

Certificate of Analysis for NR-20732

Toxoplasma gondii, Strain RUB

Catalog No. NR-20732

Product Description: *Toxoplasma gondii* RUB was isolated from the bronchoalveolar lavage of a human with toxoplasmosis from French Guyana in 1992. Strain RUB was deposited as a prototype strain for the type V haplogroup and is a reference strain for the *Toxoplasma gondii* Genome Project at the J. Craig Venter Institute's Genomic Sequencing Center for Infectious Diseases (GSCID).

Lot¹: 59758077 Manufacturing Date: 21MAR2011

TEST	SPECIFICATIONS	RESULTS
Genotyping Sequencing of uracil phosphoribosyltransferase (UPRT) intron 1 (~ 470 bp)	Consistent with <i>T. gondii</i> , haplotype V	Consistent with <i>T. gondii</i> , haplotype V ² (Figure 1)
Functional Activity by PCR Amplification ³ UPRT intron 1	~ 560 bp amplicon	~ 560 bp amplicon
Viable Cell Count by Hemacytometry (post-freeze)	> 10 ⁶ cells/mL	2.4 x 10 ⁷ 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
Mycoplasma Contamination DNA Detection by PCR	None detected	None detected

¹NR-20732 was produced by cultivation of the deposited material in human foreskin fibroblast cells (ATCC[®] CRL-1634™) with cell cultivation medium for parasites (ATCC medium 2222: adjusted to contain 10% heat-inactivated fetal bovine serum). The culture was propagated in 95% air, 5% CO₂ for 4 days at 37°C, until lysis of the host cell monolayer was reached.

Figure 1: Toxoplasma gondii, Strain RUB - UPRT Intron 1 Sequence

GTAATCCTTC	AACCGAAGTT	TGCTTTCCGT	GACTCTGCCT	CTTGGTTATA	CTGCGTGGCC	TTCCCGTCCT	GCCGCTCCCT	
TTCCTCCGCT	TGCTGTTTAA	ATGCTCGTCC	TCGTTTTCCT	TCCTGCCGCA	TCCCCGTATA	TTTTAAGGAG	AGGGAAACAG	
GCGTGAGTTG	GACGGAATGA	AAGTTCTCGG	CCTGTACGCC	GGTTGTCGCG	GTCGTCTGCA	GATTGCTTTT	TTCTTCGAAT	
CGGTGCTGTA	ACCCTCGCGA	AGAACGACGC	TGCAAACGAC	TTCTCGAACT	CTCAGTCGTG	TACTTTACGT	GCTTCCTTTC	
AGGGACCTCC	CCCCGCGTTA	CTCATTTGTA	TTCACAGCTA	CGAAGTGTCT	TGCAAGGTGG	ATTCGTGACA	GGCTCCATGT	
CTCACTCGGT	GCATTTTCGG	AAAAGTTCAT	TGTGAACGTT	GCCCTTGCGT	GTCATGACTT	TATCAG		

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²Haplotype identification is based on a sequence alignment of NR-20732, lot 59758077, to a reference sequence provided by the depositor. See figure 1 for sequence.

³Primer sequences and conditions for PCR are available upon request.

⁴Viable cells and signs of infection were seen after 13 days under cultivation conditions at 37°C.

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



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Date: 14 SEP 2015 Signature:

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