FOR RESEARCH USE ONLY!

EZCut™ SUMO Protease 1 (His-tagged), Yeast Recombinant

rev.05/17

Store at -80°C.

Cat. No.: 7868-500	**500 U**
Cat. No.: 7868-2500	**2500 U**
Cat. No.: 7868-10000	**10000 U**

ALTERNATE NAMES: Ulp1 peptidase, SUMO Protease

SOURCE: *E. coli*

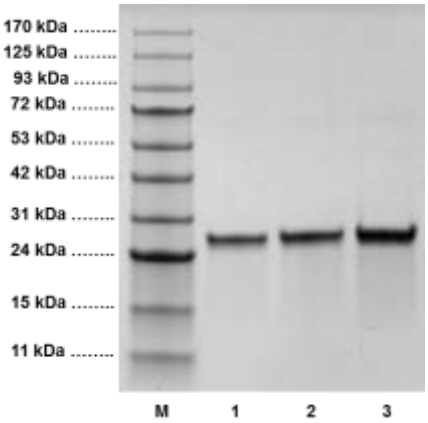
FORM: Liquid

FORMULATION: 0.1 mg/ml (100 U/µl) solution in 50 mM Tris-HCl, 100 mM NaCl, 5 mM DTT and 20% glycerol pH 8.0

PURITY: ≥ 90% by SDS-PAGE

MOL. WT.: 28.7 kDa (403-621 aa + N-terminal Poly-His tag).

STORAGE CONDITIONS: Store at -80°C. Stable for at least 1 year as supplied. It may be further diluted to 0.01-0.05 mg/ml with 50 mM Tris-HCl, 100 mM NaCl, 5 mM DTT and 20% glycerol pH 8.0 and stored at –20°C in aliquots. Avoid repeated freezing and thawing cycles.



SDS-PAGE (4-20%) of EZCut™ His-SUMO Protease 1:
M: Protein Marker
1: His-SUMO 1 (10 µg)
2: His-SUMO 1 (15 µg)
3: His-SUMO 1 (20 µg)

BACKGROUND: SUMO (**S**mall **U**biquitin-like **MO**difiers) Protease 1 (Ulp1, Ubl-specific protease 1 from *Saccharomyces cerevisiae*) is a highly active cysteine protease. It is highly specific as it recognizes the tertiary structure of the ubiquitin-like (UBL) protein, SUMO (Smt3), rather than its amino acid sequence. SUMO fusion tag, as an N-terminal fusion partner, has been shown to enhance functional protein production in prokaryotic and eukaryotic expression systems with significantly improved protein stability and solubility. BioVision's Recombinant EZCut™ SUMO Protease 1 containing an N-terminal His-tag is expressed in *E. coli* and purified by proprietary chromatographic techniques. The EZCut™ SUMO Protease 1 can be used to cleave SUMO protein tag from recombinant SUMO-fusion proteins. The optimal temperature for cleavage is 30°C; however, the enzyme is active over wide ranges of temperature and pH. After the completion of the cleavage reaction, the protease can be easily removed from the reaction by affinity chromatography using the Ni chelating resin (**Cat. # 6562-1, 10, 100, 500**).

SPECIFIC ACTIVITY: The EZCut™ SUMO Protease 1 (His-tagged) has an activity of ≥ 1 X 10^6 units/mg.

UNIT DEFINITION: One unit is defined as the amount of EZCut™ SUMO Protease 1 required to cleave >90% of 5 µg a control protein substrate (**SUMO-GFP**) in 1 h at 37°C.

Note: The EZCut™ SUMO Protease 1 (His-tagged) has also been tested with respect to its ability to cleave a fluorogenic substrate (SUMO-AMC, **Cat. # 6412-50**). The cleavage reactions was carried out by using different amounts of EZCut™ SUMO Protease 1 in 50 mM Tris-HCl, 100 mM NaCl, 5 mM DTT, pH 8.0 buffer containing 0.5 µM of the fluorogenic substrate at 37°C.

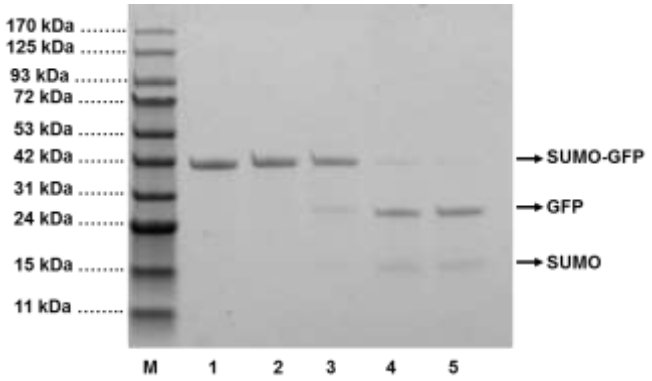
APPLICATIONS: Recombinant EZCut™ SUMO Protease 1 can be used to cleave SUMO-protein tags from recombinant SUMO-fusion proteins resulting in the target protein with the native protein sequence.

CLEAVAGE PROTOCOL: In order to find the optimum cleavage conditions for a target fusion protein, it is recommended to run preliminary cleavage reactions at a small scale. The target SUMO-fusion protein (~0.5-1 mg/ml) should be purified to homogeneity. Optimum temperature for the cleavage reaction is 30°C. However, the amount of SUMO protease 1 should be optimized if the cleavage reaction is being carried out at different temperature (4, 22 or 37°C) or pH (7-9).

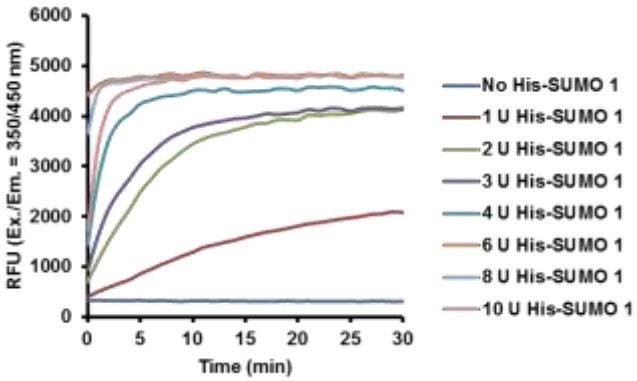
1. To the target protein (in cleavage buffer, 50 mM Tris, buffer, 0.1 M NaCl, pH 8.0 containing 10 mM DTT, pH 8) add different amounts of 1-10 ng (0.1-10 U) of EZCut™ SUMO protease 1.

2. Incubate at 30/37°C for 1 h. Analyze by SDS-PAGE.

3. Once optimum cleavage conditions are obtained, scale up the reaction to cleave the entire amount of the target fusion protein. After the cleavage reaction, remove EZCut™ SUMO Protease 1 by passing the entire mixture through a Ni chelating resin (**Cat. # 6562-1,10,100,500**). Collect the flow through and wash the resin with cleavage buffer. Combine the washes with the flow through to obtain cleaved protein product.



Cleavage of a SUMO-GFP fusion protein (5 µg) by different amounts (0.001-10 U) of Recombinant EZCut™ His-SUMO Protease 1 for 1 h at 37°C:
M: Protein Marker
1: SUMO-GFP only, 2-6: Cleavage reaction of SUMO-GFP with 0.001, 0.01, 0.1, 1 and 10 Units of Recombinant His-SUMO Protease 1 respectively



Cleavage reactions of a fluorogenic substrate (SUMO-AMC) in the presence of different amounts of Recombinant EZCut™ His-SUMO Protease 1

RELATED PRODUCTS:

- EZCut™ SUMO Protease 1 (GST-tagged), Yeast Recombinant (**Cat. No. 7869-100,500**)
- SUMO1, human recombinant (**Cat. No. 4941-100,1000**)
- SUMO2, human recombinant (**Cat. No. 4942-100,1000**)
- SUMO3, human recombinant (**Cat. No. 4943-100,1000**)
- Human recombinant UBE2H (UbcH2), HIS6SUMO **(Cat. No. 6441-3)**
- Recombinant hSUMO1-AMC (**Cat. No. 6412-50**)
- Recombinant hSUMO2-AMC **(Cat. No. 6413-50)**
- Recombinant hSUMO2-Rhodamine (**Cat. No. 6414-50**)
- EZCut™ TEV Protease, Recombinant (**Cat. No. 7847-1000,10000**)
- TurboTEV Protease, Recombinant (**Cat. No. 9205-1**)
- TEV Protease Activity Assay Kit (Fluorometric) **(Cat. No. K842-100)**
- TEV Protease Inhibitor Screening Kit (Fluorometric) (**Cat. No. K843-100**)
- Active HIV1 Protease Recombinant (GST-tagged) (Cat. No. 7849-20,100)
- HIV-1 Protease Activity Assay Kit (Fluorometric) (**Cat. No. K825-100**)
- HIV-1 Protease Inhibitor Screening Kit (Fluorometric) (**Cat. No. K826-100**)
- Active HIV2 Protease Recombinant (GST-tagged) (**Cat. No. 7851-20,100**)
- HIV-2 Protease Activity Assay Kit (Fluorometric) (**Cat. No. K845-100**)
- Glutathione Sepharose (**Cat. No. 6555-1,10,50**)
- Anacardic Acid **(1849)**
- Ginkgolic Acid C15:1 **(9523)**

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