

**Datasheet for 200-301-428****SUMO Antibody****Overview**

|                      |                                                     |
|----------------------|-----------------------------------------------------|
| <b>Description:</b>  | Anti-SUMO (MOUSE) Monoclonal Antibody - 200-301-428 |
| <b>Item No.:</b>     | 200-301-428                                         |
| <b>Size:</b>         | 100 µg                                              |
| <b>Applications:</b> | ELISA, WB, Biochemical Assay, Functional Assay, IF  |
| <b>Reactivity:</b>   | Broad                                               |
| <b>Host Species:</b> | Mouse                                               |

**Product Details****Background:**

Covalent modification of cellular proteins by the ubiquitin-like modifier SUMO (small ubiquitin-like modifier) regulates various cellular processes, such as nuclear transport, signal transduction, stress responses and cell cycle progression. But, in contrast to ubiquitination, sumoylation does not tag proteins for degradation by the 26S proteasome, but rather seems to enhance stability or modulate their subcellular compartmentalization. Ubiquitin-like proteins fall into two classes: the first class, ubiquitin-like modifiers (UBLs) function as modifiers in a manner analogous to that of ubiquitin. Examples of UBLs are SUMO, Rub1 (also called Nedd8), Apg8 and Apg12. Proteins of the second class include parkin, RAD23 and DSK2, are designated ubiquitin-domain proteins (UDPs). These proteins contain domains that are related to ubiquitin but are otherwise unrelated to each other. In contrast to UBLs, UDPs are not conjugated to other proteins. Once covalently attached to cellular targets, SUMO regulates protein:protein and protein:DNA interactions, as well as localization and stability of the target protein. Sumoylation occurs in most eukaryotic systems, and SUMO is highly conserved from yeast to humans. Where invertebrates have only a single SUMO gene termed SMT3, three members of the SUMO family have been identified in vertebrates: SUMO-1 and the close homologues SUMO-2 and SUMO-3. SUMO has been called SMT3 (yeast), sentrin, PIC1, GMP1 and UBL1. SUMO has been shown to bind and regulate mammalian SP-RINGS (such as Mdm2, PIAS and PML), RanGAP1, RanBP2, p53, p73, HIPK2, TEL, c-Jun, Fas, Daxx, TNFR1, Topo-I, Topo-II, WRN, Sp100, IκB-α, Androgen receptor (AR), GLUT1/4, Drosophila Ttk69, Dorsal, CaMK, yeast Septins, and viral CMV-IE1/2, EBV-BZLF1, HPV/BPV-E1. These bindings implicate SUMO in the stabilization of the target proteins and/or their localization to subcellular complexes. SUMO has an apparent molecular weight of ~12kDa and human SUMO-1 (a 101 amino acid polypeptide) shares 50% sequence identity with SUMO-2 and SUMO-3 and with yeast SMT3. SUMO and ubiquitin only show about 18% homology, but both possess a common three-dimensional structure characterized by a tightly packed globular fold with β-sheets wrapped around an α-helix.

|                      |                                                                               |
|----------------------|-------------------------------------------------------------------------------|
| <b>Synonyms:</b>     | mouse anti-SUMO Antibody, Ubiquitin-like protein SMT3 antibody, SMT3 antibody |
| <b>Host Species:</b> | Mouse                                                                         |
| <b>Clonality:</b>    | Monoclonal                                                                    |
| <b>Clone ID:</b>     | 4F2.F5.G2                                                                     |
| <b>Format:</b>       | IgG1                                                                          |

## Target Details

|                            |                                                                                                                                                                                                                        |
|----------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Gene Name:</b>          | SMT3                                                                                                                                                                                                                   |
| <b>Reactivity:</b>         | Broad                                                                                                                                                                                                                  |
| <b>Immunogen Type:</b>     | Recombinant Protein                                                                                                                                                                                                    |
| <b>Immunogen:</b>          | This antibody was produced in mice by repeated immunizations with full-length recombinant yeast SUMO protein.                                                                                                          |
| <b>Purity/Specificity:</b> | This product is a monoclonal antibody purified from tissue culture supernatant fluid by Protein A chromatography.                                                                                                      |
| <b>Relevant Links:</b>     | <ul style="list-style-type: none"> <li>• <a href="#">200-301-428 SDS</a></li> <li>• <a href="#">UniProtKB - Q12306</a></li> <li>• <a href="#">NCBI - 6320718</a></li> <li>• <a href="#">GeneID - 852122</a></li> </ul> |

## Application Details

|                                |                                                                                                                                                                                                                                                                                                                                                                                      |
|--------------------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Tested Applications:</b>    | ELISA, WB                                                                                                                                                                                                                                                                                                                                                                            |
| <b>Suggested Applications:</b> | Biochemical Assay, Functional Assay, IF (Based on references)                                                                                                                                                                                                                                                                                                                        |
| <b>Application Note:</b>       | This monoclonal antibody reacts with yeast SUMO (Smt3) tested by western blot and ELISA. Although not tested, this antibody is likely functional in immunohistochemistry and immunoprecipitation. Using the specified conditions, this antibody may recognize other prominent intrinsic bands (UBLs or conjugates). Other intrinsic bands are readily detectable at lower dilutions. |
| <b>Assay Dilutions:</b>        | All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.                                                                                                                                                                                                                                                                                      |
| <b>ELISA:</b>                  | 1:20,000                                                                                                                                                                                                                                                                                                                                                                             |
| <b>IHC:</b>                    | 1:1,000                                                                                                                                                                                                                                                                                                                                                                              |
| <b>WB:</b>                     | 1:500 - 1:2,000                                                                                                                                                                                                                                                                                                                                                                      |

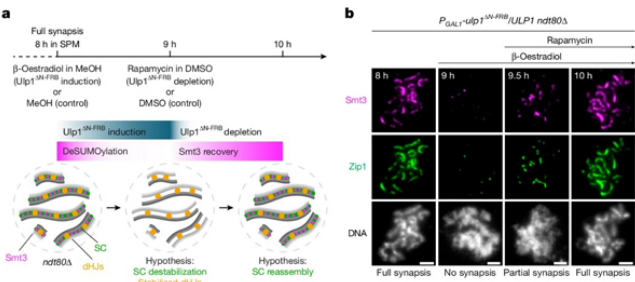
# Formulation

|                 |                                                            |
|-----------------|------------------------------------------------------------|
| Physical State: | Liquid (sterile filtered)                                  |
| Concentration:  | 1.006 mg/mL by UV absorbance at 280 nm                     |
| Buffer:         | 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2 |
| Preservative:   | 0.01% (w/v) Sodium Azide                                   |
| Stabilizer:     | None                                                       |

# Shipping & Handling

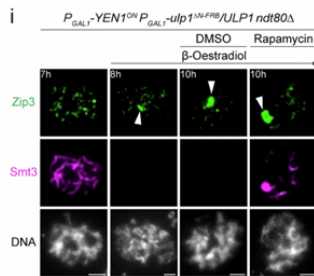
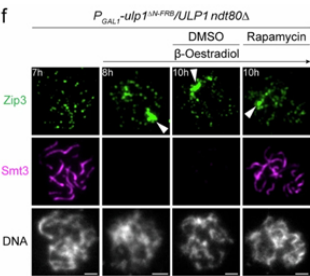
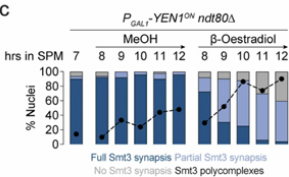
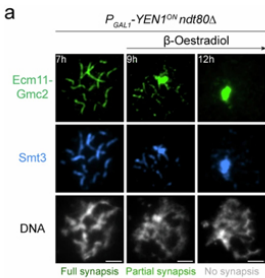
|                     |                                                                                                                                                                                                                                                                                                                                               |
|---------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Shipping Condition: | Dry Ice                                                                                                                                                                                                                                                                                                                                       |
| Storage Condition:  | Store vial at -20° C prior to opening. Aliquot contents and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use. |
| Expiration:         | Expiration date is one (1) year from date of receipt.                                                                                                                                                                                                                                                                                         |

# Images



## Immunofluorescence Microscopy

dHJs enable reversible SC disassembly. A. The experimental set-up for reversible SC disassembly in pachytene-arrested *ndt80Δ* cells through conditional protein deSUMOylation using the Smt3 isopeptidase mutant *ulp1ΔN-FRB*. B. Representative images of meiotic chromosome spreads from the experiment in a at the indicated times in SPM, immunostained for Zip1 (green) and Smt3 (magenta). Fig 3. PMID: 40993383.



### Immunofluorescence Microscopy

HJs stabilize the SC central region but are dispensable for the axis-loop organization of pachytene chromosomes.

A. Representative images of meiotic chromosome spreads at indicated times in SPM, immunostained for Ecm11-Gmc2 (green) and Smt3 (blue). Yen1ON expression was induced by β-oestradiol addition (or MeOH as control) at 7 h in SPM. Scale bars = 2 μm.

C. Quantification of SC-associated Smt3 (c) from (a) (n = 50 nuclei per time point; representative of two biological replicates).

Extended Data. Fig 2. PMID: 40993383.

### Immunofluorescence Microscopy

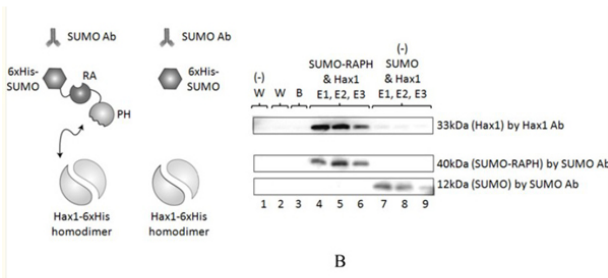
7 dHJ-ZMM protein interplay enables SC reassembly upon Ulp1ΔN-FRB depletion. F. Representative images of meiotic chromosome spreads at indicated times in SPM, immunostained for Zip3 (green) and Smt3 (magenta).

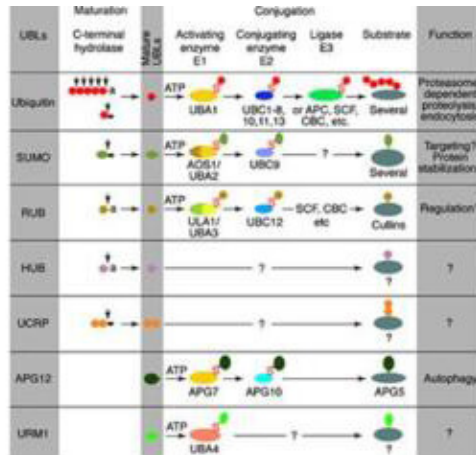
Ulp1ΔN-FRB expression was induced by β-oestradiol addition at 7 h in SPM, and nuclear depletion was initiated by rapamycin addition at 9 h in SPM. Arrowheads mark Zip3 aggregates. Scale bars = 2 μm. I. As in (f), for the simultaneous expression of Ulp1ΔN-FRB and Yen1ON by β-oestradiol addition at 7 h in SPM. Extended Data Fig 7. PMID: 40993383.

### Western Blot

B) Western Blot results for the in vitro binding assay of purified SUMO-Grb7-RAPH domains and purified Hax1. Lane 1: Last wash sample from the negative control. Lane 2: Last wash sample from the in vitro binding assay of SUMO-RAPH and Hax1. Lane 3: Blank. Lane 4–6: Elution samples (three elutions) from the in vitro binding assay of SUMO-RAPH and Hax1. Lane 7–9: Elution samples (three elutions) from the negative control.

Fig 1. PMID: 26869103



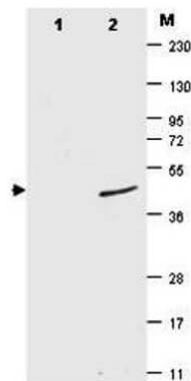


### Pathway

Most modifiers mature by proteolytic processing from inactive precursors (a; amino acid). Arrowheads point to the cleavage sites. Ubiquitin is expressed either as polyubiquitin or as a fusion with ribosomal proteins. Conjugation requires activating (E1) and conjugating (E2) enzymes that form thioesters (S) with the modifiers. Modification of cullins by RUB involves SCF(SKP1/cullin-1/F-box protein) /CBC(cullin-2/elongin B/elonginC)-like E3 enzymes that are also involved in ubiquitination. In contrast to ubiquitin, the UBLs do not seem to form multi-UBL chains. UCRP(ISG15) resembles two ubiquitin moieties linked head-to-tail. Whether HUB1 functions as a modifier is currently unclear. APG12 and URM1 are distinct from the other modifiers because they are unrelated in sequence to ubiquitin. Data contributed by S.Jentsch.

### Western Blot

Western blot of  $\gamma$ SUMO fusion protein. Anti- $\gamma$ SUMO antibody, generated by immunization with recombinant yeast SUMO, was tested by western blot against a SUMO-GFP fusion protein (lane 2). While the actual molecular weight of the fusion protein is 39 kDa, the protein migrates as a 49 kDa band (arrowhead). No reactivity is seen for lane 1 which contains His-tagged GFP protein. The membrane was blocked using BLOTTO. Primary antibody was used at a 1:1,000 dilution in BLOTTO. The membrane was washed and reacted with a 1:10,000 dilution of IRDye® 800 Conjugated Affinity Purified Goat-anti-Mouse IgG (H&L) MX10 (800 nm channel). Molecular weight estimation was made by comparison to prestained MW markers indicated at the right (lane M, 700 nm channel). Other detection systems will yield similar results.



## References

- Orlić L et al. Distinct and overlapping roles of MutL $\gamma$ , Mus81-Mms4, and STR in meiotic Holliday junction processing. *Nat Commun.* (2026)
- Henggeler A, Orlić L, Velikov D, and Matos J. Holliday junction–ZMM protein feedback enables meiotic crossover assurance. *Nature.* (2025)
- Qian et al. Grb7 and Hax1 may colocalize partially to mitochondria in EGF-treated SKBR3 cells and their interaction can affect Caspase3 cleavage of Hax1. *Journal of Molecular Recognition* (2016)
- Meyer, MR et al. Evidence for intermolecular interactions between the intracellular domains of the arabidopsis receptor-like kinase ACR4, its homologs and the Wox5 transcription factor. *PLoS One* (2015)

## Disclaimer

This product is for research use only and is not intended for therapeutic or diagnostic applications. Please contact a technical service representative for more information. All products of animal origin manufactured by Rockland Immunochemicals are derived from starting materials of North American origin. Collection was performed in United States Department of Agriculture (USDA) inspected facilities and all materials have been inspected and certified to be free of disease and suitable for exportation. All properties listed are typical characteristics and are not specifications. All suggestions and data are offered in good faith but without guarantee as conditions and methods of use of our products are beyond our control. All claims must be made within 30 days following the date of delivery. The prospective user must determine the suitability of our materials before adopting them on a commercial scale. Suggested uses of our products are not recommendations to use our products in violation of any patent or as a license under any patent of Rockland Immunochemicals, Inc. If you require a commercial license to use this material and do not have one, then return this material, unopened to: Rockland Inc., P.O. BOX 5199, Limerick, Pennsylvania, USA.