

Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of His-UBA7

<u>Enzyme description:-</u>	UBA7 1-1012
<u>Clone number:-</u>	DU32194
<u>Source:-</u>	Sf21 recombinant
<u>Tag:-</u>	N-terminal His ₆
<u>Purification method:-</u>	Ni-Sepharose
<u>Expression level:-</u>	15mg/L
<u>Calculated molecular mass:-</u>	
Monoisotopic	116433 Da
Average Mass	116500 Da
[cysteines reduced, methionines have not been oxidised]	
<u>Theoretical pI:-</u>	5.92
<u>Purity:-</u>	90%
<u>Enzyme storage buffer:-</u>	
50 mM HEPES pH 7.5, 10% glycerol, 150mM NaCl, 1mM DTT	
<u>Storage temperature:-</u>	-80°C
<u>Assay:-</u>	
Loading of ISG15 onto UBE2L6 in the absence of DTT	

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Clone Data Sheet

Protein name

Protein	UBA7; UBE1L
Clone Number	DU32194
Species	Human
Accession Number	Protein: P41226 or NP_003326
Tags	N-terminal His ₆
Aminoacid sequence of the expressed protein	MSYYHHHHHDYDIPTTENLYFOGAMGSGIQRPSTSTSSLVAAAMDALDASKLLD EELYSRQLYVLGSPAMQRIQGARVLVSGLQGLGAEVAKNLVLMGVGSLTLHDPH PTCWSDLAAQFLLSEQDLERSRAEASQELLAQLNRAVQVVVHTGDI TEDLLLD QVVVLTAAKLEEQLKVGTLCHKHGVCFLAADTRGLVGQLFCDFGEDFTVQDPTE AEPLTAAIQHISQSGPGILTLRKGANTHYFRDGLVTFSGIEGMVELNDCPRS IHVREDGSLEIGDTTTTFSRYLRGGAITEVKRPKTVRHKSLDTALLQPHVVAQSS QEVHHAHCLHQAFALHKFQHLHGRPPQPWDPVDAETVVGLARDLEPLKRTEEE PLEEPLDEALVRTVALSSAGVLSPMVAMLGAVAAQEVKAI SRKFMPLDQWLYF DALDCLPEDGELLSPEDCALRGSRYDGQIAVFGAGFQEKLRROHYLLVGAGAI GCELLKVFALVGLGAGNSGGLTVVDMDH IERSNLSRQFLFRSQDVGRPKAEVAA AAARGLNPDLOVIPLTYPLDPTTEHI YGDNFFSRVDGVAAALDSFQARRYVAAR CTHYLKPLLEAGTSGTWGSATVFMPHVTEAYRAPASAAASEDAPYPVCTVRYFP STAEHTLQWARHEFEELFRLSAETINHHQQAHTSLADMDEPQTLTLLKPVLGVL RVRPQNWQDCVAWALGHWKLCFHYGIKQLLRHFPPNKVLEDGTPFWSGPKQCPQ PLEFDTNQDTHLLYVLAANLYAQMHGLPGSQDWTALRELLKLLPQDPQOMAP IFASNLELASASAEFGPEQQKELNKALEVWSVGPPLKPLMFEKDDDSNFHVDFV VAAASLRCQNYGIPPVNRAQSKRIVGQI I PAIATTTAAVAGLLGLELYKVVSGP RPRSAFRHSYLHLAENYLI RYMPFAPAIQTFHHLKWTSDRLKVPAGQPPTLE SLLAHLQEQHGLRVRILLHGSALLYAAGWSPEKQAQHLPLRVTELVQQLTGQAP APGQRVLVLELSCEGDDETAFPPLHYEL
Native sequence	full length UBA7 in bold
Protease cleavage	TEV underlined
Cloning sites	Not1

DNA sequence of insert	gcggccgcgATGGATGCCCTGGACGCTTCGAAGCTACTGGATGAGGAGCTGTAT TCAAGACAGCTGTATGTGCTGGGCTCACCTGCCATGCAGAGGATTCAGGGAGCC AGGGTCTTGGTGTGACGGCTGCAGGGCCTGGGGGCCGAGGTGGCCAAGA ACTTG GTTCTGATGGGTGTGGGCAGCCTCACTCTGCATGATCCCCACCCACCTGCTGG TCCGACCTGGCTGCCAGTTTCTCCTCTCAGAGCAGGACTTGGAAAGGAGCAGA GCCGAGCCTCTCAAGAGCTCTTGGCTCAGCTCAACAGAGCTGTCCAGGTCGTC GTGCACACGGGTGACATCACTGAGGACCTGCTGTTGGACTTCCAGGTGGTGGTG CTGACTGCTGCAAAGCTGGAGGAGCAGCTGAAGGTGGGCACCTTGTGTCATAAG CATGGAGTTTGCTTTCTGGCGCTGACACCCGGGGCCTCGTGGGGCAGTTGTTTC
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