

MRC PPU Reagents and Services

Standard Operating Procedure

Preparation of Nucleocapsid Protein [1 - 448] Human CoV OC43

Enzyme description:- Human CoV OC43 N Protein [1 - 448]

Clone number:- DU 67725

Source:- Recombinant

Expression system:- *E. coli*

Tag:- N-terminal MBP

Purification method:- Amylose Resin

Calculated molecular mass:-

Monoisotopic 93,470.99 daltons

Average Mass 93,528.69 daltons

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 7.58

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 - 448] Human CoV OC43

Protein Human CoV OC43 N Protein [1 – 448]

Clone number DU 67725

Accession number AEN19370.1

Tags N-terminal MBP

**Bacterially
expressed protein**

MKIEEGKLVIIWINGDKGYNGLAEVGGKFEKDTGIKVTVEHPDKLE
EKFPQVAATGDGPDIIFWAHDRFGGYAQSGLLAEITPDKAFQDKL
YPFTWDAVRYNGKLIAYPIAVEALSLIYNKDLLPNPPKTWEEIPA
LDKELKAKGKSALMFNLQEPYFTWPLIAADGGYAFKYENGYDIK
DVGVDNAGAKAGLTFLVDLIKNKHMNADTDYSIAEAAFNKGETAM
TINGPWAWSNIDTSKVNYGVTVLPTFKGQPSKPFVGVLSAGINAA
SPNKELAKEFLENYLLTDEGLEAVNKDKPLGAVALKSYYEELVKD
PRIAATMENAQKGEIMPNIPOMSAFWYAVRTAVINAASGRQTVDE
ALKDAQTNSSSNNNNNNNNNLGDGDDDKVPEFLEVLFGQGPLGSM
FTPGKQSSSRASSGNRSGNGILKWADQSDQFRNVQTRGRRAQPKQ
TATSQQPSGGNVVPYYSWFSGITQFQKGKEFEFAEQGVPIAPGV
PATEAKGYWYRHNRRSFKTADGNQRQLLPRWYFYLLGTGPHAKDQ
YGTDINGVYWVASNQADVNTPADIVDRDPSSDEAIPTRFPPGTVL
PQGYIEGSGRSAPNSRSTSRSTSSRASSAGSRSRANSNRTPTSG
VTPDMADQIASLVLAKLGKDATKPOQVTKHTAKEVRQKILNKPRQ
KRSPNKQCTVQOCFGKRGNQNFGGGEMLKLGTSDPQFPILAELA
PTAGAFFFGSKLELAKVQNLSGNPDEPQKDVYELRYNGAIRFDST
LSGFETIMKVLSENLNAYQQQDGMVNMSPKPQRQRGHKNGQGEN
NISVAVPKSRVQQNKSIELTAEDISLLKMDPEPFTEDTSEI

Native sequence Amino acids M1 – I448 (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 (LEVLFQGP) residues 393 – 400