

SARS-Related Coronavirus 2, Mouse-Adapted, MA10 Variant (in isolate USA-WA1/2020 backbone), Infectious Clone (ic2019-nCoV MA10) in Calu-3 Cells

Catalog No. NR-55329

For research use only. Not for use in humans.

Contributor:

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Manufacturer:

BEI Resources

Product Description:

Virus Classification: *Coronaviridae*, *Betacoronavirus*

Species: Severe acute respiratory syndrome-related coronavirus 2

Strain/Isolate: Infectious clone of isolate MA10 (ic2019-nCoV MA10)^{1,2}

Original Source: Severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), isolate MA10 was developed by ten *in vivo* serial passages of SARS-CoV-2, isolate MA in BALB/c mice, followed by plaque purification in *Cercopithecus aethiops* kidney epithelial cells (Vero E6).^{1,2} Following deep sequencing of the plaque purified virus, an infectious clone was generated for subsequent preparation of the MA10 virus stock.¹

Note: Genome sequence information is provided on the Certificate of Analysis and includes an analysis of all sequence variations observed for each lot.

Comments: SARS-CoV-2, isolate MA was developed by reverse engineering SARS-CoV-2, isolate USA-WA1/2020 for efficient interaction between viral spike protein and the mouse orthologue of the human receptor, angiotensin-converting enzyme 2 (ACE2).^{2,3} To provide a suitable virus for use in murine models, SARS-CoV-2, isolate MA was then serially passaged 10 times in BALB/c mice, followed by plaque purification to identify SARS-CoV-2, isolate MA10.² SARS-CoV-2, isolate MA10 induces a disease course that closely resembles human disease progression in wild-type BALB/c mice.² Besides the substitution mutations engineered into the spike protein of parental isolate MA (Q498Y/P499T), isolate MA10 also includes five additional mutations [non-structural protein 4 (NSP4) C9438T; non-structural protein 7 (NSP7) A11847G; non-structural protein 8 (NSP8) A12159G; Spike (S) C23039A; and Open reading frame 6 (ORF6) T27221C]. The complete genome of SARS-CoV-2, isolate MA10 has been sequenced (GenBank: [MT952602](https://www.ncbi.nlm.nih.gov/nuccore/MT952602)).

Under the nomenclature system introduced by GISAID (Global Initiative on Sharing All Influenza Data),

SARS-CoV-2, isolate USA-WA1/2020, the backbone on which SARS-CoV-2, isolate MA10 was built, is assigned lineage A and GISAID clade S using Phylogenetic Assignment of Named Global Outbreak LINEages (PANGOLIN) tool.^{4,5}

In December 2019, an outbreak of a respiratory illness (COVID-19) began in Wuhan, Hubei Province, China. The outbreak is associated with a seafood market and although environmental samples from the market are positive for the novel coronavirus, an association with a particular animal has not been determined.⁶

Material Provided:

Each vial contains approximately 0.1 mL of spin clarified cell lysate and supernatant from *Homo sapiens* lung adenocarcinoma epithelial cells (Calu-3) infected with ic2019-nCoV MA10.

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

Packaging/Storage:

NR-55329 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: *Homo sapiens* lung adenocarcinoma epithelial cells (Calu-3; ATCC® HTB-55™)

Growth Medium: Eagle's Minimum Essential Medium containing Earle's Balanced Salt Solution, non-essential amino acids, 2 mM L-glutamine, 1 mM sodium pyruvate and 1500 mg per L of sodium bicarbonate supplemented with 2% fetal bovine serum, or equivalent

Infection: Cells should be 60% to 80% confluent

Incubation: 3 to 5 days at 37°C and 5% CO₂

Cytopathic Effect: Cell rounding and sloughing

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: SARS-Related Coronavirus 2, Mouse-Adapted, MA10 Variant (in isolate USA-WA1/2020 backbone), Infectious Clone (ic2019-nCoV MA10) in Calu-3 Cells, NR-55329, contributed by Ralph S. Baric."

Biosafety Level: 3

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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Patent application for SARS-CoV-2, MA10 has been filed by University of North Carolina at Chapel Hill and is currently pending.

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References:

1. Baric, R. S., Personal Communication.
2. Leist, S. R., et al. "A Mouse-Adapted SARS-CoV-2 Induces Acute Lung Injury and Mortality in Standard Laboratory Mice." *Cell* 183 (2020): 1070-1085. PubMed: 33031744.
3. Dinno, H. K., et al. "SARS-COV-2 Reverse Genetics Reveals a Variable Infection Gradient in the Respiratory Tract." *Cell* 182 (2020): 429-446. PubMed: 32526206.
4. Rambaut, A., et al. "A Dynamic Nomenclature Proposal for SARS-CoV-2 Lineages to Assist Genomic

Epidemiology." *Nat. Microbiol.* 5 (2020): 1403-1407. PubMed: 32669681.

5. Mercatelli, D. and F. M. Giorgi. "Geographic and Genomic Distribution of SARS-CoV-2 Mutations." *Front. Microbiol.* (2020): doi.org/10.3389/fmicb.2020.01800. PubMed: 32793182.
6. Gralinski, L. E. and V. D. Menachery. "Return of the Coronavirus: 2019-nCoV." *Viruses* 12 (2020): 135. PubMed: 31991541.

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