SUPPORTING INFECTIOUS DISEASE RESEARCH

Genomic RNA from SARS-Related Coronavirus 2, Isolate USA-CA2/2020

Catalog No. NR-52509

This reagent is the property of the U.S. Government.

Product Description:

Genomic RNA was extracted from a preparation of cell lysate and supernatant from *Cercopithecus aethiops* kidney epithelial cells (Vero E6; ATCC[®] CRL-1586[™]) infected with severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), isolate USA-CA2/2020 (BEI Resources NR-52387 lot 70034887), using QIAamp[®] Viral RNA Mini Kit (Qiagen[®] 52904). The viral genomic RNA is in a background of cellular nucleic acid and carrier RNA.

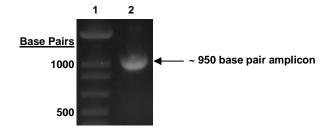
Lot: 70035260

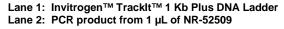
Manufacturing Date: 08JUN2020

TEST	SPECIFICATIONS	RESULTS		
Sequencing of Species-Specific Region (~ 840 nucleotides)	≥ 98% identity with SARS-CoV-2, isolate USA-CA2/2020 (GenBank: MN994468.1)	100% identity with SARS-CoV-2, isolate USA-CA2/2020 (GenBank: MN994468.1)		
Next-Generation Sequencing (NGS) of Complete Genome Using Illumina [®] iSeq™ 100 Platform (Refer to Appendix I for NGS information)	≥ 98% identity with SARS-CoV-2, isolate USA-CA2/2020 (GenBank: MN994468.1)	100% identity with SARS-CoV-2, isolate USA-CA2/2020 (GenBank: MN994468.1)		
Functional Activity by RT-PCR Amplification ¹	~ 950 base pair amplicon	~ 950 base pair amplicon (Figure 1)		
Genome Copy Number Using BioRad QX200 Droplet Digital PCR (ddPCR™) System (Post vial; 11 replicates)				
Virus Inactivation (14 Days, 2 Passages) 10% of total yield inoculated on Vero E6 cells and evaluated for cytopathic effect ²				
Passage 1	No viable virus detected	No viable virus detected		
Passage 2	No viable virus detected	No viable virus detected		
Virus Inactivation (14 Days, 2 Passages) 100% of total yield inoculated on Vero E6 cells and evaluated for cytopathic effect ²				
Passage 1	No viable virus detected	No viable virus detected		
Passage 2	No viable virus detected	No viable virus detected		

¹Amplified using iTaq[™] Universal SYBR Green One-step Kit (Bio-Rad[®] 172-5151) with 5 µL of NR-52509 in a 50 µL reaction
²Supernatant was tested by qPCR to confirm absence of CPE and no evidence of replicative RNA was detected. Samples from both passages were tested by qPCR at the end of day 14 of the passages.

Figure 1: Functional Activity of NR-52509 by RT-PCR Amplification





BEI Resources www.beiresources.org E-mail: <u>contact@beiresources.org</u> Tel: 800-359-7370 Fax: 703-365-2898

Certificate of Analysis for NR-52509

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/Heather Couch/ Heather Couch

08 OCT 2020

Program Manager or designee, ATCC Federal Solutions

ATCC[®], on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC[®]'s knowledge.

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APPENDIX I: NGS Information for NR-52509 lot 70035260

Sequence analysis resulted in the discovery of one SNP when compared to GenBank MN994468.1 (see Table below). Quality scores over 60 indicate it is improbable that the variant call is incorrect.

Position in NR-52509_ 70035260 Sequence	Position in MN994468.1	Reported MN994468.1 Sequence	Identified Alternative Base	Quality	Variant Type	Length of Variant	Frequency of Variant
23604	23607	G	Т	222	SNP	1	0.5874439