

DESCRIPTION

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|--------------------------|---|--|------------|
| Source | <i>E. coli</i> -derived sars-cov-2 NSP16/NSP10 protein | | |
| | <div><div>SARS-CoV-2 NSP16 (Ser1-Asn298) Accession # YP_009725311.1</div><div>SARS-CoV-2 NSP10 (Ala1-Gln139) Accession # YP_009725306.1</div></div> | | |
| | <div>6-His tag</div> | | |
| | N-terminus | | C-terminus |
| N-terminal Sequence | Ser1 (NSP16) & Ala1 (NSP10) | | |
| Analysis | | | |
| Predicted Molecular Mass | 33 kDa (NSP16) & 16 kDa (NSP10) | | |

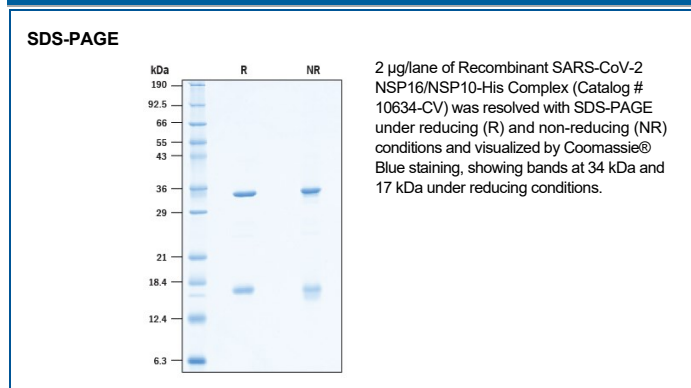
SPECIFICATIONS

| | |
|------------------------|---|
| SDS-PAGE | 34 kDa (NSP16) & 17 kDa (NSP10), under reducing conditions |
| Endotoxin Level | <1.0 EU per 1 µg of the protein by the LAL method. |
| Purity | >95%, by SDS-PAGE visualized with Silver Staining and quantitative densitometry by Coomassie® Blue Staining. |
| Formulation | Supplied as a 0.2 µm filtered solution in Tris, NaCl, TCEP and Glycerol. See Certificate of Analysis for details. |

PREPARATION AND STORAGE

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| Shipping | The product is shipped with polar packs. Upon receipt, store it immediately at the temperature recommended below. |
| Stability & Storage | <p>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</p> <ul style="list-style-type: none"> 6 months from date of receipt, -20 to -70 °C as supplied. 3 months, -20 to -70 °C under sterile conditions after opening. |

DATA



BACKGROUND

Non-structural protein 16 (NSP16) and NSP10 are two 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. NSP16 is a small monomeric 298 amino acid protein with a characteristic fold of the class I methyltransferase (MTase) family with a seven-stranded B-sheet surrounded by alpha-helices and loops that bind the cap and form a cap-binding groove and S-adenosylmethionine (SAM) cleft (2,3). NSP10 is a small 139 amino acid protein capable of forming independent dodecameric structures composed of four identical trimers where each monomer is capable of binding two zinc ions (4,5). Both the NSP16 and NSP10 SARS-CoV2 sequences are highly conserved across coronaviruses (3). NSP16 catalyzes 2'-O-MTase activity specifically on capped N7-methylated RNA through methylation of the 2'-hydroxy group of adenine using SAM as a methyl donor (6). The viral RNA cap structure protects it from degradation, promotes mRNA translation, and prevents the viral RNA from being recognized by innate immune mechanisms (7). NSP16 was shown to interact strongly with NSP10 (8) in a monomeric form via hydrophobic interactions and hydrogen bonds in the interface (3). The MTase activity of NSP16 requires interaction with NSP10 (2,7). NSP10 interaction extends and narrows the RNA-binding groove that accommodates the substrate and stabilizes NSP16 (2,3,9). The dimerization interface between NSP16 and NSP10 is critical and can be used as a target to effectively reduce replication and pathogenesis (10,11).

References:

1. Snijder, E.J. *et al.* (2016) *Adv. Virus Res* **96**:59.
2. Chen, Y. *et al.* (2011) *PLoS Pathog.* **7**:e1002294.
3. Rosas-Lemus, M. *et al.* (2020) *bioRxiv* In press.
4. Matthes, N. *et al.* (2006) *FEBS Lett.* **580**:4143.
5. Su, D. *et al.* (2006) *J. Virol.* **80**:7902.
6. Bouvet, M. *et al.* (2012) *Proc. Natl. Acad. Sci. U.S.A.* **109**:9372.
7. Decroly, E. *et al.* (2011) *PLoS Pathog.* **7**:e1002059.
8. Pan, J. *et al.* (2008) *PLoS One* **3**:e3299.
9. Bouvet, M. *et al.* (2014) *J. Biol. Chem.* **289**:25783.
10. Ma, Y. *et al.* (2015) *Proc. Natl. Acad. Sci.* **112**:9436.
11. Wang, Y. *et al.* (2015) *J. Virol* **89**:8416.