

# BAP1 [GST-tagged]

## Deconjugating enzyme: Deubiquitylase

**Alternate Names:** BRCA1 Associated Protein 1, BRCA1 Associated Protein-1 (Ubiquitin Carboxy-Terminal Hydrolase), Cerebral Protein 6, Ubiquitin Carboxy-Terminal Hydrolase 2, BRCA1-Associated Protein 1, Cerebral Protein-13

**Cat. No.** 64-0052-050  
**Lot. No.** 30421

**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 metallo-proteases. BRCA1 Associated Protein 1 (BAP1) is a cysteine protease and member of the UCH family of ubiquitin C-terminal hydrolases. Cloning of the human BAP1 gene was first described by Jensen *et al.* (1998). The nuclear DUB BAP1 is a tumour suppressor deleted and mutated in an increasing number of cancers of diverse origins thereby making BAP1 the most frequently and widely mutated DUB-encoding gene in cancer (Daou *et al.* 2015). BAP1 contains binding domains for BRCA1 (Breast cancer type 1) and BARD1 (BRCA1-associated RING domain protein 1), which form a tumour suppressor heterodimeric complex, and HCFC1 (Host cell factor 1), which interacts with histone-modifying complexes

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### Physical Characteristics

**Species:** human

**Protein Sequence:** Please see page 2

**Source:** *E. coli*

**Quantity:** 50 µg

**Concentration:** 0.18 mg/ml

**Formulation:** 50 mM Tris/HCl pH 7.5,  
0.1 mM EGTA, 150 mM NaCl,  
0.1% β-Mercaptoethanol,  
270 mM Sucrose, 0.03% Brij-35

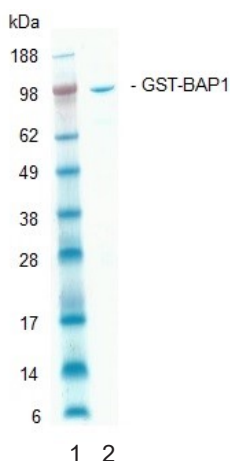
**Molecular Weight:** ~107 kDa

**Purity:** >80% 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-BAP1



**Protein Identification:**

Confirmed by mass spectrometry.

**Deubiquitylase Enzyme Assay:**

The activity of GST-BAP1 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-BAP1 was compared confirming the deubiquitylating activity of GST-BAP1.



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Lot-specific COA version tracker: v1.0.0

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### Background

Continued from page 1

during cell division. BAP1 also interacts with ASXL1 (Putative Polycomb group protein) to form the Polycomb group repressive de-ubiquitylase complex (PR-DUB), which is involved in stem cell pluripotency and other developmental processes (Harbour *et al.* 2010). More recently, BAP1 has been identified as a DUB for Krüppel-like zinc-finger transcription factor 5 (KLF5), a transcription factor that is highly expressed in certain types of breast cancers. KLF5 has been identified as an unstable protein that is ubiquitinated by WWP1 (NEDD4-like E3 ligase), SCF<sup>FBW7</sup> (a SKP1-cullin-1-F-box complex that contains FBW7 as the F-box protein) and Smurf2 (SMAD Specific E3 ligase 2) and degraded. BAP1 promotes breast cancer cell proliferation and migration *in vitro* and tumour growth and lung metastasis *in vivo*. The results from this study suggest that BAP1 and KLF5 are potential therapeutic targets for breast cancer (Qin *et al.* 2015).

#### References:

Daou S, *et al.* (2015) The BAP1/ASXL2 histone H2A deubiquitinase complex regulates cell proliferation and is disrupted in cancer. *The Journal of biological chemistry* **290**:28643-28663.

Harbour JW, *et al.* (2010) Frequent mutation of BAP1 in metastasizing uveal melanomas. *Science* **330**:1410-1413.

Jensen DE, *et al.* (1998) BAP1: a novel ubiquitin hydrolase which binds to the BRCA1 RING finger and enhances BRCA1-mediated cell growth suppression. *Oncogene* **16**:1097-1112.

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

Qin J, *et al.* (2015) BAP1 promotes breast cancer cell proliferation and metastasis by deubiquitinating KLF5. *Nature communications* **6**:8471.

Reyes-Turcu FE *et al.* (2009) Regulation and cellular roles of ubiquitin-specific deubiquitinating enzymes. *Annual review of biochemistry* **78**:363-397.

### Physical Characteristics

Continued from page 1

#### Protein Sequence:

**MSPILGYWKIKGLVQPTRLLLEYLEEKYEEH**  
**LYERDEGEDKWRNKKFELGLEFPNLPYY**  
**IDGDVKLTQSMAIIRYIADKHNMLGGCPKER**  
**AEISMLEGAVLDIRYGVSRAYSKDFETLKVD**  
**FLSKLPEMLKMFEDRLCHKTYLNGDHVTHP**  
**DFMLYDALDVVLYMDPMCLDAFPKLVCFK**  
**KRIEAI PQIDKYLKSSKYIAWPLQGWQATF**  
**GGGDHPKSDLEVL FQG PLGSMNKGWLELES**  
**DPGLFTLLVEDFGVKGQVEEIIYDLQSKCQGPVY**  
**GFIFLFWIEERRSRKRVSTLVDDTSVIDDDIVN**  
**NMFFAHQLIPNSCATHALLSVLLNCSSVDLGP**  
**SRMKDFTKGFSPESKGYAIGNAPELAKAHNSHAR**  
**PEPRHLPEKQNGLSAVRTMEAFHFVSVVPI TGRL**  
**FELDGLKVYPIDHGPWGEDEEWTDKARRVIMERI**  
**GLATAGEPYHDIRFNLMAVVPDRRIKYEARLHV**  
**KVNRQTVLEALQQLIRVTQPELIQTHKSQESQLP**  
**EESKSASNKSPLVLEANRAPAASEGNDGAEAA**  
**AGSCAQAPSHSPPNPKPLVVKPPGSSSLNGVHPNP**  
**TPIVQRLPAFLDNHNYAKSPMQEEDLAAGVGRS**  
**RVPVPPPPQYSDDEDDYEDDEEDDVQNTNSALRY**  
**KGKGTGKPGALSGSADGQLSVLQPNNTINVLAEKL**  
**KESQKDLISPLSIKTSSGAGSPAVAVPTHSQPSP**  
**TPSNESDTASEIGSAFNSPLRSPIRSANPTRPS**  
**SPVTSHISKVLFGEEDSLLRVDCIRYNRAVRDLG**  
**PVISTGLLHLAEDGVLSPALTEGGKGS SPSIRP**  
**IQGSQGS S S PVEKEVVEATDSREKTGMVRPGEPL**  
**SGEKYSPEKELLALLKCVEAEIANYEACLKEEVEK**  
**RKKFKIDDQRRTNHYDEFICTFISMLAQEGMLAN**  
**LVEQNI SVRRRQGVSI GR LHKQRK PDRRKR SRPY**  
**KAKRQ**

Tag (**bold text**): N-terminal GST

Protease cleavage site: PreScission™ (LEVL FQ ▼ GP)

BAP1 (regular text): Start **bold italics** (amino acid residues

1-729) Accession number: AAH01596



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