

Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of GST-UBE2J1 1-282

Enzyme description:- GST-UBE2J1 1-282

Clone number:- DU20687

Source:- BL21 recombinant

Tag:- N-terminal GST-tag

Purification method:- GSH-Sepharose

Expression level:- 3mg/L

Calculated molecular mass:-

Monoisotopic 57952 Da

Average Mass 57987 Da

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 6.5

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

GST-UBE2J1 1-282

<u>Protein</u>	UBE2J1 1-282
<u>Synonyms</u>	Ubc6e
<u>Clone Number</u>	DU20687
<u>Species</u>	Human
<u>Accession Number</u>	Protein: Q9Y385 DNA: NM_016021
<u>Tags</u>	N-terminal GST-tag
Aminoacid sequence of the expressed protein	MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELGLEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAIEISMLEGAVLDIRYGVSRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDHVTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPOIDKYLKSSKYIAWPLQGWQATFGGGDHPPKSDLEVLFOGPLGSMETRYNLKSPAVKRLMKEA AELKDPTDHYHAQPLEDNLFWEHFTVRGPPDSDFDGGVYHGRIVLPPEYPMKPPSIILLTANGRFEVGKKICLSISGHPETWQPSWSIRTALLAIIIGFMP PTKGEGAIGSLDYTPERRALAKKSQDFCCEGCGSAMKDVLPLKSGSDS SQADQEAKELARQISFKAEVNSSGKTI SESDLNHSFSLTDLQDDIPTTFQ GATASTSYGLQNSSAASFHQPTQP VAKNTSMSPRQRRRQQSQRRRLSTSP DVIQGHQPRDNHT
Native sequence	in bold, missing the C-terminal transmembrane domain
Protease cleavage	Prescission Protease site underlined
Cloning sites	BamH1 / NotI
<u>DNA sequence of insert</u>	<u>GGATCCATGGAGACCCGCTACAACCTGAAGAGTCCGGCTGTAAACGTTT</u> <u>AATGAAAGAAGCGCAGAATTGAAAGATCCAACAGATCATTACCATGCGC</u> <u>AGCCTTTAGAGGATAACCTTTTTGAATGGCACTTCACGGTTAGAGGGCCC</u> <u>CCAGACTCCGATTTTGATGGAGGAGTTTATCACGGGCGGATAGTACTGCC</u> <u>ACCAGAGTATCCCATGAAACCACCAAGCATTATTCTCCTAACGGCTAATG</u> <u>GTCGATTTGAAGTGGCAAGAAAATCTGTTTGAGCATCTCAGGCCATCAT</u> <u>CCTGAAACTTGGCAGCCTTCGTGGAGTATAAGGACAGCATTATTAGCCAT</u> <u>CATTGGGTTTATGCCAACAAAAGGAGAGGGAGCCATAGGTTCTCTAGATT</u> <u>ACACTCCTGAGGAAAGAAGAGCACTTGCCAAAAAATCACAAGATTTCTGT</u> <u>TGTGAAGGATGTGGCTCTGCCATGAAGGATGTCTGTTGCCTTTAAAATC</u> <u>TGGAAGCGATTCAAGCCAAGCTGACCAAGAAGCCAAAGAAGCTGGCTAGGC</u> <u>AAATAAGCTTTAAGGCAGAAGTCAATTCATCTGGAAAGACTATCTCTGAG</u> <u>TCAGACTTAAACCACTCTTTTTCACTAACTGATTTACAAGATGATATAACC</u> <u>TACAACATTCCAGGGTGCTACGGCCAGTACATCGTACGGACTCCAGAATT</u> <u>CCTCAGCAGCATCCTTTCATCAACCTACCCAACCTGTAGCTAAGAATACC</u> <u>TCCATGAGCCCTCGACAGCGCCGGGCCAGCAGCAGAGTCAGAGAAGGTT</u> <u>GTCTACTTCACCAGATGTAATCCAGGGCCACCAGCCAAGAGACAACCACA</u> <u>CTTAGGCGGCCGC</u>

