

Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of His-UBE2I

<u>Enzyme description:-</u>	His-UBE2I
<u>Clone number:-</u>	DU12799
<u>Source:-</u>	BL21 recombinant
<u>Tag:-</u>	N-terminal His ₆ -tag
<u>Purification method:-</u>	Ni ⁺⁺ -NTA-Sepharose
<u>Expression level:-</u>	50mg/L
<u>Calculated molecular mass:-</u>	
Monoisotopic	20438 Da
Average Mass	20450 Da
[cysteines reduced, methionines have not been oxidised]	
<u>Theoretical pI:-</u>	9.19
<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

Assay:-

Loading assay with SUMO and SAE1/SAE2 in the presence of Mg-ATP

Division of Signal Transduction Therapy

Clone Data Sheet

His-UBE2I

<u>Protein</u>	UBE2I
<u>Synonyms</u>	Ubc9
<u>Clone Number</u>	DU12799
<u>Species</u>	Human
<u>Accession Number</u>	Protein: NP_003336
<u>Tags</u>	N-terminal His ₆ tag
Aminoacid sequence of the expressed protein	MGSSHHHHHHSSGLEVL <u>FQGP</u> SGMSGIALSRLAQERKAWRKDHPFGFVAV PTKNPDGTMNLMNWECAIPGKKGTPWEGGLFKLRMLFKDDYPSSPPKCKF EPPLFHPNVYPSGTVCLSI LEEDKDWRPAITIKQILLGIQELLNEPNIQD PAQAEAYTIYCQNRVEYEKRVRAQAKKFAPS
Native sequence	in bold
Protease cleavage	Prescission protease site underlined
Cloning sites	BamH1 / NotI
<u>DNA sequence of insert</u>	GGATCCATGTCTGGGGATCGCCCTCAGCAGACTCGCCCAGGAGAGGAAAGC ATGGAGGAAAGACCACCCATTTGGTTTCGTGGCTGTCCCAACAAAAAATC CCGATGGCACGATGAACCTCATGAAC TGGGAGTGCGCCATTCCAGGAAAG AAAGGGACTCCGTGGGAAGGAGGCTTGTTTAAACTACGGATGCTTTTCAA AGATGATTATCCATCTTCGCCACCAAAATGTAAATTCTGAACCACCATTAT TTCACCCGAATGTGTACCCTTCGGGGACAGTGTGCCTGTCCATCTTAGAG GAGGACAAGGACTGGAGGCCAGCCATCACAATCAAACAGATCCTATTAGG AATACAGGAACTTCTAAATGAACCAAATATCCAAGACCCAGCTCAAGCAG AGGCCTACACGATT TACTGCCAAAACAGAGTGGAGTACGAGAAAAGGGTC CGAGCACAAGCCAAGAAGTTTGCGCCCTCATAAGCGGCCGC