Agrisera AB | Box 57 | SE-91121 Vännas | Sweden | +46 035 33033 | www.agrisera.com

product **AS03 037**

RbcL | Rubisco large subunit, form I and form II (100 µg)

product information

background

This antibody is especially suitable for quantifying of Rubisco in plant and algal samples.

Rubisco (Ribulose-1,5-bisphosphate carboxylase/oxygenase) catalyzes the rate-limiting step of CO2 fixation in photosynthetic organisms. It is demonstrably homologous from purple bacteria to flowering plants and consists of two protein subunits, each present in 8 copies. In plants and green algae, the large subunit (~55 kDa) is coded by the chloroplast rbcL gene, and the small subunit (15 kDa) is coded by a family of nuclear rbcS genes.

immunogen

KLH-conjugated synthetic peptide conserved across all known plant, algal and (cyano)bacterial RbcL protein sequences (form I L8S8 and form II L2), including *Arabidopsis thaliana* AtCg00490, *Hordeum vulgare* P05698, *Oryza sativa* P0C510, *Chlamydomonas reinhardtii* P00877, *Synechococcus* PCC 7920 A5CKC5

antibody format

rabbit polyclonal, affinity purified serum in PBS pH 7.4, lyophilized

quantity

100 µg - for reconstitution add 100 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), tissue printing (TP), immunofluorescence/confocal microscopy (IF), immunolabelling (IL)

additional information

anti-RbcL can be used as a cellular [**compartment marker**] of plastid stroma (cytoplasm in cyanobacteria) and detects RbcL protein from 31.25 fmoles. As both forms (I and II) are detected it is suitable for work with samples from Dinoflagellates, Haptophytes and Ochrophytes (diatoms, Raphidophytes, brown algae) as well as higher plants. **This antibody together with Agrisera Rubisco protein standard is very suitable to quantify Rubisco in plant and algal samples.**

application information

recommended dilution

1: 5000 - 10 000 with standard ECL (WB), 1: 800 (TP), immunofluorescence/confocal microscopy (IF), 1: 250 for images see Prins et al. (2008), detailed protocol available on request (IL)

expected | apparent MW

52.7 kDa (*Arabidopsis thaliana*), 52.5 kDa (cyanobacteria), 52.3 (*Chlamydomonas reinhardtii*)

confirmed reactivity

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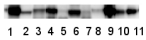
	Arabidopsis thaliana, Apium graveolens, *Bienertia sinuspersici, Chlamydomonas reinhardtii, Cyanophora paradoxa, Emiliana huxleyi, Euglena gracilis, Gonyaulax polyedra, Guzmania* hybrid*, Heterosigma akashiwo, Micromonas pusila, Physcomitrella patens, Porphyra sp. Spinacia oleracea,* lichens*, Synechococcus PCC 7942, Thalassiosira pseudonana, Prochlorococcus sp.* (surface and deep water ecotype), dinoflagellate endosymbionts (genus Symbiodinium), extreme acidophilic verrucomicrobial methanotroph *Methylacidiphilum fumariolicum* strain SolV
predicted reactivity	di and monocots, conifers, mosses, liverworts, welwitschia, green algae, red alge, brown algae, cryptomonad, cyanobacteria including prochlorophytes, gamma-proeobacteria, beta-proteobacteria, alpha proteobacteria
not reactive in	no confirmed exceptions from predicted reactivity known in the moment
additional information	This antibody has been used for immunocytochemical staining of diatoms according to Schmid (2003) J Phycol 39: 139-153 and Wordemann et al. (1986) J Cell Biol 102: 1688-1698 Antibody used in a student course in western blot and tissue printing Ma et al. (2009).
selected references	Heckwolf et al. (2011). The *Arabidopsis thaliana* aquaporin AtPIP1;2 is a physiologically relevant CO(2) transport facilitator. Plant J. doi: 10.1111/j.1365-313X.2011.04634.x. [Epub ahead of print] Johnson (2011). Manipulating RuBisCO accumulation in the green alga, *Chlamydomonas reinhardtii*. Plant Mol Biol. May 24. Kubien et al. (2011). Quantification of the amount and activity of Rubisco in leaves. Methods Mol Biol. 2011;684:349-62. Nicolardi et al. (2011). The adaptive response of lichens to mercury exposure involves changes in photosynthetic machinery. Environmental Pollution (16): 1-10. Zilliges et al (2011) The Cyanobacterial Hepatotoxin Microcystin Binds to Proteins and Increases the Fitness of *Microcystis* under Oxidative Stress Conditions. PLoS ONE.

application example



0.25 µg of chlorophyl a/lane from *Spinacia oleracea* (1), *Synechococcus* PCC 7942 (2), C*yanophora paradoxa* (3), *Heterosigma akashiwo* (4), *Thalassiosira pseudonana* (5), *Euglena gracilis* (6), *Micromonas pusilla* (7), *Chlamydomonas reinhardtii* (8), *Porphyra* sp (9), *Gonyaulax polyedra* (10), *Emiliania huxleyi* (11) extracted with PEB (**AS08 300**), were separated on **4-12%** NuPage (Invitrogen) **LDS-PAGE** and blotted 1h to **nitrocellulose**. Filters were blocked 1h with 2% low-fat **milk powder** in TBS-T (0.1% TWEEN 20) and probed

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with anti-RbcL antibody (AS03 037, **1:50 000**, 1h) and secondary anti-rabbit (**1:20000,** 1 h) antibody (HRP conjugated, recommended secondary antibody AS09 602) in TBS-T containing 2% low fat milk powder. Antibody incubations were followed by washings in TBS-T. All steps were performed at RT with agitation. Blots were developed for 5 min with ECL Advance detection reagent according the manufacturers instructions (GE Healthcare). Images of the blots were obtained using a CCD imager (FluorSMax, Bio-Rad) and Quantity One software (Bio-Rad).