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Product no AS04 043

cFBPase | Cytosolic fructose-1,6-bisphosphatase (cytoplasm marker in photosynthetic tissues)

Product information

Immunogen	Overexpressed cytosolic fructose 1,6 bisphosphatase (cFBPase) derived from the sequence from *Arabidopsis thaliana* cFBPase UniProt: Q9MA79, TAIR: AT1G43670
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 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	Kinetic and allosteric properties of the plant cytosolic FBPase are remarkably similar to the mammalian and yeast FBPase, but differ greatly from those of the chloroplastic FBPase. The antibody could detect FBPase from the human COS-7 cell line transfected with FBP1 expressing vector. This product can be sold containing ProClin if requested.

Application information

Recommended dilution	1: 500 (IL), 1 : 5 000 (WB)
Expected | apparent MW	45 | 37 kDa (*Arabidopsis thaliana*)
Confirmed reactivity	*Arabidopsis thaliana, Brassica napus, Macroptilium atropurpureum, Nicotiana benthamiana, Pinus silvestris, Pinus yunanniensis, Oryza sativa*, *Petunia hybrida* cv. Mitchell, *Solanum tuberosum*, *Zea mays*
Predicted reactivity	*Capsella rubella, Pisum sativum, Ricinus communis, Glycine max, Phaseolus vulgaris, Sesamum indicum, Spinacia oleracea, Populus trichocarpa, Vitis vinifera* Species of your interest not listed? Contact us
Not reactive in	*Chlamydomonas reinhardtii*
Additional information	This antibody does not react with chloroplastic form of FBPase. Will this antibody be good as a cytosolic (non-microsomal control) in *Arabidopsis thaliana* roots? Although it has never been tested there is every likelihood that cFBPase will be expressed at reasonable levels even in roots. Even though the biosynthetic flux through to Sucrose may not be high as in mesophyll cells, central metabolism will still be active in young roots and the Sucrose etc being supplied externally still needs to be utilised.
Selected references	Singh, Muthamilarasan, Prasad (2022). SiHSFA2e regulated expression of SisHSP21.9 maintains chloroplast proteome integrity under high temperature stress. Cell Mol Life Sci. 2022;79(11):580. Published 2022 Nov 3. doi:10.1007/s00018-022-04611-12 Cui, Liu, Li, et al. (2022) The cellulose--lignin balance affects the twisted growth of Yunnan pine trunk. Authorea. October 10, 2022. DOI: 10.22541/au.166538021.18232197/v2 Wang et al. (2022), Arabidopsis Ubiquitin-Conjugating Enzymes UBC4, UBC5, and UBC6 Have Major Functions in Sugar Metabolism and Leaf Senescence, Int. J. Mol. Sci. 2022, 23(19), 11143; https://doi.org/10.3390/ijms231911143 He, Gao, Luo, et al. (2022) VAMP724 and VAMP726 are involved in autophagosome formation in Arabidopsis thaliana [published online ahead of print, 2022 Oct 13]. Autophagy. 2022;1-18. doi:10.1080/15548627.2022.2127240 Lim et al (2022). Arabidopsis guard cell chloroplasts import cytosolic ATP for starch turnover and stomatal opening. Nat Commun. 2022 Feb 3;13(1):652. doi: 10.1038/s41467-022-28263-2. PMID: 35115512; PMCID: PMC8814037.

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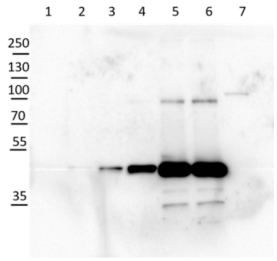
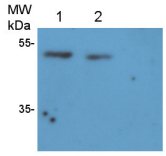
Application example



10 µg of *Arabidopsis thaliana* Col-0 WT chloroplast total protein **(1)**, 10µg Col-0 WT chloroplast stroma protein **(2)**, Col-0 WT total leaf sample 1:10 dilution **(3)**, Col-0 WT total leaf sample 1:2 dilution **(4)**, Col-0 WT total leaf sample undiluted **(5)**, Col-0 WT total leaf sample undiluted **(6)** and recombinat plastidial FBPase 0.05 µg, expressed in E.coli with no cTP present in the sequence **(7)**, extracted with 2x Laemmli buffer and denatured at 95°C for 5 min. were separated on 10% SDS-PAGE and blotted to Millipore Immobilon-P membrain (carried out at 100 V for 90 min at 4°C in blotting buffer (25 mM Tris-HCl, 192 mM glycine, 10 % [v/v] methanol). Blots were blocked with blocking solution (TBST buffer (20 mM Tris-HCl pH 7.4, 150 mM NaCl, 0.05 % [v/v] Tween-20) supplemented with 5 % [w/v] milk powder) for 1h at room temperature (RT) with agitation. Blot was incubated in the primary antibody at a dilution of 1: 5 000 for 15h (over night) at 4°C with agitation in TBST. The antibody solution was decanted and the blot was washed 6 times for 10 min in TBS-T at RT with agitation. Blot was incubated in secondary antibody (anti-rabbit IgG horse radish peroxidase conjugated) diluted to 1:20 000 in for 1h at RT with agitation. The blot was washed as above and ChemiGlow West Chemiluminescence substrate was used for development according to the manufacturer's instructions and imaged using the ChemiDoc imaging system (Biorad, Cressier, France). Exposure time was 15 seconds.

Courtesy of Zanella Martina, ETH Zürich, Switzerland



2 µg of total protein from *Arabidopsis thaliana* roots crude extract **(1)**, supernatant after 10 0000 g centrifugation **(2)**, extracted with ice-cold extraction buffer [50 mM Tris-HCl pH 7.5, 0.33 M Sucrose, 5 mM EDTA, 1x proteinase inhibitor] and denatured gradually with [50 mM Tris (pH 6.8), 10% Glycerol, 2% SDS, 2.5 M Urea, 0.005% Bromophenol Blue] at first with 55°C for 15 min followed by 95°C for 5 min. Proteins were separated on 12 % SDS-PAGE. Proteins were blotted 1h to PVDF using semi-dry transfer. Blots were blocked with 5 % nonfat milk + 0.1 % BSA for 1h at room temperature (RT) with agitation. Blot was incubated in the primary antibody at a dilution of 1: 35 000 for ON/4°C with agitation in TBS-T. 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 RT with agitation. Blot was incubated in secondary antibody (anti-rabbit IgG horse radish peroxidase) diluted to 1:1 000 in for 2h at RT with agitation. The blot was washed as above and developed for 5 min withchemiluminescent detection reagent. Exposure time was 20 min.

Courtesy of Dr. Joanna Jeleńska and Dequantarius Speed, University of Chicago, USA