# **USP25** [GST-tagged]

Deconjugating enzyme: Deubiquitylase

Alternate Names: Ubiquitin specific protease 25

Cat. No. 64-0023-050 Quantity: 30063 Lot. No. Storage:

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**CERTIFICATE OF ANALYSIS Page 1 of 2** 

Protein Sequence: Please see page 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 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 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. Ubiquitin carboxyl-terminal hydrolase 25 (Ubiquitin Specific Protease 25; USP25) is a member of the cysteine protease enzyme family and cloning of the human gene was first described by Valero et al. (1999). USP25 was originally called USP21 as it was found on chromosome 21, region 21q11-q21. The chromosome 21 trisomy (Down syndrome) is the most frequent human birth defect, and an increase in USP25 gene dosage in Down syndrome patients could seriously disturb the balance between ubiquitylated and deubiquitylated substrates (Valero et al., 1999). Instability and allelic deletions of the 21q11-q21

## **Physical Characteristics**

50 µg

-70°C

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: ~149 kDa

Purity: >74% by InstantBlue™ SDS-PAGE

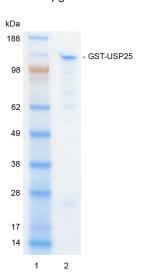
Stability/Storage: 12 months at -70°C;

aliquot as required

## Quality Assurance

## **Purity:**

4-12% gradient SDS-PAGE InstantBlue™ staining lane 1: MW markers lane 2: 1 µg GST-USP25



### **Protein Identification:**

Confirmed by mass spectrometry.

### Deubiquitylase Enzyme Assay:

The activity of GST-USP25 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-USP25 was compared confirming the deubiquitylating activity of GST-USP25.

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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

region of the chromosome have been found in 13 of 34 squamous non-small cell lung carcinomas (Groet et al., 2000). USP25 is a target for SUMOylation, being more efficient with SUMO2/3. USP25 SUMOylation impairs binding to and hydrolysis of ubiquitin chains (Meulmeester et al., 2008). USP25 is deSUMOylated by SENP1 (Mohideen and Lima, 2008).

#### References:

Groet J, Ives JH, Jones TA, Danton M, Flomen RH, Sheer D, Hrascan R, Pavelic K, Nizetic D (2000) Narrowing of the region of allelic loss in 21q11-21 in squamous non-small cell lung carcinoma and cloning of a novel ubiquitin-specific protease gene from the deleted seament. *Genes Chromosomes Cancer* 27, 153-161.

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

Meulmeester E, Kunze M, Hsiao HH, Urlaub H, Melchior F (2008) Mechanism and consequences for paralog-specific sumoylation of ubiquitin-specific protease 25. *Mol Cell* **30**, 610-619.

Mohideen F, Lima CD (2008) SUMO takes control of a ubiquitinspecific protease. *Mol Cell 30*, 539-540.

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

Valero R, Marfany G, Gonzalez-Angulo O, Gonzalez-Gonzalez G, Puelles L, Gonzalez-Duarte R (1999) USP25, a novel gene encoding a deubiquitinating enzyme, is located in the gene-poor region 21q11.2. *Genomics* **62**, 395-405.

## **Physical Characteristics**

Continued from page 1

#### **Protein Sequence:**

MSPILGYWKIKGLVQPTRLLLEYLEEKY EEHLYERDEGDKWRNKKFELGLEFPN LPYYIDGDVKLTQSMAIIRYIADKHNML GGCPKERAEISMLEGAVLDIRYGVSRIAY SKDFETLKVDFLSKLPEMLKMFEDRLCHK TYLNGDHVTHPDFMLYDALDVVLYMDPM CLDAFPKLVCFKKRIEAIPQIDKYLKSSKY  $\textbf{IAWPLQGWQATFGGGDHPPKSD} \underline{\texttt{LEVLFQG}}$ PLGSMTVEQNVLQQSAAQKHQQTFLNQL REITGINDTQILQQALKDSNGNLELAVAFL TAKNAKTPQQEETTYYQTALPGNDRYISVG SQADTNVIDLTGDDKDDLQRAIALSLAES NRAFRETGITDEEQAISRVLEASIAENKA CLKRTPTEVWRDSRNPYDRKRQDKAPVGLKN VGNTCWFSAVIQSLFNLLEFRRLVLNYK PPSNAODLPRNOKEHRNLPFMRELRYLFALL VGTKRKYVDPSRAVEILKDAFKSNDSQQQD VSEFTHKLLDWLEDAFQMKAEEETDEEKP KNPMVELFYGRFLAVGVLEGKKFENTEMF GQYPLQVNGFKDLHECLEAAMIEGEIESLH SENSGKSGQEHWFTELPPVLTFELSRFEFN OALGRPEKIHNKLEFPOVLYLDRYMHRNRE ITRIKREEIKRLKDYLTVLQQRLERYLSYGS GPKRFPLVDVLQYALEFASSKPVCTSPVDDI DASSPPSGSIPSQTLPSTTEQQGALSSELPST SPSSVAAISSRSVIHKPFTQSRIPPDLPMHPA PRHITEEELSVLESCLHRWRTEIENDTRDLOE SISRIHRTIELMYSDKSMIQVPYRLHAV LVHEGOANAGHYWAYIFDHRESRWMKYNDIA VTKSSWEELVRDSFGGYRNASAYCLMYIND KAQFLIQEEFNKETGQPLVGIETLPPDLRD FVEEDNQRFEKELEEWDAQLAQKALQEKL LASQKLRESETSVTTAQAAGDPEYLEQPSRS DFSKHLKEETIQIITKASHEHEDKSPETVLQ SAIKLEYARLVKLAOEDTPPETDYRLHHVVVY FIQNQAPKKIIEKTLLEQFGDRNLSFDERCHN IMKVAQAKLEMIKPEEVNLEEYEEWHQDYRK FRETTMYLIIGLENFORESYIDSLLFLICAY QNNKELLSKGLYRGHDEELISHYRRECLLKL NEQAAELFESGEDREVNNGLIIMNEFIVPFL PLLLVDEMEEKDILAVEDMRNRWCSYLGOE MEPHLQEKLTDFLPKLLDCSMEIKSFHEP PKLPSYSTHELCERFARIMLSLSRTPADGR

Tag (**bold text**): N-terminal GST
Protease cleavage site: PreScission™ (<u>LEVLFQ▼GP</u>)
USP25 (regular text): Start **bold italics** (amino acid residues 1-1055)

Accession number: NP\_037528



Dundee, Scotland, UK

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