

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 70818

Source:- Recombinant

Expression system:- *E.coli*

Tag:- N-terminal GST

Purification method:- GSH Agarose

Calculated molecular mass:-

Monoisotopic 109, 341.91 daltons

Average Mass 109, 412.52 daltons

[cysteines reduced, methionines have not been oxidised

Theoretical pI:- 5.46

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]

<u>Protein</u>	IAV PA [1 - 716]
<u>Clone number</u>	DU 70818
<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>	MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHYERDEGDKWRNKKFELG LEFPNLPPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAESMLEGA VLDIIRYGVSRRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDH VTHPDFMLYDALDVLYMDPMCLDAFPKLVCFKKRIEAIPOQIDKYLKSS KYIAWPLQGWQATFGGGDHPPKS <u>DLEVLFQGPLGS</u> MEDFVRQCFNPMIV E LA E K T M K E Y G E D L K I E T N K F A A I C T H L E V C F M Y S D F H F I N E Q G E S I I V E L G D P N A L L K H R F E I I E G R D R T M A W T V V N S I C N T T G A E K P K F L P D L Y D Y KENRFIEIGVTRREVHIYYLEKANKIKSEKTHIHFISFTGEEMATKADY TLDEESRARIKTRLFTIRQEMASRGLWDSFRQSERGEETIEERFEITGT MRKLADQSLPPNFSSLLENFRAYVDGFEPNGYIEGKLSQMSKEVNARIEP FLKTTPRPLRLPNPPCSQRSKFLLMDALKSIEDPSHEGEGIPLYDAI KCMRTFFGWKEPNVVKPHEKGINPNYLLSWKQVLAELQDIENEKIPKT KNMKKTSQLKWALGENMAPEKVDFFDDCKDVGDLKQYDSDPELRSLASW IQNEFNKACELTDSSWIELDEIGEDVAPIEHIASMRRNYFTSEVSHCRA TEYIMKGVYINTALLNASCAAMDDFQLIPMISKCRTKEGRRTNLYGFI IKGRSHLRNDTDVVNFVSMEFSLTDPRLEPHKWEKYCVLEIGDMLIRSA IGQVSRPMFLYVRTNGTSKIKMKWGMEMRRCLLQSLQQIESMIEAESSV KEKDMDTKEFFENKSETWPIGESPKGVEESSIGKVCRTLLAKSVFNSLYA SPQLEGFSAESRKLLLIVQALRDNLEPGTFDLGGLYEAIEECLINDPWV LLNASWFNSFLTHALS
<u>Native sequence</u>	Amino acids M1 – S716 (end residue) of IAV PA protein. Residue M232 of the fusion protein is equivalent to M1 of the native enzyme. The GST tag is located at residues 1 – 220.
<u>Protease cleavage</u>	PreScission (<u>LEVLFQGP</u>) residues 221 - 229