

Certificate of Analysis for NR-19974

Macrophage Cell Line Derived from TLR3 Knockout Mice

Catalog No. NR-19974

This reagent is the property of the U.S. Government.

Product Description: This murine macrophage cell line was derived using primary bone marrow cells from TLR3 knockout mice. The macrophage cells were immortalized by infection with the ecotropic transforming replication-deficient retrovirus J2 using techniques described in the literature.

Lot: 63819365 Manufacturing Date: 19OCT2015

TEST	SPECIFICATIONS	RESULTS
Growth Properties	Adherent	Adherent
PCR Amplification of Extracted DNA (Figure 1) ¹ TLR3 wild type primers TLR3 knockout primers	No amplicon Expected amplicon	No amplicon Expected amplicon
Stimulation of RANTES ¹	Report results	See Figure 2
Multiplex PCR Amplification of Cytochrome C Oxidase I (COI) Gene	Murine origin No evidence of another species	Murine origin No evidence of another species
Total Cell Count	> 1.0 x 10 ⁶ cells per vial	2.4 x 10 ⁶ cells per vial
Post-Freeze Viability	≥ 50%	72.2%
Sterility (21-day incubation) Harpo's HTYE broth ² , 37°C and 26°C, aerobic Trypticase soy broth, 37°C and 26°C, aerobic Sabouraud broth, 37°C and 26°C, aerobic Blood agar, 37°C, aerobic Blood agar, 37°C, anaerobic Thioglycollate broth, 37°C, anaerobic DMEM with 10% FBS, 37°C and 5% CO ₂	No growth	No growth
Mycoplasma Contamination Hoechst DNA stain Agar and broth culture (14-day incubation at 37°C) DNA Detection by PCR of Test Article nucleic acid	None detected None detected None detected	None detected None detected None detected

Depositor QC is NOT lot specific; same data as NR-19974 lot 60243377

Date: 11 APR 2016

Signature:

BEI Resources Authentication

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected by ATCC® and the contributor 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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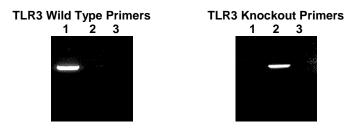
Fax: 703-365-2898

²Atlas, Ronald M. <u>Handbook of Microbiological Media</u>. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.



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Figure 1 - PCR Amplification

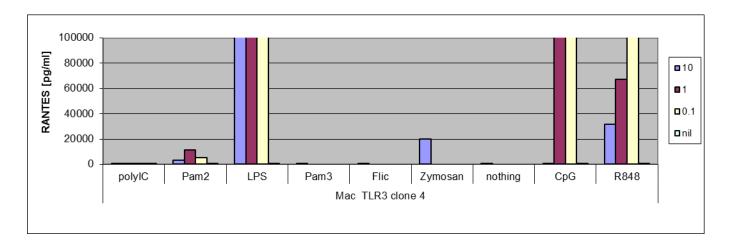


Lane 1: Wild type macrophages

Lane 2: TLR3 knockout macrophages (NR-19974)

Lane 3: Negative control (H₂O)

Figure 2 – Stimulation of RANTES



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