

Product no **AS10 710****H3 | Histone H3 (rabbit antibody) (nuclear marker)****Product information**

<b>Immunogen</b>	KLH-conjugated synthetic peptide derived from known H3 sequences, including <i>Arabidopsis thaliana</i> H3.3 <a href="#">P59169</a> ( <a href="#">At4g40030</a> , <a href="#">At4g40040</a> , <a href="#">At5g10980</a> ), H3.2 <a href="#">P59226</a> ( <a href="#">At1g09200</a> , <a href="#">At3g27360</a> , <a href="#">At5g10390</a> , <a href="#">At5g10400</a> , <a href="#">At5g65360</a> ), H3-like 2 <a href="#">Q9FXI7</a> ( <a href="#">At1g19890</a> )
<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.

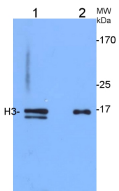
**Additional information** Cellular **[compartment marker]** of nucleoplasm, loading control antibody for *Chlamydomonas reinhardtii*

**Application information**

<b>Recommended dilution</b>	1 : 100-1 : 500 (ICC), 2 µl of antibody/500 µl solution (Chlp-qPCR), 1: 500 (IF), 1 : 5000 (WB)
<b>Expected   apparent MW</b>	15   17 kDa
<b>Confirmed reactivity</b>	<i>Arabidopsis thaliana</i> , <i>Botrytis cinerea</i> , <i>Brassica oleracea</i> , <i>Capsicum annuum</i> , <i>Cicer arietinum</i> L., <i>Chlamydomonas acidophila</i> , <i>Chlamydomonas reinhardtii</i> , <i>Cucumis sativus</i> L cv <i>Suyo</i> , <i>Cucurbita pepo</i> L. var. <i>cylindrica</i> , <i>Hordeum vulgare</i> , human, <i>Nicotiana benthamiana</i> , <i>Phalaenopsis equestris</i> , <i>Phaeodactylum tricornutum</i> , <i>Physcomitrium patens</i> , <i>Salicornia europaea</i> , <i>Solanum lycopersicum</i> , <i>Solanum soganandinum</i> , <i>Solanum tuberosum</i> , <i>Vicia faba</i> , <i>Zea mays</i>
<b>Predicted reactivity</b>	<i>Brachypodium distachyon</i> , <i>Brassica napus</i> , <i>Emiliana huxleyi</i> , <i>Fagopyrum tataricum</i> Gaertn., <i>Glycine max</i> , <i>Nannochloropsis gaditana</i> , <i>Nicotiana tabacum</i> , <i>Malus domestica</i> , <i>Medicago sativa</i> , <i>Oryza sativa</i> , <i>Ostreococcus</i> sp., <i>Pinus pinaster</i> , <i>Pisum sativum</i> , <i>Porphyridium purpureum</i> , <i>Saccharum</i> sp., <i>Sorghum bicolor</i> , <i>Triticum aestivum</i> , <i>Vitis vinifera</i> , <i>Volvox</i> sp.
	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>	Specific fluorescence in ICC has been observed for interphase nuclei as well as around centromer region (where Ser10 of histone H3 is phosphorylated) in mitotic chromosomes
<b>Selected references</b>	<p><a href="#">Kimura and Kanegae</a> (2024). A phytochrome/phototropin chimeric photoreceptor promotes growth of fern gametophytes under limited light conditions. <i>Exp Bot</i> . 2024 Apr 15;75(8):2403-2416.</p> <p><a href="#">Woo et al.</a> (2024). Attenuation of phytofungus pathogenicity of Ascomycota by autophagy modulators. <i>Nat Commun</i>. 2024 Feb 29;15(1):1621. doi: 10.1038/s41467-024-45839-2.</p> <p><a href="#">Liu et al.</a> (2023). RBPome identification in egg-cell like callus of Arabidopsis. <i>Biol Chem</i>. 2023 Sep 29;404(11-12):1137-1149. doi: 10.1515/hsz-2023-0195.</p> <p><a href="#">Hyun et al.</a> (2023). Functional demonstration of Aureochrome 1a proteasomal degradation after blue light incubation in the diatom <i>Phaeodactylum tricornutum</i>. <i>J Plant Physiol</i>. 2023 Dec 1:292:154148. doi: 10.1016/j.jplph.2023.154148</p> <p><a href="#">Skalický et al.</a> (2023). Fluorescence-activated multi-organelle mapping of subcellular plant hormone distribution. <i>Plant J</i>. 2023 Dec;116(6):1825-1841. doi: 10.1111/tpj.16456. Epub 2023 Sep 8.</p> <p><a href="#">Farago et al.</a> (2022). Small paraquat resistance proteins modulate paraquat and ABA responses and confer drought tolerance to overexpressing Arabidopsis plants. <i>Plant Cell Environ</i>. 2022 Jul;45(7):1985-2003. doi: 10.1111/pce.14338. Epub 2022 Apr 29. PMID: 35486392; PMCID: PMC9324991.</p> <p><a href="#">Margaritopoulou et al.</a> (2021) Enriched HeK4me3 marks at Pm-0 resistance-related genes prime courgette against <i>Podosphaera xanthii</i>. <i>Plant Physiol</i>. 2021 Sep 21:kiab453. doi: 10.1093/plphys/kiab453. Epub ahead of print. Erratum in: <i>Plant Physiol</i>. 2021 Nov 11;: PMID: 34597395.</p> <p><a href="#">Perlaza</a> (2021). Organelle Size and Quality Control in <i>Chlamydomonas Reinhardtii</i>. UCSF. ProQuest ID: Perlaza_ucsf_0034D_12217. Merritt ID: ark:/13030/m5257z1d. Retrieved from <a href="https://escholarship.org/uc/item/1jg3874h">https://escholarship.org/uc/item/1jg3874h</a></p> <p><a href="#">Sun et al.</a> (2021) The epigenetic factor FVE orchestrates cytoplasmic SGS3-DRB4-DCL4 activities to promote transgene silencing in Arabidopsis. <i>Sci Adv</i>. 2021 Aug 4;7(32):eabf3898. doi: 10.1126/sciadv.abf3898. PMID: 34348894; PMCID: PMC8336953.</p>

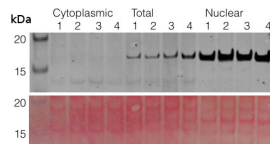
[Chen et al. \(2019\)](#). Phalaenopsis LEAFY COTYLEDON1-Induced Somatic Embryonic Structures Are Morphologically Distinct From Protocorm-Like Bodies. *Front Plant Sci.* 2019 Nov 29;10:1594. doi: 10.3389/fpls.2019.01594. PMID: 31850050; PMCID: PMC6896055.

## Application example



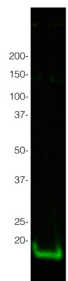
1.2  $\mu\text{g}$  of *Arabidopsis thaliana* chromatin-enriched fraction (1) and 3.75  $\mu\text{g}$  of total protein from 4-week-old *Arabidopsis thaliana* leaves (2), and were separated on 12% SDS-PAGE and blotted 50 mins to Immobilon-P (Millipore, semi-dry) PVDF membrane. Blots were blocked immediately following transfer in MTBS-T (5% milk) for 30 mins at room temperature with agitation. Blots were incubated in the primary antibody at a dilution of 1:5000 for 1h at room temperature with agitation. The antibody solution was decanted and the blot was rinsed briefly twice, then washed 3 times for 3 min in TBS-T at room temperature with agitation. Blots were incubated in secondary antibody (anti- IgG horse radish peroxidase conjugated, from Agrisera, [AS09 602](#)) diluted to 1:20 000 for 30 mins at room temperature with agitation. The blots were washed as above and developed for 5 min with ECL detection reagent according to the manufacturers instructions. Exposure time was 30 seconds. Double band in chromatin-enriched fraction (1) has been outcompeted in peptide neutralization assay by peptide used to elicit H3 antibodies. Chromatin isolation was carried out as described ([Zilberman et al. 2008](#)) with minor modifications.

Courtesy of Weronika Sura and Dr. Piotr A. Ziolkowski, Department of Biotechnology, Adam Mickiewicz University, Poznan, Poland



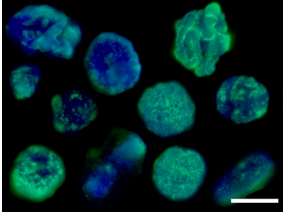
30  $\mu\text{g}$  of 5  $\mu\text{l}$  of *Chlamydomonas reinhardtii* protein saturated in 8M urea were separated on 15% SDS-PAGE and blotted for 1 hour to 0.2  $\mu\text{m}$  nitrocellulose at 100V using wet transfer system. Blots were blocked with 0.5% cold fish gelatin for 1hr at room temp with agitation. Blot was incubated in the primary antibody (anti-H3) at a dilution of 1:2500 for an hour at RT with agitation. The blots were washed with 3X 15min TBS-TT at RT with agitation. Blots as incubated in the secondary antibody (DayLight 800) 1:5000 dilution for 30min at RT with agitation and washed 1X with TBSTT for 15min, 1X with TBST for 15min before scanning with the ODyssey IRD scanner.

Courtesy of Dr. Betty Chung, University of Cambridge, United Kingdom



5  $\mu\text{l}$  of 15 $\mu\text{g}/\mu\text{l}$  *Solanum lycopersicum* protein saturated in 8M urea were separated on 15% SDS-PAGE and blotted for 1 hour to 0.2  $\mu\text{m}$  nitrocellulose at 100V using wet transfer system. Blots were blocked with 0.5% cold fish gelatin for 1hr at room temp with agitation. Blot was incubated in the primary antibody (anti-H3) at a dilution of 1:2500 for an hour at RT with agitation. The blots were washed with 3X 15min TBS-TT at RT with agitation. Blots as incubated in the secondary antibody, fluorescent antibody ([AS12 2460](#), Agrisera) 1:5000 dilution for 30min at RT with agitation and washed 1X with TBSTT for 15min, 1X with TBST for 15min before scanning with the ODyssey IRD scanner.

Courtesy of Dr. Betty Chung and Dr Zhengming Wang, University of Cambridge, United Kingdom



**Immunocytochemical** assays were performed according to the method described earlier (Rybaczek and Maszewski 2006). Excised apical parts of *Vicia faba* roots (1.5 mm long) were fixed for 45 min (18 °C) in PBS-buffered 3.7% paraformaldehyde, washed several times with PBS and placed in a citric acid-buffered digestion solution (pH 5.0; 37 °C for 45 min) containing 2.5% pectinase (Fluka), 2.5% cellulase (Onozuka R-10; Serva) and 2.5% pectoliase (ICN). After removing the digestion solution, root tips were washed 3 times in PBS, rinsed with distilled water and squashed onto Super Frost Plus glass slides (Menzel-Gläser). Air-dried slides were pretreated with PBS-buffered 5% BSA at 20 °C for 50 min and incubated overnight in a humidified atmosphere (4 °C) with rabbit antibody raised against H3 histone (Agrisera), dissolved in PBS containing 1% BSA (at a dilution of 1:50). Following incubation, slides were washed 3 times with PBS and incubated for 1 h (18 °C) with Agrisera secondary goat anti-rabbit IgG DyLight®488 antibody ([AS09\\_633](#), 1:1000). Nuclear DNA was stained with 4',6-diamidino-2-phenyl-indole (DAPI, 0.4 µg/ml; Sigma-Aldrich). Following washing with PBS, slides were air dried and embedded in Vectashield Mounting Media for Fluorescence (Vector Laboratories). Observations were made using Optiphot-2 fluorescence microscope (Nikon) equipped with B-2A filter (blue light; 495 nm) for DyLight-conjugated antibodies and UV-2A filter (UV light; 365 nm) for DAPI. All images were recorded at exactly the same time of integration using DXM 1200 CCD camera.

Courtesy Dr. Dorota Rybaczek, Łódź University, Poland