Recombinant SARS-CoV-2 Spike (Mink Cluster-5) His-tag

Catalog Number: 10736-CV

DESCRIPTION

Source	Human embryonic kidney cell, HEK293-derived sars-cov-2 Spike protein Val16-Lys1211 (His69-Val70 del & Tyr453Phe, Asp614Gly, Ile692Val)(Arg682Ser, Arg685Ser, Lys986Pro, Val987Pro), with a C-terminal 6-His tag Accession # YP_009724390.1
N-terminal Sequence Analysis	Val 16
Predicted Molecular Mass	134 kDa

SPECIFICATIONS

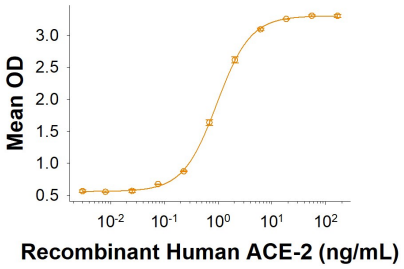
SDS-PAGE	154-170 kDa, under reducing conditions
Activity	Measured by its binding ability in a functional ELISA with Recombinant Human ACE-2 His-tag (Catalog # 933-ZN).
Endotoxin Level	<0.10 EU per 1 µg of the protein by the LAL method.
Purity	>95%, by SDS-PAGE visualized with Silver Staining and quantitative densitometry by Coomassie® Blue Staining.
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 500 µg/mL in PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 3 months, -20 to -70 °C under sterile conditions after reconstitution.

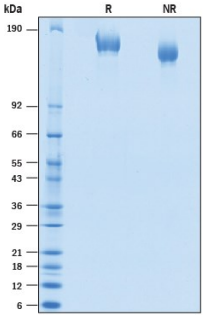
DATA

Bioactivity



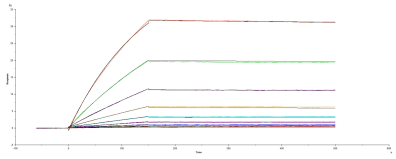
Recombinant SARS-CoV-2 Spike (Mink Cluster-5) His-tag Protein Bioactivity Recombinant SARS-CoV-2 Spike (Mink Cluster-5) His-tag (Catalog # 10736-CV) binds Recombinant Human ACE-2 His-tag (Catalog # 933-ZN) in a functional ELISA.

SDS-PAGE



Recombinant SARS-CoV-2 Spike (Mink Cluster-5) His-tag Protein SDS-PAGE. 2 µg/lane of Recombinant SARS-CoV-2 Spike (Mink Cluster-5) His-tag (Catalog # 10736-CV) was resolved with SDS-PAGE under reducing (R) and non-reducing (NR) conditions and visualized by Coomassie® Blue staining, showing bands at 154-170 kDa.

Surface Plasmon Resonance (SPR)



Binding of ACE-2 to Mink Cluster-5 variant Spike protein by surface plasmon resonance (SPR). Recombinant SARS-CoV-2 Mink Cluster-5 variant Spike protein His-tag was immobilized on a Biacore Sensor Chip CM5, and binding to recombinant human ACE-2 (Catalog # 933-ZN) was measured at a concentration range between 0.18 nM and 47.2 nM. The double-referenced sensorgram was fit to a 1:1 binding model to determine the binding kinetics and affinity, with an affinity constant of KD=0.3411 nM.

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BACKGROUND

SARS-CoV-2, which causes the global pandemic coronavirus disease 2019 (Covid-19), belongs to a family of viruses known as coronaviruses that are commonly comprised of four structural proteins: Spike protein (S), Envelope protein (E), Membrane protein (M), and Nucleocapsid protein (N) (1). SARS-CoV-2 Spike Protein (S Protein) is a glycoprotein that mediates membrane fusion and viral entry. The S protein is homotrimeric, with each ~180-kDa monomer consisting of two subunits, S1 and S2 (2). In SARS-CoV-2, as with most coronaviruses, proteolytic cleavage of the S protein into the S1 and S2 subunits is required for activation. The S1 subunit is focused on attachment of the protein to the host receptor while the S2 subunit is involved with cell fusion (3-5). A SARS-CoV-2 variant carrying the S protein amino acid (aa) change D614G has become the most prevalent form in the global pandemic and has been associated with greater infectivity and higher viral load (6, 7). The S protein of SARS-CoV-2 shares 75% and 29% aa sequence identity with S protein of SARS-CoV-1 and MERS, respectively. The S Protein of the SARS-CoV-2 virus, like the SARS-CoV-1 counterpart, binds Angiotensin-Converting Enzyme 2 (ACE2), but with much higher affinity and faster binding kinetics through the receptor binding domain (RBD) located in the C-terminal region of S1 (8). It has been demonstrated that the S Protein can invade host cells through the CD147/EMMPRIN receptor and mediate membrane fusion (9, 10). Cluster 5, also known as DFVI-spike is a variant of the virus strain that was first discovered in Denmark and infected more than 200 mink farms by the end of November 2020 (11). Evidence shows virus has been transmitted from mink farms to human in a zoonotic manner (12). WHO claims that this mutant has moderately decreased sensitivity to the neutralization antibody.

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