

*Division of Signal Transduction Therapy*

**Standard Operating Procedure**

**Preparation of hnRNP K isoform b [2 - 463]**

**Enzyme description:-** hnRNP K isoform b [2 – 463]

**Clone number:-** DU 4040

**Source:-** Recombinant

**Expression system:-** *E.coli*

**Tag:-** N-terminal GST

**Purification method:-** GSH Sepharose

**Expression level:-** 7 mg/L

**Calculated molecular mass:-**

Monoisotopic        77, 993.16 daltons  
Average Mass        78, 042.63 daltons  
[cysteines reduced, methionines have not been oxidised]

**Theoretical pI:-** 5.47

**Purity:-** 90 %

**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, 0.2 mM PMSF, 1 mM Benzamidine.

**Storage temperature:-** -70 °C

**Assay:-** Substrate for ERK1 or ERK2

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**Clone Data Sheet**

**hnRNP K isoform b [2 - 463]**

<b><u>Protein</u></b>	hnRNP K isoform b [2 - 463]
<b><u>Clone Number</u></b>	DU 4040
<b><u>Species</u></b>	Human
<b><u>Accession number</u></b>	AAH14980
<b><u>Tags</u></b>	N-terminal GST
<b><u>Bacterially expressed protein</u></b>	<p>MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELG LEFPNLPYYIDGDVKLQTQSMAIIRYIADKHNMLGGCPKERAIEISMLEGA VLDIRYGVSRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDH VTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPOIDKYLKSS KYIAWPLQGWQATFGGGDHPPKSDLEVLFGGPLGSPEF<b>ETEQPEETFPN</b> <b>TETNGEFGKRPAEDMEEEQAFKRSRNTDEMVELRILLOSKNAGAVIGKG</b> <b>GKNIKALRTDYNASVSVDPSSGPERILSISADIETIGEILKKIIP TLEE</b> <b>GLQLPSPTATSQPLPLESDAVECLNYQHYKGSDFDCELRLLIHQSLAGGI</b> <b>IGVKGAKIKELRENTQTTIKL FQEC PHSTDRVVLIGGKPDRVVECIKI</b> <b>ILDLISESPIKGRAQPYDPNFYDETYDYG GFTMMFDDRGRPVGFPMRG</b> <b>RGGFDRMPPGRGGRPMPPSRRDYDDMSPRGPPPPPPGRGGRGGSRARN</b> <b>LPLPPPPPPRGGDLMAYDRRGRPGDRYDGMVGFSADETWDSAIDTWSPS</b> <b>EWQMAYEPQGGSGYDYSYAGGRGSYGDLGGPIIT TQVTIPKDLAGSIIG</b> <b>KGGQRIKQIRHESGASIKIDEPLEGSEDRIITITGTQDQIQNAQYLLQN</b> <b>SVKQYSGKFF</b></p>
<b><u>Native sequence</u></b>	<p>Amino acids E2 – F463 (end) of human hnRNP K isoform b. Residue E235 of fusion protein is equivalent to E2 of the native enzyme. The GST tag is located at residues 1 – 220.</p>
<b><u>Protease cleavage</u></b>	PreScission ( <u>LEVLFQGPL</u> ) residues 221 - 229
<b><u>Cloning sites</u></b>	<i>Eco</i> R1 and <i>Not</i> I site of pGex6P-1

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Nucleotide  
sequence

ATGTCCCCTATACTAGGTTATTGGAAAATTAAGGGCCTTGTGCAACCCA  
CTCGACTTCTTTTGGAAATATCTTGAAGAAAAATATGAAGAGCATTTGTA  
TGAGCGCGATGAAGGTGATAAATGGCGAAACAAAAAGTTTGAATTGGGT  
TTGGAGTTTCCCAATCTTCCTTATTATATTGATGGTGATGTTAAATTAA  
CACAGTCTATGGCCATCATACGTTATATAGCTGACAAGCACAACATGTT  
GGGTGGTTGTCCAAAAGAGCGTGCAGAGATTTCAATGCTTGAAGGAGCG  
GTTTTGGATATTAGATACGGTGTTCGAGAATTGCATATAGTAAAGACT  
TTGAAACTCTCAAAGTTGATTTTCTTAGCAAGCTACCTGAAATGCTGAA  
AATGTTTGAAGATCGTTTATGTCATAAAACATATTTAAATGGTGATCAT  
GTAACCCATCCTGACTTCATGTTGTATGACGCTCTTGATGTTGTTTTAT  
ACATGGACCCAATGTGCCTGGATGCGTTCCAAAATTAGTTTGTTTTTAA  
AAAACGTATTGAAGCTATCCACAAATTGATAAGTACTTGAAATCCAGC  
AAGTATATAGCATGGCCTTTGCAGGGCTGGCAAGCCACGTTTGGTGGTG  
GCGACCATCCTCCAAAATCGGATCTGGAAGTTCTGTTCCAGGGGCCCT  
GGGATCCCCGGAATTC**GAAACTGAACAGCCAGAAGAAACCTTCCCTAAC**  
**ACTGAAACCAATGGTGAATTTGGTAAACGCCCTGCAGAAGATATGGAAG**  
**AGGAACAAGCATTTAAAAGATCTAGAAACACTGATGAGATGGTTGAATT**  
**ACGCATTCTGCTTCAGAGCAAGAATGCTGGGGCAGTGATTGGAAAAGGA**  
**GGCAAGAATATTAAGGCTCTCCGTACAGACTACAATGCCAGTGTTCAG**  
**TCCAGACAGCAGTGGCCCCGAGCGCATATTGAGTATCAGTGCTGATAT**  
**TGAAACAATTGGAGAAATTCTGAAGAAAATCATCCCTACCTTGAAGAG**  
**GGCCTGCAGTTGCCATCACCCACTGCAACCAGCCAGCTCCCGCTCGAAT**  
**CTGATGCTGTGGAATGCTTAAATTACCAACACTATAAAGGAAGTGACTT**  
**TGACTGCGAGTTGAGGCTGTTGATTCATCAGAGTCTAGCAGGAGGAATT**  
**ATTGGGGTCAAAGGTGCTAAAATCAAAGAACTTCGAGAGAACACTCAA**  
**CCACCATCAAGCTTTTCCAGGAATGCTGTCCCTCATTCCACTGACAGAGT**  
**TGTTCTTATTGGAGGAAAACCCGATAGGGTTGTAGAGTGCATAAAGATC**  
**ATCCTTGATCTTATATCTGAGTCTCCCATCAAAGGACGTGCACAGCCTT**  
**ATGATCCCAATTTTTACGATGAAACCTATGATTATGGTGGTTTTACAAT**  
**GATGTTTGATGACCGTTCGCGGACGCCAGTGGGATTTCCCATGCGGGGA**  
**AGAGGTGGTTTTTGACAGAATGCCTCCTGGTTCGGGGTGGGCGTCCCATGC**  
**CTCCATCTAGAAGAGATTATGATGATATGAGCCCTCGTCGAGGACCACC**  
**TCCCCCTCTCCCGGACGAGGCGGCCGGGGTGGTAGCAGAGCTCGGAAT**  
**CTTCTCTTCCCTCCACCACCACCCTAGAGGGGGAGACCTCATGGCCT**  
**ATGACAGAAGAGGGAGACCTGGAGACCGTTACGACGGCATGGTTGGTTT**  
**CAGTGCTGATGAACTTGGGACTCTGCAATAGATACATGGAGCCCATCA**  
**GAATGGCAGATGGCTTATGAACCACAGGGTGGCTCCGGATATGATTATT**  
**CCTATGCAGGGGGTTCGTGGCTCATATGGTGATCTTGGTGGACCTATTAT**  
**TACTACACAAGTAACTATTCCCAAAGATTTGGCTGGATCTATTATTGGC**  
**AAAGGTGGTCAAGGATTAACAAATCCGTCATGAGTCGGGAGCTTCGA**  
**TCAAATGATGAGCCTTTAGAAGGATCCGAAGATCGGATCATTACCAT**  
**TACAGGAACACAGGACCAGATACAGAATGCACAGTATTTGCTGCAGAAC**  
**AGTGTGAAGCAGTATTCTGGAAGTTTTTCT**taagcggccgc