OTUB2 [GST-tagged]

Deubiquitylating Enzyme

Alternate Names: OTU Domain-Containing Ubal-Binding Protein 2
Otubain 2; OTU2; OTB2

Cat. No.	**64-0012-050**	Quantity:	50 µg
Lot. No.	**1853**	Storage:	-70˚C

FOR RESEARCH USE ONLY

NOT FOR USE IN HUMANS

Background

The deubiquitylating enzymes (DUBs) regulate ubiquitin dependent signaling 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 protein (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 enzyme the cysteine proteases and metalloproteases. OTUB2 is a cysteine protease and member of the Ovarian Tumor (OTU) protein super family. Cloning of the human OTUB2 gene was first described by Balakirev *et al*., (2003). OTUB2 contains Ubiquitin Interaction Motifs (UIMs) and Ubiquitin Associated (UBA) domains, as well as putative nuclear localization signals and a consensus LxxLL motif (Balakirev *et al*., 2003). De-ubiquitylation has emerged as a post translational mechanism for the activation of virus triggered type I interferon (IFN) (2) induction pathways. OTUB2 has been found to negatively regulate virus triggered type I IFN activation of IRF3 and NF-kappaB by mediating the de-ubiquitylation of TRAF3 and TRAF6, two ubiquitin E3 ligases involved in virus-triggered IRF3 and NF-kappaB activation pathways (Li *et al*., 2010).

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Physical Characteristics

Species: human

Source: *E. coli* expression

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: ~54 kDa

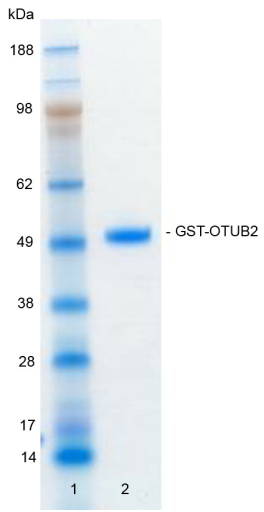
Purity: >93% by InstantBlue™ SDS-PAGE

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-OTUB2



Protein Identification:
Confirmed by mass spectrometry.

Deubiquitylating Enzyme Assay:
The activity of GST-OTUB2 was validated by determining the increase in fluorescence measured as a result of the enzyme catalysed cleavage of the fluorogenic substrate Ubiquitin-Rhodamine110-Glycine generating Ubiquitin and Rhodamine110-Glycine. Incubation of the substrate in the presence or absence of GST-OTUB2 was compared confirming the deubiquitylating activity of GST-OTUB2.

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Lot-specific COA version tracker: v1.0.0

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Background

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Integrative gene tissue array analysis has been used to locate disease relevant proteins to a linkage region associated with familial Amyotrophic Lateral Sclerosis (ALS)/frontotemporal dementia. Using this approach OTUB2 was identified as a disease relevant protein involved in TAU-1 and SOD1 induced motor neuron degeneration found in human sporadic ALS (SALS) (Kudo *et al.*, 2010).

References:

Balakirev MY, Tcherniuk SO, Jaquinod M, Chroboczek J (2003) Otubains: a new
family of cysteine proteases in the ubiquitin pathway. *EMBO Rep* **4**, 517-22.

Komander D, Clague MJ, Urbe S (2009) Breaking the chains: structure and function
of the deubiquitinases. *Nat Rev Mol Cell Biol* **10**, 550-63.

Kudo LC, Parfenova L, Vi N, Lau K, Pomakian J, Valdmanis P, Rouleau GA, Vinters HV, Wiedau-Pazos M, Karsten S (2010) Integrative gene-tissue microarray-based approach for identification of human disease biomarkers: application to amyotrophic lateral sclerosis. *Hum Mol Genet* **19**, 3233 - 3253.

Li S, Zheng H, Mao AP, Zhong B, Li Y, Liu Y, Gao Y, Ran Y, Tien P, Shu HB (2010) Regulation of virus-triggered signaling by OTUB1- and OTUB2-mediated deubiquitination of TRAF3 and TRAF6. *J Biol Chem* **285**, 4291-7.

Physical Characteristics

Continued from page 1

Protein Sequence:

MSPILGYWKIKGLVQPTRLLLEYLEEKY
EEHLYERDEGDKWRNKKFELGLEFPN
LPYYIDGDVKLTQSMAIIRYIADKHNMLG
GCPKERAEISMLEGAVLDIRYGVSRIAY
SKDFETLKVDFLSKLPEMLKMFEDRLCH
KTYLNGDHVTHPDFMLYDALDVVLYM
DPMCLDAFPKLVCFKKRIEAIPQIDKY
LKSSKYIAWPLQGWQATFGGGDHPPKS
DLEVLFQGPLGS***M***SETSFNLISEKCDIL
SILRDHPENRIYRRKIEELSKRFTAIRKT
KGDGNCFYRALGYSYLESLLGKSREIFK
FKERVLQTPNDLLAAGFEEHKFRNFFNA
FYSVVELVEKDGSVSSLLKVFNDQSAS
DHIVQFLRLLTSAFIRNRADFFRH
FIDEEMDIKDFCTHEVEPMATECDHIQI
TALSQALSIALQVEYVDEMDTALNHHVF
PEAATPSVYLLYKTSHYNILYAADKH

Tag (**bold text**): N-terminal GST
Protease cleavage site: PreScission™ (LEVLFQ▼GP)
OTUB2 (regular text): Start ***bold italics*** (amino acid residues 1-234)
Accession number: NP_075601.1

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