Modified pαH Vector Containing the Middle East Respiratory Syndrome Coronavirus Spike Glycoprotein

Catalog No. NR-54980
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Manufacturer:
BEI Resources

Product Description:
The vector for the spike (S) glycoprotein gene from middle east respiratory syndrome coronavirus (MERS-CoV), England 1 (GenBank: NC_038294) was designed by codon optimizing the full-length S sequence (residues 1 to 1291) for mammalian expression and subcloning into the pαH mammalian expression vector, which was modified by subcloning a T4 foldon trimerization domain, a HRV3C protease cleavage site and the octa-histidine and 2X Strep-tag® II tags downstream of the open reading frame.¹,² The recombinant protein is stabilized by substitution at the furin S1/S2 cleavage site (RVSR->ASVG; residues 748 to 751) and VL→PP mutations (residues 1060 and 1061). NR-54980 contains the beta-lactamase gene, bla, to provide transformant selection through ampicillin resistance in Escherichia coli (E. coli). NR-54980 is also referred to as VRC7578.¹ The resulting size of the plasmid is approximately 8000 base pairs. The complete plasmid sequence and map are provided on the BEI Resources webpage. The plasmid was produced in E. coli and extracted.

The S glycoprotein mediates viral binding to the host angiotensin converting enzyme 2 (ACE2). This protein forms a trimer, and when bound to a host receptor allows fusion of the viral and cellular membranes. The S protein is a target for neutralizing antibodies.³

Material Provided:
Each vial contains plasmid DNA in TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8.0). The DNA concentration and volume provided are shown on the Certificate of Analysis. The vial should be centrifuged prior to opening. Note: The contents of the vial should be used to replicate the plasmid in E. coli prior to mammalian expression.

Packaging/Storage:
NR-54980 was packaged aseptically in screw-capped plastic cryovials. The product is provided frozen on dry ice and should be stored at -20°C or colder immediately upon arrival. Freeze-thaw cycles should be minimized.

Citation:
Acknowledgment for publications should read “The following reagent was obtained through BEI Resources, NIAID, NIH: Modified pαH Vector Containing the Middle East Respiratory Syndrome Coronavirus Spike Glycoprotein, NR-54980.”

Biosafety Level: 1

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References:

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