂. NR-46445 is provided in cell lysate and supernatant supplemented with 20% heat-inactivated fetal bovine serum and 10% DMSO.

Lot: 70018415 Manufacturing Date: 09APR2019

TEST	SPECIFICATIONS	RESULTS
Identification of Infectivity in DH82 Cells by Indirect Fluorescent Antibody (IFA) Assay <sup>1</sup>	Fluorescence observed	Fluorescence observed
Genotypic Analysis Sequencing of ECH_0849 gene (~ 920 base pairs)	≥ 98% identity with  E. chaffeensis, strain St. Vincent (GenBank: CP007478.1)	100% identity with  E. chaffeensis, strain St. Vincent (GenBank: CP007478.1)
Titer by TCID₅ Assay in DH82 Cells by IFA¹,²	Report results	1.58 × 10 <sup>5</sup> TCID <sub>50</sub> per mL in 19 days at 37°C with 5% CO <sub>2</sub>
Amplification of <i>E. chaffeensis</i> Sequence by PCR	~ 1000 base pair amplicon	~ 1000 base pair amplicon
Sterility (21-day incubation)		
Harpo's HTYE broth, 37°C and 26°C, aerobic <sup>3</sup>	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 and 5% CO <sub>2</sub>	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

<sup>&</sup>lt;sup>1</sup>Ehrlichia chaffeensis IFA IgG reagent kit (Fuller Laboratories ECHG-120)

## /Heather Couch/

Heather Couch 13 APR 2020

Program Manager or designee, ATCC Federal Solutions

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<sup>&</sup>lt;sup>2</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 organism 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 the organism preparation.

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