

#### DESCRIPTION

**Source** Human embryonic kidney cell, HEK293-derived sars-cov-2 Spike RBD protein  
Arg319-Phe541 (Phe486Ile), with a C-terminal 6-His tag  
Accession # YP\_009724390.1

**N-terminal Sequence Analysis** Arg319

**Predicted Molecular Mass** 26 kDa

#### SPECIFICATIONS

**SDS-PAGE** 31-38 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.

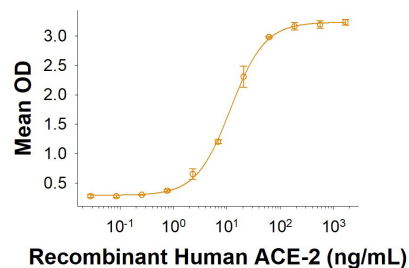
**Shipping** The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.

**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.

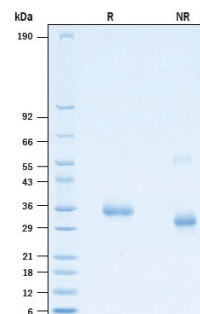
#### DATA

##### Binding Activity



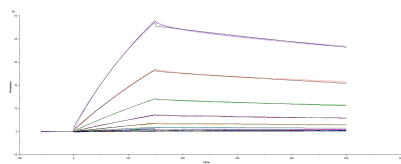
**Recombinant SARS-CoV-2 F486I Spike RBD His-tag Protein Binding Activity.** Recombinant SARS-CoV-2 F486I Spike RBD His-tag (Catalog # 10790-CV) binds Recombinant Human ACE-2 His-tag (Catalog # 933-ZN) in a functional ELISA.

##### SDS-PAGE



**Recombinant SARS-CoV-2 F486I Spike RBD His-tag Protein SDS-PAGE.** 2 µg/lane of Recombinant SARS-CoV-2 F486I Spike RBD His-tag (Catalog # 10790-CV) was resolved with SDS-PAGE under reducing (R) and non-reducing (NR) conditions and visualized by Coomassie® Blue staining, showing bands at 31-38 kDa.

##### Surface Plasmon Resonance (SPR)



**Binding of ACE-2 to SARS-CoV-2 Spike RBD protein with F486I mutation by surface plasmon resonance (SPR).** Recombinant SARS-CoV-2 Spike RBD protein mutant F486I 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  $K_D = 7.806$  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 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 metalloprotease, angiotensin-converting enzyme 2 (ACE-2), has been identified as a functional receptor for SARS-CoV-2 through interaction with a receptor binding domain (RBD) located at the C-terminus of S1 subunit (6,7). 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. SARS-CoV-2 variant carrying the aa substitution F486I was observed in linkage with E484A and might escape binding from certain therapeutic antibodies (8).

**References:**

1. Wu, F. *et al.* (2020) *Nature* **579**:265.
2. Tortorici, M.A. and D. Vesler (2019) *Adv. Virus Res.* **105**:93.
3. Bosch, B.J. *et al.* (2003) *J. Virol.* **77**:8801.
4. Belouzard, S. *et al.* (2009) *Proc. Natl. Acad. Sci.* **106**:5871.
5. Millet, J.K. and G.R. Whittaker (2015) *Virus Res.* **202**:120.
6. Li, W. *et al.* (2003) *Nature* **426**:450.
7. Wong, S.K. *et al.* (2004) *J. Biol. Chem.* **279**:3197.
8. Starr, T.N. *et al.* (2021) *Science* **371**:850.