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SUPPORTING INFECTIOUS DISEASE RESEARCH

# Recombinant Murine Coronavirus, icA59ns2dm

Catalog No. NR-43001

# For research use only. Not for use in humans.

## **Contributor:**

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## Manufacturer:

BEI Resources

#### **Product Description:**

<u>Virus Classification</u>: Coronaviridae, Coronavirinae, Betacoronavirus

<u>Species</u>: Murine coronavirus, [formerly murine hepatitis virus (MHV), formerly mouse hepatitis virus]<sup>1</sup>

Strain/Isolate: icA59-ns2dm

- Original Source: NR-43001 is a recombinant murine coronavirus that was produced using a vaccinia virus-based reverse genetics system, and derived from cloned, fulllength MHV-A59 cDNA.<sup>2</sup> The icA59-ns2dm virus carries two site-directed mutations in the catalytic region of the cyclic phosphodiesterase ns2, resulting in amino acid substitutions at positions 46 (H46A) and 126 (H126R).<sup>3</sup> Viruses carrying either of these two mutations are attenuated for replication in mouse liver following intrahepatic inoculation, but not in mouse brain following intracranial inoculation.4 The ns2 phosphodiesterase inhibits the interferon-inducible oligoadenylate synthase (OAS)-RNase L pathway by cleaving 2',5'-oligoadenylate, the product of OAS, preventing activation of the cellular endoribonuclease L, thus blocking viral RNA degradation and facilitating hepatitis development.5
- <u>Comments</u>: The complete genome of the recombinant double mutant strain has been sequenced (GenBank: <u>KF268339</u>). In order to remove contaminating mycoplasma, the second viral passage at BEI Resources was performed by lipofectamine-mediated transfection of extracted viral RNA.

## **Material Provided:**

Each vial contains approximately 1 mL of cell lysate and supernatant from *Mus musculus* liver epithelial cells infected with recombinant murine coronavirus, icA59-ns2dm.

<u>Note</u>: If homogeneity is required for your intended use, please purify prior to initiating work.

## Packaging/Storage:

NR-43001 was packaged aseptically in screw-capped plastic cryovials. The product is provided frozen and should be stored at -60°C or colder immediately upon arrival. For long-term storage, the vapor phase of a liquid nitrogen freezer is recommended. Freeze-thaw cycles should be avoided.

**Growth Conditions:** 

<u>Host</u>: NCTC clone 1469 cells (ATCC<sup>®</sup> CCL-9.1<sup>™</sup>) <u>Growth Medium</u>: Dulbecco's Modified Eagle's Medium supplemented with 10% fetal bovine serum <u>Infection</u>: Cells should be 80% to 90% confluent <u>Incubation</u>: 1 to 9 days at 37°C and 5% CO<sub>2</sub> <u>Cytopathic Effect</u>: Cell enlargement and detachment

## Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: Recombinant Murine Coronavirus, icA59-ns2dm, NR-43001."

## **Biosafety Level: 2**

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. <u>Biosafety in Microbiological and Biomedical Laboratories</u>. 6th ed. Washington, DC: U.S. Government Printing Office, 2020; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

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# **References:**

- 1. ICTV Taxonomy History for Murine coronavirus
- Coley, S. E., et al. "Recombinant Mouse Hepatitis Virus from Cloned, Full-Length cDNA Replicates to High Titers *in vitro* and is Fully Pathogenic *in vivo*." <u>J. Virol.</u> 79 (2005): 3097-3196. PubMed: 15709029.
- 3. Weiss, S. R., Personal Communication.
- 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." <u>J. Virol.</u> 83 (2009): 3743-3753. PubMed: 19176619.
- Zhao, L., et al. "Antagonism of the Interferon-Induced OAS-RNase L Pathway by Murine Coronavirus ns2 Protein is Required for Virus Replication and Liver Pathology." <u>Cell Host. Microb.</u> 11 (2012): 607-616. PubMed: 22704621.

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