

MRC PPU Reagents and Services

Standard Operating Procedure

Preparation of Influenza A Virus PA-X [192 - 252]

Enzyme description:- IAV PA-X [192 - 252]

Clone number:- DU 73892

Source:- Recombinant

Expression system:- *E.coli*

Tag:- N-terminal MBP

Purification method:- Amylose Resin

Calculated molecular mass:-

Monoisotopic 51, 180.14 daltons

Average Mass 51, 148.10 daltons

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 5.61

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-X [192 – 253]

<u>Protein</u>	IAV PA-X [192 - 253]
<u>Clone number</u>	DU 73892
<u>Species</u>	Influenza A virus (IAV) strain A/Puerto Rico/8/1934(H1N1) ('PR8')
<u>Tags</u>	N-terminal GST
<u>Bacterially expressed protein</u>	MKIEEGKLVIIWINGDKGYNGLAEVGGKFEKDTGIKVTVEHPDKLEEKFP QVAATGDGPDIIIFWAHDRFGGYAQSGLLAEITPDKAFQDKLYPFTWDAV RYNGKLIAYPIAVEALSIIYNKDLLPNPPKTWEEIPALDKELKAKGKSA LMFNLQEPYFTWPLIAADGGYAFKYENGGYDIKDVGVNDAGAKAGLTF VDLIKKNKHMNADTDYSIAEAAFNKGETAMTINGPWAWNSNIDTSKVNYGV TVLPTFKGQPSKPFVGVLSAGINAASPNKELAKEFLENYLLTDEGLEAV NKDKPLGAVALKSYYEELVKDPRIAATMENAQKGEIMPNI PQMSAFWYA VRTAVINAASGRQTVDEALKDAQTNSSSNNNNNNNNNNLGDDDDKVPEF <u>LEVLFQGPLGSVSPREEKRQLKKGLKSQEQCASLPTKVSRRRTSPALKIL</u> EPMWMSNRATLRASCLKCPKK
<u>Native sequence</u>	Amino acids V192 – K252 (end residue) of IAV PA-X protein. Residue V404 of the fusion protein is equivalent to V192 of the native enzyme. The MBP tag is located at residues 1 – 367.
<u>Protease cleavage</u>	PreScission (<u>LEVLFQGP</u>) residues 393 - 400