

MRC PPU Reagents and Services

Standard Operating Procedure

Preparation of Nucleocapsid Protein [1 - 413] MERS CoV

Enzyme description:- MERS N Protein [1 - 413]

Clone number:- DU 67733

Source:- Recombinant

Expression system:- *E. coli*

Tag:- N-terminal MBP

Purification method:- Amylose Resin

Calculated molecular mass:-

Monoisotopic 89, 204.14 daltons

Average Mass 89, 259.14 daltons

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 8.85

Purity:- 85 %

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

Nucleocapsid Protein [1 – 413] MERS CoV

Protein MERS N Protein [1 - 413]

Clone number DU 67733

Accession number ANF29169.1

Tags N-terminal MBP

**Bacterially
expressed protein**

MKIEEGKLVIWINGDKGYNGLAEVGGKFEKDTGIKVTVEHPDKLE
EKFPQVAATGDGPDIIFWAHDRFGGYAQSGLLAEITPDKAFQDKL
YPFTWDAVRYNGKLIAYPIAVEALSLIYNKDLLPNPPKTWEEIPA
LDKELKAKGKSALMFNLQEPYFTWPLIAADGGYAFKYENGGYDIK
DVGVDNAGAKAGLTFVLVDLIKKNHMNADTDYSIAEAAFNKGETAM
TINGPWAWSNIDTSKVNYGVTVLPTFKGQPSKPFVGVLSAGINAA
SPNKELAKEFLENYLLTDEGLEAVNKDKPLGAVALKSYYYEELVKD
PRIAATMENAQKGEIMPNI PQMSAFWYAVRTAVINAASGRQTVDE
ALKDAQTNSSNNNNNNNNNNLGDGDDDKVPEFLEVLFGQPLGSM
**SPAAPRAVSFADNNDITNTNLSRGRGRNPKPRAAPNNTVSWYTG
LTOHGKVPLTFPPGQGVPLNANSTPAQNAGYWRRQDRKINTGNGIK
QLAPRWYFYTTGTGPEAALPFRVAVKDGIVVHEDGATDAPSTFTGT
RNPNDSAIVTQFAPGTKLPKNFHIEGTGGNSQSSSRASSASRNS
SRSSSQSRSRGNSTRGTSPPGSGIGAVGGDLLYDLLNRLQALES
GKVKQSOPKVIITKDAAAAANKMRHKRTSTKSFNMVQAFGLRGP
DLQGNFGDLQLNKLGTEDPRWPQIAELAPTASAFMGMSQFKLTHQ
NNDDHGPNPVYFLRYSGAIKLDPKPNPNYKWLLELLEQNIDAYKTFP
KKEKKQKAPKEESTDQMSPEPPKEQRVQGSITQRTTRTRPSVQPGPM
IDVNTD**

Native sequence Amino acids M1 – N389 (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 (LEVLFGQP) residues 393 – 400