b|e|i RESOURCES

SUPPORTING INFECTIOUS DISEASE RESEARCH

Certificate of Analysis for NR-9456

Macrophage Cell Line Derived from Wild Type Mice

Catalog No. NR-9456

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

Product Description: The murine macrophage cell line, NR-9456, was derived using primary bone marrow cells from wild type mice. The primary bone marrow cells were immortalized by infection with the ecotropic transforming replication-deficient retrovirus J2 using techniques described in the literature. Characterization based on immunofluorescence, stimulation assays and flow cytometry demonstrated that the immortalized cell line retains its macrophage-specific morphological, functional and surface expression properties.

Lot: 63466416 **Manufacturing Date: 05MAY2015**

TEST	SPECIFICATIONS	RESULTS
Growth Properties	Adherent	Adherent
Morphology by Immunofluorescence[1]	Macrophage	Macrophage
Surface Marker Expression by Flow Cytometry Using Specific Antibodies[1]	Characteristic of bone marrow-derived macrophages	Characteristic of bone marrow-derived macrophages
CD11b	Positive	CD11b+++
F4/80	Positive	F4/80++
CD45	Positive	CD45+
CD80	Positive	CD80+
CD86	Positive	CD86+
CD14	Positive	CD14+
MHC	Positive	MHC++
CD11c (dendritic cell marker)	Negative	CD11c-
B220 (lymphocyte marker)	Negative	B220-
Multiplex PCR Amplification of Cytochrome C Oxidase I (COI) Gene	Murine origin No evidence of another species	Murine origin No evidence of another species
Cell Count	> 1.0×10^6 cells/vial	7.5×10^6 cells per vial
Post-Freeze Viability	≥ 50%	95.1%
Sterility (BacT/ALERT® 3D Microbial Detection System)		
21-day incubation		
i NST culture bottle, 32°C, anaerobic	No growth	No growth
i AST culture bottle, 32°C, aerobic	No growth	No growth
Sterility (21-day incubation)		
Harpo's HTYE broth[1], 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_2	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

[1]Performed on immortalized cell line prior to growth of this distribution lot

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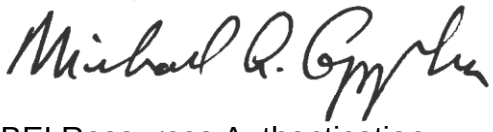
Date: 20 JUL 2015 **Signature:** 

BEI Resources Authentication

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