Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of GST-UBE2D1

Enzyme description:- UBE2D1 (2-147)

Clone number:- DU4151

Source:- human recombinant

Tag:- N-terminal GST-tag

Purification method:- GSH-Sepharose

Expression system:- *E.coli*

Calculated molecular mass:-

Monoisotopic	43266 Da
Average Mass	43293 Da

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 6.47

Purity:- 90%

Enzyme storage buffer:-

50mM HEPES pH 7.5, 150mM NaCl, 10% glycerol, 1mM DTT

Storage temperature:- -80ºC

Assay:-

Loading with Ubiquitin and UBE1 in the presence of Mg-ATP

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Clone Data Sheet

GST-UBE2D1

Protein	UBE2D1 (2-147)
Synonyms	UbcH5a, E2D1, E2-17K-1
Clone Number	DU4151
Species	Human
Accession Number	Protein: P51668 DNA: NM_003338
Tags	N-terminal GST-tag, cleaved
Aminoacid sequence of the expressed protein	MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELGL EFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLEGAVL DIRYGVSRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDHVTH PDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPQIDKYLKSSKYIA WPLQGWQATFGGGDHPPKSDLEVLFQGPLGS**ALKRIQKELSDLQRDPPAH CSAGPVGDDLFHWQATIMGPPDSAYQGGVFFLTVHFPTDYPFKPPKIAFT TKIYHPNINSNGSICLDILRSQWSPALTVSKVLLSICSLLCDPNPDDPLV PDIAQIYKSDKEKYNRHAREWTQKYAM**
Native sequence	in bold
Protease cleavage	Prescission Protease site underlined
Cloning sites	BamH1 / Not1

DNA sequence of insert

ATGGGCAGCAGCCATCATCATCATCACAGCAGCGGCCTGGTGCCGCG
CGGCAGCCATATGGCTAGCATGACTGGTGGACAGCAAATGGGTCGCGGAT
CCGCGCTGAAGAGGATTCAGAAAGAATTGAGTGATCTACAGCGCGATCCA
CCTGCTCACTGTTCAGCTGGACCTGTGGGAGATGACTTGTTCCACTGGCA
AGCCACTATTATGGGGCCTCCTGATAGCGCATATCAAGGTGGAGTCTTCT
TTCTCACTGTACATTTTCCGACAGATTATCCTTTTAAACCACCAAAGATT
GCTTTCACAACAAAAATTTACCATCCAAACATAAACAGTAATGGAAGTAT
TTGTCTCGATATTCTGAGGTCACAATGGTCACCAGCTCTGACTGTATCAA
AAGTTTTATTGTCCATATGTTCTCTACTTTGTGATCCTAATCCAGATGAC
CCCTTAGTACCAGATATTGCACAAATCTATAAATCAGACAAAGAAAAATA
CAACAGACATGCAAGAGAATGGACTCAGAAATATGCAATGTAAGCGGCCG
C