

Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of GST-Ub tetramer (linear)

Enzyme description:- GST-Ubiquitin-Ubiquitin-Ubiquitin-Ubiquitin

Clone number:- DU20766

Source:- BL21 recombinant

Tag:- N-terminal GST

Purification method:- GSH-Sepharose

Expression level:- 10mg/L

Calculated molecular mass:-

Monoisotopic 61119 Da

Average Mass 61156 Da

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 6.4

Purity:- 90%

Enzyme storage buffer:-

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

Storage temperature:- -80°C

Assay:-

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Clone Data Sheet

GST-Ubiquitin-tetramer (linear)

<u>Protein</u>	GST-Ubiquitin tetramer (linear)
<u>Synonyms</u>	Ubiquitin-60S ribosomal protein L40; AltName: CEP52; Ubiquitin A-52 residue ribosomal protein fusion product 1
<u>Clone Number</u>	DU20766
<u>Species</u>	Human
<u>Accession Number</u>	Protein: P62987
<u>Tags</u>	GST- at the N-terminus
Aminoacid sequence of the expressed protein	MSPILGYWKIKGLVQPTRLLEYLEEKYEEHLYERDEGDKWRNKKFE LGFLEFPNLPYYIDGDVKLTQSMAIRYIADKHNMLGGCPKERAIEISM LEGAVLDIRYGVSRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKT YLNQDHVTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPO IDKYLKSSKYIAWPLQGWQATFGGGDHPPKSDLEVLFGPLGSAGMQ IFVKTLTGKTITLEVEPSDTIENVKAKIQDKEGIPPDQORLIFAGKQ LEDGRTLSDYNIQESTLHLVLRRLRGGMQIFVKTLTGKTITLEVEPS DTIENVKAKIQDKEGIPPDQORLIFAGKQLEDGRTLSDYNIQESTL HLVLRRLRGGMQIFVKTLTGKTITLEVEPSDTIENVKAKIQDKEGIPP DQORLIFAGKQLEDGRTLSDYNIQESTLHLVLRRLRGGMQIFVKTLT GKTITLEVEPSDTIENVKAKIQDKEGIPPDQORLIFAGKQLEDGRTL SDYNIQESTLHLVLRRLRGG
Native sequence	
Protease cleavage	Prescission Protease
Cloning sites	NaeI / Sfo1

DNA sequence of
expression cassette

ATGTCCCCTATACTAGGTTATTGGAAAATTAAGGGCCTTGTGCAACC
CACTCGACTTCTTTTGGAAATATCTTGAAGAAAAATATGAAGAGCATT
TGTATGAGCGCGATGAAGGTGATAAATGGCGAAACAAAAAGTTTGAA
TTGGGTTTGGAGTTTCCCAATCTTCTTATTATATTGATGGTGATGT
TAAATTAACACAGTCTATGGCCATCATACGTTATATAGCTGACAAGC
ACAACATGTTGGGTGGTGTCCAAAAGAGCGTGCAGAGATTTCAATG
CTTGAAGGAGCGGTTTTGGATATTAGATACGGTGTTCGAGAATTGC
ATATAGTAAAGACTTTGAAACTCTCAAAGTTGATTTTCTTAGCAAGC
TACCTGAAATGCTGAAAATGTTTCGAAGATCGTTTATGTCATAAAACA
TATTTAAATGGTGATCATGTAACCCATCCTGACTTCATGTTGTATGA
CGCTCTTGATGTTGTTTTATACATGGACCCAATGTGCCTGGATGCGT
TCCCAAATTAGTTTTGTTTTAAAAAACGTATTGAAGCTATCCCACAA
ATTGATAAGTACTTGAAATCCAGCAAGTATATAGCATGGCCTTTGCA
GGGCTGGCAAGCCACGTTTGGTGGTGGCGACCATCCTCCAAAATCGG
ATCTGGAAGTTCTGTTCCAGGGGCCCTGGGATCC**GCCGGCATGCAG**
ATCTTCGTGAAGACCCTGACTGGTAAGACCATCACTCTCGAAGTGGA
GCCGAGTGACACCATTGAGAATGTCAAGGCAAAGATCCAAGACAAGG
AAGGCATCCCTCCTGACCAGCAGAGGTTGATCTTTGCTGGGAAACAG
CTGGAAGATGGACGCACCCTGTCTGACTACAACATCCAGAAAGAGTC
CACCCTGCACCTGGTCTCCGTCTCAGAGGCGGCATGCAGATCTTCG
TGAAGACCCTGACTGGTAAGACCATCACTCTCGAAGTGGAGCCGAGT
GACACCATTGAGAATGTCAAGGCAAAGATCCAAGACAAGGAAGGCAT
CCCTCCTGACCAGCAGAGGTTGATCTTTGCTGGGAAACAGCTGGAAG
ATGGACGCACCCTGTCTGACTACAACATCCAGAAAGAGTCCACCCTG
CACCTGGTCCCTCCGTCTCAGAGGCATGCAGATCTTCGTGAAGAC
CCTGACTGGTAAGACCATCACTCTCGAAGTGGAGCCGAGTGACACCA
TTGAGAATGTCAAGGCAAAGATCCAAGACAAGGAAGGCATCCCTCCT
GACCAGCAGAGGTTGATCTTTGCTGGGAAACAGCTGGAAGATGGACG
CACCCTGTCTGACTACAACATCCAGAAAGAGTCCACCCTGCACCTGG
TCCTCCGTCTCAGAGGCGGCATGCAGATCTTCGTGAAGACCCTGACT
GGTAAGACCATCACTCTCGAAGTGGAGCCGAGTGACACCATTGAGAA
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TCTGACTACAACATCCAGAAAGAGTCCACCCTGCACCTGGTCTCCG
TCTCAGAGGCGGCTGATAAGCGGCCGC