

Certificate of Analysis for NR-52382

SARS-Related Coronavirus 2, Isolate USA-CA1/2020

Catalog No. NR-52382

Product Description:

Severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), isolate USA-CA1/2020 was isolated from a nasopharyngeal swab from a human patient in California, USA on January 23, 2020. NR-52382 lot 70034877 was produced by infecting *Cercopithecus aethiops* kidney cells (Vero E6; ATCC[®] CRL-1586[™]) with the deposited material in Eagle's Minimum Essential Medium (ATCC 30-2003[™]) supplemented with 2% fetal bovine serum (ATCC 30-2020[™]) for 4 days at 37°C with 5% CO₂.

Passage History:

V(2)/VE6(3) (Centers for Disease Control and Prevention/BEI Resources); V = Vero cells; VE6 = Vero E6 cells

Lot: 70034877 Manufacturing Date: 18MAY2020

TEST	SPECIFICATIONS	RESULTS	
Identification by Infectivity in Vero E6 Cells	Cell rounding and detachment	Cell rounding and detachment	
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-CA1/2020 (GenBank: MN994467.1)	99.9% identity with SARS-CoV-2, isolate USA-CA1/2020 (GenBank: MN994467.1)	
Titer by TCID ₅₀ Assay in Vero E6 Cells by Cytopathic Effect ¹ (5 days at 37°C and 5% CO ₂)	Report results	2.8 × 10 ⁵ TCID ₅₀ per mL	
Sterility (21-day incubation)			
Harpo's HTYE broth, 37°C and 26°C, aerobic ²	No growth	No growth	
Trypticase Soy broth, 37°C and 26°C, aerobic	No growth	No growth	
Sabouraud broth, 37°C and 26°C, aerobic	No growth	No growth	
Sheep blood agar, 37°C, aerobic	No growth	No growth	
Sheep blood agar, 37°C, anaerobic	No growth	No growth	
Thioglycollate broth, 37°C, anaerobic	No growth	No growth	
DMEM with 10% FBS, 37°C, aerobic	No growth	No growth	
Mycoplasma Contamination			
Agar and broth culture (14-day incubation at 37°C)	None detected	None detected	
DNA detection by PCR of extracted Test Article nucleic acid	None detected	None detected	

¹The Tissue Culture Infectious Dose 50% (TCID₅₀) endpoint is the 50% infectious endpoint in cell culture. The TCID₅₀ is the dilution of virus that under the conditions of the assay can be expected to infect 50% of the culture vessels inoculated, just as a Lethal Dose 50% (LD₅₀) is expected to kill half of the animals exposed. A reciprocal of the dilution required to yield the TCID₅₀ provides a measure of the titer (or infectivity) of a virus preparation. ²Atlas, Ronald M. <u>Handbook of Microbiological Media</u>. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.

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Program Manager or designee, ATCC Federal Solutions

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APPENDIX I: NGS Information for NR-52382 lot 70034877

Sequence analysis resulted in the discovery of one SNP and two deletions when compared to GenBank MN994467.1 (see Table below). Quality scores over 60 indicate it is improbable that the variant call is incorrect.

Position in NR-52382_70034877 Sequence	Position in MN994467.1	Reported MN994467.1 Sequence	Identified Alternative Base	Quality	Variant Type	Length of Variant	Frequency of Variant
495	507	ATGGTCATGTT ATGGTTG	ATG	228	Indel	15	0.8965517
21772	21799	Т	Α	225	SNP	1	1.0000000
23555	23582	TATCAGACTCA GACTAAT	TAT	222	Indel	15	0.6134454

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