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SUPPORTING INFECTIOUS DISEASE RESEARCH

Certificate of Analysis for NR-18960

Trypanosoma cruzi, Strain Peru, Clone C7 (+*lacZ*)

Catalog No. NR-18960

Product Description: *Trypanosoma cruzi* (*T. cruzi*), strain Peru, clone C7 (+*lacZ*) is a transgenic clone derived from the Peru strain.

Lot[1]: 59490918 **Manufacturing Date: 12OCT2010**

TEST	SPECIFICATIONS	RESULTS
Genotyping		
Sequencing of 18S rRNA gene (~ 920 bp)	Consistent with *T. cruzi*	Consistent with *T. cruzi*
Phenotypic Analysis[2]		
NR-18960 β-Galactosidase activity	Positive (red)	Positive (red)
Negative control (ATCC® CCL-163™)	Negative (yellow)	Negative (yellow)
Viable Cell Count by Hemacytometry (pre-freeze)[3]	> 10^6 cells/mL	1.3 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	No growth	No growth
Trypticase soy broth, 37°C and 26°C, aerobic	No growth	No growth
Sabouraud broth, 37°C and 26°C, aerobic	No growth	No growth
Brain heart infusion, 37°C and 26°C, aerobic	No growth	No growth
Sheep blood agar, 37°C, aerobic	No growth	No growth
Sheep blood agar, 37°C, anaerobic	No growth	No growth
Thioglycollate broth, 37°C, anaerobic	No growth	No growth
Mycoplasma Contamination		
DNA Detection by PCR	None detected	None detected

[1]NR-18960 was produced by cultivation of the deposited material BALB/3T3 clone A31 mouse embryonic fibroblasts (ATCC® CCL-163™) 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_2 for 4 days at 37°C, until emergence of trypomastigotes from host cells was observed.
[2]Parasites (~ 10^7) were lysed in 0.1% Triton X-100 and PBS and 100 µM chlorophenol red-β-D-galactoside (CPRG) and incubated for 20 min. at 37°C [Buckner, F. S., et al. "Efficient Technique for Screening Drugs for Activity against *Trypanosoma cruzi* using Parasites Expressing β-Galactosidase." Antimicrob. Agents Chemother. 40 (1996): 2592-2597. PubMed: 8913471.].
[3]Only trypomastigote-stage parasites were counted
[4]Viable cells and signs of infection were seen after 7 days under cultivation conditions at 37°C
[5]Atlas, Ronald M. Handbook of Microbiological Media. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.

Date: 06 APR 2011 **Signature:**

Title: Technical Manager, BEI Authentication or designee

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