

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: Cell lysate and supernatant from *Canis familiaris* macrophage-monocyte cells¹ infected with *Ehrlichia chaffeensis* (*E. chaffeensis*), strain St. Vincent, containing 13% fetal bovine serum and 2% DMSO (final concentrations).

Lot²: 62795202 Manufacturing Date: 21FEB2015

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in DH82 Cells ¹	Report results	Cell rounding and granularity
Identification by Sequencing of Species-Specific Region (918 nucleotides)	Consistent with <i>E. chaffeensis</i> , strain St. Vincent	100% identity with <i>E.</i> chaffeensis, strain St. Vincent (GenBank: CP007478)
Titer by TCID ₅₀ Assay ^{3,4} in DH82 Cells ¹ by IFA ⁵	Report results	1.6 × 10 ³ TCID ₅₀ per mL
Sterility (21-day incubation) Harpo's HTYE broth ⁶ , 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 ₂	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

¹DH82 cells: ATCC[®] CRL-10389™

Signature:

Date: 06 APR 2016

BEI Resources Authentication

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

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²Grown in Dulbecco's Modified Eagle's Medium containing 4 mM L-glutamine, 4500 mg per L glucose, 1 mM sodium pyruvate, and 1500 mg per L sodium bicarbonate (ATCC® 30-2002), supplemented with cycloheximide (Sigma C 4859), 10% fetal bovine serum (ATCC® 30-2020), and an additional 2 mM L-glutamine (Corning® Mediatech 25-005-CL) for 7 days at 37°C with 5% CO₂

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3The 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.

⁴6 days at 37°C and 5% CO₂

⁵Using Ehrlichia chaffeensis IFA IgG reagent kit (Fuller Laboratories ECHG-120)

⁶Atlas, Ronald M. Handbook of Microbiological Media. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.