

Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of His-UBE2J1 1-282

Enzyme description:- His-UBE2J1 1-282

Clone number:- DU20686

Source:- BL21 recombinant

Tag:- N-terminal His₆-tag

Purification method:- Ni⁺⁺-NTA-Sepharose

Expression level:- 2mg/L

Calculated molecular mass:-

Monoisotopic 34686 Da

Average Mass 34707 Da

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 7.7

Purity:- 90%

Enzyme storage buffer:-

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

Storage temperature:- -80°C

Assay:-

Loading with Ubiquitin and UBE1 in the presence of Mg-ATP

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Clone Data Sheet

His-UBE2J1 1-282

<u>Protein</u>	UBE2J1 1-282
<u>Synonyms</u>	Ubc6e
<u>Clone Number</u>	DU20686
<u>Species</u>	Human
<u>Accession Number</u>	Protein: Q9Y385 DNA: NM_016021
<u>Tags</u>	N-terminal His ₆ tag
Aminoacid sequence of the expressed protein	MGSSHHHHHHSSGLVPRGSHMASMTGGQ <u>Q</u> MGRGSMETRYNLKSPAVKRLM KEAAELKDPTDHYHA <u>Q</u> PLEDNLF <u>EW</u> HFTVRGPPDSD <u>F</u> DGGVYHGRIVLPP EYPMKPPSIILLTANGRF <u>EV</u> GKKICLSISGHH <u>P</u> ETWQPSWSIR <u>T</u> ALLAI GFMP <u>T</u> KG <u>G</u> E <u>A</u> IGSLDY <u>T</u> PEERRALAKKS <u>Q</u> DFCCEGCGSAMKDVLLPLKSG SDSS <u>Q</u> AD <u>Q</u> EAKELAR <u>Q</u> ISFKA <u>E</u> VN <u>S</u> SGKTISESDLNHSFSLTDL <u>Q</u> DDIPT TF <u>Q</u> GATA <u>S</u> TSYGL <u>Q</u> NSSAAS <u>F</u> H <u>Q</u> PT <u>Q</u> PVAKNTSMSPR <u>Q</u> RR <u>Q</u> Q <u>S</u> Q <u>R</u> RLS TSPDV <u>I</u> QGH <u>Q</u> PRDNHT
Native sequence	in bold, missing the C-terminal transmembrane domain.
Protease cleavage	Thrombin site underlined
Cloning sites	BamH1 / NotI
<u>DNA sequence of insert</u>	GGATCCATGGAGACCCGCTACAACCTGAAGAGTCCGGCTGTTAAACGTTT AATGAAAGAAGCGGCAGAATTGAAAGATCCAACAGATCATTACCATGCGC AGCCTTTAGAGGATAACCTTTTTGAATGGCACTTCACGGTTAGAGGGCCC CCAGACTCCGATTTTGATGGAGGAGTTTATCACGGGCGGATAGTACTGCC ACCAGAGTATCCCATGAAACCACCAAGCATTATTCTCCTAACGGCTAATG GTCGATTTGAAGTGGGCAAGAAAATCTGTTTGAGCATCTCAGGCCATCAT CCTGAAACTTGGCAGCCTTCGTGGAGTATAAGGACAGCATTATTAGCCAT CATTGGGTTTATGCCAACAAAAGGAGAGGGAGCCATAGGTTCTCTAGATT ACACTCCTGAGGAAAGAAGAGCACTTGCCAAAAAATCACAAGATTTCTGT TGTGAAGGATGTGGCTCTGCCATGAAGGATGTCTGTTCCTTTAAATC TGGAAGCGATTCAAGCCAAGCTGACCAAGAAGCCAAAGAAGCTGGCTAGGC AAATAAGCTTTAAGGCAGAAGTCAATTCATCTGGAAAGACTATCTCTGAG TCAGACTTAAACCACTCTTTTCACTAACTGATTACAAAGATGATATACC TACAACATTCCAGGGTGCTACGGCCAGTACATCGTACGGACTCCAGAATT CCTCAGCAGCATCCTTTTCATCAACCTACCCAACCTGTAGCTAAGAATACC TCCATGAGCCCTCGACAGCGCCGGGCCAGCAGCAGAGTCAGAGAAGGTT GTCTACTTCACCAGATGTAATCCAGGGCCACCAGCCAAGAGACAACCACA CTTAGGCGGCCCGC