Division of Signal Tranduction Therapy

Standard Operating Procedure

Preparation of HP1 alpha (mouse) [1 – 191]

Enzyme description:- HP1 alpha (mouse) [1 – 191]

Clone number:- DU 36261

Source:- Recombinant

Expression system:- *E.coli*

Tag:- N-terminal GST

Purification method:- GSH Sepharose

Calculated molecular mass:-

Monoisotopic	48, 978.63 daltons
Average Mass	49, 010.12 daltons

[cysteines reduced, methionines have not been oxidised

Theoretical pI:- 5.73

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

Clone Data Sheet

HP1 alpha (mouse) [1 – 191]

Protein	HP1 alpha (mouse) [1 – 191]
Clone number	DU 36261
Species	Mouse
Accession number	NM_007626.3
Tags	N-terminal GST
Bacterially expressed protein	MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELG LEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLEGA VLDIRYGVSRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDH VTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPQIDKYLKSS KYIAWPLQGWQATFGGGDHPPKSDLEVLFQGPLGS**MGKKTKRTADSSSS** **EDEEEYVVEKVLDRRMVKGQVEYLLKWKGFSEEHNTWEPEKNLDCPELI** **SEFMKKYKKMKEGENNKPREKSEGNKRKSSFSNSADDIKSKKKREQSND** **IARGFERGLEPEKIIGATDSCGDLMFLMKWKDTDEADLVLAKEANVKCP** **QIVIAFYEERLTWHAYPEDAENKEKESAKS**
Native sequence	Amino acids M1 – S191 (end) of mouse HP1 alpha. Residue M232 of the fusion protein is equivalent to M1 of the native enzyme. The GST tag is located at residues 1 – 220.
Protease cleavage	PreScission (<u>LEVLFQGP</u>) residues 221 - 228
Cloning sites	*Bam*H1 and *Not*1 sites of pGEX6P-1

Nucleotide Sequence of insert	ggatccATGGGAAAGAAGACCAAGAGGACAGCCGACAGCTCTTCTTCAG AGGATGAGGAGGAATATGTGGTGGAAAAGGTGTTGGACAGGCGCATGGT TAAGGGGCAAGTGGAATATCTGTTGAAGTGGAAAGGCTTTTCTGAGGAG CACAATACTTGGGAACCTGAGAAGAACTTGGATTGTCCTGAACTAATTT CTGAGTTTATGAAAAAGTATAAGAAGATGAAGGAGGGTGAAAACAATAA GCCCAGGGAGAAATCAGAAGGAAACAAGAGGAAATCCAGTTTCTCCAAC AGCGCTGATGATATTAAATCTAAAAAAAAGAGAGAGCAAAGCAATGATA TCGCTCGGGGCTTTGAGAGAGGACTGGAACCAGAAAAGATCATCGGAGC AACAGATTCCTGCGGTGACTTAATGTTCTTAATGAAATGGAAAGACACA GATGAAGCTGACCTGGTTCTTGCAAAAGAAGCTAACGTGAAGTGTCCAC AGATTGTGATAGCATTTTATGAAGAGAGACTGACGTGGCACGCATATCC AGAGGATGCGGAAAACAAAGAAAAAGAAAGCGCGAAGAGCtaagcggcc gc