

DESCRIPTION

Species Reactivity	SARS-CoV-2
Specificity	Detects SARS-CoV-2 NSP8 in direct ELISAs.
Source	Monoclonal Mouse IgG _{2B} Clone # 1044736
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	<i>E. coli</i> -derived SARS-CoV-2 NSP8 protein Ala1-Gln198 Accession # YP_009725304.1
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.

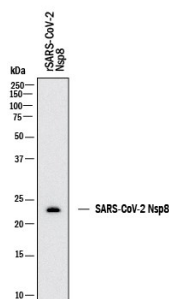
APPLICATIONS

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
Western Blot	1 µg/mL	Recombinant SARS-CoV-2 Nsp8 protein
Simple Western	20 µg/mL	Recombinant SARS-CoV-2 Nsp8 protein

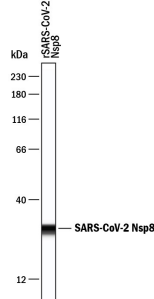
DATA

Western Blot



Detection of SARS-CoV-2 Nsp8 by Western Blot. Western blot shows recombinant SARS-CoV-2 Nsp8 protein. PVDF membrane was probed with 1 µg/mL of Mouse Anti-SARS-CoV-2 Nsp8 Monoclonal Antibody (Catalog # MAB12502) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF018). A specific band was detected for Nsp8 at approximately 24 kDa (as indicated). This experiment was conducted under reducing conditions and using Western Blot Buffer Group 1.

Simple Western



Detection of SARS-CoV-2 Nsp8 by Simple Western™. Simple Western lane view shows recombinant SARS-CoV-2 Nsp8 protein, loaded at 0.2 mg/mL. A specific band was detected for Nsp8 at approximately 29 kDa (as indicated) using 20 µg/mL of Mouse Anti-SARS-CoV-2 Membrane Monoclonal Antibody (Catalog # MAB12502). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.

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	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <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

Non-structural protein 8 (NSP8) is one of several functional proteins released by ORF1a-encoded protease cleavage of the pp1a and pp1ab replicase polyproteins expressed from the coronavirus (CoV) genome (1). The NSPs are involved in the replication and transcription of the viral RNA and not incorporated within the virion particles. Coronaviruses include various highly pathogenic strains such as SARS-CoV, MERS-CoV and SARS-CoV2 that have had significant impact on humans as well as strains that have negatively impacted livestock. NSP8 is a small 198 amino acid protein that forms a dimer with NSP7 and subsequently assembles into a large hexadecameric structure (2). The NSP8 sequence is highly conserved across coronaviruses (2). The NSP8 monomers in each of two asymmetric units can adopt two different conformations: a "golf-club like" structure with the N-terminal shaft and C-terminal head or a bent N-terminal shaft and C-terminal head domain (2). The units stack to form a supercomplex like bricks with layers of NSP7 filling the spaces in between. The supercomplexes are stacked to form a channel with electrostatic properties that would allow RNA to pass through the channel, likely to facilitate efficient replication and transcription. NSP8 was also shown to be able to polymerize small oligomers in a sequence-specific fashion and was consequently proposed to act as an RNA primase for the viral RNA-dependent RNA polymerase (RdRp), NSP12, in SARS-CoV (3). In SARS-CoV-2, RdRp has been shown to have little activity without NSP8/7 acting as cofactors to form a complex (4) making NSP8 critical for viral polymerase activity. NSP8 was shown to interact with several other viral NSP proteins, including NSP2 and NSP9, (5) as well as multiple host cell proteins (6). NSP8 showed interaction with 3 different signal recognition particle (SRP) host cell proteins suggesting the virus hijacks the SEC61-mediated protein translocation pathway for entry into the endoplasmic reticulum and NSP8 may be involved in targeting (6).

References:

1. Snijder, E.J. *et al.* (2016) Adv. Virus Res. **96**:59.
2. Zhai, Y. *et al.* (2005) Nat. Struct. Mol. Bio. **12**:980.
3. Subissi, L. *et al.* (2014) Antiviral Res. **101**:122.
4. Yin, W. *et al.* (2020) Science **368**:1499.
5. von Brunn, A. *et al.* (2007) PLoS One **2**:e459.
6. Gordon, D.E. *et al.* (2020) Nature **583**:459.