Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of UBE2L6

Enzyme description:- UBE2L6

Clone number:- DU32148

Source:- BL21 recombinant

Tag:- cleaved from N-terminal His_6-tag

Purification method:- Ni^{++}-NTA-Sepharose, protease cleavage, SEC

Expression level:- 5mg/L

Calculated molecular mass:-

Monoisotopic	18055 Da
Average Mass	18066Da

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 8.72

Purity:- 90%

Enzyme storage buffer:-

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

Storage temperature:- -80ºC

Assay:-

Loading assay with ISG15 and UBA7 in the presence of Mg-ATP

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Clone Data Sheet

UBE2L6

Protein	UBE2L6
Synonyms	UbcH6, RIG-B
Clone Number	DU32148
Species	Human
Accession Number	Protein: O14933 DNA: NM_004223
Tags	N-terminal His_6 tag
Aminoacid sequence of the expressed protein	GPGS**MMASMRVVKELEDLQKKPPPYLRNLSSDDANVLVWHALLLPDQPPYHL KAFNLRISFPPEYPFKPPMIKFTTKIYHPNVDENGQICLPIISSENWKPCTK TCQVLEALNVLVNRPNIREPLRMDLADLLTQNPELFRKNAEEFTLRFGVDRP S**
Native sequence	in bold
Protease cleavage	Prescission site underlined
Cloning sites	BamH1 / Not1

DNA sequence of insert

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GGATCCATGATGGCGAGCATGCGAGTGGTGAAGGAGCTGGAGGATCTTCA
GAAGAAGCCTCCCCCATACCTGCGGAACCTGTCCAGCGATGATGCCAATG
TCCTGGTGTGGCACGCTCTCCTCCTACCCGACCAACCTCCCTACCACCTG
AAAGCCTTCAACCTGCGCATCAGCTTCCCGCCGGAGTATCCGTTCAAGCC
TCCCATGATCAAATTCACAACCAAGATCTACCACCCCAACGTGGACGAGA
ACGGACAGATTTGCCTGCCCATCATCAGCAGTGAGAACTGGAAGCCTTGC
ACCAAGACTTGCCAAGTCCTGGAGGCCCTCAATGTGCTGGTGAATAGACC
GAATATCAGGGAGCCCCTGCGGATGGACCTCGCTGACCTGCTGACACAGA
ATCCGGAGCTGTTCAGAAAGAATGCCGAAGAGTTCACCCTCCGATTCGGA
GTGGACCGGCCCTCCTAAGCGGCCGC
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