

DESCRIPTION

Source	Human embryonic kidney cell, HEK293-derived sars-cov-2 Spike RBD protein Arg319-Phe541 (Glu484Lys, Ser494Pro, Asn501Tyr), with a C-terminal 6-His tag Accession # YP_009724390.1
N-terminal Sequence Analysis	Arg319
Predicted Molecular Mass	26 kDa

SPECIFICATIONS

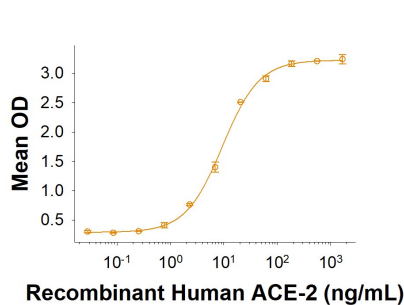
SDS-PAGE	33-40 kDa, under reducing conditions.
Activity	Measured by its binding ability in a functional ELISA with Recombinant Human ACE-2 His-tag (Catalog # 933-ZN).
Endotoxin Level	<0.10 EU per 1 µg of the protein by the LAL method.
Purity	>95%, by SDS-PAGE visualized with Silver Staining and quantitative 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	<p>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</p> <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.

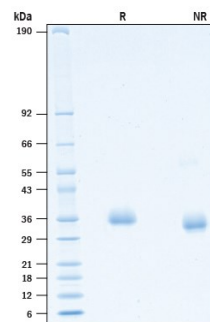
DATA

Binding Activity



Recombinant SARS-CoV-2 B.1.324.1 Spike RBD His-tag Protein Binding Activity.
Recombinant SARS-CoV-2 B.1.324.1 Spike RBD His-tag (Catalog # 10830-CV) binds Recombinant Human ACE-2 His-tag (Catalog # 933-ZN) in a functional ELISA.

SDS-PAGE



Recombinant SARS-CoV-2 B.1.324.1 Spike RBD His-tag Protein SDS-PAGE. 2 µg/lane of Recombinant SARS-CoV-2 B.1.324.1 Spike RBD His-tag Protein (Catalog # 10830-CV) was resolved with SDS-PAGE under reducing (R) and non-reducing (NR) conditions and visualized by Coomassie® Blue staining, showing bands at 33-40 kDa.

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 metalloproteinase, 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 ACE2 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 with mutations in the RBD compared to the Wuhan-Hu-1 SARS-CoV-2 reference sequence. The B.1.324.1 variant was identified as a Variant of Concern (VOC) as it contains several mutations of interest in the RBD domain that effect viral fitness and transmissibility: E484K, S494P, and N501Y (13). Structural analysis points to E484K as a potentially crucial mutation as it creates a new site for hACE-2 binding and may enhance binding affinity (14). S494P is also located within the ACE2 receptor binding domain and experimental evidence suggests that mutations at this position decrease antibody binding affinity (15). N501Y is thought to enhance binding affinity for hACE-2 and make the virus more easily transmissible (16, 17). Additionally, the E484K substitution alone has been shown to confer resistance to several monoclonal antibodies and is responsible for the first confirmed SARS-CoV-2 reinfection (18).

References:

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