

Vector pHDM Containing the SARS-Related Coronavirus 2, Wuhan-Hu-1 Spike Glycoprotein Gene with C-Terminal Deletion

Catalog No. NR-53742

Product Description:

Note: The vial label indicates this product has a 21 base pair deletion but is a 21 amino acid deletion. The vector for the spike (S) glycoprotein gene from severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), Wuhan-Hu-1 (GenBank: [MN908947](#)) was designed by codon optimization of the S glycoprotein sequence (residues 1 to 1252), and deletion of the C-terminal 21 amino acids, and subcloned into the pHDM vector under the CMV promoter. NR-53742 contains the beta-lactamase gene, *bla*, to provide transformant selection through ampicillin resistance in *Escherichia coli* (*E. coli*). The deposited plasmid was transformed into One Shot™ TOP10 *E. coli* (Invitrogen™ C404003), grown in Luria-Bertani broth with ampicillin (100 µg per mL) for 1 day at 37°C in an aerobic atmosphere, extracted using a Plasmid Plus Maxi Kit (QIAGEN® 12963) and vialled in TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8.0).

Lot: 70038289

Manufacturing Date: 12AUG2020

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 <i>bla</i>) ³	<i>bla</i> sequence present	<i>bla</i> 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
OD₂₆₀/OD₂₈₀ 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.

²NR-53742 insert was codon optimized for mammalian expression, but otherwise is consistent with 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.

/Heather Couch/

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22 JAN 2021

Program Manager or designee, ATCC Federal Solutions

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