

OTULIN [GST-tagged]

Deconjugating enzyme: Deubiquitylase

Alternate Name: FAM105B

Cat. No. 64-0048-050
Lot. No. 30164

Quantity: 50 µg
Storage: -70°C



FOR RESEARCH USE ONLY

NOT FOR USE IN HUMANS

CERTIFICATE OF ANALYSIS Page 1 of 2

Background

Deconjugating enzymes (DCEs) are proteases that process ubiquitin or ubiquitin-like gene products, reverse the modification of proteins by a single ubiquitin or ubiquitin-like protein (UBL) and remodel polyubiquitin (or poly-UBL) chains on target proteins (Reyes-Turcu *et al.*, 2009). The deubiquitylating – or deubiquitinating – enzymes (DUBs) represent the largest family of DCEs and regulate ubiquitin dependent signalling pathways. The activities of the DUBs include the generation of free ubiquitin from precursor molecules, the recycling of ubiquitin following substrate degradation to maintain cellular ubiquitin homeostasis and the removal of ubiquitin or ubiquitin-like proteins (UBL) modifications through chain editing to rescue proteins from proteasomal degradation or to influence cell signalling events (Komander *et al.*, 2009). There are two main classes of DUB, cysteine proteases and metalloproteases. Otulin is a cysteine protease and a member of the OTU (ovarian tumour) superfamily of proteins (Balakirev *et al.*, 2003). Cloning of the human gene was first described by Ota *et al.*, (2004). OTU enzymes play important roles as negative-feedback regulators in NF-κB signalling, interferon signalling and in p97 (cdc48)-mediated processes although the cellular functions of most OTU enzymes remain to be discovered. Ovarian tumour family DUBs contain a papain-like catalytic core of ~180 amino acids. In addition to their catalytic domain, many OTU members have additional ubiquitin-binding

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Physical Characteristics

Species: human

Source: *E. coli*

Quantity: 50 µg

Concentration: 0.5 mg/ml

Formulation: 50 mM HEPES pH 7.5,
150 mM sodium chloride,
2 mM dithiothreitol, 10% glycerol

Molecular Weight: ~67 kDa

Purity: >98% by InstantBlue™ SDS-PAGE

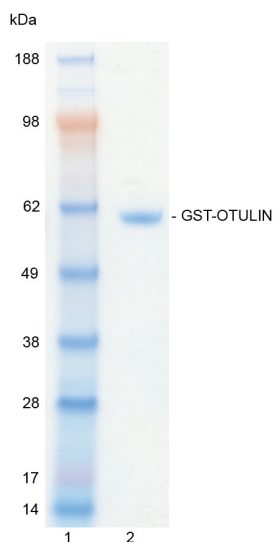
Stability/Storage: 12 months at -70°C;
aliquot as required

Protein Sequence: Please see page 2

Quality Assurance

Purity:

4-12% gradient SDS-PAGE
InstantBlue™ staining
Lane 1: MW markers
Lane 2: 1 µg GST-OTULIN



Protein Identification:

Confirmed by mass spectrometry.

Deubiquitylase Enzyme Assay:

The activity of GST-OTULIN was validated by the monitoring of mono-ubiquitin generation as a result of the enzyme catalysed cleavage of linear di-ubiquitin. Incubation of the substrate in the presence or absence of GST-OTULIN was compared confirming the deubiquitylating activity of GST-OTULIN.



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Lot-specific COA version tracker: v1.0.0

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CERTIFICATE OF ANALYSIS Page 2 of 2

Background

Continued from page 1

domains (UBDs). At least 20 different UBD families have been described, and knowledge of linkage-specific UBDs have provided the means to understand the roles of different ubiquitin linkages in cells (Licchesi *et al.*, 2012).

References:

Balakirev MY, Tcherniuk SO, Jaquinod M and Chroboczek J (2003) Otubains: a new family of cysteine proteases in the ubiquitin pathway. *EMBO Rep* **4**, 517-522.

Komander D, Clague MJ and Urbe S (2009) Breaking the chains: structure and function of the deubiquitinases. *Nat Rev Mol Cell Biol* **10**, 550-563.

Licchesi JD, Mieszczanek J, Mevissen TE, Rutherford TJ, Akutsu M, Virdee S, et al. (2012) An ankyrin-repeat ubiquitin-binding domain determines TRABID's specificity for atypical ubiquitin chains. *Nature Structural & Molecular Biology* **19**, 62-71.

Ota T, Suzuki Y, Nishikawa T, Otsuki T, Sugiyama T, Irie R, et al. (2004) Complete sequencing and characterization of 21,243 full-length human cDNAs. *Nature Genetics* **36**, 40-45.

Reyes-Turcu FE, Ventii KH and Wilkinson KD (2009) Regulation and cellular roles of ubiquitin-specific deubiquitinating enzymes. *Ann Rev Biochem* **78**, 363-397.

Physical Characteristics

Continued from page 1

Protein Sequence:

MSPILGYWKIKGLVQPTRLLLEYLEEKYEEH
LYERDEGDKWRNKKFELGLEFPNLPYY
IDGDVKLTQSMAIRYIADKHNMLGGCP
KERAEISMLEGAVLDIRYGVSR IAYSKD
FETLKVDFLSKLPEMLKMFEDRLCHKTYLNGD
HVTHPDFMLYDALDVVLYMDPMCLDAFP
KLVCFFKKRIEAIPOIDKYLKSSKYIAWPLQG
WQATFGGGDHPKSDENLYFQGGMSRGT
MPQPEAWPGASCAETPAREAAATARDG
GKAAASGQPRPEMOCPAEHEEDMYRAA
DEIEKEKELLIHERGASEPRLSVAPEMDIM
DYCKKEWRGNTQKATCMKMGYEEVVSQKFT
SIRRVRGDNYCALRATLFOAMSQAVGLP
PWLQDPELMLLPEKLI SKYNWIKQWKL
GLKFDGKNEDLVDKIKESLTLRKKWAGLAE
MRTAEARQIACDELFTNEAEEYSLYEAVK
FLMLNRAIELYNDKEKGEVPPFFSVLL
FARDTSDNPGQLLRNHLNQGHTGGLEQVEM
FLLAYAVRHTIQVYRLSKYNTTEEFITVYPTDP
PKDWPVVTLIAEDDRHYNIPVRVCEETS

Tag (**bold text**): N-terminal GST
Protease cleavage site: TEV™ (ENLYFQ▼GS)
OTULIN (regular text): Start **bold italics** (amino acid residues 1-352)
Accession number: NP_612357



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