

DESCRIPTION

Source	Human embryonic kidney cell, HEK293-derived sars-cov-2 Spike RBD protein		
	SARS-CoV-2 B.1.1.529 Spike RBD (Arg319-Phe541) (Gly339Asp, Ser371Leu, Ser373Pro, Ser375Phe, Lys417Asn, Asn440Lys, Gly446Ser, Ser477Asn, Thr478Lys, Glu484Ala, Gln493Arg, Gly496Ser, Gln498Arg, Asn501Tyr, Tyr505His) Accession # YP_009724390.1	IEGRMD	Human IgG ₁ (Pro100-Lys330)
	N-terminus		C-terminus
N-terminal Sequence Analysis	Arg319		
Structure / Form	Disulfide-linked homodimer		
Predicted Molecular Mass	52 kDa		

SPECIFICATIONS

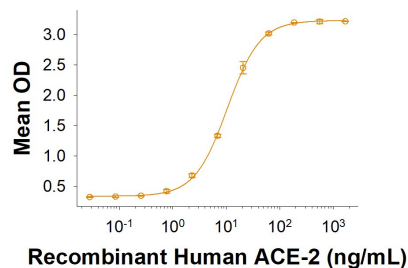
SDS-PAGE	57-67 kDa, under reducing conditions.
Activity	Measured by its binding ability in a functional ELISA with Recombinant Human ACE-2 His-tag (Catalog # 933-ZN).
Endotoxin Level	<0.10 EU per 1 µg of the protein by the LAL method.
Purity	>90%, by SDS-PAGE visualized with Silver Staining and quantitative 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. <ul style="list-style-type: none"> • 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.

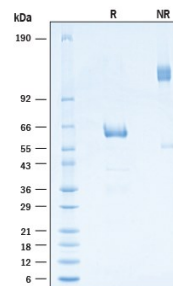
DATA

Binding Activity



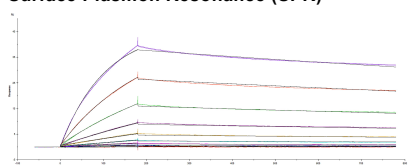
Recombinant SARS-CoV-2 B.1.1.529 Spike RBD Fc Chimera Protein Binding Activity. Recombinant SARS-CoV-2 B.1.1.529 Spike RBD Fc Chimera (Catalog # 11057-CV) binds Recombinant Human ACE-2 His-tag (Catalog # 933-ZN) in a functional ELISA.

SDS-PAGE



Recombinant SARS-CoV-2 B.1.1.529 Spike RBD Fc Chimera Protein SDS-PAGE. 2 µg/lane of Recombinant SARS-CoV-2 B.1.1.529 Spike RBD Fc Chimera Protein (Catalog # 11057-CV) was resolved with SDS-PAGE under reducing (R) and non-reducing (NR) conditions and visualized by Coomassie® Blue staining, showing bands at 57-67 kDa and 114-134 kDa, respectively.

Surface Plasmon Resonance (SPR)



Recombinant SARS-CoV-2 B.1.1.529 Spike RBD Fc Chimera Protein binds to Recombinant Human ACE-2 by surface plasmon resonance
Recombinant SARS-CoV-2 B.1.1.529 Spike RBD Fc Chimera Protein (Catalog #11057-CV) 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 0.021 nM and 53.1 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 $K_d=3.63$ nM.

BACKGROUND

SARS-CoV-2, which causes the global pandemic coronavirus disease 2019 (Covid-19), belongs to a family of viruses known as coronaviruses that also include MERS-CoV and SARS-CoV-1. Coronaviruses are commonly comprised of four structural proteins: Spike protein (S), Envelope protein (E), Membrane protein (M) and Nucleocapsid protein (N) (1). The SARS-CoV-2 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 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 receptor binding domain (RBD) in the C-terminus of the S1 subunit has been identified and the RBD of SARS-CoV-2 shares 73% amino acid (aa) identity with the RBD of the SARS-CoV-1, but only 22% aa identity with the RBD of MERS-CoV (6, 7). The low aa sequence homology is consistent with the finding that SARS and MERS-CoV bind different cellular receptors (8). The RBD of SARS-CoV-2 binds a metalloprotease, angiotensin-converting enzyme 2 (ACE-2), similar to SARS-CoV-1, but with much higher affinity and faster binding kinetics (9). Before binding to the ACE-2 receptor, structural analysis of the S1 trimer shows that only one of the three RBD domains is in the "up" conformation. This is an unstable and transient state that passes between trimeric subunits but is nevertheless an exposed state to be targeted for neutralizing antibody therapy (10). Polyclonal antibodies to the RBD of the SARS-CoV-2 protein have been shown to inhibit interaction with the ACE-2 receptor, confirming RBD as an attractive target for vaccinations or antiviral therapy (11). There is also promising work showing that the RBD may be used to detect presence of neutralizing antibodies present in a patient's bloodstream, consistent with developed immunity after exposure to the SARS-CoV-2 (12). Several emerging SARS-CoV-2 genomes have been identified including the Omicron, or B.1.1.529, variant. First identified in November 2021 in South Africa, the Omicron variant quickly became the predominant SARS-CoV-2 variant and is considered a variant of concern (VOC). The Omicron variant contains 15 mutations in RBD domain that potentially affect viral fitness and transmissibility. The majority of the mutations are involved in ACE-2 binding and Omicron binds ACE-2 with greater affinity, potentially explaining its increased transmissibility (13, 14). Several of these mutations are also identified in facilitating immune escape and reducing neutralization activity to several monoclonal antibodies (13). Additionally, a series of new mutations are present in the RBD which have unknown impacts on receptor binding or antibody neutralization.

References:

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