

## *Toxoplasma gondii*, Clone CL18

Catalog No. NR-10155

**Product Description:** *Toxoplasma gondii*, clone CL18 is a recombinant F1 clone selected from progeny of two parallel genetic crosses between a Type II parental strain [ME49 (clone B7)] and a Type III parental strain (CTG ARA-SYN).

Lot<sup>1</sup>: 58242544

Manufacturing Date: 17JUL2008

TEST	SPECIFICATIONS	RESULTS
<b>Genotyping<sup>2</sup></b> 850 locus ( <i>Sfa</i> NI digestion) <sup>3</sup> SAG1 locus <sup>4</sup>	Consistent with parental Type III strain Consistent with parental Type II strain	Consistent with parental Type III strain Consistent with parental Type II strain
<b>Drug susceptibility<sup>5</sup></b> Sinefungin Ara-A	Susceptible Resistant	Susceptible Resistant
<b>Viable Cell Count by Hemacytometry (pre-freeze)</b>	> 10 <sup>6</sup> cells/mL	2.2 x 10 <sup>7</sup> cells/mL
<b>Viability (post-freeze)<sup>6</sup></b>	Growth	Growth
<b>Sterility (21-day incubation)</b> Harpo's HTYE broth <sup>7</sup> , 37°C and 26°C, aerobic Trypticase soy broth, 37°C and 26°C, aerobic Sabouraud broth, 37°C and 26°C, aerobic Sheep blood agar, 37°C, aerobic Sheep blood agar, 37°C, anaerobic Thioglycollate broth, 37°C, anaerobic DMEM with 10% FBS, 37°C and 5% CO <sub>2</sub>	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-10155 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 medium 2222: adjusted to contain 10% heat-inactivated fetal bovine serum). The culture was propagated in 95% air, 5% CO<sub>2</sub> for 8 days at 37°C, until lysis of the host cell monolayer was reached.

<sup>2</sup>PCR amplification was performed separately for the two loci 850 and SAG1. Where appropriate, samples were subjected to restriction enzyme digestion typing by agarose gel electrophoresis.

<sup>3</sup>Primer sequences, annealing temperatures, and conditions for restriction enzyme digestion may be obtained at the *Toxoplasma* Genome Map website ([http://toxomap.wustl.edu/Toxo\\_Genetic\\_Map\\_Table.html](http://toxomap.wustl.edu/Toxo_Genetic_Map_Table.html)). <sup>4</sup>Primer sequences and conditions for PCR are available upon request.

<sup>5</sup>Sinefungin was used at a concentration of 2.7 x 10<sup>-7</sup> M and ara-A was used at a concentration of 1.3 x 10<sup>-4</sup> M, as described (Sibley, L. D., et al. "Generation of a Restriction Fragment Length Polymorphism Linkage Map for *Toxoplasma gondii*." *Genetics* 132 (1992): 1003-1015. PubMed: 1360931.)

<sup>6</sup>Incubated under cultivation conditions for 22 days at 37°C.

<sup>7</sup>Atlas, Ronald M. *Handbook of Microbiological Media*. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.

Date: 15 OCT 2009

Signature: Signature on File

Title: Technical Manager, BEI Authentication or designee

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