

MRC PPU Reagents and Services

Standard Operating Procedure

Preparation of NSP1 [1 – 180] SARS CoV2

Enzyme description:- NSP1 [1 – 180]

Clone number:- DU 67714

Source:- Recombinant

Expression system:- *E. coli*

Tag:- N-terminal MBP

Purification method:- Amylose resin

Calculated molecular mass:-

Monoisotopic 63,974.47 daltons

Average Mass 64,014.22 daltons

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 5.01

Purity:- 90 %

Enzyme storage buffer:-

50 mM Tris-HCl pH 7.5, 270 mM Sucrose, 150 mM NaCl, 0.1 mM EGTA,
0.1 % 2-mercaptoethanol, 0.03 % Brij-35

Storage temperature:- -70 °C

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Clone Data Sheet

NSP1 [1 – 180] SARS CoV2

Protein NSP1 [1 – 180]

Clone number DU 67714

Accession number QHD43415.1

Tags N-terminal MBP

**Bacterially
expressed protein**

MKIEEGKLVWINGDKGYNGLAEVGGKFEKDTGIKVTVEHPDKLE
EKFPQVAATGDGPDIIFWAHDRFGGYAQSGLLAEITPDKAFQDKL
YPFTWDAVRYNGKLIAYPIAVEALSLIYNKDLLPNPPKTWEEIPA
LDKELKAKGKSALMFNLQEPYFTWPLIAADGGYAFKYENGYDIK
DVGVDNAGAKAGLTFLVDLIKNKHMNADTDYSIAEAAFNKGETAM
TINGPWAWSNIDTSKVNYGVTVLPTFKGQPSKPFVGVLSAGINAA
SPNKELAKEFLENYLLTDEGLEAVNKDKPLGAVALKSYYEELVKD
PRIAATMENAQKGEIMPNI PQMSAFWYAVRTAVINAASGRQTVDE
ALKDAQTNSSNNNNNNNNNNLGD~~DDDKVPEF~~LEVL~~FQGP~~LSME
SLVPGFNEKTHVQLSLPVLQVRDVLVRGFGDSVEEVLSEARQHLK
DGTCGLVEVEKGVLPQLEQPYVFIKRSDARTAPHGHVMVELVAEL
EGIQYGRSGETLGVLVPHVGEIPVAYRKVLLRKNGNKGAGGHSYG
ADLKSFDLGDDELGTD~~PYEDFQENWNTKHSSGVTRELMREL~~NGG

Native sequence Amino acids M1 – G180 (end).
Residue M404 of the fusion protein is equivalent to M1 of the
native enzyme. The MBP tag is located at residues 1 – 367.

Protease cleavage Enterokinase (DDDDK) residues 384 – 388
PreScission (LEVL~~FQGP~~) residues 393 – 400