

Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of UBE2M

<u>Enzyme description:-</u>	GST-UBE2M
<u>Clone number:-</u>	DU15805
<u>Source:-</u>	BL21 recombinant
<u>Tag:-</u>	N-terminal GST-tag
<u>Purification method:-</u>	GSH-Sepharose
<u>Expression level:-</u>	2mg/L
<u>Calculated molecular mass:-</u>	
Monoisotopic	48671 Da
Average Mass	48702 Da
[cysteines reduced, methionines have not been oxidised]	
<u>Theoretical pI:-</u>	6.61
<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 assay with NEDD8 and NAE1/NAE2 in the presence of Mg-ATP	

Division of Signal Transduction Therapy

Clone Data Sheet

GST-UBE2M

<u>Protein</u>	GST-UBE2M
<u>Synonyms</u>	Ubc12
<u>Clone Number</u>	DU15805
<u>Species</u>	Human
<u>Accession Number</u>	Protein: P61081 DNA: NM_003969
<u>Tags</u>	N-terminal GST-tag
Aminoacid sequence of the expressed protein	MSPILGYWKIKGLVQPTRLLEYLEEKYEELHYERDEGDKWRNKKFELGLEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAIEISMLEGAVLDIRYGVSRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDHVTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPOIDKYLKSSKYIAWPLQGWQATFGGGDHPPKSDLEVLFOGPLGSPGIPGSTRAAAMIKL FSLKQOKKEESAGG TKGSSKKASAAQLRIQKDINELNLPK TCDISFSDPDDL NFKLVIC PDEGFYKSGKFVFSFKVGGQGYPHDPPKVKCETMVYHPNIDLEG NVCLNILREDWKPVL TINSIIYGLQYLFLEPNPEDPLNKEAAEVLQNNRR LFEQNVQRSMRGGYIGSTYFER CLK
Native sequence	in bold
Protease cleavage	Prescission site underlined
Cloning sites	NotI
<u>DNA sequence of insert</u>	GCGGCCGCGATGATCAAGCTGTTCTCGCTGAAGCAGCAGAAGAAGGAGGAGGAGTTCGGCGGGCGGCACCAAGGGCAGCAGCAAGAAGGCGTTCGGCGGGCGCAGCTGCGGATCCAGAAGGACATAAACGAGCTGAACCTGCCCAAGACGTGTGATATCAGCTTCTCAGATCCAGACGACCTCCTCAACTTCAAGCTGGTCATCTGTCTGATGAGGGCTTCTACAAGAGTGGGAAGTTTGTGTTTCAGTTTTAAGGTGGGCCAGGGTTACCCGCATGATCCCCCAAGGTGAAGTGTGAGACAATGGTCTATCACCCCAACATTGACCTCGAGGGCAACGTCTGCCTCAACATCCTCAGAGAGGACTGGAAGCCAGTCCTTACGATAAACTCCATAATTTATGGCCTGCAGTATCTCTTCTTGGAGCCCAACCCCGAGGACCCACTGAACAAGGAGGCCGAGAGGTCCTGCAGAACAACCGGGCGCTGTTTGTGAGCAGAACGTGCAGCGCTCCATGCGGGGTGGCTACATCGGCTCCACCTACTTTGAGCGCTGCCTGAAATAGGCGGCCGC