

Certificate of Analysis for NR-53816

SARS-Related Coronavirus 2, Wuhan-Hu-1 Spike-Pseudotyped Lentiviral Kit V2

Catalog No. NR-53816

Product Description:

Note: The NR-53742 vial label indicates this product has a 21 base pair deletion, but it is a 21 amino acid deletion. The severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), Wuhan-Hu-1 (GenBank: NC 045512) spike-pseudotyped lentiviral kit version 2 (NR-53816) is designed to generate pseudotyped lentiviral particles with the spike (S) glycoprotein gene, as well as luciferase (Luc2) and green fluorescent protein (GFP).

The deposited plasmids were transformed into One Shot™ TOP10 *E. coli* (Invitrogen™ C404003), grown in Luria-Bertani broth with ampicillin (50-100 µg per mL) for 1 day at 37°C in an aerobic atmosphere, extracted using a Plasmid *Plus* Maxi Kit (QIAGEN® 12963) and vialed in TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8.0).

Table 1: Lentiviral Kit Components

COMPONENT NUMBER	DESCRIPTION	LOT NUMBER	DATE OF MANUFACTURE
NR-53742	Viral entry protein encoding for S glycoprotein ∆Cter	70038289	12AUG2020
NR-52516	Lentiviral Backbone encoding for Luc2 and ZsGreen	70035474	29APR2020
NR-52517	Helper plasmid encoding for Gag and Pol	70035478	29APR2020
NR-52518	Helper plasmid encoding for Tat1b	70035480	29APR2020
NR-52519	Helper plasmid encoding for Rev1b	70035482	29APR2020

Table 2: Viral Entry Protein (NR-53742)

TEST	SPECIFICATIONS	RESULTS
Next-Generation DNA Sequencing	~ 8310 base pairs	8316 base pairs ¹
Genotypic Analysis		
Sequencing of S glycoprotein insert (~ 3760 base pairs)	≥ 99% sequence identity to depositor's sequence	100% sequence identity to depositor's sequence ²
Antibiotic Resistance		
Ampicillin (encoded by beta-lactamase gene bla)3	bla sequence present	bla sequence present
Concentration by PicoGreen® Measurement	≥ 2 µg/mL	0.7 μg in 30 μL per vial (22 μg/mL)
Amount per Vial	Report results	0.7 μg per vial
OD ₂₆₀ /OD ₂₈₀ Ratio (pre-vial)	1.7 to 2.1	1.9
Effective Bacterial Transformation Invitrogen™ One Shot™ TOP10 <i>E. coli</i>	≥ 50 colonies per ng	248 colonies per ng

¹The sequence was assembled pre-vial using the depositor's predicted sequence as the reference sequence. The complete plasmid sequence and map are provided on the BEI Resources webpage.

BEI Resources www.beiresources.org E-mail: contact@beiresources.org Tel: 800-359-7370

Fax: 703-365-2898

²The NR-53742 insert was codon optimized for mammalian expression, but otherwise is 100% identical to the SARS-CoV-2, Wuhan-Hu-1 S protein (GenPept: YP_009724390; residues 1-1252).

³The antibiotic ampicillin degrades quickly during growth. Bacterial stationary phase should be minimized during plasmid replication to avoid plasmid loss and increased antibiotic concentrations may be necessary.



Certificate of Analysis for NR-53816

Table 3: Lentiviral Backbone (NR-52516)

TEST	SPECIFICATIONS	RESULTS
Next-Generation DNA Sequencing	~ 9370 base pairs	9369 base pairs ¹
Genotypic Analysis		
Sequencing of Luc2 gene (~ 1650 base pairs)	≥ 99% sequence identity to depositor's sequence	100% sequence identity to depositor's sequence
Sequencing of ZsGreen1 gene (~ 700 base pairs)	≥ 99% sequence identity to depositor's sequence	100% sequence identity to depositor's sequence
Antibiotic Resistance Ampicillin (encoded by beta-lactamase gene bla) ²	bla sequence present	bla sequence present
Concentration by PicoGreen® Measurement	≥ 2 µg/mL	0.2 μg in 20 μL per vial (11 μg/mL)
Amount per Vial	Report results	0.2 μg per vial
OD ₂₆₀ /OD ₂₈₀ Ratio (pre-vial)	1.7 to 2.1	1.8
Effective Bacterial Transformation Invitrogen™ One Shot™ TOP10 <i>E. coli</i>	≥ 50 colonies per ng	> 500 colonies per ng

¹The sequence was assembled pre-vial using the depositor's predicted sequence as the reference sequence. The complete plasmid sequence and map are provided on the BEI Resources webpage.

Table 4: Helper Plasmids (NR-52517 to NR-52519)

TEST	SPECIFICATIONS	RESULTS	
Next-Generation DNA Sequencing	Report results	Consistent with depositor reported size ¹	
Genotypic Analysis			
Sequencing of insertion	≥ 99% sequence identity to depositor's sequence	100% sequence identity to depositor's sequence	
Antibiotic Resistance			
Ampicillin (encoded by beta-lactamase gene bla)2	bla sequence present	bla sequence present	
Neomycin [NR-52519, encoded by aminoglycoside	aph(3')-II sequence present	aph(3')-II sequence present	
3'-phosphotransferase gene aph(3')-II]			
Concentration by PicoGreen® Measurement			
NR-52517	≥ 2 µg/mL	0.2 μg in 20 μL per vial (12 μg/mL)	
NR-52518	≥ 2 µg/mL	0.8 μg in 50 μL per vial (15 μg/mL)	
NR-52519	≥ 2 µg/mL	0.9 μg in 70 μL per vial (13 μg/mL)	
Amount per Vial			
NR-52517	Report results	0.2 μg	
NR-52518	Report results	0.8 μg	
NR-52519	Report results	0.9 μg	
OD ₂₆₀ /OD ₂₈₀ Ratio (pre-vial)	1.7 to 2.1	1.7 to 2.1	
Effective Bacterial Transformation			
Invitrogen™ One Shot™ TOP10 <i>E. coli</i>	≥ 50 colonies per ng	> 500 colonies per ng	

¹The sequence was assembled pre-vial using the depositor's predicted sequence as the reference sequence. The complete plasmid sequence and map are provided on the BEI Resources webpage.

BEI Resources www.beiresources.org E-mail: contact@beiresources.org
Tel: 800-359-7370

Fax: 703-365-2898

²The antibiotic ampicillin degrades quickly during growth. Bacterial stationary phase should be minimized during plasmid replication to avoid plasmid loss and increased antibiotic concentrations may be necessary.

²The antibiotic ampicillin degrades quickly during growth. Bacterial stationary phase should be minimized during plasmid replication to avoid plasmid loss and increased antibiotic concentrations may be necessary.



Certificate of Analysis for NR-53816

/Heather Couch/ Heather Couch

22 JAN 2021

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 by ATCC® and the contributor 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.

ATCC® is a trademark of the American Type Culture Collection.

You are authorized to use this product for research use only. It is not intended for human use.

BEI Resources www.beiresources.org E-mail: contact@beiresources.org Tel: 800-359-7370

Fax: 703-365-2898