Division of Signal Tranduction Therapy

Standard Operating Procedure

Preparation of ABIN1 R480Q [1 - 636]

Enzyme description:- ABIN1 R480Q [1 – 636]

Clone number:- DU 8878

Source:- Recombinant

Expression system:- *E.coli*

Tag:- N-terminal GST

Purification method:- GSH Sepharose

Calculated molecular mass:-
Monoisotopic 98, 624.19 daltons
Average Mass 98, 686.38 daltons
[cysteines reduced, methionines have not been oxidised

Theoretical pI:- 6.01

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

ABIN1 R480Q [1 – 636]

Protein ABIN1 R480Q [1 – 636]

Clone number DU 8878

Species Human

Accession number NM_006058.4

Tags N-terminal GST

Bacterially expressed protein

MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELG
LEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLEGA
VLDIRYGVSRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDH
VTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPQIDKYLKSS
KYIAWPLQGWQATFGGGDHPPKSDLEVLFQGPLGS**MEGRGPYRIYDPG**
GSVPSGEASAAFERLVKENSRLKEKMQGIKMLGELLEESQMEATRLRQK
AEELVKDNELLPPPSPSLGSFDPLAELTGKDSNVTASPTAPACPSDKPA
PVQKPPSSGTSSEFEVVTPEEQNSPESSSHANAMALGPLPREDGNLMLH
LQRLETTLSVCAEEPDHGQLFTHLGRMALEFNRLASKVHKNEQRTSILQ
TLCEQLRKENEALKAKLDKGLEQRDQAAERLREENLELKKLLMSNGNKE
GASGRPGSPKMEGTGKKAVAGQQQASVTAGKVPEVVALGAPEKKVKMLE
QQRSELLEVNKQWDQHFRSMKQQYEQKITELRQKLADLQKQVTDLEAER
EQKQRDFDRKLLLAKSKIEMEETDKEQLTAEAKELRQKVKYLQDQLSPL
TRQREYQEKEIQRLNKALEEALSIQTPPSSPPTAFGSPEGAGALLRKQE
LVTQNELLKQQVKIFEEDFQRERSDQERMNEEKEELKKQVEKLQAQVTL
SNAQLKAFKDEEKAREALRQQKRKAKASGERYHVEPHPEHLCGAYPYAY
PPMPAMVPHHGFEDWSQIRYPPPPMAMEHPPPLPNSRLFHLPEYTWRLP
CGGVRNPNQSSQVMDPPTARPTEPESPKNDREGPQ

Native sequence Amino acids M1 – Q636 (end) of human ABIN1.
Residue M232 of the fusion protein is equivalent to M1 of the native enzyme. The GST tag is located at residues 1 – 220. The protein has a R480Q mutation. Residue R480 is equivalent to residue Q717 of the fusion protein.

Protease cleavage PreScission (LEVLFQGP) residues 221 - 228

Cloning sites *Bam*H1 and *Not*1 sites of pGEX6P-1

Nucleotide Sequence Of Insert

ggatccATGGAAGGGAGAGGACCGTACCGGATCTACGACCCTGGGGGCA
GCGTGCCCTCAGGAGAGGCATCCGCAGCTTTTGAGCGCCTAGTGAAGGA
GAATTCCCGGCTGAAGGAAAAAATGCAAGGGATAAAGATGTTAGGGGAG
CTTTTGGAAGAGTCCCAGATGGAAGCGACCAGGCTCCGGCAGAAGGCAG
AGGAGCTAGTGAAGGACAACGAGCTGCTCCCACCACCTTCTCCCTCCTT
GGGCTCCTTCGACCCCCTGGCTGAGCTCACAGGAAAGGACTCAAATGTC
ACAGCATCTCCCACAGCCCCTGCATGCCCCAGTGACAAGCCAGCACCAG
TCCAGAAGCCTCCATCCAGTGGCACCTCCTCTGAATTTGAAGTGGTCAC
TCCTGAGGAGCAGAATTCACCAGAGAGCAGCAGCCATGCCAATGCGATG
GCGCTGGGCCCCCTGCCCCGTGAGGACGGCAACCTGATGCTGCACCTGC
AGCGCCTGGAGACCACGCTGAGTGTGTGTGCCGAGGAGCCGGACCACGG
CCAGCTCTTCACCCACCTGGGCCGCATGGCCCTGGAGTTCAACCGACTG
GCATCCAAGGTGCACAAGAATGAGCAGCGCACCTCCATTCTGCAGACCC
TGTGTGAGCAGCTTCGGAAGGAGAACGAGGCTCTGAAGGCCAAGTTGGA
TAAGGGCCTGGAACAGCGGGATCAGGCTGCCGAGAGGCTGCGGGAGGAA
AATTTGGAGCTCAAGAAGTTGTTGATGAGCAATGGCAACAAAGAGGGTG
CGTCTGGGCGGCCAGGCTCACCGAAGATGGAAGGGACAGGCAAGAAGGC
AGTGGCTGGACAGCAGCAGGCTAGTGTGACGGCAGGTAAGGTCCCAGAG
GTGGTGGCCTTGGGCGCACCCGAGAAGAAGGTGAAGATGCTGGAGCAGC
AGCGCAGTGAGCTGCTGGAAGTGAACAAGCAGTGGGACCAGCATTTCCG
GTCCATGAAGCAGCAGTATGAGCAGAAGATCACTGAGCTGCGTCAGAAG
CTGGCTGATTTGCAGAAGCAGGTGACTGACCTGGAGGCCGAGCGGGAGC
AGAAGCAGCGTGACTTTGACCGCAAGCTCCTCCTGGCCAAGTCCAAGAT
TGAAATGGAGGAGACCGACAAGGAGCAGCTGACAGCAGAGGCCAAGGAG
CTGCGCCAAAAGGTCAAGTACCTGCAGGATCAGCTGAGCCCACTCACCC
GACAGCGTGAGTACCAGGAAAAGGAGATCCAGCGGCTCAACAAGGCCCT
GGAGGAAGCACTGAGCATCCAAACCCCGCCATCATCTCCACCAACAGCA
TTTGGGAGCCCAGAAGGAGCAGGGGCCCTCCTAAGGAAACAGGAGCTGG
TCACGCAGAATGAGTTGCTGAAACAGCAGGTGAAGATCTTCGAGGAGGA
CTTCCAGAGGGAGCGCAGTGATCAAGAGCGCATGAATGAGGAGAAGGAA
GAGCTGAAGAAGCAAGTGGAGAAGCTGCAGGCCCAGGTCACCCTGTCAA
ATGCCCAGCTAAAAGCATTCAAAGATGAGGAGAAGGCAAGAGAAGCCCT
CAGACAGCAGAAGAGGAAAGCAAAGGCCTCAGGAGAGCGTTACCATGTG
GAGCCCCACCCAGAACATCTCTGCGGGGCCTACCCCTACGCCTACCCGC
CCATGCCAGCCATGGTGCCACACCATGGCTTCGAGGACTGGTCCCAGAT
CCGCTACCCCCCTCCCCCCATGGCCATGGAGCACCCGCCCCCACTCCCC
AACTCGCGCCTCTTCCATCTGCCGGAATACACCTGGCGTCTACCTTGTG
GAGGGGTTCGAAATCCAAATCAGAGCTCCCAAGTGATGGACCCTCCCAC
AGCCAGGCCTACAGAACCAGAGTCTCCAAAAAATGACCGTGAGGGGCCT
CAGtgagcggccgc