b|e|i RESOURCES
SUPPORTING INFECTIOUS DISEASE RESEARCH

Certificate of Analysis for NR-48595

Anaplasma phagocytophilum, Strain ApNYW

Catalog No. NR-48595

Product Description:

Anaplasma phagocytophilum (A. phagocytophilum), strain ApNYW was isolated from a human in New York.

Lot: 70026733[1,2] **Manufacturing Date: 29JUL2019**

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in HL-60 Cells by Indirect Fluorescent Antibody (IFA) Assay[1,3]	Fluorescence observed	Fluorescence observed
Genotypic Analysis		
Sequencing of 16S ribosomal RNA gene (~ 1280 base pairs)	≥ 99% identity with *A. phagocytophilum*, strain ApNYW (GenBank: LAOG01000008.1)	100% identity with *A. phagocytophilum*, strain ApNYW (GenBank: LAOG01000008.1)[4]
Titer by $TCID_{50}$ Assay in HL-60 Cells by IFA[1,3,5,6]	Report results	2.8×10^4 $TCID_{50}$ per mL
Sterility (21-day incubation)		
Harpo's HTYE broth[7], 37°C and 26°C, aerobic	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% CO2	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

[1]HL-60; ATCC® CCL-240™
[2]NR-48595 was produced by infecting HL-60 cells with the depositor's vial and incubating in RPMI-1640 medium containing 10% fetal bovine serum (ATCC® 30-2020) for 3 days at 37°C with 5% CO_2.
[3]Using *A. phagocytophilum* IFA IgG reagent kit (Fuller Laboratories EEG-120)
[4]Also consistent with *Ehrlichia equi* and "HGE agent", however, these species were recently recognized as *A. phagocytophilum*. For more information, please see Dumler, J. S., et al. "Reorganization of Genera in the Families *Rickettsiaceae* and *Anaplasmataceae* in the Order Rickettsiales: Unification of Some Species of *Ehrlichia* with *Anaplasma*, *Cowdria* with *Ehrlichia* and *Ehrlichia* with *Neorickettsia*, Descriptions of Six New Species Combinations and Designation of *Ehrlichia equi* and 'HGE agent' as Subjective Synonyms of *Ehrlichia phagocytophila*." Int. J. Syst. Evol. Microbiol. 51 (2001): 2145-2165. PubMed: 11760958.
[5]The Tissue Culture Infectious Dose 50% ($TCID_{50}$) endpoint is the 50% infectious endpoint in cell culture. The $TCID_{50}$ 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_{50}) is expected to kill half of the animals exposed. A reciprocal of the dilution required to yield the $TCID_{50}$ provides a measure of the titer (or infectivity) of the organism preparation.
[6]Assay plates were incubated 8 days at 37°C with 5% CO_2.
[7]Atlas, Ronald M. Handbook of Microbiological Media. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798

/Heather Couch/
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Program Manager or designee, ATCC Federal Solutions

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