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# Product no AS05 084A PsbA | D1 protein of PSII, C-terminal (affinity purified)

### **Product information**

Immunogen	<u>KLH</u> -conjugated synthetic peptide derived from available plant, algal and cyanobacterial PsbA sequences, including Arabidopsis thaliana UniProt: <u>A4QJR4</u> , TAIR: <u>AtCg00020</u> , <i>Oryza sativa<u>P0C434</u>, Populus alba<u>Q14FH6</u>, Physcomitrella patens <u>Q6YXN7</u>, Chlamydomonas reinhardtii <u>P07753</u>, Synechocystis sp. <u>P14660</u> and many others</i>
Host	Rabbit
Clonality	Polyclonal
Purity	Immunogen affinity purified serum in PBS pH 7.4.
Format	Lyophilized
Quantity	50 µg
Reconstitution	For reconstitution add 50 $\mu$ l 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	Due to biology of PsbA (D1) protein a number of degradation products can apprear in a sample and 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 : 15 000 (WB)
Expected   apparent MW	38   28-30 kDa
Confirmed reactivity	Anabaena 7120, Arabidopsis thaliana, Artemisia annua, Arundo sp., Chlamydomonas reinhardtii, Colobanthus quitensis Kunt Bartl, Craterostigma sp., Coscinodiscus wailesii, Cynara cardunculus var altilis, Ditylum brightwellii, Glycine max, Hordeum vulgare, Lindernia sp., Miscanthus x giganteus, Marchantia polymorpha (liverwort), Nicotiana benthamiana,Panicum miliaceum, Panax ginseng, Panicum maximum, Paulinella chromatophora (amoeba), Pinus strobus, Physcomitrium patens, Prochlorococcus sp. (surface and deep water ecotype), Synechococcus sp. PCC 7942, Spirodela polyrhiza, Symbiodinium sp., Zea mays
Predicted reactivity	Algae (brown and red), <i>Brassica napus</i> , Conifers, Cyanobacteria, Dictos, <i>Cannabis sativa</i> , <i>Galdieria sulphuraria</i> , <i>Lactuca sativa</i> , <i>Lycopersicum esculentum</i> , <i>Medicago sativa</i> , <i>Nannochloropsis sp.</i> , <i>Oryza sativa</i> , <i>Ostreococcus sp.</i> <i>Pisum sativum</i> , <i>Porphyridium purpureum</i> , <i>Sesamum indicum</i> , <i>Thalassiosira pseudonana</i> , <i>Zosteria marina</i> , <i>Vitis vinifera</i> cellular [compartment marker] of thylakoid membrane Species of your interest not listed? Contact us
Not reactive in	No confirmed exceptions from predicted reactivity are currently known
Additional information	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.
	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.
	This antibody will detect the phosphorylated form of D1 as an alternate band to the main band on a high resolution gel.
	The antibody will bind to cross-linked proteins: D1/D2, D1/cyt b559, D1/CP43.
Selected references	<u>Kafri</u> et al. (2023). Systematic identification and characterization of genes in the regulation and biogenesis of photosynthetic machinery. Cell. 2023 Dec 7;186(25):5638-5655.e25.doi: 10.1016/j.cell.2023.11.007. <u>Wada</u> et al. (2021) Identification of a Novel Mutation Exacerbated the PSI Photoinhibition in pgr5/pgrl1 Mutants;

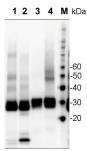


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Caution for Overestimation of the Phenotypes in Arabidopsis pgr5-1 Mutant. Cells. 2021 Oct 26;10(11):2884. doi: 10.3390/cells10112884. PMID: 34831107; PMCID: PMC8616342. <u>Sorrentino</u> et al. (2018). Performance of three cardoon cultivars in an industrial heavy metal-contaminated soil: Effects on morphology, cytology and photosynthesis. J Hazard Mater. 2018 Jun 5;351:131-137. doi: 10.1016/j.jhazmat.2018.02.044. <u>Kanazawa</u> et al. (2017). Chloroplast ATP Synthase Modulation of the Thylakoid Proton Motive Force: Implications for Photosystem I and Photosystem II Photoprotection. Front Plant Sci. 2017 May 3;8:719. doi: 10.3389/fpls.2017.00719. <u>Li</u> et al. (2016). A Hard Day's Night: Diatoms Continue Recycling Photosystem II in the Dark. Front. Mar. Sci., 08 November 2016

### **Application example**



2 μg of total protein from samples such as *Arabidopsis thaliana* leaf (1), *Hordeum vulgare* leaf (2), *Chlamydomonas reinhardtii* total cell (3), *Synechococcus* sp. PCC7942 total cell (4), were extracted with Protein Extraction Buffer PEB (AS08 300). Samples were diluted with 1X sample buffer (NuPAGE LDS sample buffer (Invitrogen) supplemented with 50 mM DTT and heat at 70 °C for 5 min and keept on ice before loading. Protein samples were separated on 4-12% Bolt Plus gels, LDS-PAGE and blotted for 70 minutes to PVDF using tank transfer. Blots were blocked immediately following transfer in 2% blocking reagent or 5% non-fat milk dissolved 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: 10 000 (in blocking reagent) for 1h at room temperature with agitation. Blots were incubated and the blot was rinsed briefly twice, and then washed 1x15 min and 3x5 min with TBS-T at room temperature with agitation. Blots were incubated in secondary antibody (anti-rabbit IgG horse radish peroxidase conjugated, recommended secondary antibody <u>AS09 602</u>, Agrisera) diluted to 1:25 000 in blocking reagent for 1h at room temperature with agited on 5 min with ECL detection reagent according to the manufacturer's instructions. Images of the blots were obtained using a CCD imager (VersaDoc MP 4000) and Quantity One software (Bio-Rad). Exposure time was 30 seconds.

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