

Toxoplasma gondii, Clone CL12

Catalog No. NR-10169

Product Description: *Toxoplasma gondii*, clone CL12 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).

Lot¹: 58364310

Manufacturing Date: 01DEC2008

TEST	SPECIFICATIONS	RESULTS
Genotyping² 850 locus (<i>Sfa</i> NI digestion) ³ SAG1 locus ⁴	Consistent with parental Type II strain Consistent with parental Type III strain	Consistent with parental Type II strain Consistent with parental Type III strain
Drug susceptibility⁵ Sinefungin Ara-A	Resistant Resistant	Resistant Resistant
Viable Cell Count by Hemacytometry (pre-freeze)	> 10 ⁶ cells/mL	3.2 x 10 ⁷ cells/mL
Viability (post-freeze)⁶	Growth	Growth
Sterility (21-day incubation) Harpo's HTYE broth ⁷ , 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 ₂	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
Mycoplasma Contamination DNA Detection by PCR	None detected	None detected

¹NR-10169 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 7 days at 37°C, in a humidified atmosphere until lysis of the host cell monolayer was reached.

²PCR 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.

³Primer sequences, annealing temperatures, and conditions for restriction enzyme digestion may be obtained at the *Toxoplasma* Genome Map website ([Toxoplasma Genome Map](#)).

⁴Primer sequences and conditions for PCR are available upon request.

⁵Sinefungin was used at a concentration of 2.7 x 10⁻⁷ M and ara-A was used at a concentration of 1.3 x 10⁻⁴ 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.)

⁶Viable cells and signs of infection were seen after 7 days under cultivation conditions at 37°C.

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

Date: 16 OCT 2009

Signature: Signature on File

Title: Technical Manager, BEI Authentication or designee

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