

## Product Information

## Anti-SARS-CoV-2-Spike-RBD Region Antibody

Mouse Monoclonal, Clone Sp-10, Purified from Hybridoma Cell Culture

**SAB4200875**

### Product Description

Monoclonal Anti-SARS-CoV-2-Spike-RBD region antibody (mouse IgG1 isotype) is derived from the SP-10 hybridoma, produced by the fusion of mouse myeloma cells and splenocytes from a mouse immunized with synthetic peptide corresponding to the SARS-CoV-2 Spike protein RBD region (GeneID: QHD43416.1), conjugated to KLH as immunogen. The isotype is determined by ELISA using Mouse Monoclonal Antibody Isotyping Reagents (Sigma ISO-2). The antibody is purified from culture supernatant of hybridoma cells.

Monoclonal Anti-SARS-CoV-2-Spike-RBD region antibody specifically recognizes Spike from COVID-19 virus origin. The antibody may be used in various immunochemical techniques including immunoblotting and ELISA. Detection of the Spike RBD protein band by immunoblotting is specifically inhibited by the immunogen.

Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) or (2019-nCoV) is a novel coronavirus that had spread on December 2019 in Hubei province of China and infected millions of people worldwide.<sup>1</sup> The causative agent of COVID-19, the SARS-CoV-2 virus is a positive-strand RNA virus. The mature SARS-CoV-2 contains 4 structural proteins: Envelope (E), Membrane (M), Nucleocapsid (N), and the Spike protein (S), E and M proteins help in viral assembly and N protein is needed for RNA synthesis. The main receptor for SARS-CoV and SARS-CoV-2 on the membrane of the target cells is the Angiotensin 2 Converting Enzyme (ACE2). ACE2 is a metallopeptidase present on the membrane of many cells, including type-I and -II pneumocytes, small intestine enterocytes, kidney proximal tubules cells, the endothelial cells of arteries and veins, and the arterial smooth muscle, among other tissues.<sup>15-16</sup>

It has been shown that SARS-CoV-2 virus employs transmembrane protease serine 2 (TMPRSS2) for S protein priming and it is speculated that furin-mediated cleavage at the S1/S2 site in infected cells, may promote subsequent TMPRSS2-dependent entry into target cells. The Spike protein (S) is responsible for virus binding and entry into the host cells. SARS-CoV-2 S protein precursor is cleaved into S1 subunit (685 amino acids), and S2 (588 amino acids) subunits. S1 subunit harbor the receptor binding domain (RBD) that mediates virus entry into susceptible cells through the peptidase domain of host ACE2 with high affinity ( $K_d = 15$  nM). S2 protein, which is reported to be well-conserved and showing 99% identity with bat coronavirus, is responsible for the membrane fusion. The Spike protein is the most studied between the coronaviruses proteins, due to its crucial role in the host cell entry, it contains the RBD for the ligand on the host cell membrane (the ACE2 protein), and also has epitopes recognized by T and B cells, which induce the production of neutralizing antibodies.<sup>2</sup>

Anti-SARS-CoV-2-Spike protein antibodies are important tools in the COVID-19 research field and can be used for detection of Spike protein in different samples and in cell culture assays.<sup>17</sup>

### Reagent

Supplied as a solution in 0.01 M phosphate buffered saline pH 7.4, containing 15 mM sodium azide as a preservative.

Antibody Concentration: ~ 1.0 mg/mL

## Precautions and Disclaimer

For research use only. Not for use in diagnostic procedure. Unless otherwise stated in our catalogue or other company documentation accompanying the product(s), our products are intended for research use only and are not to be used for any other purpose, which includes but is not limited to, unauthorized commercial uses, in vitro diagnostic uses, ex vivo or in vivo therapeutic uses or any type of consumption or application to humans or animals. Data presented is the available current product information and provided as-is. This product has not been tested or verified in any additional applications, sample types, including any clinical use. Experimental conditions must be empirically derived by the user. Our Antibody Guarantee only covers tested applications stated herein and conditions presented in our product information and is not extended to publications.

## Storage/Stability

For continuous use, store at 2-8 °C for up to one month. For extended storage, freeze in working aliquots. Repeated freezing and thawing is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilution samples should be discarded if not used within 12 hours.

## Product Profile

### Immunoblotting

A working concentration of 0.1-0.2 µg/mL is recommended using SARS-CoV-2 RBD (Cat. No. SAE1000).

### ELISA

A working concentration of 0.25-0.5 µg/mL is recommended using SARS-CoV-2 RBD (Cat. No. SAE1000) for coating.

## Neutralizing ELISA

A working concentration of 20-40 µg/mL is recommended blocking the Spike RBD-ACE2 interaction using 2 µg/mL SARS-CoV-2 RBD (Cat. No. SAE1000) for coating and 2 µg/mL ACE2-Biotin (Cat. No. SAE0171) for competition. Applying antibody concentration of 20 µg/mL on a Spike RBD-coated well, will reduce > 65% of ACE2-Biotin binding signal, added to the well after Spike RBD-antibody incubation.

**Note:** In order to obtain best results in different techniques and preparations it is recommended to determine optimal working concentration by titration test.

## References

1. García, Luis F., *Frontiers in immunology*, **11**: 1441 (2020).
2. Walls AC, et al., *Cell*. **181**: 281–92.e6 (2020).
3. Shang J., et al., *Nature*. **581**: 221–4 (2020).
4. Tai W., et al. *Cell Mol Immunol*. **17**: 621–30 (2020).
5. Chen Y., et al., *Biochem Biophys Res Commun*. **525**:135–40 (2020).
6. Hoffmann M., et al. *Cell*. **181**: 270–81.e8 (2020).
7. Lan J., et al. *Nature*. **581**: 215–20 (2020).
8. Liu Z., et al. *J Med Virol*. **92**: 595–601 (2020).
9. Luan J., et al., *Biochem Biophys Res Commun*. **526**:165–9 (2020).
10. Yan R., et al., *Science*. **367**: 1444 (2020).
11. Fung T.S., et al., *Annu Rev Microbiol*. **73**: 529–57 (2019).
12. Wrapp D., et al., *Science*. **367**: 1260 (2020).
13. Ou X., et al., *Nat Commun*. **11**: 1620 (2020).
14. Shang J., et al., *PLoS Pathog*. **16**: e1008392 (2020).
15. Hamming I., et al., *J Pathol*. **203**: 631–7 (2004).
16. Zou X., et al., *Front Med*. **14**:185–92 (2020).
17. Abe K.T., et al. *JCI insight* **5**:19 (2020)

---

## Notice

We provide information and advice to our customers on application technologies and regulatory matters to the best of our knowledge and ability, but without obligation or liability. Existing laws and regulations are to be observed in all cases by our customers. This also applies in respect to any rights of third parties. Our information and advice do not relieve our customers of their own responsibility for checking the suitability of our products for the envisaged purpose.

The information in this document is subject to change without notice and should not be construed as a commitment by the manufacturing or selling entity, or an affiliate. We assume no responsibility for any errors that may appear in this document.

### Technical Assistance

Visit the tech service page at [SigmaAldrich.com/techservice](https://SigmaAldrich.com/techservice).

### Standard Warranty

The applicable warranty for the products listed in this publication may be found at [SigmaAldrich.com/terms](https://SigmaAldrich.com/terms).

### Contact Information

For the location of the office nearest you, go to [SigmaAldrich.com/offices](https://SigmaAldrich.com/offices).

The life science business of Merck KGaA, Darmstadt, Germany operates as MilliporeSigma in the U.S. and Canada.

MilliporeSigma, and Sigma-Aldrich are trademarks of Merck KGaA, Darmstadt, Germany or its affiliates. All other trademarks are the property of their respective owners. Detailed information on trademarks is available via publicly accessible resources.  
© 2021 Merck KGaA, Darmstadt, Germany and/or its affiliates. All Rights Reserved.

SAB4200875dat Rev 07/21

For research use only. Not for use in diagnostic procedures.