

## *MRC PPU Reagents and Services*

### **Standard Operating Procedure**

#### **Preparation of Influenza A Virus PB1-F2 [1 – 87]**

**Enzyme description:-** IAV PB1-F2 [1 - 87]

**Clone number:-** DU 73898

**Source:-** Recombinant

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

**Tag:-** N-terminal MBP

**Purification method:-** Amylose Resin

**Calculated molecular mass:-**

Monoisotopic 54, 744.93 daltons

Average Mass 54, 779.22 daltons

[cysteines reduced, methionines have not been oxidised]

**Theoretical pI:-** 5.94

**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 A Virus PB1-F2 [1 – 87]**

<b><u>Protein</u></b>	IAV PB1-F2 [1 - 87]
<b><u>Clone number</u></b>	DU 73898
<b><u>Species</u></b>	Influenza A virus (IAV) strain A/Puerto Rico/8/1934(H1N1) ('PR8')
<b><u>Tags</u></b>	N-terminal MBP
<b><u>Bacterially expressed protein</u></b>	<p>MKIEEGKLVIIWINGDKGYNGLAEVGGKFEKDTGIKVTVEHPDKLEEKFP QVAATGDGPDIIIFWAHDRFGGYAQSGLLAEITPDKAFQDKLYPFTWDAV RYNGKLIAYPIAVEALSLIYNKDLLPNPPKTWEEIPALDKELKAKGKSA LMFNLQEPYFTWPLIAADGGYAFKYENGGYDIKDVGVNDNAGAKAGLTFL VDLIKKNKHMNADTDYSIAEAAFNKGETAMTINGPWAWSNIDTSKVNYGV TVLPTFKGQPSKPFVGVLSAGINAASPNKELAKEFLENYLLTDEGLEAV NKDKPLGAVALKSYEEELVKDPRIAATMENAQKGEIMPNI PQMSAFWYA VRTAVINAASGRQTVDEALKDAQTNSSNNNNNNNNNNLGDDDDKVPEF <u>LEVL</u><u>FQGP</u><u>LGSMGQEQDTPWILSTGHI</u><u>STQKRE</u><u>DGQQT</u><u>PKLEHRNSTR</u> <u>MGHCQKTMNQVVMPKQIVYWRRWLSLRNPILVFLKTRVLKRWRLFSKHE</u></p>
<b><u>Native sequence</u></b>	<p>Amino acids M1 – E87 (end residue) of IAV PB1-F2 protein. Residue M404 of the fusion protein is equivalent to M1 of the native enzyme. The MBP tag is located at residues 1 – 367.</p>
<b><u>Protease cleavage</u></b>	PreScission ( <u>LEVL</u> <u>FQGP</u> ) residues 393 - 400