Division of Signal Tranduction Therapy

Standard Operating Procedure

Preparation of hnRNP H [2 - 449]

Enzyme description:- hnRNP H [2 – 449]

Clone number:- DU 1823

Source:- Recombinant

Expression system:- *E.coli*

Tag:- N-terminal GST

Purification method:- GSH Sepharose

Calculated molecular mass:-
Monoisotopic 75, 874.00 daltons
Average Mass 75, 922.44 daltons
[cysteines reduced, methionines have not been oxidised

Theoretical pI:- 5.83

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

hnRNP H [2 – 449]

Protein hnRNP H [2 – 449]

Clone number DU 1823

Species Human

Accession number AAA91346.1

Tags N-terminal GST

Bacterially expressed protein

MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELG
LEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLEGA
VLDIRYGVSRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDH
VTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPQIDKYLKSS
KYIAWPLQGWQATFGGGDHPPKSDLEVLFQGPLGS**MLGTEGGEGFVVKV**
RGLPWSCSADEVQRFFSDCKIQNGAQGIRFIYTREGRPSGEAFVELESE
DEVKLALKKDRETMGHRYVEVFKSNNVEMDWVLKHTGPNSPDTANDGFV
RLRGLPFGCSKEEIVQFFSGLEIVPNGITLPVDFQGRSTGEAFVQFASQ
EIAEKALKKHKERIGHRYIEIFKSSRAEVRTHYDPPRKLMAMQRPGPYD
RPGAGRGYNSIGRGAGFERMRRGAYGGGYGGYDDYNGYNDGYGFGSDRF
GRDLNYCFSGMSDHRYGDGGSTFQSTTGHCVHMRGLPYRATENDIYNFF
SPLNPVRVHIEIGPDGRVTGEADVEFATHEDAVAAMSKDKANMQHRYVE
LFLNSTAGASGGAYEHRYVELFLNSTAGASGGAYGSQMMGGMGLSNQSS
YGGPASQQLSGGYGGGYGGQSSMSGYDQVLQENSSDFQSNIA

Native sequence Amino acids M2 – A449 (end) of human hnRNP H.
Residue M232 of the fusion protein is equivalent to M2 of the native enzyme. The GST tag is located at residues 1 – 220.

Protease cleavage PreScission (LEVLFQGP) residues 221 - 228

Cloning sites *Bam*H1 sites of pGEX6P-1

Nucleotide Sequence Of Insert

ggatccATGTTGGGCACGGAAGGTGGAGAGGGATTCGTGGTGAAGGTCC
GGGGCTTGCCCTGGTCTTGCTCGGCCGATGAAGTGCAGAGGTTTTTTTC
TGACTGCAAAATTCAAAATGGGGCTCAAGGTATTCGTTTCATCTACACC
AGAGAAGGCAGACCAAGTGGCGAGGCTTTTGTTGAACTTGAATCAGAAG
ATGAAGTCAAATTGGCCCTGAAAAAAGACAGAGAAACTATGGGACACAG
ATATGTTGAAGTATTCAAGTCAAACAACGTTGAAATGGATTGGGTGTTG
AAGCATACTGGTCCAAATAGTCCTGACACGGCCAATGATGGCTTTGTAC
GGCTTAGAGGACTTCCCTTTGGATGTAGCAAGGAAGAAATTGTTCAGTT
CTTCTCAGGGTTGGAAATCGTGCCAAATGGGATAACATTGCCGGTGGAC
TTCCAGGGGAGGAGTACGGGGGAGGCCTTCGTGCAGTTTGCTTCACAGG
AAATAGCTGAAAAGGCTCTAAAGAAACACAAGGAAAGAATAGGGCACAG
GTATATTGAAATCTTTAAGAGCAGTAGAGCTGAAGTTAGAACTCATTAT
GATCCACCACGAAAGCTTATGGCCATGCAGCGGCCAGGTCCTTATGACA
GACCTGGGGCTGGTAGAGGGTATAACAGCATTGGCAGAGGAGCTGGCTT
TGAGAGGATGAGGCGTGGTGCTTATGGTGGAGGCTATGGAGGCTATGAT
GATTACAATGGCTATAATGATGGCTATGGATTTGGGTCAGATAGATTTG
GAAGAGACCTCAATTACTGTTTTTCAGGAATGTCTGATCACAGATACGG
GGATGGTGGCTCTACTTTCCAGAGCACAACAGGACACTGTGTACACATG
CGGGGATTACCTTACAGAGCTACTGAGAATGACATTTATAATTTTTTTT
CACCGCTCAACCCTGTGAGAGTACACATTGAAATTGGTCCTGATGGCAG
AGTAACTGGTGAAGCAGATGTCGAGTTCGCAACTCATGAAGATGCTGTG
GCAGCTATGTCAAAAGACAAAGCAAATATGCAACACAGATATGTAGAAC
TCTTCTTGAATTCTACAGCAGGAGCAAGCGGTGGTGCTTACGAACACAG
ATATGTAGAACTCTTCTTGAATTCTACAGCAGGAGCAAGCGGTGGTGCT
TATGGTAGCCAAATGATGGGAGGCATGGGCTTGTCAAACCAGTCCAGCT
ACGGGGGCCCAGCCAGCCAGCAGCTGAGTGGGGGTTACGGAGGCGGCTA
CGGTGGCCAGAGCAGCATGAGTGGATACGACCAAGTTTTACAGGAAAAC
TCCAGTGATTTTCAATCAAACATTGCAtagggatcc

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