Recombinant Murine Coronavirus MHV-A59-MHV2Spike with Enhanced Green Fluorescent Protein (eGFP)

Catalog No. NR-53717

For research use only. Not for use in humans.

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Manufacturer:
BEI Resources

Product Description:
Virus Classification: *Coronaviridae*, *Betacoronavirus*
Species: Murine coronavirus [formerly murine hepatitis virus (MHV)]
Isolate: MHV-A59-MHV2Spike-eGFP
Original Source: MHV, isolate MHV-A59-MHV2Spike-eGFP is a recombinant MHV-A59 virus with spike genes from strain MHV-2 in which open reading frame 4 (ORF4) was replaced by a gene encoding the enhanced green fluorescent protein (eGFP).[1,2]
Comments: Targeted recombination and selection was used to construct MHV, isolate MHV-A59-MHV2Spike-eGFP. Spike genes in MHV, isolate MHV-A59-MHV2Spike-eGFP are derived by switching mildly virulent strain A59 spike genes with spike genes from highly neurovirulent strain MHV-2. All other genes in the virus genome are derived from strain MHV-A59. The eGFP gene was inserted into the virus genome in place of the nonessential gene ORF4.[2] eGFP is a modified version of GFP gene designed for brighter fluorescence, in which the codon utilization has been maximized for translation in eukaryotic cells.[3,4] Expression of eGFP is stable over multiple passages *in vitro* and at a level high enough to be readily detected in cultured cells and in the central nervous system of infected animals.[2]

Material Provided:
Each vial contains approximately 1.0 mL of cell lysate and supernatant from murine 17Cl-1 cells infected with MHV, isolate MHV-A59-MHV2Spike-eGFP.

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

Packaging/Storage:
NR-53717 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:
Host: Murine 17Cl-1 cells (BEI Resources NR-53719)
Growth Medium: Dulbecco's Modified Eagle's Medium (DMEM) modified to contain 4 mM L-glutamine, 4500 mg per L glucose, 1 mM sodium pyruvate, and 1500 mg per L sodium bicarbonate supplemented with 2% fetal bovine serum or equivalent
Infection: Cells should be 70% to 80% confluent
Incubation: 2 to 4 days at 37°C and 5% CO_2
Cytopathic Effect: Cell rounding and sloughing

Citation:
Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: Recombinant Murine Coronavirus MHV-A59-MHV2Spike with Enhanced Green Fluorescent Protein (eGFP), NR-53717."

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. Biosafety in Microbiological and Biomedical Laboratories. 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. Weiss, S., Personal Communication.
2. Sarma, J. D., et al. "Enhanced Green Fluorescent Protein Expression May be Used to Monitor Murine Coronavirus Spread *in vitro* and in the Mouse Central Nervous System." J. Neurovirol. 8 (2002): 381-391. PubMed: 12402164.
3. Cormack, B. P., R. H. Valdivia and S. Falkow. "FACS-Optimized Mutants of the Green Fluorescent Protein (GFP)." Gene 173 (1996): 33-38. PubMed: 8707053.
4. Haas, J., E. C. Park and B. Seed. "Codon Usage Limitation in the Expression of HIV-1 Envelope Glycoprotein." Curr. Biol. 6 (1996): 315-324. PubMed: 8805248.

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