pUNO1-SpikeV13
Expression vector encoding the SARS-CoV-2 Omicron variants (BA.4/BA.5 lineages) Spike gene
Catalog code: p1-spike-v13
https://www.invivogen.com/omicron-ba4ba5-spike-expression-vectors

For research use only
Version 22G19-AK

PRODUCT INFORMATION
Contents
• 20 µg of lyophilized pUNO1-SpikeV13 (plasmid DNA)
• 2 x 1 ml of Blasticidin (10 mg/ml)

Storage and Stability
• Product is shipped at room temperature.
• Lyophilized DNA should be stored at -20°C.
• Resuspended DNA is stable for 1 year at -20°C.
• Store Blasticidin at 4°C or -20°C. The expiry date is specified on the product label.

Quality control
• Plasmid construct is confirmed by restriction analysis and full-length open reading frame (ORF) sequencing.
• After purification by ion exchange chromatography, predominant supercoiled confirmation is verified by electrophoresis.

PLASMID FEATURES
Omicron Variant (BA.4/BA.5 lineages) SARS-CoV-2 Spike cassette
• EF-1α/HTLV hybrid promoter is a composite promoter comprised of the Elongation Factor-1α (EF-1α) core promoter and the 5’ untranslated region of the Human T-Cell Leukemia Virus (HTLV). EF-1α utilizes a type 2 promoter that encodes a “house-keeping” gene. It is expressed at high levels in all cell cycles and lower levels during the G0 phase. Additionally, since the promoter is not tissue-specific it is highly expressed in all cell types. The R segment and part of the U5 sequence (R-US5) of the HTLV Type 1 Long Terminal Repeat has been coupled to the EF-1α promoter to enhance stability of DNA and RNA. This modification not only increases steady state transcription, but also significantly increases translation efficiency.
• Codon-optimized Spike ORF
pUNO1-SpikeV13 encodes the Spike protein from the SARS-CoV-2 Omicron BA.4 and BA.5 variants, first reported in South Africa in early 2022. These variants share the same Spike protein. They are characterized by several of mutations and deletions within the Spike coding sequence (see below)3,4. Additionally, to improve expression of the S protein in cell lines, the gene is codon-optimized and the last 19 amino acids, which contain an endoplasmic reticulum (ER)-retention motif (KxHxx), have been removed3,5.
- S1/S2 boundary: Functional furin cleavage site
- S2 domain: N764K, D796Y, Q954H, N969K

Spike (S) is a structural glycoprotein expressed on the surface of SARS-CoV-2. It mediates membrane fusion and viral entry into target cells upon binding to the host receptor ACE2 and the proteolytic activity of host proteases such as furin and TMPRSS22.

For more information visit: https://www.invivogen.com/sars2-spike

METHODS
• Plasmid resuspension
  - Quickly spin the tube containing the lyophilized plasmid to pellet the DNA.
  - To obtain a plasmid solution at 1 µg/µl, resuspend the DNA in 20 µl of sterile water.
  - Store the resuspended plasmid at -20°C.
• Plasmid amplification and cloning

Plasmid amplification and cloning can be performed in E. coli GT116 or other commonly used laboratory E. coli strains, such as DH5α.

Blasticidin usage
Blasticidin should be used at 25-100 µg/ml in bacteria and 1-30 µg/ml in mammalian cells. Blasticidin is supplied as a 10 mg/ml colorless solution in HEPES buffer.

TECHNICAL SUPPORT
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REFERENCES


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For a complete list of InvivoGen’s COVID-19 related products visit: https://www.invivogen.com/covid-19