

*Division of Signal Transduction Therapy*

**Standard Operation Procedure**

**Preparation of GST-UBE2L3**

<b><u>Enzyme description:-</u></b>	GST-UBE2L3
<b><u>Clone number:-</u></b>	DU3772
<b><u>Source:-</u></b>	BL21 recombinant
<b><u>Tag:-</u></b>	N-terminal GST-tag
<b><u>Purification method:-</u></b>	GSH-Sepharose
<b><u>Expression level:-</u></b>	5mg/L
<b><u>Calculated molecular mass:-</u></b>	
Monoisotopic	44656 Da
Average Mass	44684Da
[cysteines reduced, methionines have not been oxidised]	
<b><u>Theoretical pI:-</u></b>	6.87
<b><u>Purity:-</u></b>	90%
<b><u>Enzyme storage buffer:-</u></b>	
50mM HEPES pH 7.5, 150mM NaCl, 10% glycerol, 1mM DTT	
<b><u>Storage temperature:-</u></b>	-80°C
<b><u>Assay:-</u></b>	
Loading with Ubiquitin and UBE1 in the presence of Mg-ATP	

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**Clone Data Sheet**

**GST-UBE2L3**

<b>Protein</b>	UBE2L3
<b>Synonyms</b>	UbcH7
<b>Clone Number</b>	DU3772
<b>Species</b>	Human
<b>Accession Number</b>	Protein: P68036      DNA: NM_003347
<b>Tags</b>	N-terminal GST-tag
Aminoacid sequence of the expressed protein	MSPILGYWKIKGLVQPTTRLLEYLEEKYEEHLYERDEGDKWRNKKFELGL EFPNLPLYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAESMLEGAVL DIRYGVSRRIAYSKDFETLKVDFLSKPEMLKMFDRLCHKTYLNGDHVTH PDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPQIDKYLKSSKYIA WPLQGWQATFGGGDHPPKSDELVLFQGPLGSMAASRRLMKELEEIRKCGM <b>KNFRNIQVDEANLLTWQGLIVPDNPPYDKGAFRIEINFPAEYPFKPPKIT</b> <b>FTKIYHPNIDEKGQVCLPVI</b> SAENWKPATKTDQVIQSLIALVNDPQPEH <b>PLRADLAEEYSKDRKKFCKNAEEFTKKYGEKRPVD</b>
Native sequence	in bold
Protease cleavage	Prescission site underlined
Cloning sites	BamH1 / Not1
<b>DNA sequence of insert</b>	GGATCCATGGCGGCCAGCAGGAGGCTGATGAAGGAGCTTGAAGAAATCCG CAAATGTGGATGAAAAACTTCCGTAACATCCAGGTTGATGAAGCTAATT TATTGACTTGGCAAGGGCTATTGTTCCCTGACAACCCCTCATATGATAAG GGAGCCTTCAGAACATCGAAACTTTCAAAGATCTATCACCCAAACATCGACGAAA ACCGAAGATCACATTAAAACAAAGATCTATCACCCAAACATCGACGAAA AGGGGCAGGTCTGTCTGCCAGTAATTAGTGCGAAAACTGGAAGCCAGCA ACCAAAACCGACCAAGTAATCCAGTCCCTCATAGCACTGGTGAATGACCC CCAGCCTGAGCACCGCTTCGGGCTGACCTAGCTGAAGAATACTCTAAGG ACCGTAAAAAAATTCTGTAAGAATGCTGAAGAGTTACAAAGAAATATGGG GAAAAGCGACCTGTGGACTAAGCGGCCGC