

Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of His-UBE2Q1

<u>Enzyme description:-</u>	UBE2Q1
<u>Clone number:-</u>	DU20176
<u>Source:-</u>	BL21 recombinant
<u>Tag:-</u>	N-terminal His ₆ -tag
<u>Purification method:-</u>	Ni ⁺⁺ -NTA-Sepharose
<u>Expression level:-</u>	3 mg/L
<u>Calculated molecular mass:-</u>	
Monoisotopic	49422 Da
Average Mass	49451 Da
[cysteines reduced, methionines have not been oxidised]	
<u>Theoretical pI:-</u>	5.68
<u>Purity:-</u>	90%
<u>Enzyme storage buffer:-</u>	
50mM HEPES pH 7.5, 150mM NaCl, 10% glycerol, 1mM DTT	
<u>Storage temperature:-</u>	-80°C
<u>Assay:-</u>	
Loading with Ubiquitin and UBE1 in the presence of Mg-ATP	

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Clone Data Sheet

His-UBE2Q1

Protein UBE2Q1
Synonyms UBE2Q, GTAP, NICE-5
Clone Number DU20176
Species Human
Accession Number Protein: NP_060052 DNA: NM_017582
Tags N-terminal His₆-tag

Aminoacid sequence of the expressed protein
HHHHHSSGLVPRGSHMASMTGGQQMGRGSEF**QQPQPQGGQQPQPGQQLG**
GQGAAPGAGGGPGGGPGPCLRRELKLLSIFHRGHERFRIASACLDL
SCEFLLAGAGGAGAGAAPGPHLPPRGSVPGDPVRIHCNITESYPVPIW
SVESDDPNLAAVLERLVDIKKGNLTLLOHLKRIISDLCKLYNLPQHPDVE
MLDQPLPAEQCTQEDVSSSEDEDEEMPEDETLDDHYEMKEEPAEGKSED
DGIGKENLAILEKIKKNQRQDYLNQAVSGSVQATDRLMKELRDIYRSQSF
KGGNYAVELVNDSLYDWNVKKLVQDSALHNDLQILKEKEGADFILLNF
SFKDNFPFDPFVVRVSPVLSGGYVLGGGAICMELLTKQGWSSAYSIESV
IMQISATLVKVKARVQFGANKSQYSLTRAQOSYKSLVQIHEKNGWYTPPK
EDG

Native sequence in bold, Start Methionine is missing
Protease cleavage Thrombin site underlined
Cloning sites EcoR1 / NotI

DNA sequence of the insert
GAATTCAGCAGCCGAGCCGAGCCGAGGGGCAGCAGCAGCCGGGGCCGGGGCA
GCAGCTGGGGGGCCAGGGGGCGGCGCCGGGGGGCCGGGGCCAGGGG
GGGGCCCGGGCCGGGGCCCTGCCTGAGGCGAGAGCTGAAGCTGCTCGAG
TCCATCTTCCACCGCGGCCACGAGCGCTTCCGCATTGCCAGCGCCTGCCT
GGACGAGCTGAGCTGCGAGTTCCTGCTGGCTGGGGCCGAGGGGCCGGGG
CGGGGGCCGCGCCCGGACCGCATCTCCCCCACGGGGTTCGGTGCCTGGG
GATCCTGTCCGCATCCACTGCAACATCACGGAGTCATACCCTGCTGTGCC
CCCCATCTGGTTCGGTGGAGTCTGATGACCCTAACTTGGCTGCTGTCTTGG
AGAGGCTGGTGGACATAAAGAAAGGAATACTCTGCTATTGCAGCATCTG
AAGAGGATCATCTCCGACCTGTGTAAACTCTATAACCTCCCTCAGCATCC
AGATGTGGAGATGCTGGATCAACCCTTGCCAGCAGAGCAGTGCACACAGG
AAGACGTGTCTTCAGAAGATGAAGATGAGGAGATGCCTGAGGACACAGAA
GACTTAGATCACTATGAAATGAAAGAGGAAGAGCCAGCTGAGGGCAAGAA
ATCTGAAGATGATGGCATTGGAAAAGAAAAGTGGCCATCCTAGAGAAAA
TTAAAAGAACCAGAGGCAAGATTACTTAAATGGTGCAGTGTCTGGCTCG
GTGCAGGCCACTGACCGGCTGATGAAGGAGCTCAGGGATATATACCGATC
ACAGAGTTTCAAAGGCGGAAACTATGCAGTCGAACTCGTGAATGACAGTC
TGTATGATTGGAATGTCAAACCTCCTCAAAGTTGACCAGGACAGCGCTTTG
CACACGATCTCCAGATCCTCAAAGAGAAAGAAGGAGCCGACTTCATTCT

ACTTAACTTTTCCTTTAAAGATAACTTTCCCTTTGACCCACCATTTGTCA
GGGTGTGTCTCCAGTCCTCTCTGGAGGGTATGTTCTGGGCGGAGGGGCC
ATCTGCATGGAACCTCTCACCAAACAGGGCTGGAGCAGTGCCTACTCCAT
AGAGTCAGTGATCATGCAGATCAGTGCCACACTGGTGAAGGGGAAAGCAC
GAGTGCAGTTTGGAGCCAACAAATCTCAATACAGTCTGACAAGAGCACAG
CAGTCCTACAAGTCCTTGGTGCAGATCCACGAAAAAACGGCTGGTACAC
ACCCCAAAGAAGACGGCTAAGCGGCCGC