

EZCut™ SUMO Protease 1 Agarose Beads

CATALOG #: M1028-1 1 ml
 M1028-5 5 ml
 M1028-25 25 ml

LIGAND DENSITY: 50 µg/ml of the resin.

FORMULATION: Provided as 50% slurry in 100% glycerol.

STORAGE CONDITIONS: Store at -20°C

SHELF LIFE: Stable for 6 months when properly stored.

DESCRIPTION: SUMO (**S**mall **U**biquitin-like **M**odifiers) 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, 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 EZCut™ SUMO Protease 1 Agarose is prepared by covalent coupling of SUMO Protease 1 to activated 4% cross-linked agarose beads. EZCut™ SUMO Protease 1 Agarose Beads are designed for efficient cleavage of recombinant fusion proteins, containing a SUMO tag, circumventing the need for additional chromatographic step(s) to remove the protease after cleavage reaction is completed. The optimal temperature for cleavage is 30°C; however, the enzyme is active over wide ranges of temperature and pH. These beads can be regenerated for repeated use and the reusability depending on the usage and handling of the product.

APPLICATIONS: Efficient and convenient cleavage of SUMO tag containing recombinant fusion proteins.

ACTIVITY: >30 U/ml. (25 µl of beads (or 50 µl of the 50% slurry) is sufficient to cleave >80% of 100 µg of control SUMO-GFP protein at room temperature in 24 h.)

GENERAL 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. Successful cleavage with SUMO Protease 1 is dependent upon proper folding of the fusion protein that enables access of the SUMO tag by the enzyme. Once optimum cleavage conditions are obtained, the reaction can be scaled up to cleave the entire amount of the target fusion

protein. The target fusion protein should be purified to homogeneity and dialyzed against 50 mM Tris buffer, 0.1 M NaCl, 10 mM DTT, pH 8.0 before setting up the cleavage reaction.

CLEAVAGE PROTOCOL:

1. Re-suspend the beads by gentle swirling. Do not Vortex.
2. Aliquot 50 µl of the suspended slurry and add to 100 µg of the fusion protein (0.25-1 mg/ml) in an Eppendorf tube.
3. Mix gently by inverting the tube (do not vortex) and gently shake on a rotary shaker at room temperature. At regular time intervals spin down the tube to aliquot a test sample and freeze it immediately. At the end of the reaction, analyze the samples by SDS-PAGE.
4. After the fusion protein is completely cleaved, spin down the reaction mixture for 2-3 min at 5000 rpm. Remove the supernatant and wash the beads with 0.1 ml of 50 mM Tris buffer, pH 8. Further chromatography may be necessary to remove the cleaved fragments from the target protein.

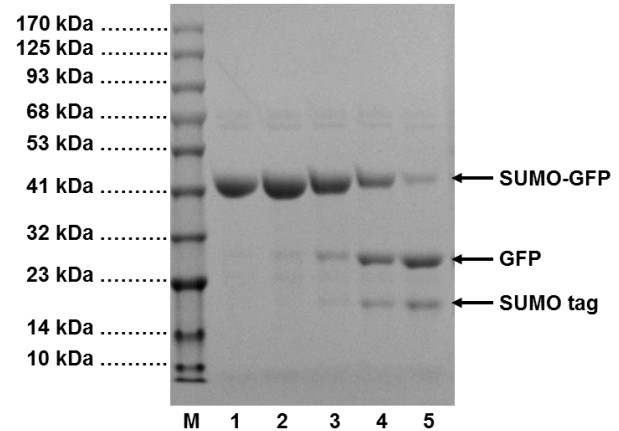


Figure: Analysis of the cleavage activity on SUMO-GFP control protein by EZCut™ SUMO Protease 1 Agarose Beads on 4-20% SDS-PAGE after 0 h (1), 2 h (2), 4 h (3), 6 h (4) and 24 h (5) of reaction at RT (M: Protein Marker).

RELATED PRODUCTS:

- EZCut™ SUMO Protease 1 (GST-tagged), Yeast Recombinant (Cat No. 7869-100, -500)
- EZCut™ SUMO Protease 1 (His-tagged), Yeast Recombinant (Cat No. 7868-500, -2500, -10000)
- SUMO1, human recombinant (Cat No. 4941-100, -1000)
- Urokinase Sepharose Beads (Cat No. 7927-1, -5, -25)
- Hi-Bind Ni QR Agarose Beads (Cat No. 6562-1, -10, -100, -500)
- Thrombin Sepharose Beads (Cat No. 7925-1, -5, -25)

FOR RESEARCH USE ONLY! Not to be used in humans.