

product **AS01 016**

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

product information

background	The PsbA (D1) protein of Photosystem II is rapidly cycled under illumination in all oxygenic photobionts. Disruption of PsbA cycling or losses of PsbA pools are central to photoinhibition of photosynthesis in cyanobacteria, algae and plants under a wide range of conditions including excess light, low temperature and UV exposure. Tracking PsbA pools using the Global PsbA antibody can show the functional content of Photosystem II in a wide range of samples.
immunogen	<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>Populus alba</i> <u>Q14FH6</u> , <i>Synechocystis</i> sp. <u>P14660</u> and many others
antibody format	hen polyclonal total IgY in PBS pH 8.0+ 0.02% sodium azide liquid
quantity	100 µl
storage	store at 4 °C; make aliquots to avoid working with a stock. Please, Remember to spin tubes briefly prior to opening them to avoid any losses that might occur from liquid material adhering to the cap or sides of the tubes.
tested applications	western blot (WB)
additional information	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. Anti-PsbA antibodies will not detect D2 protein, as the peptide used to generate PsbA antibodies has no homology to the D2 sequence.

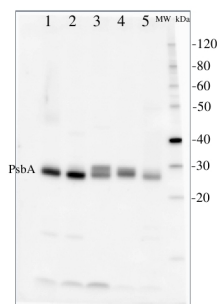
application information

recommended dilution	1:4000 - 1: 8000 on 5 µg of total protein, detected with standard ECL (WB)
expected apparent MW	38 28-30 kDa
confirmed reactivity	<i>Alaria esculenta</i> , <i>Amphidinium carterae</i> , <i>Arabidopsis thaliana</i> , <i>Chlamydomonas reinhardtii</i> , <i>Chlamydomonas raudensis</i> (both Antarctic and mesophilic strains), <i>Cyanophora</i> sp., <i>Gonyaulax polyedra</i> , <i>Fucus vesiculosus</i> , <i>Hordeum vulgare</i> , <i>Lobaria pulmonaria</i> , <i>Petunia</i> sp., <i>Pinus sylvestris</i> , <i>Synechococcus</i> sp. PCC 7942,

	<p><i>Ulva</i> sp., symbiotic dinoflagellates of <i>Stylophora pistillata</i> and <i>Turbinaria reniformis</i>.</p>
predicted reactivity	<p>dicots including legumes, monocots, conifers, brown algae, red algae, cryptomonads, stramenopiles, euglenoids, xanthophytes, prochlorophytes</p>
not reactive in	<p>no confirmed exceptions from predicted reactivity known in the moment</p>
additional information	<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>This antibody will also detect the phosphorylated form of D1 as an alternate band to the main band on a high resolution gel.</p>
selected references	<p>Hooogenboom et al. (2012). Effects of Light, Food Availability and Temperature Stress on the Function of Photosystem II and Photosystem I of Coral Symbionts. POLS one</p> <p>Tits et al. (2006) PDX1 is essential for vitamin B6 biosynthesis, development and stress tolerance in Arabidopsis. The Plant J. 48:933-946.</p> <p>Wang et al (2005). Synthesis and degradation of dinoflagellate plastic-encoded PsbA proteins are light regulated, not circadian-regulated. PNAS 102:2844-2849.</p>

application example

2 µg of total protein from (1) *Arabidopsis thaliana* leaf extracted with PEB (**AS08 300**), (2) *Hordeum vulgare* leaf extracted with PEB (**AS08 300**), (3) *Chlamydomonas reinhardtii* total cell extracted with PEB (**AS08 300**), (4) *Synechococcus* sp. 7942 total cell extracted with PEB (**AS08 300**), (5) *Anabaena* sp. total cell extracted with PEB (**AS08 300**) 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-hen IgY horse radish peroxidase conjugated, recommended secondary antibody **AS09 603**) diluted to 1:50 000 in 2% ECL Advance blocking solution for 1h at room temperature with agitation. The blots were washed as



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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).