

MRC PPU Reagents and Services

Standard Operating Procedure

Preparation of Spike Protein [1 – 1353] MERS CoV

Enzyme description:- MERS S Protein [1 - 1353]

Clone number:- DU 67745

Source:- Recombinant

Expression system:- *E. coli*

Tag:- N-terminal GST

Purification method:- GSH Agarose

Calculated molecular mass:-

Monoisotopic 176, 107.10 daltons

Average Mass 176, 220.26 daltons

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 5.75

Purity:- 65 %

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

Spike Protein [1 – 1353] MERS CoV

Protein MERS S Protein [1 - 1353]

Clone number DU 67745

Accession number AHI48572.1

Tags N-terminal GST

Bacterially
expressed protein

MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKK
FELGLEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERA
EISMLEGAVLDIRYGVSRIAYSKDFETLKVDVFLSKLPEMLKMFED
RLCHKTYLNGDHVTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFK
KRIEAIPOIDKYLKSSKYIAWPLQGWQATFGGGDHPPKSDLEVL
FQGPLGSMIHSVFLLMFLLTPTESYVDVGPDSVKSACIEVDIQQTF
FDKTWPRPIDVSKADGIIYPQGRYTSNITITYQGLFPYQGDHGM
YVYSAGHATGTTPOKLFVANYSQDVKQFANGFVVRIGAAANSTGT
VIISPSTSATIRKIYPAPMLGSSVGNFSDGKMGRFFNHTLVLLPD
GCGTLLRAFYCILEPRSGNHCPAGNSYTSFATYHTPATDCSDGNY
NRNASLNSFKEYFNLRNCTFMYTYNITEDEILEWFGITQTAQGVH
LFSSRYVDLYGGNMFQFATLPVYDTIKYYSIIPHSIRSIOQSDRKA
WAAFYVYKLQPLTFLDFSDVGYIRRAIDCGFNDLSQLHCSYESF
DVEGVSYSVSSFEAKPSGSVVEQAEGVECDFSPLLSGTPPQVYNF
KRLVFTNCNYNLTKLLSLFSVNDFTCSQISPAAIASNCYSSLILD
YFSYPLSMKSDLVSSAGPISQFNYKQSFNPTCLILATVPHNLT
TITKPLKYSYINKCSRLSDDRTEVPQLVNAVQYSPCVSIVPSTV
WEDGDYRQQLSPLEGGWLVASGSTVAMTEQLQMGFGITVQYGT
DTNSVCPKLEFANDTKIASQLGNCVEYSLYGVSGRGVFNQCTAVG
VRQRFVYDAYQNLVGYYSDDGNYCLRACVSVVSVIYDKETKT
HATLFGSVACEHISSTMSQYSRSTRSMLKRRDSTYGPLQTPVGCV
LGLVNSSLFVEDCKLPLGQSLCALPDTPTSTLTPRSVRSVPGEMRL
ASIAFNHPIQVDQLNSSYFKLSIPTNFSFGVTQEYIQTTIQKVTV
DCKQYVCNGFQKCEQLLREYGQFCSKINQALHGANLRQDDSVRNL
FASVKSSQSSPIIPGFGGDFNLTLEPVSI STGSRARSARSAIEDLL
FDKVTIADPGYMQYDDCMQOGPASARDLICAQYVAGYKVLPLM
DVNMEAAYTSSLLGSIAGVGWTAGLSSFAAIPFAQSIFYRLNGVG
ITQQVLSNQKLIANKFNQALGAMQTGFTTTNEAFRKVQDAVNNN
AQALSKLASELSNTFGAISASIGDIIQRLDVLEQDAQIDRLINGR
LTTLNAFVAQQLVRSESAALSAQLAKDKVNECVKAQSKRSGFCGQ
GTHIVSFVFNAPNGLYFMHVGYYPSNHIEVVSAYGLCDAANPTNC
IAPVNGYFIKTNNTTRIVDEWSYTGSSFYAPEPITSLNNTKYVAPQV

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**TYQNI STNLPPPLLGNSTGIDFQDELDEFFKNVSTSI PNFGSLTQ
INTLLDLTYEMLSLOQVVKALNESYIDLKELGNYTYYNKWPWYI
WLGFIAGLVALALCVFFILCCTGCGTNCMGKLCNRCCDRYEEYD
LEPHKVHVH**

Native sequence

Amino acids M1 – H1353 (end).

Residue M232 of the fusion protein is equivalent to M1 of the native enzyme. The GST tag is located at residues 1 – 220.

Protease cleavage

PreScission (LEVLFQGP) residues 221 – 228