

Division of Signal Transduction Therapy

Standard Operation Procedure

Dac-USP45

| | |
|-------------------------------------|----------------------|
| <u>Enzyme description:-</u> | Dac-USP45 |
| <u>Clone number:-</u> | DU23041 |
| <u>Source:-</u> | Sf21 Recombinant |
| <u>Tag:-</u> | N-terminal Dac tag |
| <u>Purification method:-</u> | Ampicillin sepharose |
| <u>Expression level:-</u> | 8 mg/L |

Calculated molecular mass:-

| | |
|---|-----------|
| Monoisotopic | 121269 Da |
| Average Mass | 121343 Da |
| [cysteines reduced, methionines have not been oxidised] | |

Theoretical pI:- 8.57

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

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Clone Data Sheet

Dac-USP45

Protein Dac-USP45

Synonyms

Clone Number DU23041

Species Human

Accession Number Protein: Q70EL2 DNA: NM_001080401.1

Tags N-terminal Dac tag

Amino acid sequence of expressed protein MSAIPGVPOIDAESYILIDYNSGKVLAEQNADVRRDPASLTKMMTSYVIGQAM
KAGFKETDLVTIGNDAWATGNPVFKGSSLMFLKPGMQVPVSQLIRGINLQSG
NDACVAMADFAAGSQDAFVGLMNSYVNALGLKNTHFQTVHGLDADGOYSSARD
MALIGQALIRDVPNEYSIYKEKEFTFNGIRQLNRNGLLWDNSLNVLDGDKTGT
DKAGYNLVASATEGQMLISAVMGGRTFKGREAESKLLTWGFRFFETVNPEN
LYFOGGSMRVKDPTKALPEKAKRSKRPTVPHDEDSSDDIAVGLTCQHVSHAIS
VNHVKRAIAENLWSVCSECLKERRFYDQQLVLTSDIWLCLKCGFQCGKNS
QHSLKHFKSSRTEPHCI I INLSTWI IWCYECDEKLSTHCNKKVLAQIVDFLOK
HASKTQTSAFSRIMKLCCEKCETDEIQKGGKCRNLSVRGITNLGNTCFFNAVM
QNLAQTYTLTDLMNEIKESSTKLKIFPSSDSQLDPLVVELSRPGPLTSALFLF
LHSMKETEKGPLSPKVLFNQLCQKAPRFKDFQQQDSQELLHYLLDAVRTEETK
RIQASILKAFNNPTTKTADDETRKKVKAYGKEGVKMNFI DRIFIGELTSTVMC
EECANISTVKDPFIDISLPIIEERVSKPLLWGRMNKYRSLRETDHDRYSGNVT
IENIHQPRAAKKHSSSKDKSQLIHDRKCI RKLSSGETVVTYQKNENLEMNGDSL
MFASLMNSESRLESPTDDSEKEASHSESNDADSEPESESESASKQTGLFRSS
SGSGVQPDGPLYPLSAGKLLYTKETDSGDKEMAEAI SELRLSSTVTGDQDFDR
ENQPLNISNNLCFLEGKHLRSYSPQNAFQTL SQSYITTSKECSIQSCLYQFTS
MELLMGNKLLCENCTKNKQYQEETSFAEKKVEGVYTNARKQLLISAVPAVL
ILHLKRFHQAGLSLRKVNHRVDFPLMLDLAPFCSATCKNASVGDKVLYGLYGI
VEHSGSMREGHYTAYVKVRTPSRKLSEHNTKKKNVPGKAADNESAGQWVHVS
DTYLQVVPESTRALSQAAYLLFYERVL

Native sequence in bold

Protease cleavage TEV site underlined

Cloning sites BglIII / NotI

**DNA sequence of
insert**

ATGCGGGTGAAAGATCCAAC TAAAGCTTTACCTGAGAAAGCCAAAAGAAGTAA
AAGGCCTACTGTACCTCATGATGAAGACTCTTCAGATGATATTGCTGTAGGTT
TAAC TTGCCAACATGTAAGTCATGCTATCAGCGTGAATCATGTAAAGAGAGCA
ATAGCTGAGAATCTGTGGTCAGTTTGCTCAGAATGTTTAAAAGAAAGAAGATT
CTATGATGGGCAGCTAGTACTTACTTCTGATATTTGGTTGTGCCTCAAGTGTG
GCTTCCAGGGATGTGGTAAAACTCAGAAAGCCAACATTCATTGAAGCACTTT
AAGAGTTCCAGAACAGAGCCCCATTGTATTATAATTAATCTGAGCACATGGAT
TATATGGTGTTATGAATGTGATGAAAAATTATCAACGCATTTGTAATAAGAAGG
TTTTGGCTCAGATAGTTGATTTTCTCCAGAAACATGCTTCTAAAACACAAACA
AGTGCATTTTCTAGAATCATGAACTTTGTGAAGAAAAATGTGAAACAGATGA
AATACAGAAGGGAGGAAAATGCAGAAATTTATCTGTAAGAGGAATTACAAATT
TAGGAAATACTTGCTTTTTTAATGCAGTCATGCAGAACTTGGCACAGACTTAT
ACTCTTACTGATCTGATGAATGAGATCAAAGAAAGTAGTACAAAACCTCAAGAT
TTTTCCCTTCCCTCAGACTCTCAGCTGGACCCATTGGTGGTGGAACCTTCAAGGC
CTGGACCACTGACTTCTGCCTTGTTCCTGTTTCTTACAGCATGAAGGAGACT
GAAAAGGACCACCTTCTCCTAAAGTTCTTTTTAATCAGCTTTGTGAGAAGGC
ACCTCGATTTAAAGATTTCCAGCAACAGGACAGTCAGGAGCTTCTTCATTATC
TTCTGGATGCAGTGAGGACAGAAGAAACAAAGCGAATACAAGCTAGCATTTCTA
AAAGCATTTAAACAACCCAACTACTAAAACCTGCTGATGATGAACTAGAAAAAA
AGTCAAAGCATATGGAAAAGAAGGTGTGAAAATGAACTTCATAGATCGGATCT
TTATTGGTGAATTAAC TAGCACGGTCATGTGTGAAGAATGTGCAAATATCTCC
ACGGTGAAAGATCCATTTCATTGATATTTCACTTCTATAATAGAAGAAAGGGT
TTCAAACCTTTACTTTGGGGAAGAATGAATAAATATAGAAGTTTACGGGAGA
CAGATCATGATCGATACAGTGGCAATGTTACTATAGAAAATATTCATCAACCT
AGAGCTGCCAAGAAGCATTCTTCATCTAAAGATAAGAGTCAACTAATTCATGA
CCGAAAATGTATTAGAAAATGTGCATCTGGAGAAACTGTCACATAACCAGAAAA
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TCTGAGTCACGTC TGAATGAAAGCCCTACTGATGACAGTGAAAAAGAAGCCAG
CCATTCTGAAAGCAATGTTGATGCTGACAGTGAGCCTTCAGAATCTGAAAGTG
CTTCAAAGCAGACTGGGCTGTT CAGATCCAGTAGTGGATCCGGTGTGCAGCCA
GATGGACCCCTTTACCCTCTGTCAGCAGGTAAACTGCTGTACACCAAGGAGAC
TGACAGTGGTGATAAGGAAAATGGCAGAAGCTATTTCTGAACTTCGTTTGAGCA
GCACTGTAACCTGGAGATCAAGATTTTGACAGAGAAAATCAGCCACTAAAATATT
TCAAATAATTTATGTTTTTTGGAGGGGAAGCATTTGAGGTCTTATAGTCCCCA
AAATGCTTTTTCAGACCCTTCTCAGAGCTATATAACTACTTCTAAAAGATGTT
CAATTCAGTCCGTCTCTACCAGTTTACATCTATGGAATTACTAATGGGGAAT
AATAAGCTTCTATGTGAGAATTGTACTAAAAACAAACAGAAGTACCAAGAAGA
AACCAGTTTTGCAGAAAAGAAAGTAGAAGGAGTTTATACTAATGCCAGGAAGC
AATTGCTCATTTCTGCTGTTCCAGCTGTCCTAATTTCTCCACCTGAAAAGATTT
CATCAGGCTGGCTTGAGTCTTCGTAAAGTAAACAGACATGTAGATTTTCCACT
TATGCTCGATTTAGCACCATTCCTGCTCTGCTACTTGTAAGAATGCAAGTGTGG
GAGATAAAGTTCTCTACGGTCTCTATGGCATAAGTGAACATAGTGGCTCGATG
AGAGAAGGCCACTACACTGCTTATGTGAAAGTGAGAACACCCTCCAGGAAATT
ATCGGAACATAACACTAAAAAGAAAATGTGCCTGGTTTGAAGCGGCTGATA
ATGAATCAGCAGGCCAGTGGGTCCATGTTAGTGACACTTACTTACAGGTGGTT
CCAGAATCAAGAGCACTTAGTGCACAAGCCTACCTTCTTTTCTATGAAAGAGT
ATTATAA