

***MRC PPU Reagents and Services***

**Standard Operating Procedure**

**Preparation of Influenza A Virus PA [1 – 716]**

**Enzyme description:-** IAV PA [1 - 716]

**Clone number:-** DU 70826

**Source:-** Recombinant

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

**Tag:-** N-terminal MBP

**Purification method:-** Amylose Resin

**Calculated molecular mass:-**

Monoisotopic 127, 746.64 daltons

Average Mass 126, 827.27 daltons

[cysteines reduced, methionines have not been oxidised

**Theoretical pI:-** 5.18

**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 PA [1 – 716]

<b><u>Protein</u></b>	IAV PA [1 - 716]
<b><u>Clone number</u></b>	DU 70826
<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>	MKIEEGKLVIWINGDKGYNGLAEVGKKFEKDTGIKVTVEHPDKLEEKFPQVAATGDGPDIIFWAHDRFGGYAQSGLLAEITPDKAQDKLYPFTWDARYNGKLIAPIAVEALSLIYNKDLLPNPPKTWEETIPALDKELAKGKSA LMFnLQE PYFTWPLIAADGGYAFKYENGKYD IKDVGVDNAGAKAGLTFL VDLIKNKHMNADTDYSIAEAAFNKG ETAMT INGPWAWSNI DTSKVNYGV TVLPTFKGQPSKPFVGVL SAGINAASPNKELAKEFLENYLLTDEGLEAV NDKPLGAVALKS YEEELVKDPRIAATMENA QKGEIMPNI PQMSAFWYA VRTAVINAASGRQTVDEALKDAQTNTSSNNNNNNNNNLGDDDKVPEF LEVLFQGPLGS MEDFVRQCFNPMIVELAEKTMKEYGEDLKIE TNKFAAI CTHLEVCFMYSDFH FINEQGES I IVELGDPN ALLKHRFE II EGRDRTMA WTVVNSICNTTGAEPKPKFLPDLYD YKENRFIEIGVTRREVHIYYLEKAN KIKSEKTHIHIFSFTGEEMATKADYTLDEESRARIKTRLFTIRQEMASR GLWDSFRQSERGEETIEERFEITGTMRKLADQSLPPNFSSL ENFRAYVD GFEPNGYIEGKLSQMSKEVNARIEPFLKTTPRPLRPNGPPCSQR SKFL LMDALKL SIEDPSHEGEGIPLYDAIKCMRTFGWKEPNVVKPHEKGINP NYLLSWKQVLAELQDIENEKIPKTKNMKKTSQLKWALGENMAPEKVD F DDCKDVGDLKQYDSDEPELRSLASWIQNEFNKACELTDSSWIELDEIGE DVAPIEHIASMRRNYFTSEVSHCRATEYIMKGVYINTALLNASCAAMDD FQLIPMISKCRTKEGRRKTNLYGFIIKGRSHLRNDTDVVNFVSMEFSLT DPRLEPHKWEKYCVLEIGDMLIRSAIGQVS RPMFLYVRTNGTSKIKMKW GMEMRRCLLQSLQQIESMIEAESSVKEKDMTKEFFENKSETWPIGESPK GVEESSIGKVCRTILLAKSVFNSLYASPQLEGFSAESRKLLLIVQALRDN LEPGTFDLGGLYEAIEECLINDPWVLLNASWFNSFLTHALS
<b><u>Native sequence</u></b>	Amino acids M1 – S716 (end residue) of IAV PA protein. Residue M404 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