

product **AS05 084**

**PsbA | D1 protein of PSII, C-terminal (100 µl)**

### product information

<b>background</b>	The psbA gene has been cloned from many species of plants, green algae, and cyanobacteria. The psbA gene is located in the chloroplast genome and encodes for the D1 protein, a core component of Photosystem II. PsbA/D1 is rapidly cycled under illumination in all oxygenic photobionts. Tracking PsbA pools using the Global PsbA antibody can show the functional content of Photosystem II in a wide range of samples. Alternative names: 32 kDa thylakoid membrane protein, photosystem II protein D1
<b>immunogen</b>	<u>KLH</u> -conjugated synthetic peptide derived from available plant, algal and cyanobacterial PsbA sequences, including <i>Arabidopsis thaliana</i> <u>AtCg00020</u> , <i>Oryza sativa</i> <u>P0C434</u> , <i>Physcomitrella patens</i> <u>Q6YXNZ</u> , <i>Chlamydomonas reinhardtii</i> <u>P07753</u> , <i>Synechococcus</i> sp. <u>P14660</u>
<b>antibody format</b>	rabbit polyclonal, serum, lyophilized
<b>quantity</b>	100 µl - for reconstitution add 100 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 tubes briefly prior to opening them to avoid any losses that might occur from lyophilized material adhering to the cap or sides of the tubes.
<b>tested applications</b>	western blot (WB)
<b>additional information</b>	<p>A number of degradation products may be observed when using anti-PsbA antibodies, including products having apparent molecular weights of 24kDa and 16kDa. D1 degradation is a complex set of events and the products observed can be influenced by both the extraction procedure and the physiology of the cells prior to harvest. Third, cross-linking may occur between D1 and cytochrome b559, shifting the protein higher in the gel. In cyanobacteria (PCC7942), three different bands were competed out by preincubating the antibody with the PsbA free peptide, indicating that all bands are indeed PsbA and its precursors or breakdown products. Competition assays were also performed with spinach and <i>Chlamydomonas</i>, confirming the identity of PsbA bands.</p> <p>Anti-PsbA antibodies will not detect D2 protein, as the peptide used to generate PsbA antibodies has no homology to the D2 sequence.</p>

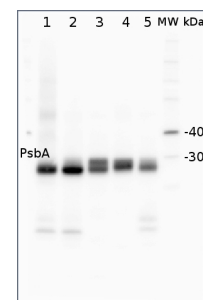
### application information

<b>recommended dilution</b>	1: 10 000 with standard ECL (WB)
<b>expected   apparent MW</b>	38   28-30 kDa
<b>confirmed reactivity</b>	

	<p><i>Arabidopsis thaliana</i>, <i>Hordeum vulgare</i>, <i>Nicotiana benthamiana</i>, <i>Panicum miliaceum</i>, <i>Panicum maximum</i>, <i>Pisum sativum</i>, <i>Zea mays</i>, <i>Chlamydomonas reinhardtii</i>, <i>Physcomitrella patens</i>, <i>Synechococcus</i> sp. PCC 7942, <i>Anabaena</i> 7120, <i>Prochlorococcus</i> sp. (surface and deep water ecotype), <i>Symbiodinium</i> sp., <i>Coscinodiscus wailesii</i></p>
<b>predicted reactivity</b>	di and monocots, conifers, brown and red algae, cyanobacteria; cellular <b>[compartment marker]</b> of thylakoid membrane
<b>not reactive in</b>	no confirmed exceptions from predicted reactivity known in the moment
<b>additional information</b>	<p>The antibody is appropriate for detecting both, 24 kDa or the 10 kDa C-terminal fragments, whichever is generated under given treatment conditions. In our analysis we have seen both, ca. 24 kDa and ca. 10 kDa fragments from different samples, depending on treatments and isolation procedures.</p> <p>Rabbit anti-PsbA antibody can detect more than one band of PsbA protein, e.g. precursor and mature protein as compare to the hen anti-PsbA antibodies AS01 016.</p> <p>This antibody will detect the phosphorylated form of D1 as an alternate band to the main band on a high resolution gel.</p>
<b>selected references</b>	<p><a href="#">Rodríguez-Herva et al. (2012)</a>. A bacterial cysteine protease effector protein interferes with photosynthesis to suppress plant innate immune responses. <i>Cell Microbiol</i> (ahead of print)</p> <p><a href="#">Petrou et al. (2010)</a>. Rapid photoprotection in sea-ice diatoms from the East Antarctic pack ice- <i>Limnol Oceanogr.</i> 3: 1400-1407. (PsbA quantitation in diatoms)</p> <p><a href="#">Fristedt et al. (2009)</a>. Phosphorylation of photosystem II controls functional macroscopic folding of photosynthetic membranes in <i>Arabidopsis</i>. <i>Plant Cell</i> 21:3950-3964</p> <p><a href="#">Brown et al (2007)</a> Resource dynamics during infection of <i>Micromonas pusilla</i> by virus MpV-Sp1. <i>Environmental Microbiol</i> 11: 2720-2727</p>

## application example

2 µg of total protein from (1) *Arabidopsis thaliana* leaf extracted with Protein Extraction Buffer, PEB (**AS08 300**), (2) *Hordeum vulgare* leaf extracted with PEB, (3) *Chlamydomonas reinhardtii* total cell extracted with PEB, (4) *Synechococcus* sp. 7942 total cell extracted with PEB, (5) *Anabaena* sp. total cell extracted with PEB were separated on 4-12% NuPage (Invitrogen) **LDS-PAGE** and blotted 1h to **PVDF**. Blots were blocked immediately following transfer in 2% ECL Advance blocking reagent (GE Healthcare) in 20 mM Tris, 137 mM sodium chloride pH 7.6 with 0.1% (v/v) Tween-20 (TBS-T) for 1h at room temperature with agitation. Blots were incubated in the primary antibody at a dilution of 1: 50 000 for 1h at room temperature 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 room temperature with agitation. Blots were incubated in secondary antibody (anti-rabbit IgG horse radish peroxidase



conjugated, recommended secondary antibody [AS09 602](#)) diluted to 1:50 000 in 2% ECL Advance blocking solution for 1h at room temperature with agitation. The blots were washed as above and developed for 5 min with ECL Advance detection reagent according the manufacturers instructions. Images of the blots were obtained using a CCD imager (FluorSMax, Bio-Rad) and Quantity One software (Bio-Rad).