

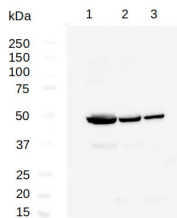
Product no **AS10 680****Tubulin alpha chain (polyclonal antibodies)****Product information**

<b>Immunogen</b>	KLH-conjugated peptide derived from available tubulin alpha chain sequences including <i>Arabidopsis thaliana</i> tubulin alpha-1-chain <a href="#">P11139(At1g64740)</a> , alpha-2/alpha-4 chain <a href="#">B9DGT7(At1g50010)</a> , alpha-5 chain <a href="#">B9DHQ0(At5g19780)</a> , alpha-6-chain <a href="#">P29511(At4g14960)</a> Peptide used to elicit this antibody is not present in tubulin beta.
<b>Host</b>	Rabbit
<b>Clonality</b>	Polyclonal
<b>Purity</b>	Serum
<b>Format</b>	Lyophilized
<b>Quantity</b>	50 µl
<b>Reconstitution</b>	For reconstitution add 50 µl of sterile water
<b>Storage</b>	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**

<b>Recommended dilution</b>	1 : 500 (IF), 1 : 1000 (WB)
<b>Expected   apparent MW</b>	49   52 kDa ( <i>Arabidopsis thaliana</i> )
<b>Confirmed reactivity</b>	<i>Arabidopsis thaliana</i> , <i>Chlamydomonas reinhardtii</i> , <i>Euglena gracilis</i> , <i>Hordeum vulgare</i> , <i>Oryza sativa</i> , <i>Setaria italica</i> , <i>Zea mays</i>
<b>Predicted reactivity</b>	<i>Brassica napus</i> , <i>Chlorella vulgaris</i> , <i>Chlorella variabilis</i> , <i>Cucumis sativus</i> , <i>Eragrostis tef</i> , <i>Euglena gracilis</i> , <i>Glycine max</i> , <i>Micromonas pusilla</i> , <i>Nannochloropsis gaditana</i> , <i>Ostreococcus lucimarinus</i> , <i>Pisum sativum</i> , <i>Physcomitrium patens</i> , <i>Picea sitchensis</i> , <i>Populus trichocarpa</i> , <i>Solanum tuberosum</i> , <i>Sorghum bicolor</i> , <i>Ricinus communis</i> , <i>Triticum aestivum</i> , <i>Vigna radiata</i> , <i>Vitis vinifera</i> Species of your interest not listed? <a href="#">Contact us</a>
<b>Not reactive in</b>	No confirmed exceptions from predicted reactivity are currently known
<b>Additional information</b>	Metal induced stress affected the expression of tubulin, and that therefore, this protein cannot be used as a loading control under that type of conditions. More information can be found <a href="#">here</a> .
<b>Selected references</b>	<a href="#">Graf et al. (2024)</a> . D6PK plasma membrane polarity requires a repeated CXX (X) P motif and PDK1-dependent phosphorylation. <i>Nat Plants</i> . 2024 Feb;10(2):300-314. <a href="#">Kafri et al. (2023)</a> . Systematic identification and characterization of genes in the regulation and biogenesis of photosynthetic machinery. <i>Cell</i> . 2023 Dec 7;186(25):5638-5655.e25.doi: 10.1016/j.cell.2023.11.007. <a href="#">Kumar et al. (2022)</a> . Proteomic dissection of rice cytoskeleton reveals the dominance of microtubule and microfilament proteins, and novel components in the cytoskeleton-bound polysome. <i>Plant Physiology and Biochemistry</i> , Volume 170,2022,Pages 75-86,ISSN 0981-9428, <a href="https://doi.org/10.1016/j.plaphy.2021.11.037">https://doi.org/10.1016/j.plaphy.2021.11.037</a> . <a href="#">Gu et al. (2021)</a> A Lipid Bodies-Associated Galactosyl Hydrolase Is Involved in Triacylglycerol Biosynthesis and Galactolipid Turnover in the Unicellular Green Alga <i>Chlamydomonas reinhardtii</i> <a href="#">Sakuraba et al. (2020)</a> . Multilayered regulation of membrane-bound ONAC054 is essential for abscisic acid-induced leaf senescence in rice. <i>Plant Cell</i> . 2020 Jan 6. pii: tpc.00569.2019. doi: 10.1105/tpc.19.00569. <a href="#">Roustan et al. (2020)</a> . Protein sorting into protein bodies during barley endosperm development is putatively regulated by cytoskeleton members, MVBs and the HvSNF7s. <i>Sci Rep</i> . 2020 Feb 5;10(1):1864. doi: 10.1038/s41598-020-58740-x. <a href="#">Roustan et al. (2020)</a> . Protein sorting into protein bodies during barley endosperm development is putatively regulated by cytoskeleton members, MVBs and the HvSNF7s. <i>Sci Rep</i> . 2020 Feb 5;10(1):1864. doi: 10.1038/s41598-020-58740-x.

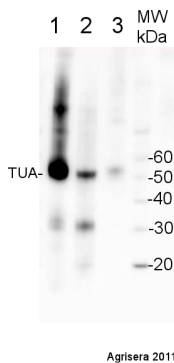
## Western Blot



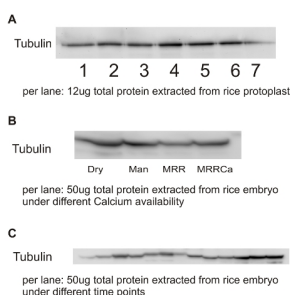
- 1 - 20 µg of *Arabidopsis thaliana* whole rosette leaves extract  
 2 - 10 µg of *Arabidopsis thaliana* whole rosette leaves extract  
 3 - 5 µg of *Arabidopsis thaliana* whole rosette leaves extract

20, 10 and 5 µg of total protein from *Arabidopsis thaliana* rosette leaves extracted with extraction buffer 100 mM Tris-HCl, pH 8.5, 4% [w/v] SDS, 2% [v/v] -mercapthoethanol, and 2 mM phenylmethylsulfonyl fluoride and separated on 10% SDS-PAGE and blotted 1h to PVDF. Blots were blocked with 1% non-fat milk in TBS for 1 h at room temperature with agitation. Membranes were incubated in the primary antibody at a dilution 1:5000 (A) or 1:1000 (B) in 1% non-fat milk in TBS at 4°C overnight. Then, membranes were washed 3 Times for 15 min in TBS at RT with agitation. Membranes were incubated in secondary antibody (Anti-rabbit IgG horse radish peroxidase conjugated from Agrisera AS09 602) diluted to 1: 25 000 in 1% non-fat milk in TBS for 1h at RT with agitation. The blots were washed as above and developed for 5 min with ECL (Agrisera ECL Bright AS16 ECL-N) according to the manufacturer's instructions. Exposure time was 2 seconds.

Courtesy of Dr. Agnieszka Zienkiewicz, Centre for Modern Interdisciplinary Technologies, Nicolaus Copernicus University, Toruń, Poland



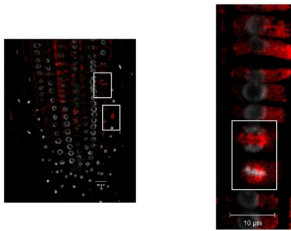
**5 µg of total protein** from *Arabidopsis thaliana* (1), *Hordeum vulgare* (2), *Zea mays* (3) extracted with [Agrisera PEB extraction buffer](#) were separated on 4-12 % **SDS-PAGE** and blotted 1h to **PVDF**. Blots were blocked with for 1h at room temperature (RT) with agitation. Blot was incubated in the primary antibody at a dilution of 1: 10 000 for 1h at RT with agitation. The antibody solution was decanted and the blot was rinsed briefly twice, then washed once for 15 min and 3 times for 5 min in TBS-T at RT with agitation. Blot was incubated in secondary antibody (anti-rabbit IgG horse radish peroxidase conjugated, from Agrisera [AS09 602](#) ) diluted to 1:25 000 in for 1h at RT with agitation. The blot was washed as above and developed for 5 min with ECL Advance according to the manufacturers instructions (GE Health care). Exposure time was 120 seconds.



Total protein from either rice embryos or rice protoplasts extracted with buffer containing 60mM Tris-HCl (pH8.0), 2% SDS(w/v), 15% Sucrose (w/v) and protease inhibitor 1X were separated on 12 % SDS-PAGE and blotted 1h to PVDF. Blots were blocked with skimmed milk containing TBS 1X for 1h at room temperature (RT) with agitation. Blot was incubated in the primary antibody at a dilution of 1: 1 000 overnight at 4 degree with shaking about 40 rpm. The antibody solution was decanted and the blot was rinsed briefly twice, then washed once for 15 min and 3 times for 5 min in TBS-T at RT with agitation. Blot was incubated in secondary antibody (anti-rabbit IgG horse radish peroxidase conjugated) diluted to 1:20 000 in for 1h at RT with agitation. The blot was washed as above and developed for 5 min with ECL according to the manufacturers instructions. Exposure time was 10 seconds.

Courtesy of Ho Viet The, PhD Student, Scuola Superiore Sant'Anna, Pisa, Italy

### Immunolocalization



Tubulin alpha localization in roots of *Arabidopsis thaliana*. Tubulin alpha (red), nucleus (DAPI white). Plant material has been fixed in para-formaldehyde for 30 minutes. Tissue cleaning has been performed before immunolocalization. Primary antibodies: Agrisera anti-tubulin alpha 1: 500. Secondary antibody: goat anti-rabbit IgG Alexa conjugated (red color), dilution 1: 500. Scale bar – 10 µm.

Courtesy Dr. Taras Pasternak, Freiburg University, Germany