Recombinant SARS-CoV-2 Spike RBD His-tag Alexa Fluor® 488

Catalog Number: AFG10500

DESCRIPTION

Source	Human embryonic kidney cell, HEK293-derived sars-cov-2 Spike RBD protein Arg319-Phe541, with a C-terminal 6-His tag Accession # YP_009724390.1
N-terminal Sequence Analysis	Arg319
Structure / Form	Labeled with Alexa Fluor® 488 via amines Excitation Wavelength: 488 nm Emission Wavelength: 515-545 nm
Predicted Molecular Mass	26 kDa

SPECIFICATIONS

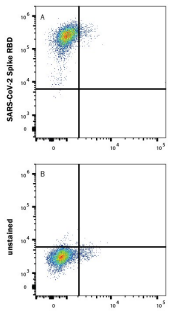
SDS-PAGE	28-38 kDa, under reducing conditions.
Activity	Measured by flow cytometry for its ability to bind HEK293 human embryonic kidney cells transfected with human ACE-2 at 0.50-3.00 µg/mL (100 µL/well). Please Note: Optimal dilutions should be determined by each laboratory for each application.
Endotoxin Level	<1.0 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	Supplied as a 0.2 µm filtered solution in PBS with BSA as a carrier protein. See Certificate of Analysis for details.

PREPARATION AND STORAGE

Shipping	The product is shipped with dry ice or equivalent. Upon receipt, store it immediately at the temperature recommended below.
Stability & Storage	**Protect from light. Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** • 6 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after opening. • 3 months, -20 to -70 °C under sterile conditions after opening.

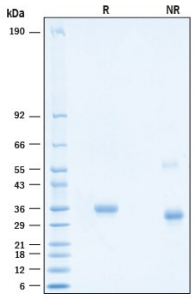
DATA

Flow Cytometry



Detection of ACE-2 on HEK293 Transfectants with Recombinant SARS-CoV-2 Spike RBD His-tag Alexa Fluor® 488 by Flow Cytometry. HEK293 human embryonic kidney cells transfected with human ACE-2 were stained with (A) 1 µg/mL (100 µL/well) Recombinant SARS-CoV-2 Spike RBD His-tag Alexa Fluor® 488 Protein (Catalog # AFG10500) or (B) unstained.

SDS-PAGE



Recombinant SARS-CoV-2 Spike His-tag RBD Alexa Fluor® 488 Protein SDS-PAGE 2 µg/lane of Recombinant SARS-CoV-2 Spike RBD Alexa Fluor® 488 Protein (Catalog # AFG10500) was resolved with SDS-PAGE under reducing (R) and non-reducing (NR) conditions and visualized by Coomassie® Blue staining, showing bands at 28-38 kDa.

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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 two distinct peptides, 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). Based on structural biology studies, the receptor binding domain (RBD), located in the C-terminal region of S1, can be oriented either in the up/standing or down/lying state (6). The standing state is associated with higher pathogenicity and both SARS-CoV-1 and MERS can access this state due to the flexibility in their respective RBDs. A similar two-state structure and flexibility is found in the SARS-CoV-2 RBD (7). Based on amino acid (aa) sequence homology, the SARS-CoV-2 S1 subunit RBD has 73% identity with the RBD of the SARS-CoV-1 S1 RBD, but only 22% homology with the MERS S1 RBD. The low aa sequence homology is consistent with the finding that SARS and MERS bind different cellular receptors (8). 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 (9). Before binding to the ACE2 receptor, structural analysis of the S1 trimer shows that only one of the three RBD domains in the trimeric structure 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 virus (12). Lastly, it has been demonstrated the S Protein can invade host cells through the CD147/EMMPRIN receptor and mediate membrane fusion (13).

References:

1. Wu, F. *et al.* (2020) Nature **579**:265.
2. Tortorici, M.A. and D. Veesler (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. Yuan, Y. *et al.* (2017) Nat. Commun. **8**:15092.
7. Walls, A.C. *et al.* (2010) Cell **180**:281.
8. Jiang, S. *et al.* (2020) Trends. Immunol. **41**:355.
9. Ortega, J.T. *et al.* (2020) EXCLI J. **19**:410.
10. Wrapp, D. *et al.* (2020) Science **367**:1260.
11. Tai, W. *et al.* (2020) Cell. Mol. Immunol. **17**:613.
12. Okba, N. M. A. *et al.* (2020). Emerg. Infect. Dis. **26**:1478
13. Wang, K. *et al.* (2020) Signal Transduct. Target Ther. **5**:283.

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