Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of Halo-Ubiquitin (pSer65) dimer

Enzyme description:- Ubiquitin (pSer65) dimer (1-76) full length

Clone number:- DU24951

Source:- human recombinant

Tag:- His-Halo

Purification method:- Ni-agarose; phosphorylated by PINK1

Expression system:- E.coli

Calculated molecular mass:-

Monoisotopic Da
Average Mass Da
[cysteines reduced, methionines have not been oxidised]

Theoretical pI:-

Purity:- 90%

Enzyme storage buffer:-

50 mM Tris pH 7.5, 150mM NaCl, 1mM DTT

Storage temperature:- -80ºC

Assay:-

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Clone Data Sheet

Protein name Ubiquitin (pSer65) dimer

Protein	Ubiquitin (pSer65) (1-76) full length mature dimer
Synonyms	
Clone Number	DU24951
Species	Human
Accession Number	P62987
Tags	N-terminal His-Halo
Aminoacid sequence of the expressed protein	MHHHHHHMAEIGTGFPFDPHYVEVLGERMHYVDVGPRDGTPVLFLHGNPT SSYVWRNIIPHVAPTHRCIAPDLIGMGKSDKPDLGYFFDDHVRFMDAFIE ALGLEEVVLVIHDWGSALGFHWAKRNPERVKGIAFMEFIRPIPTWDEWPE FARETFQAFRTTDVGRKLIIDQNVFIEGTLPMGVVRPLTEVEMDHYREPF LNPVDREPLWRFPNELPIAGEPANIVALVEEYMDWLHQSPVPKLLFWGTP GVLIPPAEAARLAKSLPNCKAVDIGPGLNLLQEDNPDLIGSEIARWLSTL EISGENLYFQGGSAG**MQIFVKTLTGKTITLEVEPSDTIENVKAKIQDKEG IPPDQQRLIFAGKQLEDGRTLSDYNIQKEpSTLHLVLRLRGGMQIFVKTL TGKTITLEVEPSDTIENVKAKIQDKEGIPPDQQRLIFAGKQLEDGRTLSD YNIQKEpSTLHLVLRLRGG**
Native sequence	mature full length ubiquitin dimer1-76 in bold.
Protease cleavage	TEV-protease site underlined
Cloning sites	BamH1 / Not1

DNA sequence of insert

ATGCATCACCATCACCATCACATGGCAGAAATCGGTACTGGCTTTCCATT
CGACCCCCATTATGTGGAAGTCCTGGGCGAGCGCATGCACTACGTCGATG
TTGGTCCGCGCGATGGCACCCCTGTGCTGTTCCTGCACGGTAACCCGACC
TCCTCCTACGTGTGGCGCAACATCATCCCGCATGTTGCACCGACCCATCG
CTGCATTGCTCCAGACCTGATCGGTATGGGCAAATCCGACAAACCAGACC
TGGGTTATTTCTTCGACGACCACGTCCGCTTCATGGATGCCTTCATCGAA
GCCCTGGGTCTGGAAGAGGTCGTCCTGGTCATTCACGACTGGGGCTCCGC
TCTGGGTTTCCACTGGGCCAAGCGCAATCCAGAGCGCGTCAAAGGTATTG
CATTTATGGAGTTCATCCGCCCTATCCCGACCTGGGACGAATGGCCAGAA
TTTGCCCGCGAGACCTTCCAGGCCTTCCGCACCACCGACGTCGGCCGCAA
GCTGATCATCGATCAGAACGTTTTTATCGAGGGTACGCTGCCGATGGGTG
TCGTCCGCCCGCTGACTGAAGTCGAGATGGACCATTACCGCGAGCCGTTC
CTGAATCCTGTTGACCGCGAGCCACTGTGGCGCTTCCCAAACGAGCTGCC
AATCGCCGGTGAGCCAGCGAACATCGTCGCGCTGGTCGAAGAATACATGG
ACTGGCTGCACCAGTCCCCTGTCCCGAAGCTGCTGTTCTGGGGCACCCCA
GGCGTTCTGATCCCACCGGCCGAAGCCGCTCGCCTGGCCAAAAGCCTGCC
TAACTGCAAGGCTGTGGACATCGGCCCGGGTCTGAATCTGCTGCAAGAAG
ACAACCCGGACCTGATCGGCAGCGAGATCGCGCGCTGGCTGTCGACGCTC
GAGATTTCCGGCGAAAACCTGTATTTTCAGGGCGGATCCGCCGGCATGCA
GATCTTCGTGAAGACCCTGACTGGTAAGACCATCACTCTCGAAGTGGAGC
CGAGTGACACCATTGAGAATGTCAAGGCAAAGATCCAAGACAAGGAAGGC
ATCCCTCCTGACCAGCAGAGGTTGATCTTTGCTGGGAAACAGCTGGAAGA
TGGACGCACCCTGTCTGACTACAACATCCAGAAAGAGTCCACCCTGCACC
TGGTCCTCCGTCTCAGAGGCGGCATGCAGATCTTCGTGAAGACCCTGACT
GGTAAGACCATCACTCTCGAAGTGGAGCCGAGTGACACCATTGAGAATGT
CAAGGCAAAGATCCAAGACAAGGAAGGCATCCCTCCTGACCAGCAGAGGT
TGATCTTTGCTGGGAAACAGCTGGAAGATGGACGCACCCTGTCTGACTAC
AACATCCAGAAAGAGTCCACCCTGCACCTGGTCCTCCGTCTCAGAGGCGG
CTGATAAGCGGCCGC