product AS09 458
PEPC | Phosphoenolpyruvate carboxylase

product information

Background
PEPC (phosphoenolpyruvate carboxylase), EC=4.1.1.31, belongs to an enzyme family of carboxy-lyases that is catalyzing adding CO2 to phosphoenolpyruvate (PEP) to form oxaloacetate. Alternative names: PEPCase 1, PEPCase 3, PEPC 1, PEPC 3

Immunogen
KLH-conjugated synthetic peptide well conserved PEPC1 and sequences from different plant species including Arabidopsis thaliana Q9MAH0, At1g53310 (PEPC 1), Q84VW9, At3g14940 (PEPC 3). The peptide chosen to elicit this antibody is also perfectly conserved in bacterial type of this enzyme NP_177043.2 (PEPC 4).

For Zea mays, the peptide is conserved in PEP1 and PEP4 isoforms.

Host
Rabbit

Clonality
Polyclonal

Purity
Affinity purified serum in PBS, pH 7.4

Format
Lyophilized

Quantity
50 µg

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. Please do not re-use this primary antibody solution. In case of cyanobacterial samples there will be no signal in your second incubation.

Tested applications
Immunolocalization (IL), Western blot (WB)

Related products
AS07 241 | anti-PEPCK | PEP carboxy kinase

Application information

Recommended dilution
1 : 500 (IL), 1 : 1000 (WB)

Expected | apparent MW
110 | 105 kDa

Confirmed reactivity
Ananas comosus, Arabidopsis thaliana, Cenchrus ciliaris, Choris gayana, Chromera velia, Hordeum vulgare, Jatropha curcas, Leptochloa fusca, Lupinus sp., Megathysrus maximus, Mesembryanthemum crystallinum, Nicotiana tabacum, Oryza sativa, Panicum antidotale, Panicum coloratum, Pinus strobus, Saccharum spp. hybrid clone C91-301, Salsola lanata, Salsola lanigera, Sorghum bicolor, Synechocystis PCC 6803, Phaeodactylum tricornutum (strain CCAP 1055/1), Pinus strobus, Thalassiosira weissflogii, Zea mays, Zostera muelleri

Predicted reactivity

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

Additional information
Antibody can be also used following 2D gel electrophoresis.

Selected references
Wen et al. (2017). Possible involvement of phosphoenolpyruvate carboxylase and NAD-malic enzyme in response to
Application example

**5 µg of total protein** from (1) *Arabidopsis thaliana* leaf extracted with Protein Extraction Buffer, PEB (AS09 300) , (2) *Spinacia oleracea* total cell, extracted with PEB, (3) *Hordeum vulgare* total cell extracted with PEB, (4) *Zea mays* total cell extracted with PEB, 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) diluted to 1: 75 000 for 1h at room temperature with agitation. The blot was 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).

10 µg of total protein extracted freshly from *Arabidopsis thaliana* wt leaf tissue (At, non-senescent leaves), *Arabidopsis thaliana* wt leaf tissue (At, senescent leaves), *Pinus strobus* needle tissue (PS, PSd) with 1 M Tris-HCl, pH 6.8, 10 % SDS, 15 % sucrose, 0.5 DTT and denatured at 75°C for 5 min. were separated on 10 % Bis-Tris Nupage Novex gel (120 V/45 min. using MES buffer system) and blotted 30 min. to PVDF. Blot was blocked with 5 % non-fat milk 45 min. at RT with agitation. Blot was incubated in the primary antibody at a dilution of 1:1000 for 1h at RT with agitation. The antibody solution was decanted and the blot was rinsed briefly twice for 10 min. in TBS at RT with agitation. Blot was incubated in Agrisera matching secondary antibody (anti-rabbit IgG horse radish peroxidase conjugated, AS09 602) diluted to 1:75 000 in for 1h/RT with agitation. The blot was washed as above and developed using chemiluminescence detection. Exposure time was 40 seconds.

 Courtesy of Dr. Christine Yao-Yun Chang and the Ensminger lab, University of Toronto, Canada