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**Product no AS09 527**

# Anti-AGO1 | Argonaute 1

## Product information

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| **Immunogen** | KLH-conjugated, N-terminal peptide of *Arabidopsis thaliana* AGO1 O04379, At1g48410 |
| **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. |
| **Additional information** | Antibody binds microRNA and tasiRNAs, preference for 21nt miRNAs with 5'U.<br><br>To detect AGO1 in *Nicotiana benthamiana*, please inquire.<br><br>Recommended for detection of AGO1: extreme low femtogram range chemiluminescent detection reagent |

## Application information

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| **Recommended dilution** | 2 µg (ChIP), 1: 200 (IF), (IL), small-RNA-IP-Seq, 1 : 5000-1 : 10 000 (WB) |
| **Expected \| apparent MW** | 116.4 \| 130 kDa |
| **Confirmed reactivity** | *Arabidopsis thaliana, Hyacinthus orientalis, Nicotiana benthamiana* |
| **Predicted reactivity** | *Brassica pekinensis, Capsella rubella, Cucumis sativus, Glycine max, Malus domestica, Pisum sativum, Ricinus communis, Solanum lycopersicum, Solanum tuberosum, Vitis vinifera*<br><br>Species of your interest not listed? Contact us |
| **Not reactive in** | *Chlamydomonas reinhardtii, Medicago sativa, Salvia sp., Spinacia oleracea, Oryza sativa, Phaseolus vulgaris, Triticum aestivum, Zea mays*<br><br>Mammals |
| **Additional information** | AGO expression may be tissue specific and using floral tissue is recommended where most of the AGOs are expressed the highest. Use of proteasome inhibitors as MG132 can help to stabilize AGO proteins during extraction procedure. Buffer for extraction of AGO proteins: Paudel et al. 2018.<br><br>The AGO1 antibody is extremely specific to AGO1 and does not cross-react with other antibodies. The evidence is 1) the peptide to which it was raised is at the very N-terminus of the protein and is not present in other AGOs 2) aAGO1 does not cross react with the AGOs which are overexpressed (AGO2, AGO3, AGO4, AGO5, AGO6, AGO9) using a western blot.<br><br>TCA acetone precipitation method |
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Delenko et al. (2022) MicroRNA biogenesis and activity in plant cell dedifferentiation stimulated by cell wall removal. BMC Plant Biol. 2022 Jan 3;22(1):9. doi: 10.1186/s12870-021-03323-9. PMID: 34979922; PMCID: PMC8722089. (immunofluorescence)
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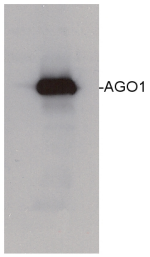
Dalmadi et al. (2021) Controlled RISC loading efficiency of miR168 defined by miRNA duplex structure adjusts ARGONAUTE1 homeostasis. Nucleic Acids Res. 2021 Dec 16;49(22):12912-12928. doi: 10.1093/nar/gkab1138. PMID: 34850097; PMCID: PMC8682782.




80 µg of *Arabidopsis thaliana* soluble total cell extract (extracted in 20 mMTris pH 7.5, 5mM $MgCl_2$, 2.5mM DTT, 300 mM NaCl, 0.1% NP-40, 1% proteasome inhibitor MG132) was separated on 6% SDS-PAGE and blotted 1h to nitrocellulose. Filters were blocked 1h with 5% low-fat milk powder in TBS-TT (0.25% TWEEN20; 0.1% Triton-X) and probed with anti-AGO1 antibody (1:10 000, 1h) and secondary anti-rabbit (1:10000, 1 h) antibody (HRP conjugated) in TBS-TT containing 5% low fat milk powder. Antibody incubations were followed by washings in TBS-TT. All steps were performed at RT with agitation. Blots were developed for 5 min with ECL-PLUS detection reagent according to the manufacturer's instructions. Exposure time was 5 seconds.

Roche's protease inhibitor cocktail (no EDTA) can also be applied in extraction buffer.

Courtesy Dr. Ericka Havecker, University of Cambridge, UK