

Division of Signal Transduction Therapy

Standard Operating Procedure

Preparation of hnRNP F [2 - 415]

Enzyme description:- hnRNP F [2 – 415]

Clone number:- DU 1895

Source:- Recombinant

Expression system:- *E.coli*

Tag:- N-terminal GST

Purification method:- GSH Sepharose

Calculated molecular mass:-

Monoisotopic 72, 318.45 daltons

Average Mass 72, 364.83 daltons

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 5.49

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 F [2 – 415]

<u>Protein</u>	hnRNP F [2 – 415]
<u>Clone number</u>	DU 1895
<u>Species</u>	Human
<u>Accession number</u>	P52597.3
<u>Tags</u>	N-terminal GST
<u>Bacterially expressed protein</u>	<p>MSPILGYWKIKGLVQPTRLLEYLEEKYEHLIERDEGDKWRNKKFELG LEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAIEISMLEGA VLDIRYGVSR IAYS KDFETLKVDFLSKLP EMLKMFEDRLCHKTYLNGDH VTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPOIDKYLKSS KYIAWPLQGWQATFGGGDHPPKSDLEVL FQGPLGSMLGPEGGEGFVVKL RGLPWSCSVEDVQNFLSDCTIHDGAAGVHF IY TREGRQSGEAFVELGSE DDVKMALKKDRESMGHRYIEVFKSHRTEM DWVLKHS GPNSADSANDGFV RLRGLPFGCTKEEIVQFFSGLEIVPNGITLPVDPEGKITGEAFVQFASQ ELA EKALGKHKERIGHRYIEVFKSSQEEVRSYSDPPLKFMSVQRPGPYD RPGTARRYIGIVKQAGLERMRPGAYSTGYGGYEEYSGLSDGYGFTTDLF GRDLSYCLSGMYDHRYGDSEFTVQSTTGHCVHMRGLPYKATENDIYNFF SPLNPVRVHIEIGPDGRVTGEADVEFATHEEAVAAMSKDRANMQHRYIE LFLNSTTGASNGAYSSQVMQGMGVSAQAQATYSGLESQSVSGCYGAGYSG QNSMGGYD</p>
<u>Native sequence</u>	<p>Amino acids M2 – D415 (end) of human hnRNP F. Residue M232 of the fusion protein is equivalent to M2 of the native enzyme. The GST tag is located at residues 1 – 220.</p>
<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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Nucleotide
Sequence Of
Insert

ggatccATGCTGGGCCCTGAGGGAGGTGAAGGCTTTGTGGTCAAGCTCC
GTGGCCTGCCCTGGTCCTGCTCTGTTGAGGACGTGCAGAACTTCCTCTC
TGACTGCACGATTCATGATGGGGCCGCAGGTGTCCATTTTCATCTACACT
AGAGAGGGCAGGCAGAGTGGTGAGGCTTTTGTGAACTTGGATCAGAAG
ATGATGTAAAAATGGCCCTGAAAAAAGACAGGGAAAGCATGGGACACCG
GTACATTGAGGTGTTCAAGTCCCACAGAACCGAGATGGATTGGGTGTTG
AAGCACAGTGGTCCCAACAGTGCCGACAGCGCCAACGATGGCTTCGTGC
GGCTTCGAGGACTCCCATTTGGATGCACAAAGGAAGAAATTGTTTCAGTT
CTTCTCAGGGTTGAAATTTGTGCCAAACGGGATCACATTGCCTGTGGAC
CCCGAAGGCAAGATTACAGGGGAAGCGTTCGTGCAGTTTGCCTCGCAGG
AGTTAGCTGAGAAGGCTCTAGGGAAACACAAGGAGAGGATAGGGCACAG
GTACATTGAGGTGTTAAGAGCAGCCAGGAGGAAGTTAGGTCATACTCA
GATCCCCCTCTGAAGTTCATGTCCGTGCAGCGGCCAGGGCCCTATGACC
GGCCCGGACTGCCAGGAGGTACATTGGCATCGTGAAGCAGGCAGGCCT
GGAAAGGATGAGGCCTGGTGCCTACAGCACAGGCTACGGGGGCTACGAG
GAGTACAGTGGCCTCAGTGATGGCTACGGCTTACCACCGACCTGTTCG
GGAGAGACCTCAGTACTGTCTCTCCGGGATGTATGACCACAGATACGG
CGACAGTGAGTTCACAGTGCAGAGCACCACAGGCCACTGTGTCCACATG
AGGGGCCTGCCGTACAAAGCGACCGAGAACGACATTTACAACCTTCTTCT
CTCCTCTCAACCTGTGAGAGTCCATATTGAGATTGGCCCAGATGGAAG
AGTGACGGGTGAAGCAGATGTTGAGTTTGCTACTCATGAAGAAGCTGTG
GCAGCTATGTCCAAAGACAGGGCCAATATGCAGCACAGATATATAGAAC
TCTTCTTGAATTCAACAACAGGGGCCAGCAATGGGGCGTATAGCAGCCA
GGTGATGCAAGGCATGGGGGTGTCTGCTGCCAGGCCACTTACAGTGGC
CTGGAGAGCCAGTCAGTGAGTGGCTGTTACGGGGCCGGCTACAGTGGGC
AGAACAGCATGGGTGGCTATGACTaggcggccgc

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