

DESCRIPTION

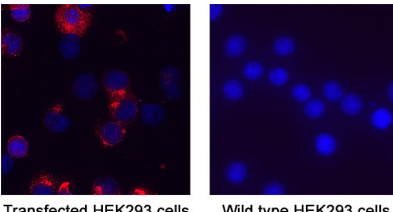
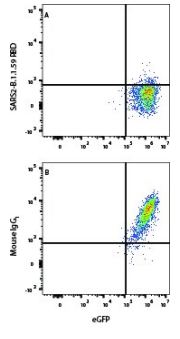
Species Reactivity	SARS-CoV-2
Specificity	It detects SARS-CoV-2 Omicron BA.1 in Direct ELISA. In ELISA, this antibody does not detect SARS-CoV-2 Alpha, Gamma or Delta variants.
Source	Monoclonal Mouse IgG ₁ Clone # 1056608
Purification	Protein A or G purified from cell culture supernatant
Immunogen	Recombinant SARS-CoV-2 B.1.1.529 (Omicron) Spike RBD domain. Arg319-Phe541 Accession # YP_009724390.1
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

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.

	Recommended Concentration	Sample
Immunocytochemistry	3-25 µg/mL	Immersion fixed HEK293 (positive) and HEK293 wild type (negative) cells
Blockade of Receptor-ligand Interaction	In a functional flow cytometry test, 25 µg/mL of Mouse Anti-SARS-CoV-2 Spike RBD Monoclonal Antibody (Catalog # MAB11227) will block the binding of Recombinant SARS-Cov-2 Spike RBD His-tagged protein (Catalog # 10500-CV) to HEK293 human embryonic kidney cell line transfected with recombinant human ACE-2 and eGFP	

DATA

<p>Immunocytochemistry</p>  <p>Transfected HEK293 cells Wild type HEK293 cells</p> <p>Detection of Spike RBD in HEK293 (positive) and HEK293 wild type (negative) cells. Spike RBD was detected in immersion fixed HEK293 (positive) and HEK293 wild type (negative) cells using Mouse Anti-SARS-CoV-2 Spike RBD Monoclonal Antibody (Catalog # MAB11227) at 3 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NL007) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Non-adherent Cells.</p>	<p>Blockade of Receptor-ligand Interaction</p>  <p>Blocking of Spike RBD in HEK293 cells transfected with Human ACE-2 and eGFP cells by Flow Cytometry. In a functional flow cytometry test, Recombinant SARS-Cov-2 Spike RBD His-tagged protein (Catalog # 10500-CV) binds to HEK293 human embryonic kidney cell line transfected with recombinant human ACE-2 and eGFP. (A) Binding is blocked by 25µg/mL of Mouse Anti-SARS-CoV-2 Spike RBD Monoclonal Antibody (Catalog # MAB11227) but not by (B) Mouse IgG₁ Isotype Control (Catalog # MAB002). Protein binding was detected with His Tag Allophycocyanin Mab (Catalog # IC050A). View our protocol for Staining Membrane-associated Proteins.</p>
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PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	<p>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</p> <ul style="list-style-type: none"> • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 6 months, -20 to -70 °C under sterile conditions after reconstitution.

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 metalloproteinase, 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. First identified in November 2021 in South Africa, the Omicron variant quickly became the predominant SARS-CoV-2 variant and is considered a variant of concern (VOC). The Omicron variant contains 15 mutations in RBD domain that potentially affect viral fitness and transmissibility. The majority of the mutations are involved in ACE-2 binding and Omicron binds ACE-2 with greater affinity, potentially explaining its increased transmissibility (13, 14). Several of these mutations are also identified in facilitating immune escape and reducing neutralization activity to several monoclonal antibodies (13). Additionally, a series of new mutations are present in the RBD which have unknown impacts on receptor binding or antibody neutralization.

References:

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