

## **Certificate of Analysis for NR-19977**

## Macrophage Cell Line Derived from TLR1 Knockout Mice

## Catalog No. NR-19977

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

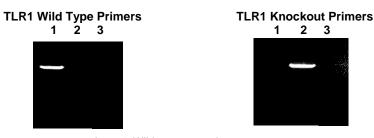
**Product Description:** This murine macrophage cell line was derived using primary bone marrow cells from TLR1 knockout mice. The macrophage cells were immortalized by infection with the ecotropic transforming replication-deficient retrovirus J2 using techniques described in the literature.

Lot: 70014858 Manufacturing Date: 04MAY2018

TEST	SPECIFICATIONS	RESULTS
Growth Properties	Adherent	Adherent
PCR Amplification of Extracted DNA <sup>1</sup>		
TLR1 wild type primers	No amplicon	No amplicon (Figure 1)
TLR1 knockout primers	Expected amplicon	Expected amplicon (Figure 1)
Stimulation of TNF-α <sup>1</sup>	Report results	Expected pattern for different ligands (Figure 2)
Multiplex PCR Amplification of Cytochrome C Oxidase I	Murine origin	Murine origin
(COI) Gene	No evidence of another species	No evidence of another species
Total Cell Count	> 1.0 x 10 <sup>6</sup> cells/vial	2.8 x 10 <sup>6</sup> cells/vial
Post-Freeze Viability	≥ 50%	68.4%
Sterility (21-day incubation)		
Harpo's HTYE broth <sup>2</sup> , 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
Blood agar, 37°C, aerobic	No growth	No growth
Blood agar, 37°C, anaerobic	No growth	No growth
Thioglycollate broth, 37°C, anaerobic	No growth	No growth
DMEM with 10% FBS, 37°C and 5% CO <sub>2</sub>	No growth	No growth
Mycoplasma Contamination		
Hoechst DNA stain	None detected	None detected
Agar and broth culture (14-day incubation at 37°C)	None detected	None detected
DNA Detection by PCR of Test Article nucleic acid	None detected	None detected

<sup>&</sup>lt;sup>1</sup>Depositor data; not lot specific.

Figure 1: PCR Amplification



Lane 1: Wild type macrophages

Lane 2: TLR1 knockout macrophages (NR-19977)

Lane 3: Negative control (H<sub>2</sub>O)

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Tel: 800-359-7370

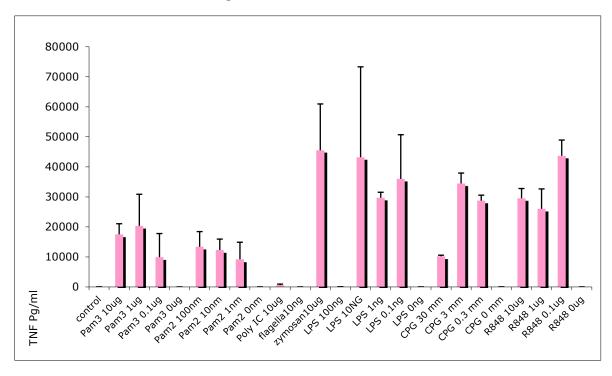
Fax: 703-365-2898

<sup>&</sup>lt;sup>2</sup>Atlas, 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 2: Stimulation of TNF-α



/Heather Couch/ Heather Couch

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