Biotinylated Recombinant SARS-CoV-2 B.1.1.7 N501Y Spike RBD His-tag Avi-tag

Catalog Number: AVI10730

DESCRIPTION

Source

Human embryonic kidney cell, HEK293-derived sars-cov-2 Spike RBD protein

SARS-CoV-2 B.1.1.7 Spike RBD (Arg319-Phe541) (Asn501Tyr) Accession # YP_009724390.1	6-His tag	Avi-tag

N-terminus C-terminus

N-terminal Sequence Analysis	Arg319
Structure / Form	Biotinylated via Avi-tag
Predicted Molecular Mass	29 kDa

SPECIFICATIONS

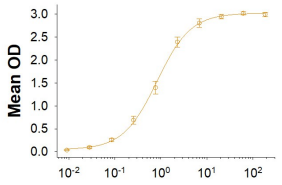
SDS-PAGE	36-40 kDa, under reducing conditions.
Activity	Measured by its binding ability in a functional ELISA with Recombinant Human ACE-2 Fc Chimera (Catalog # 10544-ZN). The biotin to protein ratio is greater than 0.7 as determined by the HABA assay.
Endotoxin Level	<0.10 EU per 1 µg of the protein 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

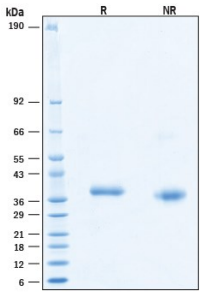
Binding Activity



Biotinylated Recombinant SARS-CoV-2 B.1.1.7 N501Y Spike RBD His-tag Avi-tag (ng/mL)

Biotinylated Recombinant SARS-CoV-2 B.1.1.7 N501Y Spike RBD His-tag Avi-tag Protein Binding Activity. Biotinylated Recombinant SARS-CoV-2 B.1.1.7 N501Y Spike RBD His-tag Avi-tag (Catalog # AVI10730) binds Recombinant Human ACE-2 Fc Chimera (Catalog # 10544-ZN) in a functional ELISA.

SDS-PAGE



Biotinylated Recombinant SARS-CoV-2 B.1.1.7 S RBD Avi-tag Protein SDS-PAGE. 2 µg/lane of Biotinylated Recombinant SARS-CoV-2 B.1.1.7 S RBD Avi-tag Protein (Catalog # AVI10730) was resolved with SDS-PAGE under reducing (R) and non-reducing (NR) conditions and visualized by Coomassie® Blue staining, showing bands at 34-38 kDa.

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Global bio-techne.com info@bio-techne.com techsupport@bio-techne.com TEL +1 612 379 2956
USA TEL 800 343 7475 Canada TEL 855 668 8722 China TEL +86 (21) 52380373
Europe | Middle East | Africa TEL +44 (0)1235 529449

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Catalog Number: AVI10730

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 B 1.1.7 (United Kingdom) variant (13). 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 (14). 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 (13, 15). Our Avi-tag Biotinylated SARS-CoV-2 Spike B.1.1.7 RBD features biotinylation at a single site contained within the Avi-tag, a unique 15 amino acid peptide. Protein orientation will be uniform when bound to streptavidin-coated surface due to the precise control of biotinylation and the rest of the protein is unchanged so there is no interference in the protein's bioactivity.

References:

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