

product **AS06 142-33**

PsbO | 33 kDa of the oxygen evolving complex (OEC) of PSII

product information

background	PSII reaction centre components are generating the redox potential required to drive highly oxidizing water splitting reaction. Four Mn atoms are present on a lumenal surface and form the catalytic site of the water-splitting reaction which is in close association with the 33 kDa (PsbO), 23 kDa (PsbP) and 17 kDa (PsbQ) extrinsic subunits of oxygen evolving complex OEC. A 33-kDa extrinsic protein is also termed the Mn-stabilizing protein (MSP), however recent evidences shown that it is C-terminal domain of PsbA (D1) protein which is involved in the assembly and stabilization of the OEC.
immunogen	native purified 33 kDa protein from <i>Spinacia oleracea</i>
antibody format	rabbit polyclonal total IgG in PBS pH 7.4 lyophilized
quantity	260 µg for reconstitution add 200 µl of sterile water.
storage	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.
tested applications	western blot (WB)
additional information	total IgG fraction has been purified by 40% ammonium sulphate precipitation followed by DEAE cellulose chromatography

application information

recommended dilution	1:2000-1: 5000 with standard ECL (WB)
expected apparent MW	35 33 kDa
confirmed reactivity	<i>Arabidopsis thaliana</i> , <i>Hordeum vulgare</i> , <i>Pinus banksiana</i> , <i>Spinacia oleracea</i> , <i>Chlamydomonas reinhardtii</i> , <i>Synechococcus</i> sp. PCC 7942
predicted reactivity	dicots including: <i>Nicotiana tabacum</i> , <i>Solanum lycopersicum</i> , trees: <i>Populus trichocarpa</i>
not reactive in	no confirmed exceptions from predicted reactivity known in the moment
additional information	to be added when available
selected references	Camm et al. (1987). Association of the 33 kDa extrinsic polypeptide (water-splitting) with PSII particles: immunochemical quantification of residual polypeptide after membrane extraction. Photos Res 13: 69-80.

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: 10 000 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, from Abcam) 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).

