

Division of Signal Transduction Therapy

Standard Operating Procedure

Preparation of C15orf41 [1 – 281]

Enzyme description:- C15orf41 [1 - 281]

Clone number:- DU 49143

Source:- Recombinant

Expression system:- *E.coli*

Tag:- N-terminal GST

Purification method:- GSH Sepharose

Calculated molecular mass:-

Monoisotopic 59, 049.99 daltons

Average Mass 59, 088.11 daltons

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 6.00

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

C15orf41 [1 - 281]

<u>Protein</u>	C15orf41 [1 -281]
<u>Clone number</u>	DU 49143
<u>Species</u>	Human
<u>Accession number</u>	NM_001130010.2
<u>Tags</u>	N-terminal GST
<u>Bacterially expressed protein</u>	MSPILGYWKIKGLVQPTRLLEYLEEKYEEHLYERDEGDKWRNKKFELG LEFPNLPYYIDGDVKLTQSMATIRYIADKHNMLGGCPKERAETSMLEGA VLDIRYGVSRAYSCKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDH VTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPOIDKYLKSS KYIAWPLQGWQATFGGGDHPPKSDLEVLFGQGPLGSMILTKAQYDEIAQC LVSVPPTRQSLRKLKQRFPSQSQATLLSIFSQEQKHAKRTHAKHHTSE AIESYYQRYLNGVVKNGAAPVLLDLANEVDYAPSLMARLILERFLQEHE ETPPSKSIINSMLRDPSPQIPDGVLANQVYQCIVNDCCYGPLVDCIKHAI GHEHEVLLRDLLLEKNLSFLDEDQLRAKGYDKTPDFILQVPVAVEGHI HWIESKASFGDECSSHAYLHDQFWSYWNRFGPGLVIYWGFIQELDCNR ERGILLKACFPTNIVTLCHSIA
<u>Native sequence</u>	Amino acids M1 – A281 (end) of human C15orf41. 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> I sites of pGEX6P-1

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Nucleotide

Sequence of insert

ggatccATGATACTGACCAAAGCTCAGTACGACGAGATAGCCCAGTGCC
TAGTGTCTGTGCCGCCTACCAGGCAGAGCCTGAGGAAGCTGAAGCAGAG
GTTTCCCAGTCAATCGCAGGCCACTCTGCTGAGCATCTTCTCCCAGGAG
TACCAGAAACACATTAAAAGAACACATGCCAAACATCATACTTCGGAAG
CAATTGAAAGTTATTACCAGAGGTACCTGAATGGAGTGGTGAAAAATGG
AGCTGCCCCAGTGCTCCTGGACCTGGCCAATGAGGTGGACTATGCGCCC
TCATTAATGGCTCGGCTTATACTGGAGAGGTTTCTACAGGAACACGAGG
AAACTCCACCCTCCAAGTCTATTATAAATAGTATGCTACGGGACCCTTC
TCAGATTCCAGATGGAGTTCTAGCAAATCAGGTCTATCAGTGCATTGTG
AACGACTGCTGTTACGGACCACTAGTGGACTGCATCAAGCATGCCATTG
GTCATGAGCATGAGGTCCTGCTGAGAGACTTGCTTCTAGAGAAAAACCT
GTCCTTCCTAGATGAAGATCAGCTTCGTGCAAAGGGTTATGACAAAACA
CCAGACTTCATTTTACAAGTACCAGTTGCTGTAGAAGGGCACATAATTC
ACTGGATTGAAAGCAAAGCCTCATTTGGTGATGAATGTAGCCACCACGC
CTACCTGCATGACCAGTTCTGGAGCTACTGGAATAGATTTGGGCCAGGC
TTAGTCATCTATTGGTATGGATTTATCCAGGAGCTGGACTGCAACCGGG
AAAGGGGCATCCTGCTCAAAGCCTGTTTCCCCACGAACATTGTCACCTT
ATGCCACAGCATAGCTtgagcggccgc