

Certificate of Analysis for NR-46466

Acanthamoeba sp., Strain CDC:V313

Catalog No. NR-46466

Product Description: *Acanthamoeba* sp., strain CDC:V313 was isolated in 1994 from the nasal tissue of a male in Georgia, USA.

Lot¹: 62709495 Manufacturing Date: 23JUN2014

TEST	SPECIFICATIONS	RESULTS
Genotyping Sequencing of 18S ribosomal RNA gene (~ 420 base pairs)	Consistent with Acanthamoeba sp.	Consistent with <i>Acanthamoeba</i> sp. ² (Genotype T4)
Functional Activity by PCR Amplification ³ 18S ribosomal RNA gene (amplicon ASA.S1)	423 to 551 base pair amplicon	~ 450 base pair amplicon
Viable Cell Count by Hemacytometry (pre-freeze)	> 10 ⁶ cells/mL	7.8 x 10 ⁶ cells/mL
Viability (post-freeze) ⁴	Growth	Growth
Sterility (21-day incubation) Harpo's HTYE broth ⁵ , 37°C and 26°C, aerobic Trypticase soy broth, 37°C and 26°C, aerobic Sabouraud broth, 37°C and 26°C, aerobic Brain heart infusion, 37°C and 26°C, aerobic Sheep blood agar, 37°C, aerobic Sheep blood agar, 37°C, anaerobic Thioglycollate broth, 37°C, anaerobic	No growth	No growth

NR-46466 was produced by cultivation of the deposited material in PYG Medium (ATCC® medium 712) for 3 days at 25°C in an aerobic atmosphere and preserved.

Date: 23 OCT 2014

Signature:

Title: Technical Manager, BEI Authentication or designee

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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²> 99.8% identical to *Acanthamoeba* sp., strain CDC:V313 (GenBank: AY702997.1)

³PCR amplification was performed using the JDP1 and JDP2 primer set (JDP1: 5'-GGCCCAGATCGTTTACCGTGAA-3' and JDP2: 5'-TCTCACAAGCTGCTAGGGAGTCA-3') as described in Schroeder, J. M. et al. "Use of Subgenic 18S Ribosomal DNA PCR and Sequencing for Genus and Genotype Identification of Acanthamoebae from Humans with Keratitis and from Sewage Sludge." J. Clin. Microbiol. 39 (2001): 1903-1911. PubMed: 11326011.

⁴Viable cells were observed after 1 day under cultivation conditions.

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