

Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of His-UBE2V1

Enzyme description:- UBE2V1 (2-170) full length without Start Met

Clone number:- DU20179

Source:- BL21 recombinant

Tag:- N-terminal His₆-tag

Purification method:- Ni⁺⁺-NTA-Sepharose

Expression level:- 2mg/L

Calculated molecular mass:-

Monoisotopic 22343 Da

Average Mass 22357 Da

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 9.59

Purity:- 90%

Enzyme storage buffer:-

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

Storage temperature:- -80°C

Assay:-

Production of free K63 linked Ub-chains with Ubiquitin, UBE1 and UBE2N in the presence of Mg-ATP

Assay buffer: 50mM HEPES, pH 7.5, 1mM DTT

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Clone Data Sheet

His-UBE2V1

Protein UBE2V1
Synonyms Uev1A, CIR1, CROC1
Clone Number DU20179
Species Human
Accession Number Protein: NP_068823 DNA: NM_021988
Tags N-terminal His₆ tag
Aminoacid sequence of the expressed protein HHHHHHSSGLVPRGSHMASMTGGQQMGRGSPGEVQASYLKLSQSKLSDEGR
LEPRKFHCKGVKVPNFRLLLEELEEGQKGVGDGTVSWGLEDDEDMTLTRW
TGMIIIGPPRTIYENRIYSLKIECGPKYPEAPPFVRFVTKINMNGVNSSNG
VVDPRAISVLAKWQNSYSIKVVLQELRRLMMSKENMKLPQPPEGQCYSN
Native sequence in bold, Start Met is missing
Protease cleavage Thrombin site underlined
Cloning sites BamH1 / Sal1

DNA sequence of insert GGATCCCCAGGAGAGGTTCAAGCGTCTTACCTGAAGTCACAAAGCAAAC
GAGTGATGAAGGAAGACTTGAACCTAGAAAATTTCACTGCAAAGGAGTAA
AAGTCCCTCGCAATTTCCGACTGTTGGAAGAAGTGAAGAAGGCCAGAAA
GGAGTAGGAGATGGCACAGTTAGCTGGGGTCTAGAAGATGACGAAGACAT
GACACTTACAAGATGGACAGGGATGATAATTGGGCCCTCAAGAACAATTT
ATGAAAACCGAATATACAGCCTTAAAATAGAATGTGGACCTAAATACCCA
GAAGCACCCCCCTTTGTAAGATTTGTAACAAAAATTAATATGAATGGAGT
AAATAGTTCTAATGGAGTGGTGGACCCAAGAGCCATATCAGTGCTAGCAA
AATGGCAGAATTCATATAGCATCAAAGTTGTCTGCAAGAGCTTCGGCGC
CTAATGATGTCTAAAGAAAATATGAAACTCCCTCAGCCGCCCGAAGGACA
GTGTTACAGCAATTAAGTCGAC