

Genomic RNA from SARS-Related Coronavirus 2, Isolate New York-PV08410/2020

Catalog No. NR-53518

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 New York-PV08410/2020 (BEI Resources NR-53514 lot 70036345), using QIAamp® Viral RNA Mini Kit (Qiagen 52904). The viral genomic RNA is in a background of cellular nucleic acid and carrier RNA.

Lot: 70037087

Manufacturing Date: 24JUN2020

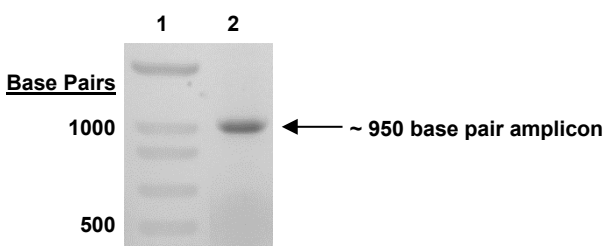
TEST	SPECIFICATIONS	RESULTS
Sequencing of Species-Specific Region (~ 850 nucleotides)	≥ 98% identity with SARS-CoV-2, isolate NY-PV08410 (GenBank: MT370900.1)	100% identity with SARS-CoV-2, isolate NY-PV08410 (GenBank: MT370900.1)
Next-Generation Sequencing (NGS) of Complete Genome Using Illumina® iSeq™ 100 Platform (Refer to Appendix I for NGS information)	≥ 98% identity with SARS-CoV-2, isolate NY-PV08410 (GenBank: MT370900.1)	100% identity with SARS-CoV-2, isolate NY-PV08410 (GenBank: MT370900.1)
Functional Activity by RT-PCR Amplification¹	~ 950 base pair amplicon	~ 950 base pair amplicon (Figure 1)
Estimated Concentration (post-dilution) by RiboGreen® Measurement (Viral, Cellular and Carrier)²	Report results	13.3 ng per 100 µL (0.13 µg/mL)
Estimated Amount per Vial²	Report results	13.3 ng
Genome Copy Number Using BioRad QX200 Droplet Digital PCR (ddPCR™) System (Post vial; 9 replicates)	Report results	9.12 × 10 ⁷ genome equivalents/mL
Virus Inactivation (14 Days, 2 Passages) 10% of total yield inoculated on Vero E6 cells and evaluated for cytopathic effect ³ Passage 1 Passage 2	No viable virus detected No viable virus detected	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 ³ Passage 1 Passage 2	No viable virus detected No viable virus detected	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-53518 in a 50 µL reaction

²Measurement is determined pre-vial prior to dilution due to the limit of detection of the quantification method

³Supernatant 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-53518 by RT-PCR Amplification



Lane 1: Invitrogen™ TrackIt™ 1 Kb Plus DNA Ladder
Lane 2: PCR product from 1 µL of NR-53518

/Heather Couch/
Heather Couch

21 JAN 2021

Program Manager or designee, ATCC Federal Solutions

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APPENDIX I: NGS Information for NR-53518 lot 70037087

Reference sequence from GenBank MT370900.1 contained ambiguous bases (N; referred to as Amb Ref in the Table below) in nine regions. Sequence analysis of NR-53518_70037087 resulted in filling-in of these ambiguous regions of the reference sequence MT370900.1 and demonstrated that these nine ambiguous regions matched GenBank MN908947.3 (SARS-CoV-2, isolate Wuhan-Hu-1, complete genome) sequence. Two SNPs were discovered in NR-53518_70037087 when compared to the reference sequence MT370900.1. Additionally, both the reference sequence MT370900.1 and NR-53518_70037087 contained six SNPs when compared to GenBank MN908947.3 (see Table below). Quality scores over 60 indicate it is improbable that the variant call is incorrect.

Position in NR-53518_70037087 Sequence	Position in MT370900 Reference Sequence	Position in MN908947 Sequence	Reported MN908947 Sequence	Reported MT370900 Reference Sequence	Identified Alternative Base	Quality	Variant Type	Length of Variant	Frequency of Variant
186	186	241	C	T	T	N/A	SNP	1	1.0000000
1004	1004	1059	C	T	T	N/A	SNP	1	1.0000000
2982	2982	3037	C	T	T	N/A	SNP	1	1.0000000
6321	6321	6376	TAATCTTG CC	N (×10)	TAATCTTG CC	N/A	Amb Ref	10	1.0000000
6373	6373	6428	*	N (×698)	*	N/A	Amb Ref	698	1.0000000
14353	14353	14408	C	T	T	N/A	SNP	1	1.0000000
21157	21157	21212	*	N (×170)	*	N/A	Amb Ref	170	1.0000000
21355	21355	21410	*	N (×58)	*	N/A	Amb Ref	58	1.0000000
21417	21417	21472	*	N (×80)	*	N/A	Amb Ref	80	1.0000000
22157	22157	22212	CT	NN	CT	N/A	Amb Ref	2	1.0000000
22173	22173	22228	TTTAGAAC C	NNNNNNN NN	TTTAGAAC C	N/A	Amb Ref	9	1.0000000
22191	22191	22246	*	N (×20)	*	N/A	Amb Ref	20	1.0000000
22218	22218	22273	GTT	NNN	GTT	N/A	Amb Ref	3	1.0000000
23348	23348	23403	A	G	G	N/A	SNP	1	1.0000000
23551	23551	23606	C	C	T	222	SNP	1	0.4391635
25508	25508	25563	G	T	T	N/A	SNP	1	1.0000000
27506	27506	27561	G	G	T	220	SNP	1	0.8291815

* If N > 10, the sequence of the variant is not reported in the table above. For all nucleotides in these variants, the identified alternative base is the same as the reported MN908947.3 sequence.