

MRC PPU Reagents and Services

Standard Operating Procedure

Preparation of NSP4 [386 - 500] SARS CoV2

Enzyme description:- NSP4 [386 - 500]

Clone number:- DU 75131

Source:- Recombinant

Expression system:- *E. coli*

Tag:- N-terminal GST

Purification method:- GSH Agarose

Calculated molecular mass:-

Monoisotopic 40, 132.26 daltons

Average Mass 40, 158.39 daltons

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 6.72

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

MRC PPU Reagents and Services

Clone Data Sheet

NSP4 [386 – 500] SARS CoV2

Protein NSP4 [386 - 500]

Clone number DU 75131

Accession number QHD43415.1

Tags N-terminal GST

Bacterially expressed protein MSPILGYWKIKGLVQPTRLLEYLEEKYEEHLYERDEGDKWRNKK
FELGLEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERA
EISMLEGAVLDIRYGVSRIAYS KDFETLKVDFLSKLPEMLKMFED
RLCHKTYLNGDHVTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFK
KRIEAI PQIDKYLKSSKYIAWPLQGWQATFGGGDHPPKSDLEVL
FQGPLGS **STKHFYWFFSNYLRVVFNGVSFSTFEEAALCTFLLNK**
EMYLKLRSDVLLPLTQYNRYLALYNKYKYFSGAMDTTSYREAACC
HLAKALNDFSNVSGSDVLYQPPQTSITSAVLQ

Native sequence Amino acids S386 – Q500 (end residue).
Residue S232 of the fusion protein is equivalent to S386 of the
native enzyme. The GST tag is located at residues 1 – 220.

Protease cleavage PreScission (LEVLFQGP) residues 221 - 228