MRC PPU Reagents and Services

Standard Operating Procedure

Preparation of ORF3B [1 - 151] SARS CoV2

Enzyme description:- ORF3B [1 - 151]

Clone number:- DU 68507

Source:- Recombinant

Expression system:- *E. coli*

Tag:- N-terminal GST

Purification method:- GSH Agarose

Calculated molecular mass:-
Monoisotopic 44, 102.73 daltons
Average Mass 44, 131.44 daltons
[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 5.14

Purity:- 75 %

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

ORF3B [1 – 151] SARS CoV2

Protein	ORF3B [1 - 151]
Clone number	DU 68507
Accession number	QHD43419.1
Tags	N-terminal GST
Bacterially expressed protein	MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKK FELGLEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERA EISMLEGAVLDIRYGVSRIAYSKDFETLKVDFLSKLPEMLKMFED RLCHKTYLNGDHVTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFK KRIEAIPQIDKYLKSSKYIAWPLQGWQATFGGGDHPPKSDLEVLF QGPLGS**MRLWLCWKCRSKNPLLYDANYFLCWHTNCYDYCIPYNSV TSSIVITSGDGTTSPISEHDYQIGGYTEKWESGVKDCVVLHSYFT SDYYQLYSTQLSTDTGVEHVTFFIYNKIVDEPEEHVQIHTIDGSS GVVNPVMEPIYDEPTTTTSVPL**
Native sequence	Amino acids M1 - L151 (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