

MRC PPU Reagents and Services

Standard Operating Procedure

Preparation of NSP13 [1 - 601] SARS CoV2

Enzyme description:- NSP13 [1 - 601]

Clone number:- DU 66419

Source:- Recombinant

Expression system:- *E. coli*

Tag:- N-terminal MBP

Purification method:- Amylose Resin

Calculated molecular mass:-

Monoisotopic 111,022.96 daltons

Average Mass 111,093.66 daltons

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 6.60

Purity:- 50 %

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

NSP13 [1 – 601] SARS CoV2

Protein NSP13 [1 - 601]

Clone number DU 66419

Accession number QHD43415.1

Tags N-terminal MBP

**Bacterially
expressed protein**

MKIEEGKLVIIWINGDKGYNGLAEVGGKFEKDTGIKVTVEHPDKLE
EKFPQVAATGDGPDIIFWAHDRFGGYAQSGLLAEITPDKAFQDKL
YPFTWDAVRYNGKLIAYPIAVEALSIIYNKDLLPNPPKTWEEIPA
LDKELKAKGKSALMFNLQEPYFTWPLIAADGGYAFKYENKGYDIK
DVGVDNAGAKAGLTFVLVDLIKKNHMNADTDYSIAEAAFNKGETAM
TINGPWAWSNIDTSKVNYGVTVLPTFKGQPSKPFVGVLSAGINAA
SPNKELAKEFLENYLLTDEGLEAVNKDKPLGAVALKSYYYEELVKD
PRIAATMENAQKGEIMPNIPOMSAFWYAVRTAVINAASGRQTVDE
ALKDAQTNSSNNNNNNNNNNLGDGDDDKVPEFLEVLFGPLGSAV
GACVLCNSQTSLRGACIRRPFLCCKCCYDHVISTSHKLVLSVNP
YVCNAPGCDVTDVTQLYLGGMSYYCKSHKPPISFPLCANGQVFG
YKNTCVGSDNVTDFNAIATCDWTNAGDYILANTCTERLKLFAAET
LKATEETFKLSYGIATVREVLSDRELHLSWEVGKPRPPLNRNYVF
TGYRVTKNSKVQIGEYTFEKGDYGDVVYRGTTTYKLVNGDYFVL
TSHTVMPLSAPTLVPQEHYVRITGLYPTLNI SDEFSSNVANYQKV
GMQKYSTLQPPGTGKSHFAIGLALYPSARIVYTACSHA AVDAL
CEKALKYLPIDKCSRIIPARARVECFDKFKVNSTLEQYVFCTVNA
LPETTADIVVFDEISMATNYDLSVVNARLRAKHVYIGDPAQLPA
PRTLLTKGTLEPEYFNSVCRMKTIGPDMFLGTCRRCPAEIVDTV
SALVYDNKLAHKDKSAQCFKMFYKGVITHDVSSAINRPQIGVVR
EFLTRNPAWRKAVFISPYNSQNAVASKILGLPTQTVDSSQGSEYD
YVIFTQTTETAHSCNVNRFNVAITRAKVGILCIMS DRDLYDKLQF
TSLEIPRRNVATLQ

Native sequence Amino acids A1 – Q601 (end).
Residue A404 of the fusion protein is equivalent to A1 of the native enzyme. The MBP tag is located at residues 1 – 367.

Protease cleavage Enterokinase (DDDDK) residues 384 – 388
PreScission (LEVLFGQP) residues 393 – 400

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