# Recombinant SARS-CoV-2 Spike S1 Subunit Protein Host Cell Receptor Binding Domain (RBD) N331Q mutant

## Source

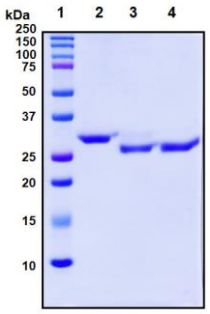
- **Species** SARS-CoV-2
- **Accession Number** QHD43416
- **Expressed Region** Arg319 - Phe541. N-glycosylation site at Asn (N) 331 was mutated to Gln (Q).
- **Synonyms** Spike protein, S Protein, S1 Subunit, Host Cell Receptor Binding Domain (RBD).

## Preparation

- **Expression System** Human embryonic kidney 293 (HEK293) cells
- **Tag** *C*-terminal his-tag
- **Purification** His-tag affinity purification by immobilized metal ion affinity chromatography (IMAC)
- **Endotoxin Level** <0.5 EU per µg of the protein as determined by the LAL method
- **Purity** >95%
- **Purity determined** By SDS-PAGE under reducing conditions and visualized by Coomassie blue staining
- **Molecular Weight** Recombinant protein product has a calculated molecular mass of 25 kDa. Due to the abundant glycosylation, it migrates as approximately 30 kDa protein bands in SDS-PAGE under DTT, beta-mercaptoethanol reducing conditions. See deglycosylation analysis image below.

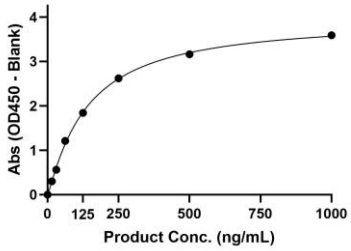
## Protein Specifications

- **Format** Liquid
- **Formulation** Supplied as a 0.2 µm filtered solution in PBS (pH 7.4)
- **Concentration** Lot specific (see the label on the vial), determined by BCA protein assay.
- **SDS-PAGE Image** Deglycosylation analysis of purified recombinant proteins. The purified proteins were untreated (*Lane 2*) or treated with Protein Deglycosylation Kit under native (*Lane 3*) or reducing (*Lane 4*) conditions. Deglycosylation treatment resulted in a mobility shift of the protein to produce one major band at the expected size (~25 kDa), thus indicating that the untreated recombinant protein (*Lane 2*) was glycosylated. **Lane 1,** protein standard ladder (kDa); **Lane 2**, untreated protein under reducing conditions; **Lane 3**, treated protein with deglycosylation enzymes under native conditions; **Lane 4**, treated protein with deglycosylation enzymes under reducing conditions.




## Binding

Product binding ability was measured by ELISA. The immobilized recombinant human ACE2 protein (Catalog #. 230-30165, coated at 0.5 ug/ml, 100 ul/well) was incubated with the serial diluted SARS CoV-2 S1 RBD N331 mutant protein (Catalog #. 230-30157). The bound mutant protein was detected by mouse anti-S1 RBD monoclonal antibody using ELISA. The calculated EC50 is 138.1 ng/ml (*right*).




## Shipping

The product is shipped with ice packs.

## Storage/Stability

Upon arrival, the protein may be stored for 2 weeks at 4 °C. For long term storage, it is recommended to store at -20 °C or -80 °C in appropriate aliquots. Avoid repeated freeze-thaw cycles.

## References

- N Dong, et al. Genomic and protein structure modelling analysis depicts the origin and infectivity of 2019-nCoV, a new coronavirus which caused a pneumonia outbreak in Wuhan, China. **bioRxiv** (2020).
- M Hoffmann, et al. SARS-CoV-2 Cell Entry Depends on ACE2 and TMPRSS2 and Is Blocked by a Clinically Proven Protease Inhibitor. **Cell**. 181, 1–10 (2020).
- W Li et al. Angiotensin-converting enzyme 2 is a functional receptor for the SARS coronavirus. **Nature**. 426, 450–454 (2003).