

MRC PPU Reagents and Services

Standard Operating Procedure

Preparation of Influenza B Virus NEP (12 – 122)

Enzyme description:- IBV NEP (12 – 122)

Clone number:- DU 75488

Source:- Recombinant

Expression system:- *E.coli*

Tag:- N-terminal GST

Purification method:- GSH Sepharose

Calculated molecular mass:-

Monoisotopic 39, 853.45 daltons

Average Mass 39, 879.36 daltons

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 6.18

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 B Virus NEP (12 – 122)

<u>Protein</u>	IBV NEP (12 – 122)
<u>Clone number</u>	DU 75488
<u>Species</u>	Influenza B virus (IBV) strain B/Florida/04/2006
<u>Tags</u>	N-terminal GST
<u>Bacterially expressed protein</u>	MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELG LEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLEGA VLDIRYGVSR IAYS KDFETLKVDFLSKLP EMLKMFEDRLCHKTYLNGDH VTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAI PQIDKYLKSS KYIAWPLQGWQATFGGGDHPPKSDLEVL <u>FQGPLGS</u> WRMKKMAIGSSIHS SSVLMKDIQSQFEQLKLRWESYPNLVKSTDYHQKRETIRLVTEELYLLS KRIDDNILFHKTVIANS SIIADMVVSLSLLETLYEMKDVVEVYSRQCL
<u>Native sequence</u>	Amino acids W12 – L122 (end residue) of IBV NEP protein. Residue W232 of the fusion protein is equivalent to D11 of the native enzyme. The GST tag is located at residues 1 – 220.
<u>Protease cleavage</u>	PreScission (<u>LEVLFQGP</u>) residues 221 - 229