MRC PPU Reagents and Services

Standard Operating Procedure

Preparation of SPIKE Protein [1 - 1273] SARS CoV2

Enzyme description:- S Protein [1 - 1273]

Clone number:- DU 67743

Source:- Recombinant

Expression system:- *E. coli*

Tag:- N-terminal MBP

Purification method:- Amylose resin

Calculated molecular mass:-
Monoisotopic 185, 299.77 daltons
Average Mass 185, 417.39 daltons
[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 5.59

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 – 1273] SARS CoV2

Protein S Protein [1 - 1273]

Clone number DU 67743

Accession number QHD43416.1

Tags N-terminal MBP

Bacterially expressed protein

MKIEEGKLVIWINGDKGYNGLAEVGKKFEKDTGIKVTVEHPDKLE
EKFPQVAATGDGPDIIFWAHDRFGGYAQSGLLAEITPDKAFQDKL
YPFTWDAVRYNGKLIAYPIAVEALSLIYNKDLLPNPPKTWEEIPA
LDKELKAKGKSALMFNLQEPYFTWPLIAADGGYAFKYENGKYDIK
DVGVDNAGAKAGLTFLVDLIKNKHMNADTDYSIAEAAFNKGETAM
TINGPWAWSNIDTSKVNYGVTVLPTFKGQPSKPFVGVLSAGINAA
SPNKELAKEFLENYLLTDEGLEAVNKDKPLGAVALKSYEEELVKD
PRIAATMENAQKGEIMPNIPQMSAFWYAVRTAVINAASGRQTVDE
ALKDAQTNSSSNNNNNNNNNLGDDDDKVPEFLEVLFQGPLGS**MF
VFLVLLPLVSSQCVNLTTRTQLPPAYTNSFTRGVYYPDKVFRSSV
LHSTQDLFLPFFSNVTWFHAIHVSGTNGTKRFDNPVLPFNDGVYF
ASTEKSNIIRGWIFGTTLDSKTQSLLIVNNATNVVIKVCEFQFCN
DPFLGVYYHKNNKSWMESEFRVYSSANNCTFEYVSQPFLMDLEGK
QGNFKNLREFVFKNIDGYFKIYSKHTPINLVRDLPQGFSALEPLV
DLPIGINITRFQTLLALHRSYLTPGDSSSGWTAGAAAYYVGYLQP
RTFLLKYNENGTITDAVDCALDPLSETKCTLKSFTVEKGIYQTSN
FRVQPTESIVRFPNITNLCPFGEVFNATRFASVYAWNRKRISNCV
ADYSVLYNSASFSTFKCYGVSPTKLNDLCFTNVYADSFVIRGDEV
RQIAPGQTGKIADYNYKLPDDFTGCVIAWNSNNLDSKVGGNYNYL
YRLFRKSNLKPFERDISTEIYQAGSTPCNGVEGFNCYFPLQSYGF
QPTNGVGYQPYRVVVLSFELLHAPATVCGPKKSTNLVKNKCVNFN
FNGLTGTGVLTESNKKFLPFQQFGRDIADTTDAVRDPQTLEILDI
TPCSFGGVSVITPGTNTSNQVAVLYQDVNCTEVPVAIHADQLTPT
WRVYSTGSNVFQTRAGCLIGAEHVNNSYECDIPIGAGICASYQTQ
TNSPRRARSVASQSIIAYTMSLGAENSVAYSNNSIAIPTNFTISV
TTEILPVSMTKTSVDCTMYICGDSTECSNLLLQYGSFCTQLNRAL
TGIAVEQDKNTQEVFAQVKQIYKTPPIKDFGGFNFSQILPDPSKP
SKRSFIEDLLFNKVTLADAGFIKQYGDCLGDIAARDLICAQKFNG
LTVLPPLLTDEMIAQYTSALLAGTITSGWTFGAGAALQIPFAMQM
AYRFNGIGVTQNVLYENQKLIANQFNSAIGKIQDSLSSTASALGK
LQDVVNQNAQALNTLVKQLSSNFGAISSVLNDILSRLDKVEAEVQ
IDRLITGRLQSLQTYVTQQLIRAAEIRASANLAATKMSECVLGQS
KRVDFCGKGYHLMSFPQSAPHGVVFLHVTYVPAQEKNFTTAPAIC

HDGKAHFPREGVFVSNGTHWFVTQRNFYEPQIITTDNTFVSGNCD
VVIGIVNNTVYDPLQPELDSFKEELDKYFKNHTSPDVDLGDISGI
NASVVNIQKEIDRLNEVAKNLNESLIDLQELGKYEQYIKWPWYIW
LGFIAGLIAIVMVTIMLCCMTSCCSCLKGCCSCGSCCKFDEDDSE
PVLKGVKLHYT

Native sequence Amino acids M1 – T1273 (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