

# *MRC PPU Reagents and Services*

## **Standard Operating Procedure**

### **Preparation of ORF3A [1 - 275] SARS CoV2**

**Enzyme description:-** ORF3A [1 - 275]

**Clone number:-** DU 67698

**Source:-** Recombinant

**Expression system:-** *E. coli*

**Tag:-** N-terminal GST

**Purification method:-** GSH Agarose

**Calculated molecular mass:-**

Monoisotopic 57, 909.46 daltons

Average Mass 57, 947.10 daltons

[cysteines reduced, methionines have not been oxidised]

**Theoretical pI:-** 5.64

**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

### ORF3A [1 – 275] SARS CoV2

**Protein** ORF3A [1 - 275]

**Clone number** DU 67698

**Accession number** QHD43417.1

**Tags** N-terminal GST

**Bacterially  
expressed protein**

MSPILGYWKIKGLVQPTRLLEYLEEKYEHLIERDEGDKWRNKK  
FELGLEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERA  
EISMLEGAVLDIRYGVSR IAYS KDFETLKVDFLSKLPEMLKMFED  
RLCHKTYLNGDHVTHPDMFLYDALDVVLYMDPMCLDAFPKLVCFK  
KRIEAIPOIDKYLKSSKYIAWPLOGWQATFGGGDHPPKSDLEVL  
**QGPLGSMDLFRIFTIGTVTLKQGEIKDATPSDFVRATATPIQA**  
**SLPFGWLI VGVALLAVFQSASKIITLKKRWQLALS KGVHFVCNLL**  
**LLFVTVYSHLLLVAAGLEAPFLYLYALVYFLQSIN FVRIIMRLWL**  
**CWKRSKNPLLYDANYFLCWHTNCYDYCIPYNSVTSSIVITSGDG**  
**TTSPISEHDYQIGGYTEKWESGVKDCVVLHSYFTSDYYQLYSTQL**  
**STD TGVEHVTFFIYNKIVDEPEEHVQIHTIDGSSGVVNPVMEPIY**  
**DEPTTTTSVPL**

**Native sequence** Amino acids M1 – L275 (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