

## *MRC PPU Reagents and Services*

### **Standard Operating Procedure**

#### **Preparation of Influenza D Virus NP [1 – 552]**

**Enzyme description:-** IDV NP [1 - 552]

**Clone number:-** DU 75220

**Source:-** Recombinant

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

**Tag:-** N-terminal MBP

**Purification method:-** Amylose Resin

**Calculated molecular mass:-**

Monoisotopic 105, 464.57 daltons

Average Mass 105, 531.18 daltons

[cysteines reduced, methionines have not been oxidised]

**Theoretical pI:-** 7.86

**Purity:-** 80 %

**Enzyme storage buffer:-**

50 mM Tris-HCl pH 7.5, 270 mM Sucrose, 150 mM NaCl, 0.1 mM EGTA, 0.5 mM TCEP

**Storage temperature:-** -70 °C

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**Clone Data Sheet**

**Influenza D Virus NP [1 – 552]**

<b><u>Protein</u></b>	IDV NP [1 - 552]
<b><u>Clone number</u></b>	DU 75220
<b><u>Species</u></b>	Influenza D virus (IDV) strain D/bovine/Oklahoma/660/2013
<b><u>Tags</u></b>	N-terminal MBP
<b><u>Bacterially expressed protein</u></b>	<p>MKIEEGKLVIIWINGDKGYNGLAEVGGKFEKDTGIKVTVEHPDKLEEKFP QVAATGDGPDIIIFWAHDRFGGYAQSGLLAEITPDKAFQDKLYPFTWDAV RYNGKLIAYPIAVEALSIIYNKDLLPNPPKTWEEIPALDKELKAKGKSA LMFNLQEPYFTWPLIAADGGYAFKYENGGYDIKDVGVNDAGAKAGLTF VDLIKKNHNMADTDYSIAEAAFNKGETAMTINGPWAWNSIDTISKVNYGV TVLPTFKGQPSKPFVGVLSAGINAASPNKELAKEFLENYLLTDEGLEAV NKDKPLGAVALKSYYEELVKDPRIAATMENAQKGEIMPNI PQMSAFWYA VRTAVINAASGRQTVDEALKDAQTNSSNNNNNNNNNNNLGDDDDKVPEF <u>LEVLFQGP</u>LGSM<b>DKAQTPEEQRAKNAKTILENIQIYERMCDLFGVSE</b> <b>DDKLI IENSI SIERMIRVVTDKKYQDKKLKNAGSDLEKIANAGKVFCRL</b> <b>VESTAGKCSARLGMALKNVEAVLTDVLDGNELDRAAVLGKRMGFTAMFK</b> <b>SNLEEVLYQRGKNQLKKRNAEFTLSQASLEARFRPIMEKHLGVGTV</b> <b>VASIKNILASKKNGNYRNKMVRKPGGNRESWSPLEREISFLNKKLFPGP</b> <b>MRQLCKKFEYLNDOEKQLALNLMLDASLILKPQVTHKMIMPWSMWLAVK</b> <b>KYAEMNKGSPSLEDLAAYSGVRAFMAFNTACYMSKFTIGKGI VGD AEIM</b> <b>ENGNDKMQTLAMACFGLAYEDTGIVAAMISQPMKKRYQLRVGNFNPPEE</b> <b>GTIKGTSAGYFHKWAEFGNRLPFNSFGTGESKQISNSGVFAVQRPSTTN</b> <b>IQRLAELMARNTGETSDNFTQLVQKIREQVGT FADQKANLREFTGGYIY</b> <b>DITDVTKSNPKIPQLGGDSFFFEFTGSDVPRTGAKRRVGGADDVTPGTS</b> <b>QPKKRGRQGAGAESSMDIETVGED</b></p>
<b><u>Native sequence</u></b>	Amino acids M1 – N552 (end residue) of IDV NP protein. Residue M408 of the fusion protein is equivalent to M1 of the native enzyme. The MBP tag is located at residues 1 – 367.
<b><u>Protease cleavage</u></b>	PreScission ( <u>LEVLFQGP</u> ) residues 393 - 400