

USP25 [GST-tagged]

Deconjugating enzyme: Deubiquitylase

Alternate Names: Ubiquitin specific protease 25

Cat. No. **64-0023-050**
Lot. No. **30063**

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 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

Continued on page 2

Physical Characteristics

Species: human

Protein Sequence: Please see page 2

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

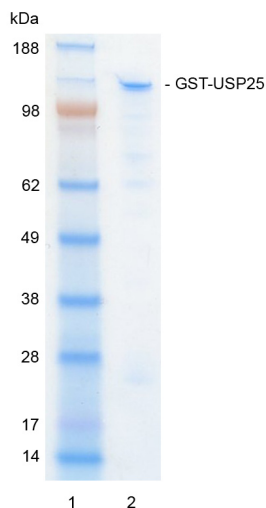
Molecular Weight: ~149 kDa

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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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 segment. *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 paralogue-specific sumoylation of ubiquitin-specific protease 25. *Mol Cell* **30**, 610-619.

Mohideen F, Lima CD (2008) SUMO takes control of a ubiquitin-specific 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:

M S P I L G Y W K I K G L V Q P T R L L L E Y L E E K Y
E E H L Y E R D E G D K W R N K K F E L G L E F P N
L P Y Y I D G D V K L T Q S M A I I R Y I A D K H N M L
G G C P K E R A E I S M L E G A V L D I R Y G V S R I A Y
S K D F E T L K V D F L S K L P E M L K M F E D R L C H K
T Y L N G D H V T H P D F M L Y D A L D V V L Y M D P M
C L D A F P K L V C F K K R I E A I P Q I D K Y L K S S K Y
I A W P L Q G W Q A T F G G G D H P P K S D L E V L F Q G
P L G S M T V E Q N V L Q Q S A A Q K H Q Q T F L N Q L
R E I T G I N D T Q I L Q Q A L K D S N G N L E L A V A F L
T A K N A K T P Q Q E E T T Y Y Q T A L P G N D R Y I S V G
S Q A D T N V I D L T G D D K D D L Q R A I A L S L A E S
N R A F R E T G I T D E E Q A I S R V L E A S I A E N K A
C L K R T P T E V W R D S R N P Y D R K R Q D K A P V G L K N
V G N T C W F S A V I Q S L F N L L E F R R L V L N Y K
P P S N A Q D L P R N Q K E H R N L P F M R E L R Y L F A L L
V G T K R K Y V D P S R A V E I L K D A F K S N D S Q Q Q D
V S E F T H K L L D W L E D A F Q M K A E E E T D E E K P
K N P M V E L F Y G R F L A V G V L E G K K F E N T E M F
G Q Y P L Q V N G F K D L H E C L E A A M I E G E I E S L H
S E N S G K S G Q E H W F T E L P P V L T F E L S R F E F N
Q A L G R P E K I H N K L E F P Q V L Y L D R Y M H R N R E
I T R I K R E E I K R L K D Y L T V L Q Q R L E R Y L S Y G S
G P K R F P L V D V L Q Y A L E F A S S K P V C T S P V D D I
D A S S P P S G S I P S Q T L P S T T E Q Q G A L S S E L P S T
S P S S V A A I S S R S V I H K P F T Q S R I P P D L P M H P A
P R H I T E E E L S V L E S C L H R W R T E I E N D T R D L Q E
S I S R I H R T I E L M Y S D K S M I Q V P Y R L H A V
L V H E G Q A N A G H Y W A Y I F D H R E S R W M K Y N D I A
V T K S S W E E L V R D S F G G Y R N A S A Y C L M Y I N D
K A Q F L I Q E E F N K E T G Q P L V G I E T L P P D L R D
F V E E D N Q R F E K E L E E W D A Q L A Q K A L Q E K L
L A S Q K L R E S E T S V T T A Q A A G D P E Y L E Q P S R S
D F S K H L K E E T I Q I I T K A S H E H E D K S P E T V L Q
S A I K L E Y A R L V K L A Q E D T P P E T D Y R L H H V V V Y
F I Q N Q A P K K I I E K T L L E Q F G D R N L S F D E R C H N
I M K V A Q A K L E M I K P E E V N L E E Y E E W H Q D Y R K
F R E T T M Y L I I G L E N F Q R E S Y I D S L L F L I C A Y
Q N N K E L L S K G L Y R G H D E E L I S H Y R R E C L L K L
N E Q A A E L F E S G E D R E V N N G L I M N E F I V P F L
P L L L V D E M E E K D I L A V E D M R N R W C S Y L G Q E
M E P H L Q E K L T D F L P K L L D C S M E I K S F H E P
P K L P S Y S T H E L C E R F A R I M L S L S R T P A D G R

Tag (**bold text**): N-terminal GST
Protease cleavage site: PreScission™ (**LEVLQ▼GP**)
USP25 (regular text): Start **bold italics** (amino acid residues 1-1055)
Accession number: NP_037528



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