Product Information Sheet for NR-54007

Vector pCAGGS Containing the SARS-Related Coronavirus 2, Beta Variant Spike Glycoprotein Receptor Binding Domain (RBD) Gene

Catalog No. NR-54007

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For research use only. Not for use in humans.

Contributor:

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Manufacturer:

BEI Resources

Product Description:

The vector for the receptor binding domain (RBD) of the spike (S) glycoprotein gene from severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), Wuhan-Hu-1 (GenBank: MN908947) was designed by fusing the N-terminal S protein signal sequence to the spike RBD (amino acids 319 to 541) with a C-terminal hexa-histidine tag.[1,2] The sequence was codon optimized for mammalian expression, mutated to include the Beta variant [also referred to as the South Africa variant; B.1.351 lineage] K417N, E484K and N501Y mutations and subcloned into the pCAGGS mammalian expression vector under the AG promoter.[2,3] NR-54007 contains the beta-lactamase gene, *bla*, to provide transformant selection through ampicillin resistance in *Escherichia coli (E. coli)*. The resulting size of the plasmid is approximately 5500 base pairs. The complete plasmid sequence and map are provided on the BEI Resources webpage. The plasmid was produced in *E. coli* and extracted.

Note: For a detailed protocol and list of related items, see https://labs.icahn.mssm.edu/krammerlab/covid-19/

The S glycoprotein mediates viral binding to the host angiotensin converting enzyme 2 (ACE2). This protein forms a trimer, and when bound to a host receptor allows fusion of the viral and cellular membranes.[4] The Beta variant includes three mutations in the RBD that may have functional significance: K417N, E484K and N501Y.[5] Structural modeling and mouse studies indicate N501Y increases S glycoprotein binding to ACE2, resulting in increased SARS-CoV-2 virulence.[6,7]

Material Provided:

Each vial contains plasmid DNA in TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8.0). The DNA concentration and volume provided are shown on the Certificate of Analysis. The vial should be centrifuged prior to opening. Note: The contents of the vial should be used to replicate the plasmid in *E. coli* prior to mammalian expression.

Packaging/Storage:

NR-54007 was packaged aseptically in screw-capped plastic cryovials. The product is provided frozen on dry ice and should be stored at -20°C or colder immediately upon arrival. Freeze-thaw cycles should be minimized.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: Vector pCAGGS Containing the SARS-Related Coronavirus 2, Beta Variant Spike Glycoprotein Receptor Binding Domain (RBD) Gene, NR-54007."

Biosafety Level: 1

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.

Disclaimers:

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Registrants interested in commercializing this product must contact Mount Sinai for a license (Frenz, Christopher christopher.frenz@mssm.edu).

References:

1. Krammer, F. and F. Amanat, Personal Communication.
2. Amanat, F., et al. "A Serological Assay to Detect SARS-CoV-2 Seroconversion in Humans." Nat. Med. 26 (2020): 1033-1036. PubMed: 32398876.
3. Wu, F., et al. "A New Coronavirus Associated with Human Respiratory Disease in China." Nature 579 (2020): 265-269. PubMed: 32015508.
4. Hulswit, R. J. G., C. A. M. de Haan and B.-J. Bosch. "Coronavirus Spike Protein and Tropism Changes." Adv. Virus Res. 96 (2016): 29-57. PubMed: 27712627.
5. WHO
6. Gu, H., et al. "Adaptation of SARS-CoV-2 in BALB/c Mice for Testing Vaccine Efficacy." Science 369 (2020): 1603-1607. PubMed: 32732280.
7. Leung, K., et al. "Early Transmissibility Assessment of the N501Y Mutant Strains of SARS-CoV-2 in the United Kingdom, October to November 2020." Euro. Surveill. 26 (2021): pii 2002106. PubMed: 33413740.

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