Division of Signal Tranduction Therapy

Standard Operating Procedure

Preparation of ABIN3 L228P [1 - 325]

Enzyme description:- ABIN3 L228P [1 – 325]

Clone number:- DU 8799

Source:- Recombinant

Expression system:- *E.coli*

Tag:- N-terminal GST

Purification method:- GSH Sepharose

Calculated molecular mass:-
Monoisotopic 66, 627.63 daltons
Average Mass 66, 670.20 daltons
[cysteines reduced, methionines have not been oxidised

Theoretical pI:- 6.63

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

ABIN3 L228P [1 – 325]

Protein ABIN3 L228P [1 – 325]

Clone number DU 8799

Species Human

Accession number NM_024873.5

Tags N-terminal GST

Bacterially expressed protein

MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELG
LEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLEGA
VLDIRYGVSRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDH
VTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPQIDKYLKSS
KYIAWPLQGWQATFGGGDHPPKSDLEVLFQGPLGSPGIPGSTRAAA**MAH**
FVQGTSRMIAAESSTEHKECAEPSTRKNLMNSLEQKIRCLEKQRKELLE
VNQQWDQQFRSMKELYERKVAELKTKLDAAERFLSTREKDPHQRQRKDD
RQREDDRQRDLTRDRLQREEKEKERLNEELHELKEENKLLKGKNTLANK
EKEHYECEIKRLNKALQDALNIKCSFSEDCLRKSRVEFCHEEMRTEMEV
LKQQVQIYEEDFKKERSDRERLNQEKEEPQQINETSQSQLNRLNSQIKA
CQMEKEKLEKQLKQMYCPPCNCGLVFHLQDPWVPTGPGAVQKQREHPPD
CQWYALDQLPPDVQHKANGLSSVKKVHP

Native sequence Amino acids M1 – P325 (end) of human ABIN3.
Residue M243 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 L228P mutation. Residue L228 is equivalent to residue P470 of the fusion protein.

Protease cleavage PreScission (LEVLFQGP) residues 221 - 228

Cloning sites *Not*1 sites of pGEX6P-2

Nucleotide Sequence Of Insert

gcggccgcgATGGCACATTTTGTACAGGGCACATCTAGAATGATTGCCG
CAGAAAGTTCTACGGAGCATAAAGAGTGTGCTGAACCATCAACAAGAAA
GAACTTGATGAATTCTCTTGAACAAAAGATAAGGTGTTTGGAAAAACAA
AGAAAAGAGCTCCTGGAAGTTAACCAGCAATGGGATCAGCAATTTAGAA
GTATGAAAGAGTTATATGAAAGAAAGGTAGCAGAGCTGAAGACGAAACT
GGACGCCGCGGAAAGATTCCTCAGCACGCGGGAGAAGGATCCGCATCAG
AGGCAGAGAAAGGACGACAGGCAGAGAGAGGACGACAGGCAGCGCGACC
TGACCCGGGACCGGCTGCAGCGGGAGGAGAAGGAAAAGGAACGCCTAAA
TGAAGAATTACATGAATTGAAAGAAGAGAATAAACTTTTAAAGGGAAAA
AATACTCTTGCGAACAAGGAAAAGGAACATTACGAATGTGAAATAAAAC
GCCTCAATAAGGCTCTTCAGGATGCCTTGAATATCAAGTGTTCATTTTC
CGAGGACTGTTTGAGGAAGTCTCGAGTGGAATTCTGCCATGAGGAGATG
AGAACAGAAATGGAAGTTCTGAAGCAGCAGGTGCAAATATACGAAGAAG
ACTTCAAAAAGGAACGATCGGATCGAGAGAGACTTAATCAAGAGAAAGA
GGAGCCACAGCAAATTAATGAAACTTCCCAATCCCAGTTGAACAGGCTG
AATTCCCAGATAAAAGCTTGTCAGATGGAGAAAGAAAAACTAGAAAAGC
AATTAAAACAGATGTATTGCCCACCCTGTAACTGCGGCTTGGTTTTCCA
CCTGCAAGATCCATGGGTACCAACAGGCCCTGGAGCTGTGCAGAAGCAA
CGGGAGCACCCACCAGACTGTCAGTGGTATGCTCTTGACCAGCTTCCGC
CAGATGTACAACACAAGGCAAATGGTTTATCCTCAGTAAAGAAAGTCCA
TCCGtaggcggccgc