

MRC PPU Reagents and Services

Standard Operating Procedure

Preparation of Influenza B Virus NS1 [12 – 281]

Enzyme description:- IBV NS1 [12 - 281]

Clone number:- DU 75476

Source:- Recombinant

Expression system:- *E.coli*

Tag:- N-terminal MBP

Purification method:- Amylose Resin

Calculated molecular mass:-

Monoisotopic 74, 999.93 daltons

Average Mass 75, 046.69 daltons

[cysteines reduced, methionines have not been oxidised

Theoretical pI:- 5.28

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 B Virus NS1 [12 – 281]

<u>Protein</u>	IBV NS1 [12 - 281]
<u>Clone number</u>	DU 75476
<u>Species</u>	Influenza B virus (IBV) strain B/Florida/04/2006
<u>Tags</u>	N-terminal MBP
<u>Bacterially expressed protein</u>	MMKIEEGKLVIWINGDKGYNGLAEVGKKFEKDTGIKVTVHPDKLEEKFPQVAATGDGPDIIFWAHDRFGGYAQSGLLAEITPDKAFAQDKLYPFTWDA VRYNGKLIAYPIAVEALSLIYNKDLPNPPKTWEETPALDKELKAKGKS ALMFNLQE PYFTWPLIAADGGYAFKYENGKYDIKVGVNDAGAKAGLTF LVDLIKNKHMNADTDYSIAEAAFNKGETAMTINGPWAWSNIDTSKVNYG VTVLPTFKGQPSKPFVGVLISAGINAASPNKELAKEFLENYLLTDEGLEA VNVDKPLGAVALSKYEEELVKDPRIAATMENAQKGEIMPNIPQMSAFWY AVR TAVINAASGRQTVD EALKDAQTNSSNNNNNNNNNLGDDDKVPE FLEVLFQGPLGSVPGATNATINFEAGILECYERLSWQRALDYPGQDRL NRLKRKLESRIKTHNKSEPE SKRMSLEERKAIGVKMMKVLLFMNPSAGIEGFE PYCMKSSNSNCPKYNWTDYPSTPGRC LDDIEEPDDVDGPTEIV LRDMNNKDARQKIKEEVNTQKEGKFRLTIKRDIRNVLSRVLVNGTFLK HPNGYKSILSTLHRLNAYDQSGRLVAKLVATDDLTV EDEEDGHRI LNSLF ERLNEGHSKPIRAAETAVGVLSQFGQEHRLSPEEGDN
<u>Native sequence</u>	Amino acids V12 – N281 (end residue) of IBV NS1 protein. Residue V404 of the fusion protein is equivalent to V12 of the native enzyme. The MBP tag is located at residues 1 – 367.
<u>Protease cleavage</u>	PreScission (<u>LEVLFQGP</u>) residues 393 - 400