product information

Background RA - Ribulose bisphosphate carboxylase/oxygenase activase is an enzyme localized to chloroplasts which activates Rubisco by promoting ATP-dependent conformational changes. Alternative name: RuBisCo activase RCA.

Immunogen Purified, recombinant Rubisco activase from *Gossypium hirsutum* Q9AXG1

Host Rabbit

Clonality Polyclonal

Purity Serum

Format Lyophilized

Quantity 50 µl

Reconstitution For reconstitution add 50 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)

Related products Antibodies to RbcL, RbcS

Plant and algal protein extraction buffer

Secondary antibodies

Additional information This product can be sold containing ProClin if requested

Application information

Recommended dilution 1 : 5000-1 : 10 000 (WB)

Expected | apparent MW 47 and 42 kDa (maize, tobacco, *Chlamydomonas*)

Confirmed reactivity *Arabidopsis thaliana, Camelina sativa, Chlamydomonas reinhardtii, Hordeum spontaneum, Festuca pratensis, Glycine max, Gossypium hirsutum, Gossypium barbadense, Lollum perenne, Nanochloropsis oceanica, Nicotiana tabacum, Oryza sativa, Populus balsamifera, Rheoe discolor, Solanum lycopersicum, Zea mays, Thellungiella salsuginea, red sulfur bacterium Thiodictyon sp. Cad16 (isolated from Lake Cadagno)

Predicted reactivity *Glycine max, Gossypium mexicanum, Hordeum vulgare, Medicago sativa, Olea europea, Picea sitchensis, Physcomitrella patens, Ricinus communis, Spinacia oleracea, Triticum aestivum, Zea mays, Vitis vinifera*

Not reactive in No confirmed exceptions from predicted reactivity are currently known.

Additional information There are two forms of activase (alpha and beta) in some species (for example Arabidopsis, camelina, spinach, rice) and only one form in other species (tobacco, maize, *Chlamydomonas*). Alpha is about 46-47 Kda, beta is about 42 kDa. Species that have only one form have the beta form.

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Selected references


Application example

3, 5 and 11 µg of total soluble protein from *Arabidopsis thaliana* (1), *Oryza sativa* (2) and *Camelina sativa* (3) extracted with 50 mM Tricine-NaOH, pH 8, 10 mM EDTA, 1% PVP-40, 20 mM β-mercaptoethanol, 1 mM PMSF and 10 µM leupeptin were separated on 12 % SDS-PAGE and blotted for 1h to PVDF. Blots were blocked with 4% non-fat milk in TBS for 1h at room temperature (RT) with agitation. Blot was incubated in the primary antibody at a dilution of 1:10 000 for over night with agitation. The antibody solution was decanted and the blot was rinsed briefly with H2O, then washed six times for 15 min in TBS-T at RT with agitation. Blot was incubated in secondary antibody (anti-rabbit IgG alkaline phosphatase conjugated) diluted to 1:3000 in 0.5% non-fat milk in TBS for 2h at RT with agitation. The blot was washed with four changes of TBS-T and developed for 5 min with NBT/BCIP according to the manufacturer’s instructions (Promega). There are two forms of activase (alpha and beta). Alpha is about 46-47 Kda, beta is about 42 kDa what is shown on the blot above.

5 µg of total protein from samples such as *Arabidopsis thaliana* leaf (1), *Hordeum vulgare* leaf (2), *Zea mays* leaf (3), *Nicotiana tabacum* (4), *Chlamydomonas reinhardtii* total cell (5), 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 keep 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 (GE RPN 2125; Healthcare) 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. The antibody solution was decanted and the blot was rinsed briefly with TBS-T and agitated. Blot was incubated in secondary antibody (anti-rabbit IgG alkaline phosphatase conjugated, recommended secondary antibody AS10 1489, Agrisera) diluted to 1:25 000 in blocking reagent for 1h at room temperature with agitation. The blots were washed as above. The blot was developed for 5 min with TMA-6 (Lumigen) detection reagent according 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.