

Division of Signal Transduction Therapy

Standard Operating Procedure

Preparation of C15orf41 Y94C [1 - 281]

Enzyme description:- C15orf41 Y94C [1 – 281]

Clone number:- DU 24443

Source:- Recombinant

Expression system:- *E.coli*

Tag:- N-terminal MBP

Purification method:- Amylose Resin

Calculated molecular mass:-

Monoisotopic 76, 525.71 daltons

Average Mass 76, 574.01 daltons

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 5.35

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, 0.2 mM PMSF, 1 mM Benzamidine.

Storage temperature:- -70

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Clone Data Sheet

C15orf41 Y94C [1 – 281]

<u>Protein</u>	C15orf41 Y94C [1 – 281]
<u>Clone number</u>	DU 24443
<u>Species</u>	Human
<u>Accession number</u>	NM_001130010.2
<u>Tags</u>	N-terminal MBP
<u>Bacterially expressed protein</u>	<p>MMKIEEGKLVIIWINGDKGYNGLAEVGGKFEKDTGIKVTVEHPDKLEEK FPQVAATGDGPDIIFWAHDRFGGYAQSGLLAEITPDKAFQDKLYPFTW DAVRYNGKLIAYPIAVEALSLIYNKDLLPNPPKTWEEIPALDKELKAK GKSALMFNLQEPYFTWPLIAADGGYAFKYENGYDIKDVGVNDNAGAKA GLTFLVDLIIKNKHMNADTDYSIAEAAFNKGETAMTINGPWAWSNIDTS KVNYGVTVLPFTFKGQPSKPFVGVLSAGINAASPNKELAKEFLENYLLT DEGLEAVNKDKPLGAVALKSYEEELVKDPRIAATMENAQKGEIMPNIIP QMSAFWYAVRTAVINAASGRQTVDEALKDAQTNSSSNNNNNNNNNNLG DDDDKVPEFLEVLFGQPLGSMILTKAQYDEIAQCLVSVPPTRQSLRKL KQRFPSQSQATLLSIFSQEYQKHIKRTHAKHHTSEAIESYYQRYLNGV VKNGAAPVLLDLANEVDCAPSLMARLILERFLQEHEETPPSKSIINSM LRDPSQIPDGVLANQVYQCI VNDCCYGPLVDCIKHAIGHEHEVLLRDL LLEKNLSFLDEDQLRAKGYDKTPDFILQVPVAVEGHIHWIESKASFG DEC SHHAYLHDQFWSYWNRF GPGLVIIYWYGF IQELDCNRERGI LLKAC FPTNIVTLCHSIA</p>
<u>Native sequence</u>	<p>Amino acids M1 – A281 (end) of human C15orf41. Residue M405 of the fusion protein is equivalent to M1 of the native enzyme. The GST tag is located at residues 1 – 393</p> <p>The protein has an Y94C mutation. Residue Y94 is equivalent to C498 of the fusion protein.</p>
<u>Protease cleavage</u>	PreScission (<u>LEVLFQGP</u>) residues 394 - 401
<u>Cloning sites</u>	<i>Bam</i> H1 and <i>Not</i> 1 site of pMEX6P-1

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**Nucleotide
sequence of
insert**

ggatccATGATACTGACCAAAGCTCAGTACGACGAGATAGCCCAGTGC
CTAGTGTCTGTGCCGCTACCAGGCAGAGCCTGAGGAAGCTGAAGCAG
AGGTTTCCCAGTCAATCGCAGGCCACTCTGCTGAGCATCTTCTCCCAG
GAGTACCAGAAACACATTTAAAGAACACATGCCAAACATCATACTTCG
GAAGCAATTGAAAGTTATTACCAGAGGTACCTGAATGGAGTGGTGAAA
AATGGAGCTGCCCCAGTGCTCCTGGACCTGGCCAATGAGGTGGACTgT
GCGCCCTCATTAATGGCTCGGCTTATACTGGAGAGGTTTCTACAGGAA
CACGAGGAAACTCCACCCTCCAAGTCTATTATAAATAGTATGCTACGG
GACCCTTCTCAGATTCCAGATGGAGTTCAGCAAATCAGGTCTATCAG
TGCATTGTGAACGACTGCTGTTACGGACCACTAGTGGACTGCATCAAG
CATGCCATTGGTCATGAGCATGAGGTCTGCTGAGAGACTTGCTTCTA
GAGAAAACCTGTCCTTCCTAGATGAAGATCAGCTTCGTGCAAAGGGT
TATGACAAAACACCAGACTTCATTTTACAAGTACCAGTTGCTGTAGAA
GGGCACATAATTCCTGGATTGAAAGCAAAGCCTCATTGGGTGATGAA
TGTAGCCACCACGCCTACCTGCATGACCAGTTCCTGGAGCTACTGGAAT
AGATTTGGCCAGGCTTAGTCATCTATTGGTATGGATTTATCCAGGAG
CTGGACTGCAACCGGGAAAGGGGCATCCTGCTCAAAGCCTGTTTCCCC
ACGAACATTGTCACCTTATGCCACAGCATAGCTtgagcggccgc