

**DESCRIPTION**

<b>Source</b>	Human embryonic kidney cell, HEK293-derived sars-cov-2 Spike protein		
	SARS-CoV-2 Spike (Val16-Lys1211)(His69del, Val70del, Tyr145del, Glu484Lys, Asn501Tyr, Ala570Asp, Asp614Gly, Pro681His, Thr716Iso, Ser982Ala, Asp1118His) (Arg682Ser, Arg685Ser, Lys986Pro, Val987Pro) Accession # YP_009724390.1	GCN4-IZ	6-His tag
	N-terminus		C-terminus

<b>N-terminal Sequence Analysis</b>	Val16
<b>Predicted Molecular Mass</b>	138 kDa

**SPECIFICATIONS**

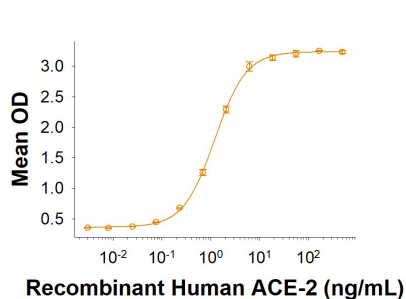
<b>SDS-PAGE</b>	140-170 kDa, under reducing conditions.
<b>Activity</b>	Measured by its binding ability in a functional ELISA with Recombinant Human ACE-2 His-tag (Catalog # <a href="#">933-ZN</a> ).
<b>Endotoxin Level</b>	<0.10 EU per 1 µg of the protein by the LAL method.
<b>Purity</b>	>95%, by SDS-PAGE visualized with Silver Staining and quantitative densitometry by Coomassie® Blue Staining.
<b>Formulation</b>	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details.

**PREPARATION AND STORAGE**

<b>Reconstitution</b>	Reconstitute at 500 µg/mL in PBS.
<b>Shipping</b>	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.
<b>Stability &amp; Storage</b>	<p><b>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</b></p> <ul style="list-style-type: none"> <li>• 12 months from date of receipt, -20 to -70 °C as supplied.</li> <li>• 1 month, 2 to 8 °C under sterile conditions after reconstitution.</li> <li>• 3 months, -20 to -70 °C under sterile conditions after reconstitution.</li> </ul>

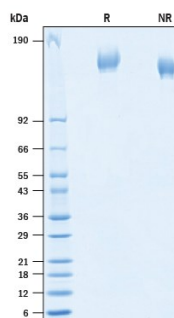
DATA

Binding Activity



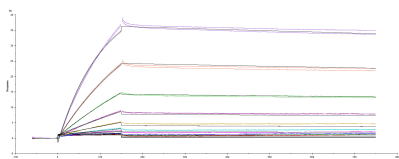
**Recombinant SARS-CoV-2 +E484K B.1.1.7 Spike (GCN4-IZ) His-tag Protein Binding Activity.** Recombinant SARS-CoV-2 +E484K B.1.1.7 Spike (GCN4-IZ) His-tag (Catalog # 10824-CV) binds Recombinant Human ACE-2 His-tag (Catalog # 933-ZN) in a functional ELISA.

SDS-PAGE



**Recombinant SARS-CoV-2 +E484K B.1.1.7 Spike (GCN4-IZ) His-tag Protein SDS-PAGE.** 2 µg/lane of Recombinant SARS-CoV-2 +E484K B.1.1.7 Spike (GCN4-IZ) His-tag Protein (Catalog # 10824-CV) was resolved with SDS-PAGE under reducing (R) and non-reducing (NR) conditions and visualized by Coomassie® Blue staining, showing bands at 140-170 kDa.

Surface Plasmon Resonance (SPR)



**Binding of ACE-2 to SARS-CoV-2 Spike protein UK variant with the E484K mutation by surface plasmon resonance (SPR).** Recombinant SARS-CoV-2 B.1.1.7 UK variant Spike protein with the E484K mutation 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 0.046 nM and 23.6 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  $K_D=0.2762$  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). The S protein of SARS-CoV-2 shares 75% and 29% amino acid 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 a metalloproteinase, Angiotensin-Converting Enzyme 2 (ACE-2), but with much higher affinity and faster binding kinetics through the receptor binding domain (RBD) located in the C-terminal region of S1 subunit (6). It has been demonstrated that the S Protein can invade host cells through the CD147/EMMPRIN receptor and mediate membrane fusion (7, 8). 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 (9). 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 (10). Several emerging SARS-CoV-2 genomes have been identified including the B.1.1.7 (United Kingdom) variant (11). The B.1.1.7 variant contains a significant mutation of interest in the RBD domain, N501Y, which has been shown to result in enhanced binding affinity for hACE-2 (12). Additionally, the B.1.1.7 variant appears to more easily transmissible, exhibit increased viral loads and, potentially, be associated with higher mortality rates compared to preexisting variants (11, 3). 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 (14). 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 (15).

**References:**

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