Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of UBE2F

Enzyme description:- UBE2F

Clone number:- DU14051

Source:- BL21 recombinant

Tag:- cleaved from N-terminal GST-tag

Purification method:- GSH-Sepharose, protease treatment

Expression level:- 3mg/L

Calculated molecular mass:-

Monoisotopic	21474 Da
Average Mass	21487 Da

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 6.7

Purity:- 90%

Enzyme storage buffer:-

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

Storage temperature:- -80ºC

Assay:-

Loading assay with NEDD8 and NAE1/NAE2 in the presence of Mg-ATP

Division of Signal Transduction Therapy

Clone Data Sheet

UBE2F

Protein	UBE2F
Synonyms	
Clone Number	DU14051
Species	Human
Accession Number	Protein: Q969M7 DNA: NM_080678
Tags	N-terminal GST-tag (cleaved)
Aminoacid sequence of the expressed protein	GPLGS**MLTLASKLKRDDGLKGSRTAATASDSTRRVSVRDKLLVKEVAELEAN LPCTCKVHFPDPNKLHCFQLTVTPDEGYYQGGKFQFETEVPDAYNMVPPKVK CLTKIWHPNITETGEICLSLLREHSIDGTGWAPTRTLKDVVWGLNSLFTDLL NFDDPLNIEAAEHHLRDKEDFRNKVDDYIKRYAR**
Native sequence	in bold
Protease cleavage	Prescission protease site underlined
Cloning sites	BamH1 / Not1

DNA sequence of insert

```
GGATCCATGCTAACGCTAGCAAGTAAACTGAAGCGTGACGATGGTCTCAA
AGGGTCCCGGACGGCAGCCACAGCGTCCGACTCGACTCGGAGGGTTTCTG
TGAGAGACAAATTGCTTGTTAAAGAGGTTGCAGAACTTGAAGCTAATTTA
CCTTGTACATGTAAAGTGCATTTTCCTGATCCAAACAAGCTTCATTGTTT
TCAGCTAACAGTAACCCCAGATGAGGGTTACTACCAGGGTGGAAAATTTC
AGTTTGAAACTGAAGTTCCCGATGCGTACAACATGGTGCCTCCCAAAGTG
AAATGCCTGACCAAGATCTGGCACCCCAACATCACAGAGACAGGGGAAAT
ATGTCTGAGTTTATTGAGAGAACATTCAATTGATGGCACTGGCTGGGCTC
CCACAAGAACATTAAAGGATGTCGTTTGGGGATTAAACTCTTTGTTTACT
GATCTTTTGAATTTTGATGATCCACTGAATATTGAAGCTGCAGAACATCA
TTTGCGGGACAAGGAGGACTTCCGGAATAAAGTGGATGACTACATCAAAC
GTTATGCCAGATGAGCGGCCGC
```