

Division of Signal Transduction Therapy

Standard Operation Procedure

Dac-USP45 C199A

Enzyme description:- Dac-USP45 C199A

Clone number:- DU22217

Source:- Sf21 Recombinant

Tag:- N-terminal Dac tag

Purification method:- Ampicillin sepharose

Expression level:- 8 mg/L

Calculated molecular mass:-

Monoisotopic 121237 Da

Average Mass 121310 Da

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 8.58

Purity:- 80%

Enzyme storage buffer:-

50 mM HEPES pH 7.5, 10% glycerol, 150mM NaCl, 1mM DTT

Storage temperature:- -80°C

Assay:-

Ub-Rho110-Gly cleavage assay monitored by Ex/Em 485/535 nm

Assay buffer:-

40 mM Tris pH 7.5, 100 mM NaCl, 5 mM DTT, 0.01% Triton X-100, 0.005% Ovalbumin, 0.5 µM Ub-Rho110-Gly

Division of Signal Transduction Therapy

Clone Data Sheet

Dac-USP45 C199A

Protein Dac-USP45 C199A

Synonyms

Clone Number DU22217

Species Human

Accession Number Protein: Q70EL2 DNA: NM_001080401.1

Tags N-terminal Dac tag

Amino acid sequence of expressed protein MSAIPGVPOIDAESYILIDYNSGKVLAEQNADVRRDPASLTKMMTSYVIGQAM
KAGFKETDLVTIGNDAWATGNPVFKGSSLMFLKPGMQVPVSQLIRGINLQSG
NDACVAMADFAAGSQDAFVGLMNSYVNALGLKNTHFQTVHGLDADGOYSSARD
MALIGQALIRDVPNEYSIYKEKEFTFNGIRQLNRNGLLWDNSLNVLDGDKTGH
DKAGYNLVSATGEGMRLISAVMGGRTFKGREAESKLLTWGFRFFETVNPEN
LYFOGGSMRVKDPTKALPEKAKRSKRPTVPHDEDSSDDIAVGLTCQHVSHAIS
VNHVKRAIAENLWSVCSECLKERRFYDQQLVLTSDIWLCLKCGFQCGKNS
QHSLKHFKSSRTEPHCI I INLSTWI IWCYECDEKLSTHCNKKVLAQIVDFLOK
HASKTQTSAFSRIMKLCCEKCETDEIQKGGKCRNLSVRGITNLGNTAFFNAV
QNLAQTYTLTDLMNEIKESSTKLKIFPSSDSQLDPLVVELSRPGPLTSALFLF
LHSMKETEKGPLSPKVLFNQLCQKAPRFKDFQQQDSQELLHYLLDAVRTEETK
RIQASILKAFNNPTTKTADDETRKKVKAYGKEGVKMNFI DRIFIGELTSTVMC
EECANISTVKDPFIDISLPIIEERVSKPLLWGRMNKYRSLRETDHDRYSGNVT
IENIHQPRAAKKHSSSKDKSQLIHDRKCI RKLSSGETVITYQKNENLEMNGDSL
MFASLMNSESRLESPTDDSEKEASHSESNDADSEPESESESASKQTGLFRSS
SGSGVQPDGPLYPLSAGKLLYTKETDSGDKEMAEAI SELRLSSTVTGDQDFDR
ENQPLNISNNLCFLEGKHLRSYSPONAFQTL SQSYITTSKECSIQSCLYQFTS
MELLMGNKLLCENCTKNKQYQEETSFAEKKVEGVYTNARKQLLISAVPAVL
ILHLKRFHQAGLSLRKVNHRHVDPLMLDLAPFCSATCKNASVGDKVLYGLYGI
VEHSGSMREGHYTAYVKVRTPSRKLSEHNTKKKNVPLKAADNESAGQWVHVS
DTYLQVVPESTRALSQAAYLLFYERVL

Native sequence in bold

Protease cleavage TEV site underlined

Cloning sites BglIII / NotI

DNA sequence of insert

agatcctATGCGGGTGAAAGATCCAACCTAAAGCTTTACCTGAGAAAGCCAAAAG
AAGTAAAAGGCCTACTGTACCTCATGATGAAGACTCTTCAGATGATATTGCTG
TAGGTTTAACTTGCCAACATGTAAGTCATGCTATCAGCGTGAATCATGTAAAG
AGAGCAATAGCTGAGAATCTGTGGTCAGTTTGCTCAGAATGTTTAAAAGAAAG
AAGATTCTATGATGGGCAGCTAGTACTTACTTCTGATATTTGGTTGTGCCTCA
AGTGTGGCTTCCAGGGATGTGGTAAAAACTCAGAAAGCCAACATTCATTGAAG
CACTTTAAGAGTTCCAGAACAGAGCCCCATTGTATTATAATTAATCTGAGCAC
ATGGATTATATGGTGTATGAATGTGATGAAAAATTATCAACGCATTGTAATA
AGAAGGTTTTGGCTCAGATAGTTGATTTTCTCCAGAAACATGCTTCTAAAACA
CAAACAAGTGCATTTTCTAGAATCATGAACTTTGTGAAGAAAAATGTGAAAC
AGATGAAATACAGAAGGGAGGAAAATGCAGAAATTTATCTGTAAGAGGAATTA
CAAATTTAGGAAATACTgcCTTTTTTAAATGCAGTCATGCAGAACTTGGCACAG
ACTTATACTCTTACTGATCTGATGAATGAGATCAAAGAAAAGTAGTACAAAAC
CAAGATTTTTCTTCCCTCAGACTCTCAGCTGGACCCATTGGTGGTGGAACTTT
CAAGGCCTGGACCACTGACTTCTGCCTTGTTCCTGTTTCTTCACAGCATGAAG
GAGACTGAAAAAGGACCACTTCTCCTAAAGTTCTTTTTAATCAGCTTTGTCA
GAAGGCACCTCGATTTAAAGATTTCCAGCAACAGGACAGTCAGGAGCTTCTTC
ATTATCTTCTGGATGCAGTGAGGACAGAAGAAACAAAGCGAATACAAGCTAGC
ATTCATAAAGCATTTAACAACCCAACTACTAAAACCTGCTGATGATGAACTAG
AAAAAAGTCAAAGCATATGGAAAAGAAGGTGTGAAAATGAACTTCATAGATC
GGATCTTTATTGGTGAATTAAC TAGCACGGTCATGTGTGAAGAATGTGCAAT
ATCTCCACGGTGAAAGATCCATTCATTGATATTTCACTTCCATAATAGAAGA
AAGGGTTTTCAAACCTTTACTTTGGGGAAAGAATGAATAAATATAGAAGTTTAC
GGGAGACAGATCATGATCGATACAGTGGCAATGTTACTATAGAAAATATTCAT
CAACCTAGAGCTGCCAAGAAGCATTCTTCATCTAAAGATAAGAGTCAACTAAT
TCATGACCGAAAATGTATTAGAAAATTGTCATCTGGAGAACTGTACATAACC
AGAAAATGAAAACCTTGAAATGAATGGGGATTCTTTAATGTTTGGCCAGCCTC
ATGAATTCTGAGTCACGTCTGAATGAAAGCCCTACTGATGACAGTGAAAAAGA
AGCCAGCCATTCTGAAAGCAATGTTGATGCTGACAGTGAGCCTTCAGAATCTG
AAAGTGCTTCAAAGCAGACTGGGCTGTTCCAGATCCAGTAGTGGATCCGGTGTG
CAGCCAGATGGACCCCTTTACCCTCTGTCAGCAGGTAAACTGCTGTACACCAA
GGAGACTGACAGTGGGTGATAAGGAAATGGCAGAAGCTATTTCTGAACTTCGTT
TGAGCAGCACTGTAACCTGGAGATCAAGATTTTGACAGAGAAAATCAGCCACTA
AATATTTCAAATAATTTATGTTTTTTGGAGGGGAAGCATTGAGGTCTTATAG
TCCCCAAAATGCTTTTTCAGACCCTTCTCAGAGCTATATAACTACTTCTAAAG
AATGTTCAATTCAGTCCTGTCTCTACCAGTTTACATCTATGGAATTACTAATG
GGGAATAATAAGCTTCTATGTGAGAATTGTACTAAAAACAAACAGAAGTACCA
AGAAGAAACCAGTTTTTGCAGAAAAGAAAAGTAGAAGGAGTTTATACTAATGCCA
GGAAGCAATTGCTCATTCTGCTGTTCCAGCTGTCTAATTTCTCCACCTGAAA
AGATTTTCATCAGGCTGGCTTGAGTCTTCGTAAAGTAAACAGACATGTAGATTT
TCCACTTATGCTCGATTTAGCACCATTCTGCTCTGCTACTTGTAAAGAAATGCAA
GTGTGGGAGATAAAGTTCTCTACGGTCTCTATGGCATAAGTGAACATAGTGGC
TCGATGAGAGAAGGCCACTACACTGCTTATGTGAAAGTGAGAACACCCCTCCAG
GAAATTCGGAACATAACACTAAAAAGAAAAATGTGCCTGGTTTTGAAAGCGG
CTGATAATGAATCAGCAGGCCAGTGGGTCCATGTTAGTGACACTTACTTACAG
GTGTTCCAGAATCAAGAGCACTTAGTGCACAAGCCTACCTTCTTTTCTATGA
AAGAGTATTATAAgcggccgc