

Certificate of Analysis for NR-9904

Microglial Cell Line Derived from MAL/My88 Double Knockout Mice

Catalog No. NR-9904

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Product Description: The murine microglial cell line, NR-9904, was derived using brain tissue from MAL (MyD88 adaptor-like)/MyD88 (myeloid differentiation primary response protein 88) double knockout mice. The microglial 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 microglial-specific morphological, functional and surface expression properties.

Lot: 58381525 Manufacturing Date: 10NOV2008

TEST	SPECIFICATIONS	RESULTS
Growth Properties	Adherent	Adherent
Morphology by Immunofluorescence ¹	Microglial	Microglial
Surface Marker Expression by Flow Cytometry Using Specific Antibodies ¹ CD11b F4/80 MHCII GFAP (astrocyte marker)	Characteristic of microglial cells Positive Positive Positive Negative	Characteristic of microglial cells Positive Positive Positive Negative
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/vial	3.3 X 10 ⁶ cells/vial
Post-Freeze Viability	≥ 50%	74%
Sterility (BacT/ALERT® 3D Microbial Detection System) 14-day incubation of NR-9904: i NST culture bottle, 32°C, anaerobic i AST culture bottle, 32°C, aerobic	No growth No growth	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

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

Date: 06 MAR 2014 Signature: Doothy C. Young

Title: Technical Manager, BEI Authentication or designee

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