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Schillerstr. 5

R1564P Polyclonal Antibody to ULP1 - Purified

Alternate names: LPB11C, Ubiquitin-like-specific protease 1, YPL020C

Quantity: 0.5 mg

Concentration: 5.0 mg/ml (by UV absorbance at 280 nm)

Background: ULP-1, ubiquitin-like protein-specific protease 1, initially processes Smt3 and also

acts as a deconjugating enzyme for Smt3 [Saccharomyces cerevisiae (Baker's yeast)]. 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 ubiquination, sumoylation does not tag proteins for degradation by the 26S proteasome, but rather seems to enhance stability or modulate their subcellular compartmentalization. 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. Three distinct steps can be distinguished in the SUMO modification pathway: 1) activation of SUMO, 2) transfer of SUMO to the conjugating enzyme, and 3) substrate modification. Since SUMO is synthesized as a precursor protein, a maturation step precedes the activation reaction. In yeast, C-terminal processing of the SUMO precursor is mediated by the processing protease Ulp1, which has an additional role in the deconjugation of SUMO-modified substrates. Mature SUMO is activated by SUMO-

activating enzyme, an E1-like heterodimeric protein complex composed of Uba2 and Aos1. Ulp1 function has provided evidence that SUMO modification in yeast, as has

been suspected for vertebrates, plays an important role in nucleocytoplasmic trafficking.

Uniprot ID: <u>Q02724</u>
NCBI: <u>NP 015305.1</u>

GeneID: 856087
Host: Rabbit

Immunogen: Prepared from rabbit serum after repeated immunizations with recombinant yeast

ULP-1 protein.

Format: State: Lyophilized purified Ig fraction.

Purification: Multi-step process which includes delipidation, salt fractionation and

ion exchange chromatography followed by extensive dialysis.

Buffer System: 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2 with

0.01% (w/v) Sodium Azide as preservative.

Reconstitution: Restore with 0.1 ml of deionized water (or equivalent).

Applications:

This purified polyclonal antibody reacts with yeast ULP-1 by Western blot

(1/500-1:2,000) and ELISA (1/4,000-1/20,000).

Although not tested, this antibody is likely functional in Immunohistochemistry and Immunoprecipitation. Expect a band approximately 72.4 kDa in size corresponding to

yeast ULP-1 by western blotting in the appropriate lysate or extract.

Other applications not tested. Optimal dilutions are dependent on conditions and

should be determined by the user.

Specificity:

Assay by immunoelectrophoresis resulted in a single precipitin arc against anti-Rabbit Serum. Reactivity against ULP-1 from other sources or ULP-2 has not been

determined.

Add. Information:

Protein Sequence: Yeast ULP-1, 621 aa, predicted MW 72.4 kDa 1 msvevdkhrn tlqyhkknpy splfspisty rcyprvlnnp sesrrsasfs giykkrtnts 61 rfnylndrrv lsmeesmkdg sdraskagfi ggiretlwns gkylwhtfvk neprnfdgse 121 veasgnsdve srssgsrssd vpyglrenys sdtrkhkfdt stwalpnkrr riesegygtp 181 stspisslas qksncdsdns itfsrdpfgw nkwktsaigs nsenntsdqk nsydrrqygt 241 afirkkkvak gninntklvs ragseevtyl rgifngeykv pkilkeerer glklmdmdke 301 kdtglkksii dltekiktil iennknrlqt rnendddlvf vkekkissle rkhkdylnqk 361 lkfdrsilef ekdfkrynei lnerkkiged lkkkkeglak kklvpelnek dddgvgkala 421 srentglmnr dnieitvrdf ktlaprrwln dtiieffmky iekstpntva fnsffytnls 481 ergyggvrrw mkrkktgidk ldkiftpinl ngshwalgii dlkkktigyv dslsngpnam 541 sfailtdlqk yvmeeskhti gedfdlihld cpqqpngydc giyvcmntly gsadapldfd 601 ykdairmrrf iahliltdal k

Storage:

Store vial at 2-8°C prior to restoration.

After restoration, store the antibody undiluted at 2-8°C for one month or (in aliquots)

at -20°C for longer.

Avoid repeated freezing and thawing.

Centrifuge product if not completely clear after standing at room temperature.

Shelf life: one year from despatch.

General Readings:

1. Li SJ, Hochstrasser M. A new protease required for cell-cycle progression in yeast.

Nature. 1999 Mar 18;398(6724):246-51. PubMed PMID: 10094048.

2. Mossessova E, Lima CD. Ulp1-SUMO crystal structure and genetic analysis reveal conserved interactions and a regulatory element essential for cell growth in yeast. Mol

Cell. 2000 May;5(5):865-76. PubMed PMID: 10882122.

3. Stade K, Vogel F, Schwienhorst I, Meusser B, Volkwein C, Nentwig B, et al. A lack of SUMO conjugation affects cNLS-dependent nuclear protein import in yeast. J Biol Chem. 2002 Dec 20;277(51):49554-61. Epub 2002 Oct 18. PubMed PMID: 12393908. 4. Li SJ, Hochstrasser M. The Ulp1 SUMO isopeptidase: distinct domains required for viability, nuclear envelope localization, and substrate specificity. J Cell Biol. 2003 Mar

31;160(7):1069-81. Epub 2003 Mar 24. PubMed PMID: 12654900.

5. Takahashi Y, Mizoi J, Toh-E A, Kikuchi Y. Yeast Ulp1, an Smt3-specific protease, associates with nucleoporins. J Biochem. 2000 Nov;128(5):723-5. PubMed PMID:

11056382.

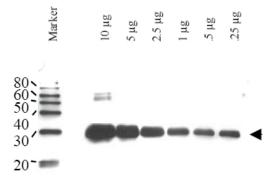


Pictures:

Figure 2. Western blot using Affinity Purified anti-Yeast ULP-1 antibody was used to confirm the specificity of the antibody. SDS-PAGE of 2 µg of ULP-1 homologues from other sources (lanes 2 through 9). After blocking for 1 hour with 5% non-fat dry milk in TTBS, the blot was probed overnight at 4°C with a 1:1,000 dilution of antiyULP1 antibody detected as above. This antibody is specific for yeast ULP1 and does not react with ULP1 from related sources including human SENP.

Figure 1. Western blot using Affinity Purified anti-Yeast ULP-1 antibody shows detection of a truncated ULP-1 fusion protein (arrowhead). Increasing concentrations of yeast ULP-1 were run on a SDS-PAGE, transferred onto nitrocellulose, and blocked for 1 hour with 5% non-fat dry milk in TTBS, and probed overnight at 4°C with a 1:1000 dilution of anti-yULP-1 antibody in 5% non-fat dry milk in TTBS. Detection occurred using a 1:1,000 dilution of HRPlabeled Donkey anti-Rabbit IgG for 1 hour at room temperature. A chemiluminescence system was used for signal detection (Roche) using a 3-sec exposure time.





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