

Division of Signal Transduction Therapy

Standard Operating Procedure

Preparation of HP1 beta (mouse) [1 – 185]

Enzyme description:- HP1 beta (mouse) [1 – 185]

Clone number:- DU 36262

Source:- Recombinant

Expression system:- *E.coli*

Tag:- N-terminal GST

Purification method:- GSH Sepharose

Calculated molecular mass:-

Monoisotopic 48, 211.21 daltons

Average Mass 48, 241.94 daltons

[cysteines reduced, methionines have not been oxidised

Theoretical pI:- 5.13

Purity:- >80 %

Enzyme storage buffer:-

50 mM Tris-HCl pH 7.5, 270 mM Sucrose, 150 mM NaCl, 0.1 mM EGTA,
0.1 % 2-mercaptoethanol, 0.02 % Brij-35, 1 mM benzamidine, 0.2 mM PMSF

Storage temperature:- -70 °C

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Clone Data Sheet

HP1 beta (mouse) [1 – 185]

<u>Protein</u>	HP1 beta (mouse) [1 – 185]
<u>Clone number</u>	DU 36262
<u>Species</u>	Mouse
<u>Accession number</u>	NM_007622.3
<u>Tags</u>	N-terminal GST
<u>Bacterially expressed protein</u>	MSPILGYWKIKGLVQPTRLLEYLEEKYEEHYERDEGDKWRNKKFELG LEFPNLPPYYIDGDVKLTQSMAIIRYIADKHNLGGCPKERAESIMLEGA VLDIHYGVSRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDH VTHPDFMLYDALDVLYMDPMCLDAFPKLVCFKKRKIEAIPQIDKYLKSS KYIAWPLQGWQATFGGGDHPPKSDELVLFQGPLGS MGKKQNKVV EVL EEEEEEYVVEKVLDRRVVKGVYLLKWKGFSDEDNTWEPEENLDCPDL IAEFLQSQKTAHETDKSEGGKRKADSDSEDKGEESKPKKKKESEKPRG FARGLEPERIIGATDSSGELMFLMKWKSDEADLVPAKEANVKCPQVVI SFYEERLTWHSYPSSEDDKKDDKN
<u>Native sequence</u>	Amino acids M1 – N185 (end) of mouse HP1 beta. 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 - 228
<u>Cloning sites</u>	<i>Bam</i> H1 and <i>Not</i> 1 sites of pGEX6P-1

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<u>Nucleotide Sequence of insert</u>	
	ggatccATGGGAAAAAGCAAAACAAGAAGAAAGTGGAGGAGGTACTAG AAGAAGAGGAAGAGGAATATGTGGTGGAAAAGTTCTGATCGCGAGT TGTCAAGGGCAAGGTGGAATATCTTCTAAAGTGGAAAGGGTTCTCAGAT GAGGACAAACACTTGGGAGCCAGAAGAGAATCTGGATTGCCCTGACCTTA TTGCTGAGTTCTACAGTCACAGAAAACAGCTCATGAGACAGATAAGTC AGAGGGAGGCAGCGCAAAGCTGATTCTGAAGATAAAAGGAGAG GAAAGCAAACCAAAGAAGAAGAAAGAAGAGTCAGAAAAGCCACGAGGCT TTGCCCGGGTTGGAGCCAGAGCGGATTATTGGAGCTACTGACTCCAG TGGAGAGCTCATGTTCTGATGAAATGGAAAAACTCTGATGAGGCTGAC CTGGTCCCTGCCAAGGAAGCCAATGTCAAGTGCCCACAGGTTGTCAATAT CCTTCTATGAGGAAAGGCTAACGTGGCATT CCTACCCCTCAGAGGATGA TGACAAAAAAGACGACAAGAATTAGCGGCCGC