

# OTUD3 [GST-tagged]

## Deconjugating enzyme: Deubiquitylase

Alternate Names: OTU domain-containing protein 3, KIAA0459

Cat. No. 64-0035-050  
Lot. No. 30103

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. OTUD3 is a cysteine protease and is a member of the OTU superfamily of proteins (Balakirev *et al.*, 2003). Cloning of the human gene was first described by Seki *et al.* (1999). 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 addi-

Continued on page 2

### 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:** ~72 kDa

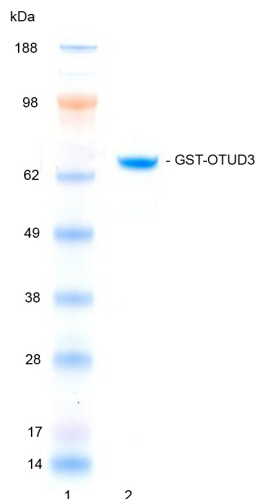
**Purity:** >98% by InstantBlue™ SDS-PAGE

**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-OTUD3



**Protein Identification:**  
Confirmed by mass spectrometry.

**Deubiquitylase Enzyme Assay:**  
The activity of GST-OTUD3 was validated by determining the increase in fluorescence measured as a result of the enzyme catalysed cleavage of the fluorogenic substrate Ubiquitin-Rhodamine 110-Glycine generating Ubiquitin and Rhodamine 110-Glycine. Incubation of the substrate in the presence or absence of GST-OTUD3 was compared confirming the deubiquitylating activity of GST-OTUD3.



www.ubiquigent.com  
Dundee, Scotland, UK

#### ORDERS / SALES SUPPORT

International: +1-617-245-0003  
US Toll-Free: 1-888-4E1E2E3 (1-888-431-3233)  
Email: sales.support@ubiquigent.com

#### UK HQ and TECHNICAL SUPPORT

International: +44 (0) 1382 381147 (9AM-5PM UTC)  
US/Canada: +1-617-245-0020 (9AM-5PM UTC)  
Email: tech.support@ubiquigent.com

Email services@ubiquigent.com for enquiries regarding compound profiling and/or custom assay development services.

© Ubiquigent 2012. Unless otherwise noted, Ubiquigent, Ubiquigent logo and all other trademarks are the property of Ubiquigent, Ltd.

Limited Terms of Use: For research use only. Not for use in humans or for diagnostics. Not for distribution or resale in any form, modification or derivative OR for use in providing services to a third party (e.g. screening or profiling) without the written permission of Ubiquigent, Ltd.

Lot-specific COA version tracker: v1.0.0

# OTUD3 [GST-tagged]

## Deconjugating enzyme: Deubiquitylase

Alternate Names: OTU domain-containing protein 3, KIAA0459

Cat. No. **64-0035-050**  
Lot. No. **30103**

Quantity: 50 µg  
Storage: -70°C



FOR RESEARCH USE ONLY

NOT FOR USE IN HUMANS

CERTIFICATE OF ANALYSIS Page 2 of 2

### Background

Continued from page 1

tional ubiquitin-binding 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.

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

Seki N, Hattori A, Hayashi A, Kozuma S, Sasaki M, Suzuki Y, *et al.* (1999) Cloning and expression profile of mouse and human genes, Rnf11/RNF11, encoding a novel RING-H2 finger protein. *Biochim Biophys Acta* **1489**, 421-427.

### Physical Characteristics

Continued from page 1

#### Protein Sequence:

**MSPILGYWKIKGLVQPTRLLLEYLEEKYEEH**  
**LYERDEGDKWRNKKFELGLEFPNLPYYIDGD**  
**VKLTQSMAIRYIADKHNMLGGCPKERAEISM**  
**LEGAVLDIRYGVSRIRIAYSKDFETLKVDFL**  
**SKLPEMLKMFEDRLCHKTYLNGDHVTHPD**  
**FMLYDALDVVLYMDPMCLDAFPKLVCFK**  
**KRIEAIPOIDKYLKSSKYIAWPLQGWQATFG**  
**GGDHPKSDLEVLFGQPLGSMRSRKQAAKSR**  
**PGSGSRKAEAEERKRDERAARRALAKERRN**  
**RPESGGGGGCEEEFVVSFANQLQALGLKL**  
**REVPDGNCLFRALGDQLEGHRSRNLKHRQET**  
**VDYMIKQREDFEFPVEDDIPFEKHVASLAK**  
**PGTAFAGNDAIIVAFARNHQLNVVIHQNLAPL**  
**WQIRGTEKSSVRELHIAIRYGEHYDSVR**  
**RINDNSEAPAHLQTDFOMLHQDESINKREIK**  
**TKGMDSEDDLREVEDAVQKVCNATGCSDF**  
**NLIVQNLEAENYNIESAIIAVLRMNQGKRN**  
**NAEENLEPSGRVLKQCGPLWEEGGSGARIFGN**  
**QGLNEGRTEENKAQASPSEENKANKNQLAKVT**  
**NKQRREQQWMEKKKRQEERHRHKALESRG**  
**SHRDNRRSEAEANTQVTLVKTFAALNI**

Tag (**bold text**): N-terminal GST  
Protease cleavage site: PreScission™ (LEVLFGQ▼GP)  
OTUD3 (regular text): Start **bold italics** (amino acid residues 1-629)  
Accession number: NP\_056022



www.ubiquigent.com  
Dundee, Scotland, UK

#### ORDERS / SALES SUPPORT

International: +1-617-245-0003  
US Toll-Free: 1-888-4E1E2E3 (1-888-431-3233)  
Email: sales.support@ubiquigent.com

#### UK HQ and TECHNICAL SUPPORT

International: +44 (0) 1382 381147 (9AM-5PM UTC)  
US/Canada: +1-617-245-0020 (9AM-5PM UTC)  
Email: tech.support@ubiquigent.com

Email services@ubiquigent.com for enquiries regarding compound profiling and/or custom assay development services.

© Ubiquigent 2012. Unless otherwise noted, Ubiquigent, Ubiquigent logo and all other trademarks are the property of Ubiquigent, Ltd.

Limited Terms of Use: For research use only. Not for use in humans or for diagnostics. Not for distribution or resale in any form, modification or derivative OR for use in providing services to a third party (e.g. screening or profiling) without the written permission of Ubiquigent, Ltd.

Lot-specific COA version tracker: v1.0.0