b|e|i RESOURCES

SUPPORTING INFECTIOUS DISEASE RESEARCH

Certificate of Analysis for NR-15431

Genomic DNA from *Toxoplasma gondii*, CTG ARA-SNF

Catalog No. NR-15431

Product Description: Genomic DNA was isolated from *Toxoplasma gondii (T. gondii),* CTG ARA-SNF, which was originally the product of a genetic cross between singly resistant parental clones of the C (also CEP and CTG) strain, that were obtained by chemical mutagenesis.

Lot[1]: 58723098 **Manufacturing Date: 18FEB2010**

TEST	SPECIFICATIONS	RESULTS
Agarose Gel Electrophoresis	High molecular weight chromosomal DNA	High molecular weight chromosomal DNA (Figure 1)
Content by PicoGreen® Measurement	1 to 3 µg in 25 to 100 µL per vial	2.4 µg in 50 µL per vial (48 µg/mL)
Functional Activity by PCR Amplification[2] 850 locus SAG1 locus	 ~ 750 bp amplicon ~ 250 bp amplicon	 ~ 750 bp amplicon ~ 250 bp amplicon
Genotyping[3] 850 locus (*Sfa*NI digestion) SAG1 locus[4]	 Consistent with parental Type III strain Consistent with parental Type III strain	 Consistent with parental Type III strain Consistent with parental Type III strain
OD_{260}/OD_{280} Ratio	1.7 to 2.0	2.0
Protozoan Inactivation 100% of vial contents inoculated on human foreskin fibroblast monolayers (ATCC® CRL-1634™)[5]	Cell line infection not detected	Cell line infection not detected

[1]NR-15431 was produced from a culture of NR-10151 grown in human foreskin fibroblasts (Hs27; ATCC® CRL-1634™) in cell cultivation medium for parasites (DMEM; ATCC® medium 2222: adjusted to contain 10% heat-inactivated fetal bovine serum). The culture was propagated in 95% air, 5% CO_2 at 37°C until lysis of the host cell monolayer was reached. Tachyzoites of NR-10150 were harvested and genomic DNA was extracted using the Qiagen DNeasy Blood and Tissue Kit (Catalog no. 69504).

[2]Primer sequences and conditions for PCR are available upon request.

[3]PCR was performed separately for the 850 and SAG1 loci. Where appropriate, samples were subjected to restriction enzyme digestion as described in the *Toxoplasma* Genome Map website (http://toxomap.wustl.edu/Toxo_Genetic_Map_Table.html).

[4]Based on migration pattern of the SAG1 amplicon from NR-15431 during agarose gel electrophoresis as compared to NR-15430 (Genomic DNA from *Toxoplasma gondii* CTG ARA-SYN)

[5]Incubated in DMEM (ATCC® 30-2002) and propagated in 95% air, 5% CO_2 for 7 days at 37°C.

Date: 22 OCT 2010 **Signature:**

Title: Technical Manager, BEI Authentication or designee

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

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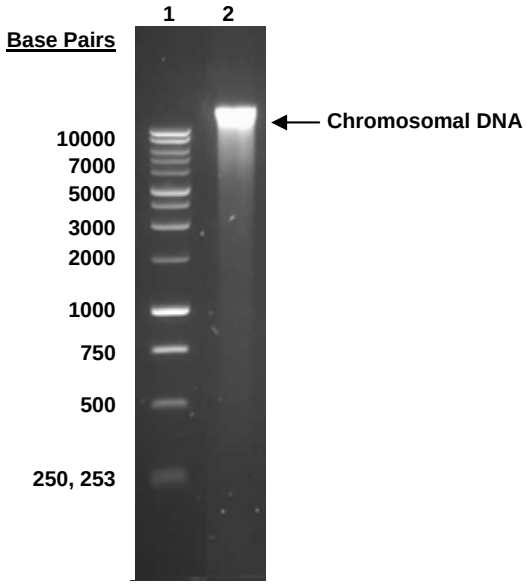
Figure 1



Lane 1: Promega 1kb DNA Ladder
Lane 2: 288 ng of NR-15431

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