Recombinant SARS-CoV-2 D614G Spike His-tag

Catalog Number: 10620-CV

DESCRIPTION

Source	Chinese Hamster Ovary cell line, CHO-derived sars-cov-2 Spike protein Val16-Lys1211 (Asp614Gly, Arg682Ser, Arg685Ser, Lys986Pro, Val987Pro), with a C-terminal 6-His tag Accession # YP_009724390.1
N-terminal Sequence Analysis	Val16
Predicted Molecular Mass	134 kDa

SPECIFICATIONS

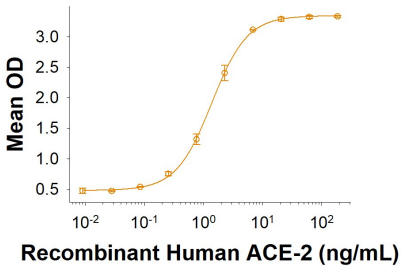
SDS-PAGE	150-170 kDa, under reducing conditions
Activity	Measured by its binding ability in a functional ELISA with Recombinant Human ACE-2 His-tag (Catalog # 933-ZN).
Endotoxin Level	<0.10 EU per 1 µg of the protein by the LAL method.
Purity	>95%, by SDS-PAGE visualized with Silver Staining and quantitative 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.

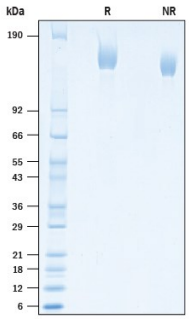
DATA

Binding Activity



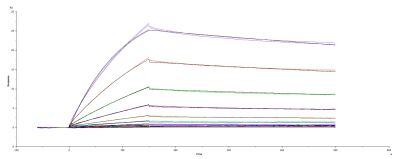
Recombinant SARS-CoV-2 D614 G Spike His-tag, Protein Binding Activity. Recombinant SARS-CoV-2 D614G Spike His-tag (Catalog # 10620-CV) binds Recombinant Human ACE-2 His-tag (Catalog # 933-ZN) in a functional ELISA.

SDS-PAGE



Recombinant SARS-CoV-2 D614G Spike His-tag, Protein SDS-PAGE. 2 µg/lane of Recombinant SARS-CoV-2 D614G Spike His-tag, Protein (Catalog # 10620-CV) was resolved with SDS-PAGE under reducing (R) and non-reducing (NR) conditions and visualized by Coomassie® Blue staining, showing bands at 150-170 kDa.

Surface Plasmon Resonance (SPR)



Binding of ACE-2 to SARS-CoV-2 Spike protein, D614G mutant by surface plasmon resonance (SPR). Recombinant SARS-CoV-2 Spike protein D614G 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 0.092 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=2.099 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). It is proposed that the D614G mutation introduces an additional elastase cleavage site near the S1-S2 junction and this additional processing helps with viral entry (11).

References:

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