USP7 [6His-tagged]

Deubiquitylating Enzyme

Alternate Names: Herpesvirus-Associated Ubiquitin-Specific Protease, HAUSP VMW110associated protein

64-0003-050 Quantity: Cat. No. Lot. No. 1736 Storage:

FOR RESEARCH USE ONLY NOT FOR USE IN HUMANS



CERTIFICATE OF ANALYSIS Page 1 of 2

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 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 specific processing protease 7 (USP-7) is a member of the cysteine protease enzyme family and cloning of the gene in humans was first described by Everett et al. (1997). Overexpression of p53 and USP7 stabilizes p53 through the removal of ubiquitin moieties from polyubiguitylated p53 (Kon et al., 2010). Inhibition of USP7 expression results in the accumulation and stabilisation of p53 protein. However, USP7 has been shown to stabilise mdm2 a ubiquitin ligase that promotes the degradation of p53 (Holowaty and Frappier. 2004; Krishna and Grishin. 2004). Thus USP7 appears to play multiple roles in regulating the p53/mdm2 pathway and maintaining steady-state levels of p53 in the cell. Due to the role USP7 has been shown to play in the regulation of p53 its importance as a thera-

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Physical Characteristics

50 µg

-70°C

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: ~130 kDa

Purity: >80% by InstantBlue™ SDS-PAGE

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

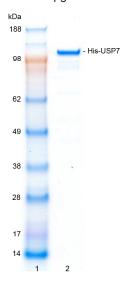
aliquot as required

Protein Sequence: Please see page 2

Quality Assurance

Purity:

4-12% gradient SDS-PAGE InstantBlue™ staining Lane 1: MW markers Lane 2: 1 µg His-USP7



Protein Identification:

Confirmed by mass spectrometry.

Deubiquitylating Enzyme Assay:

The activity of His-USP7 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 His-USP7 was compared confirming the deubiquitylating activity of His-USP7.



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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

peutic target for treating hematopoietic tumours has been highlighted (Cheon and Baek. 2006).

References:

Cheon KW, Baek KH (2006) HAUSP as a therapeutic target for hematopoietic tumors (review). *Int J Oncol* **28**, 1209-15.

Everett RD, Meredith M, Orr A, Cross A, Kathoria M, Parkinson J (1997) A novel ubiquitin-specific protease is dynamically associated with the PML nuclear domain and binds to a herpesvirus regulatory protein. *EMBO J* 16, 1519-30.

Holowaty MN, Frappier L (2004) HAUSP/USP7 as an Epstein-Barr virus target. *Biochem Soc Trans* **32**, 731-2.

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

Kon N, Kobayashi Y, Li M, Brooks CL, Ludwig T, Gu W (2010) Inactivation of HAUSP in vivo modulates p53 function. Oncogene 29, 1270-9.

Krishna SS, Grishin NV (2004) The finger domain of the human deubiquitinating enzyme HAUSP is a zinc ribbon. *Cell Cycle* **3**, 1046-9.

Physical Characteristics

Continued from page 1

Protein Sequence:

MGSSHHHHHHSSGLEVLFQGPGSMN HQQQQQQKAGEQQLSEPEDMEMEAGDTD DPPRITQNPVINGNVALSDGHNTAEEDMED DTSWRSEATFQFTVERFSRLSESVLSPP CFVRNLPWKIMVMPRFYPDRPHQKSVGF FLQCNAESDSTSWSCHAQAVLKIINYRD DEKSFSRRISHLFFHKENDWGFSNFMAWSEVT DPEKGFIDDDKVTFEVFVQADAPHGVAWDSK KHTGYVGLKNQGATCYMNSLLQTLFFTNQL RKAVYMMPTEGDDSSKSVPLALQRVFYELQHS DKPVGTKKLTKSFGWETLDSFMQHDVQEL CRVLLDNVENKMKGTCVEGTIPKLFRGKM VSYIQCKEVDYRSDRREDYYDIQLSIKGK KNIFESFVDYVAVEQLDGDNKYDAGEHGLQE AEKGVKFLTLPPVLHLQLMRFMYDPQTDQNI KINDRFEFPEQLPLDEFLQKTDPKDPANYIL HAVLVHSGDNHGGHYVVYLNPKGDGKW CKFDDDVVSRCTKEEAIEHNYGGHDDDLS VRHCTNAYMLVYIRESKLSEVLQAVTDHDI POOLVERLOEEKRIEAOKRKEROEAHLYMOVO IVAEDQFCGHQGNDMYDEEKVKYTVFKVLKNS SLAEFVQSLSQTMGFPQDQIRLWPMQARSNGT KRPAMLDNEADGNKTMIELSDNENPWTIFLET VDPELAASGATLPKFDKDHDVMLFLKMYDPK TRSLNYCGHIYTPISCKIRDLLPVMCDRAG FIQDTSLILYEEVKPNLTERIQDYDVSLDKA LDELMDGDIIVFQKDDPENDNSELPTAKEY FRDLYHRVDVIFCDKTIPNDPGFVVTLSNRM NYFQVAKTVAQRLNTDPMLLQFFKSQGYRDG PGNPLRHNYEGTLRDLLQFFKPRQPKKLYY QQLKMKITDFENRRSFKCIWLNSQFREEEIT LYPDKHGCVRDLLEECKKAVELGEKASGKL RLLEIVSYKIIGVHQEDELLECLSPATSRT FRIEEIPLDQVDIDKENEMLVTVAHFHKEV FGTFGIPFLLRIHQGEHFREVMKRIQSLL DIQEKEFEKFKFAIVMTGRHQYINEDEYEVN LKDFEPQPGNMSHPRPWLGLDHFNKAPKRSRY TYLEKAIKIHN*

Tag (bold text): N-terminal His

Protease cleavage site: PreScission™ (<u>LEVLFQ▼GP</u>)

USP7 (regular text): Start *bold italics* (amino acid residues
1-1102)

Accession number: CAA96580



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