

## Certificate of Analysis for NR-46495

## Naegleria fowleri, Strain CDC:V455

## Catalog No. NR-46495

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

**Product Description:** *Naegleria fowleri (N. fowleri)*, strain CDC:V455 is a clinical isolate collected in 2000 from a brain biopsy sample from a male in Nevada, USA.

Lot<sup>1</sup>: 70002521 Manufacturing Date: 10FEB2017

TEST	SPECIFICATIONS	RESULTS
Cellular Morphology <sup>2</sup>	Report results	Refractile
Genotyping³ Sequencing of Internal Transcribed Spacer 1 (ITS), 5.8S ribosomal RNA (rRNA) gene, ITS 2 (~ 580 base pairs)	Consistent with N. fowleri	Consistent with <i>N. fowleri</i> (genotype III) <sup>4,5</sup>
Functional Activity by PCR Amplification <sup>3,6</sup> ITS 1, 5.8S rRNA gene	~ 600 base pair amplicon	~ 600 base pair amplicon
Viable Cell Count by Hemacytometry <sup>3</sup>	> 10 <sup>6</sup> cells/mL	1.65 x 10 <sup>6</sup> cells/mL
Viability <sup>2,7</sup>	Growth	Growth
Sterility (21-day incubation) <sup>2</sup> Harpo's HTYE broth <sup>8</sup> , 37°C and 26°C, aerobic Trypticase soy broth, 37°C and 26°C, aerobic Sabouraud broth, 37°C and 26°C, aerobic DMEM with 10% FBS, 37°C, aerobic Sheep blood agar, 37°C, aerobic Sheep blood agar, 37°C, anaerobic Thioglycollate broth, 37°C, anaerobic	No growth	No growth

<sup>&</sup>lt;sup>1</sup>NR-46495 was produced by cultivation of the deposited material in modified PYNFH medium (ATCC® medium 1034) supplemented with 10% heat-inactivated fetal bovine serum for 3 days at 35°C in an aerobic atmosphere until peak density was reached.

Date: 21 JUN 2017

Signature:

**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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<sup>&</sup>lt;sup>2</sup>Testing completed on vialed, post-freeze material.

<sup>&</sup>lt;sup>3</sup>Testing completed on bulk material prior to vialing and freezing.

<sup>&</sup>lt;sup>4</sup>For genotyping details refer to Zhou, L., et al. "Genetic Variations in the Internal Transcribed Spacer and Mitochondrial Small Subunit rRNA Gene of *Naegleria* Spp." J. Eukaryot. Microbiol. 50 (2003): 522-526. PubMed: 14736150.

<sup>&</sup>lt;sup>5</sup>Also consistent with *Naegleria Iovaniensis* 

<sup>&</sup>lt;sup>6</sup>PCR amplification was performed using the NF-ITS-F1 and NT-ITS-F2 primer set as described in Zhou, L., et al. "Genetic Variations in the Internal Transcribed Spacer and Mitochondrial Small Subunit rRNA Gene of *Naegleria* Spp." <u>J. Eukaryot. Microbiol.</u> 50 (2003): 522-526. PubMed: 14736150.

<sup>&</sup>lt;sup>7</sup>Viable cells were observed after 1 day under cultivation conditions.

<sup>&</sup>lt;sup>8</sup>Atlas, Ronald M. <u>Handbook of Microbiological Media</u>. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.