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Product no AS08 349

Anti-SUMO3 | Small ubiquitin-like modifier protein 3 (peptide antibody)

Product information

Immunogen KLH-conjugated peptide derived from SUMO3 sequence of Arabidopsis thaliana, UniProt: Q9FLP5, TAIR: AT5G55170

Host Rabbit

Clonality Polyclonal

Purity Immunogen affinity purified serum in PBS pH 7.4.

Format Lyophilized

Quantity 50 μg

Reconstitution For reconstitution add 50 μl of sterile water

Storage Store lyophilized/reconstituted at -20°C; once reconstituted make aliquots to avoid repeated freeze-thaw cycles. Please remember to spin the tubes briefly prior to opening them to avoid any losses that might occur from material adhering to

the cap or sides of the tube.

Application information

Recommended dilution 1:5000 for detection of recombinant SUMO3 (WB)

Not reactive in No confirmed exceptions from predicted reactivity are currently known

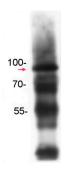
Additional information The antibody is recognizing recombinant SUMO3 and shows no cross reactivity to SUMO1/2

Selected references Saleh et al. (2015). Posttranslational Modifications of the Master Transcriptional Regulator NPR1 Enable Dynamic but

Tight Control of Plant Immune Responses. Cell Host Microbe. 2015 Aug 12;18(2):169-82. doi:

10.1016/j.chom.2015.07.005.

Application example



Protein, tagged with a 6xhis-tag, was expressed from a strain of *E. coli* that reconstituted the sumoylation system with SUMO3 (described in Elrouby et al). Cells were harvested and lysated by sonication in buffer containing 6M urea. Proteins were used to load a Nickel column under denaturing conditions with buffers containing 6M urea. A pH gradient was applied to release the specifically bound proteins and 0.5 ml aliquots were collected. The presence of the desired protein in each aliquot was checked in SDS-PAGE by Comassie staining. 3 ul of the aliquot showing the most intense signal were separated on 10% SDS-PAGE and blotted 1h to PVDF using tank transfer. Blots were blocked with TBS-T (0.1%) and 5% dry-milk powder for 4 h at room temperature (RT) with agitation. Blot was incubated in the primary antibody at a dilution of 1: 5000 overnight at 4°C with agitation. The antibody solution was decanted and the blot was rinsed 4 times for 15 min in TBS-T (0.25%) at RT with agitation. Blot was incubated in secondary antibody (anti-rabbit IgG horse radish peroxidase conjugated, <u>AS09 602</u> from Agrisera) diluted to 1:50 000 in TBS-T (0.1%) and 5% dry-milk powder for 1h at RT with agitation. The blot was washed as above and developed for 5 min with chemiluminescent detection reagent, according to the manufacturer's instructions. Exposure time was 15 second.

Courtesy of Dr. Concepción Almoguera, CSIC, Spain