

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

Catalog Number: AFG11094

DESCRIPTION	
Source	Human embryonic kidney cell, HEK293-derived sars-cov-2 Spike RBD protein Arg319-Phe541 (Gly339Asp, Ser371Phe, Ser373Pro, Ser375Phe, Thr376Ala, Asp405Asn, Arg408Ser, Lys417Asn, Asn440Lys, Ser477Asn, Thr478Lys, Glu484Ala, Gln493Arg, Gln498Arg, Asn501Tyr, Tyr505His), 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	33-39 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 0.500-2.00 μg/mL (100 μL/well). Please Note: Optimal dilutions should be determined by each laboratory for each application.
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	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.	
	<ul> <li>6 months from date of receipt, -20 to -70 °C as supplied.</li> </ul>	

- 1 month, 2 to 8 °C under sterile conditions after opening.
- 3 months, -20 to -70 °C under sterile conditions after opening.

# Flow Cytometry

DATA

Detection of ACE-2 on HEK293 Transfectants with Recombinant SARS-CoV-2 BA.2 Spike RBD His-tag Alexa Fluor® 488 Protein by Flow Cytometry. HEK293 human embryonic kidney cells transfected with Al Recombinant SARS-CoV-2 BA.2 Spike RBD His-tag Alexa Fluor® 488 (Catalog # AFG11094, filled histogram) or (B) unstained (open histogram).

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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 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 metallopeptidase, 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. Additionally, several subvariants of Omicron have been discovered, including the BA.2 or 'stealth' variant. First identified in November 2021 in South Africa, the Omicron variant quickly became the predominant SARS-CoV-2 variant, with BA.2 now the primary sub-lineage. The Omicron BA.2 variant contains 16 mutations in RBD domain, with 3 new mutations and 2 other mutations eliminated compared to the original Omicron variant. The majority of the mutations are involved in ACE-2 binding and Omicron binds ACE-2 with greater affinity, potentially explaining its increased transmissibility and viral fitness (13, 14). Several of the RBD mutations are also identified in facilitating immune escape and reducing neutralization activity to several monoclonal antibodies (13). Additionally, a series of novel mutations are present in the RBD which have unknown impacts on receptor binding or antibody neutralization. The BA.2 subvariant is predicted to be up to about 35 percent more transmissible than the original Omicron variant.

### References:

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