

Recombinant Murine Coronavirus, icA59-ns2dm

Catalog No. NR-43001

Product Description: Cell lysate and supernatant from *mus musculus* liver epithelial cells¹ infected with recombinant murine coronavirus, icA59-ns2dm

Lot²: 61556176

Manufacturing Date: 15MAY2013

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in NCTC clone 1469 Cells ¹	Cell enlargement and detachment	Cell enlargement and detachment
Sequencing of Species-Specific Region (659 nucleotides)	Consistent with murine coronavirus, icA59-ns2dm	100% identity with murine coronavirus, icA59-ns2dm (GenBank: KF268339) ^{3,4}
Titer by TCID ₅₀ Assay ^{5,6} in NCTC clone 1469 Cells ¹	Report results	2.3 × 10 ⁷ TCID ₅₀ per mL
Sterility (21-day incubation) Harpo's HTYE broth ⁷ , 37°C and 26°C, aerobic Trypticase soy broth, 37°C and 26°C, aerobic Sabouraud broth, 37°C and 26°C, aerobic Sheep blood agar, 37°C, aerobic Sheep blood agar, 37°C, anaerobic Thioglycollate broth, 37°C, anaerobic DMEM with 10% FBS, 37°C and 5% CO ₂	No growth No growth No growth No growth No growth No growth No growth	No growth No growth No growth No growth No growth No growth No growth
Mycoplasma Contamination 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

¹NCTC clone 1469 cells (ATCC® CCL-9.1™)

²Grown in Dulbecco's Modified Eagle Medium (ATCC® 30-2002™) supplemented with 10% fetal bovine serum (ATCC® 30-2020™) for 1 day at 37°C and 5% CO₂

³Note that the nomenclature used for this virus in the NCBI database differs from that used here.

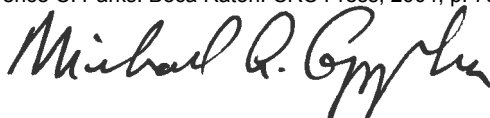
⁴Sequence analysis at BEI Resources confirmed the presence of the H46A and H126R mutations described in Roth-Cross, J. K., et al. "Organ-Specific Attenuation of Murine Hepatitis Virus Strain A59 by Replacement of Catalytic Residues in the Putative Viral Cyclic Phosphodiesterase ns2." *J. Virol.* 83 (2009): 3743-3753. PubMed: 19176619.

⁵The Tissue Culture Infectious Dose 50% (TCID₅₀) endpoint is the 50% infectious endpoint in cell culture. The TCID₅₀ is the dilution of virus that under the conditions of the assay can be expected to infect 50% of the culture vessels inoculated, just as a Lethal Dose 50% (LD₅₀) is expected to kill half of the animals exposed. A reciprocal of the dilution required to yield the TCID₅₀ provides a measure of the titer (or infectivity) of a virus preparation.

⁶9 days at 37°C and 5% CO₂

⁷Atlas, Ronald M. *Handbook of Microbiological Media*. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.

Date: 21 OCT 2013

Signature: 

Title: Technical Manager, BEI Authentication or designee

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