Store at -80°C

SARS-CoV-2 Spike (trimeric) (16-1208) Recombinant Protein (8xHis-Tag)

#65444

100 µg

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Entrez-Gene ID #43740568
UniProt ID #P0DTC2

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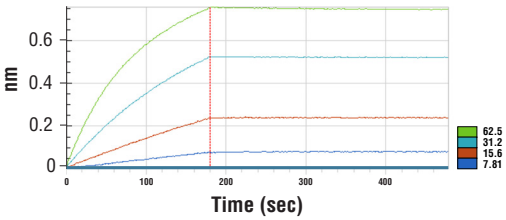
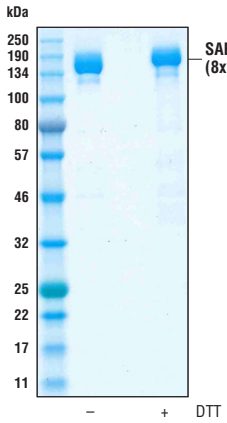
Description: SSARS-CoV-2 Spike (trimeric) (16-1208) Recombinant Protein (8xHis-Tag) is derived from a recombinant expression construct designed to generate a trimeric, cleavage-resistant variant of the SARS-CoV-2 spike protein ectodomain. The S1/S2 polybasic cleavage site (R685/S686) has been removed by substituting RRAR (R682-R685) with the non-cleavable sequence GSAS. An additional mutation (K986P/V987P) was introduced by site-directed mutagenesis, designed to constrain the coronavirus spike proteins to a pre-fusion configuration (10). Additional sequence corresponding to the foldon domain of bacteriophage T4 fibritin was introduced to the carboxy terminal region to promote trimerization of the expressed protein. The expressed protein also contains an 8xHis-Tag at its carboxy terminus.

Background: The cause of the COVID-19 pandemic is a novel and highly pathogenic coronavirus, termed SARS-CoV-2 (severe acute respiratory syndrome coronavirus-2). SARS-CoV-2 is a member of the Coronaviridae family of viruses (1). The genome of SARS-CoV-2 is similar to other coronaviruses, and is comprised of four key structural proteins: S, the spike protein, E, the envelope protein, M, the membrane protein, and N, the nucleocapsid protein (2). Coronavirus spike proteins are class I fusion proteins and harbor an ectodomain, a transmembrane domain, and an intracellular tail (3,4). The highly glycosylated ectodomain projects from the viral envelope surface and facilitates attachment and fusion with the host cell plasma membrane. The ectodomain can be further subdivided into host receptor-binding domain (RBD) (S1) and membrane-fusion (S2) subunits, which are produced upon proteolysis by host proteases at S1/S2 and S2' sites. S1 and S2 subunits remain associated after cleavage and assemble into crown-like homotrimers (2,4). In humans, both SARS-CoV and SARS-CoV-2 spike proteins utilize the angiotensin-converting enzyme 2 (ACE2) protein as a receptor for cellular entry (5-7). Spike protein subunits represent a key antigenic feature of coronavirus virions, and therefore represent an important target of vaccines, novel therapeutic antibodies, and small-molecule inhibitors (8,9).

Immobilized **Human ACE2 (18-615) Recombinant Protein (hFc-Tag) #38365**
In Solution **SARS-CoV-2 Spike (trimeric) (16-1208) Recombinant Protein (8xHis-Tag)** – *by Conc. (nM)*

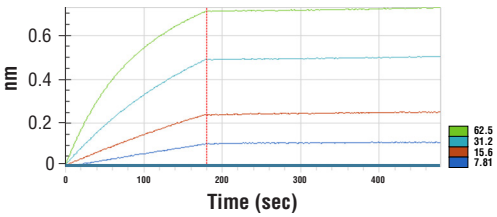


Binding response kinetics between Human ACE2 (18-615) Recombinant Protein (hFc-Tag) #38365 (immobilized) and SARS-CoV-2 Spike (trimeric) (16-1208) Recombinant Protein (8xHis-Tag) (in solution, at indicated concentrations). The vertical red line (180 sec) indicates addition of PBS to induce dissociation. Binding was detected with an anti-human Fc biosensor. Values on y-axis indicate binding response signals recorded for 4 different concentrations of SARS-CoV-2 Spike (trimeric) (16-1208) Recombinant Protein (8xHis-Tag) (7.81, 15.6, 31.2, and 62.5 nM).



The purity of SARS-CoV-2 Spike (trimeric) (16-1208) Recombinant Protein (8xHis-Tag) was determined by densitometry after SDS-PAGE of 1 µg of protein followed by staining with Coomassie Blue. Purity values were determined from the DTT-reduced samples (+).

Immobilized **Human ACE2 (18-615) Recombinant Protein (mFc-Tag) #27413**
In Solution **SARS-CoV-2 Spike (trimeric) (16-1208) Recombinant Protein (8xHis-Tag)** – *by Conc. (nM)*



Binding response kinetics between Human ACE2 (18-615) Recombinant Protein (mFc-Tag) #27413 (immobilized) and SARS-CoV-2 Spike (trimeric) (16-1208) Recombinant Protein (8xHis-Tag) (in solution, at indicated concentrations). The vertical red line (180 sec) indicates addition of PBS to induce dissociation. Binding was detected with an anti-mouse Fc biosensor. Values on y-axis indicate binding response signals recorded for 4 different concentrations of SARS-CoV-2 Spike (trimeric) (16-1208) Recombinant Protein (8xHis-Tag) (7.81, 15.6, 31.2, and 62.5 nM).

Molecular Weight: 136 kDa (reduced and non-reduced)

Formulation:
Expression Host: Hamster (ExpiCHO cells)

Supplied in a PBS solution (pH 7.2).

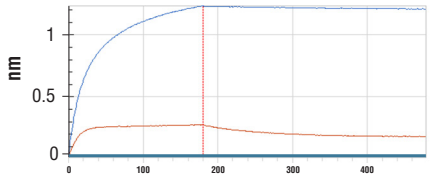
Purity: ≥94%, determined by SDS-PAGE.

Storage: Stable at -80°C for 3 years after receipt. Avoid repeated freeze-thaw cycles.

Background References:

(1) Zhou, P. et al. (2020) *Nature* 579, 270-3.
(2) Tortorici, M.A. and Veesler, D. (2019) *Adv Virus Res* 105, 93-116.
(3) Li, F. et al. (2006) *J Virol* 80, 6794-800.
(4) Li, F. (2016) *Annu Rev Virol* 3, 237-261.
(5) Shang, J. et al. (2020) *Nature* 581, 221-4.
(6) Wrapp, D. et al. (2020) *Science* 367, 1260-3.
(7) Yan, R. et al. (2020) *Science* 367, 1444-8.
(8) Yuan, Y. et al. (2017) *Nat Commun* 8, 15092.
(9) Amanat, F. and Krammer, F. (2020) *Immunity* 52, 583-9.
(10) Pallesen, J. et al. (2017) *Proc Natl Acad Sci U S A* 114, E7348-E7357.

Immobilized **Human ACE2 (18-615) Recombinant Protein (hFc-Tag) #38365**
In Solution **SARS-CoV-2 Spike (trimeric) (16-1208) Recombinant Protein (8xHis-Tag) (blue)**
SARS-CoV-2 Spike RBD (318-541) Recombinant Protein (8xHis-Tag) #48801 (red)



Binding response kinetics between Human ACE2 (18-615) Recombinant Protein (hFc-Tag) #38365 (immobilized), SARS-CoV-2 Spike (trimeric) (16-1208) Recombinant Protein (8xHis-Tag) (in solution, 250 nM, blue) and SARS-CoV-2 Spike RBD (318-541) Recombinant Protein (8xHis-Tag) #48801 (in solution, 250 nM, red). The vertical red line (180 sec) indicates addition of PBS to induce dissociation. Binding was detected with an anti-human Fc biosensor. Values on y-axis indicate binding response signals (nm) for each protein in solution.

Applications: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **FC-FP**— Flow cytometry-Fixed/Permeabilized **FC-L**— Flow cytometry-Live **E-P**—ELISA-Peptide
Species Cross-Reactivity: **H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **C**—chicken **Dm**—D. melanogaster **X**—Xenopus **Z**—zebrafish **B**—bovine **Dg**—dog **Pg**—pig **Sc**—S. cerevisiae **Ce**—C. elegans **Hr**—Horse
All—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.