

DESCRIPTION

Species Reactivity	SARS-CoV-2
Specificity	Detects SARS-CoV-2 Spike RBD in direct ELISAs.
Source	Monoclonal Mouse IgG _{2B} Clone # 1049349
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	Human embryonic kidney cell HEK293-derived SARS-CoV-2 Spike RBD Val16-Pro681 Accession # YP_009724390.1
Conjugate	Alexa Fluor 594 Excitation Wavelength: 590 nm Emission Wavelength: 617 nm
Formulation	Supplied 0.2 mg/mL in a saline solution containing BSA and Sodium Azide. *Contains <0.1% Sodium Azide, which is not hazardous at this concentration according to GHS classifications. Refer to the Safety Data Sheet (SDS) for additional information and handling instructions.

APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. [General Protocols](#) are available in the Technical Information section on our website.

Flow Cytometry	Titration recommended for optimal concentration with starting range of 0.1-1 µg/1 million cells. Sample used for this experiment was HEK293 human embryonic kidney cell line transfected with human ACE-2 and eGFP incubated with Recombinant SARS-CoV-2 CAL.20C S1 His-Tag protein (Catalog # 10779-CV)
-----------------------	--

PREPARATION AND STORAGE

Shipping	The product is shipped with polar packs. Upon receipt, store it immediately at the temperature recommended below.
Stability & Storage	Protect from light. Do not freeze. 12 months from date of receipt, 2 to 8 °C as supplied.

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 homotrimeric glycoprotein that mediates membrane fusion and viral entry. As with most coronaviruses, proteolytic cleavage of the SARS-CoV-2 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 (2-5). A SARS-CoV-2 variant (named CAL.20C) carrying the S1 subunit amino acid (aa) change W152C, L452R, and D614G emerged in Southern California (6,7). 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 (8). 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 (9). Based on amino acid (aa) sequence homology, the SARS-CoV-2 S1 subunit has 65% identity with SARS-CoV-1 S1 subunit, but only 22% homology with the MERS S1 subunit. The low aa sequence homology is consistent with the finding that SARS and MERS bind different cellular receptors (10). The S Protein of the SARS-CoV-2 virus, like the SARS-CoV-1 counterpart, binds Angiotensin-Converting Enzyme 2 (ACE-2), but with much higher affinity and faster binding kinetics (11). Before binding to the ACE-2 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 (12). Polyclonal antibodies to the RBD of the SARS-CoV-2 S1 subunit have been shown to inhibit interaction with the ACE-2 receptor, confirming S1 subunit especially the RBD as an attractive target for vaccinations or antiviral therapy (13). 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 (14). Lastly, it has been demonstrated the S Protein can invade host cells through the CD147/EMMPRIN receptor and mediate membrane fusion (15, 16).

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. Zhang, W. *et al.* (2021) *JAMA* <https://doi.org/10.1001/jama.2021.1612>.
7. Zhang, W. *et al.* (2021). *MedRxiv* <https://doi.org/10.1101/2021.01.18.21249786>.
8. Yuan, Y. *et al.* (2017) *Nat. Commun.* **8**:15092.
9. Walls, A.C. *et al.* (2010) *Cell* **180**:281.
10. Jiang, S. *et al.* (2020) *Trends. Immunol.* <https://doi.org/10.1016/j.it.2020.03.007>.
11. Ortega, J. T. *et al.* (2020) *EXCLI J.* **19**:410.
12. Wrapp, D. *et al.* (2020) *Science* **367**:1260.
13. Tai, W. *et al.* (2020) *Cell. Mol. Immunol.* <https://doi.org/10.1016/j.it.2020.03.007>.
14. Okba, N.M.A. *et al.* (2020). *Emerg. Infect. Dis.* <https://doi.org/10.3201/eid2607.200841>.
15. Wang, X. *et al.* (2020) <https://doi.org/10.1038/s41423-020-0424-9>.
16. Wang, K. *et al.* (2020) *ioRxiv* <https://www.biorxiv.org/content/10.1101/2020.03.14.988345v1>.

PRODUCT SPECIFIC NOTICES

This product is provided under an agreement between Life Technologies Corporation and R&D Systems, Inc, and the manufacture, use, sale or import of this product is subject to one or more US patents and corresponding non-US equivalents, owned by Life Technologies Corporation and its affiliates. The purchase of this product conveys to the buyer the non-transferable right to use the purchased amount of the product and components of the product only in research conducted by the buyer (whether the buyer is an academic or for-profit entity). The sale of this product is expressly conditioned on the buyer not using the product or its components (1) in manufacturing; (2) to provide a service, information, or data to an unaffiliated third party for payment; (3) for therapeutic, diagnostic or prophylactic purposes; (4) to resell, sell, or otherwise transfer this product or its components to any third party, or for any other commercial purpose. Life Technologies Corporation will not assert a claim against the buyer of the infringement of the above patents based on the manufacture, use or sale of a commercial product developed in research by the buyer in which this product or its components was employed, provided that neither this product nor any of its components was used in the manufacture of such product. For information on purchasing a license to this product for purposes other than research, contact Life Technologies Corporation, Cell Analysis Business Unit, Business Development, 29851 Willow Creek Road, Eugene, OR 97402, Tel: (541) 465-8300. Fax: (541) 335-0354.