

**Toxoplasma gondii, Strain SF20**

**Catalog No. NR-49186**

**Product Description:** *Toxoplasma gondii* (*T. gondii*), strain SF20 is a recombinant F1 clone selected from progeny of a genetic cross between a sinfungin-resistant line of the highly virulent Type I GT-1 strain (GT1-SNF<sup>R</sup>) and a 5-fluoro-2'-deoxyuridine-resistant line of the non-virulent Type 2 ME49 strain (ME49 FUDR<sup>R</sup>).

**Lot<sup>1,2</sup>: 63833058**

**Manufacturing Date: 29OCT2015**

TEST	SPECIFICATIONS	RESULTS
<b>Cell Morphology</b>	Report results	Refractile and oval-shaped
<b>PCR Assay of Extracted DNA<sup>3,4</sup></b> AK56 locus	~ 520 base pair amplicon	~ 520 base pair amplicon
<b>Genotypic Analysis<sup>3,4</sup></b> Sequencing of AK56 locus (~ 480 base pairs) AK56 locus ( <i>MfeI</i> digestion)	Consistent with <i>T. gondii</i> Consistent with parental Type II strain	Consistent with <i>T. gondii</i> (Figure 1) Consistent with parental Type II strain
<b>Viable Cell Count by Hemacytometry (pre-freeze)</b>	> 10 <sup>6</sup> cells/mL	2.6 × 10 <sup>7</sup> cells/mL
<b>Viability (post-freeze)<sup>5</sup></b>	Viable parasites	Viable parasites
<b>Sterility (21-day incubation)</b> Harpo's HTYE broth <sup>6</sup> , 37°C and 26°C, aerobic Tryptic Soy broth, 37°C and 26°C, aerobic Sabouraud Dextrose 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 No growth No growth No growth No growth No growth	No growth No growth No growth No growth No growth No growth No growth
<b>Mycoplasma Contamination</b> DNA Detection by PCR	None detected	None detected

<sup>1</sup>NR-49186 was produced by cultivation of the deposited material in human foreskin fibroblast cells (ATCC<sup>®</sup> CRL-1634<sup>™</sup>) with cell cultivation medium for parasites (ATCC<sup>®</sup> medium 2222: adjusted to contain 10% heat-inactivated fetal bovine serum). The culture was propagated 7 days at 37°C in an aerobic atmosphere with 5% CO<sub>2</sub> until lysis of the host cell monolayer was reached.

<sup>2</sup>Quality control testing completed on post-freeze material unless specified as pre-freeze.

<sup>3</sup>PCR amplification of the AK56 locus was performed. Samples were subjected to restriction enzyme digestion typing by agarose gel electrophoresis.

<sup>4</sup>Primer sequences, annealing temperatures, and conditions for restriction enzyme digestion may be obtained at the *Toxoplasma* Genome Map website ([Toxoplasma Genome Map](http://ToxoplasmaGenomeMap.org)).

<sup>5</sup>Viable cells and signs of infection were seen after 3 days under cultivation conditions at 37°C.

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

**Figure 1: AK56 (Chromosome II) Amplicon Sequence**

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TTTTATTAGG TTTTTCGGTG TTTTCGCGGA GTCGTCTGAG CTCGGCACTC GCTGCTTTCC AAAATCTCGT TTCAACGTAT
CGCGGGCGCCG TCACCGCGCG CAATCCACTG TGATGCATGA TTCTGTTTCT AAAAACTGCG CCTTTTAGCC GGCTCGTTTT
TGCATACGTT TGGACCATAA AACCTCGTAT TGTGGAAGAA GAATGCAATT TGTGTCTGTG CTGATCACC GATGAAAATC
GGCGTGTCTC GCCCCCTGCC GTGTGCGCGT CCGCTTTTTG CGACCCCGGT ACACCCGTTT TTTGTGGTCA GCGAGGAACG
CACTTTTGCT GTTATTGTTT ACTTTTCAGC GTAACACTGA CCCCTTTCAT CGTGGCAGGA AACGAACTCT CAGCAAGAAT
TTTCGAGCAC TACTGCGTCG CAGCAGCCTA GTGGGGTGA CACGCATGTG CAGGACGGAC AGAAACTGCA AGCTTGTTCC
GC
    
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**Date:** 29 MAR 2016

**Signature:**



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