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Product no AS21 4567

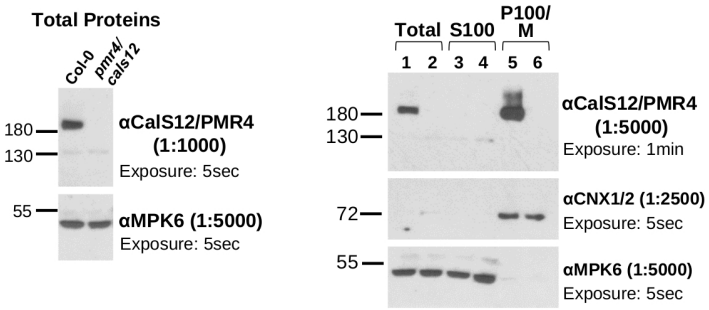
Anti-CALS12/PMR4 | Callose synthase 12

Product information

Immunogen	KLH-conjugated peptide derived from *Arabidopsis thaliana* CALS12 protein sequence, UniProt: Q9ZT82 , TAIR: At4g03550
Host	Rabbit
Clonality	Polyclonal
Purity	Antigen 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.

Application information

Recommended dilution	1 : 1000 (WB)
Expected | apparent MW	206.9 | 185 kDa
Confirmed reactivity	*Arabidopsis thaliana*
Predicted reactivity	*Capsella rubella, Camelina sativa, Eutrema salsugineum, Brassica napus, Brassica oleracea, Brassica rapa, Tarenaya hassleriana* Species of your interest not listed? Contact us
Not reactive in	*Nicotiana benthamiana, Solanum tuberosum*
Selected references	To be added when available, antibody available in September 2022.



Samples:

1: Col-0 – Total protein extract
2: pmr4-1/cals12 – Total protein extract
3: Col-0 – soluble protein fraction (S100)
4: pmr4-1/cals12 – soluble protein fraction (S100)
5: Col-0 – microsomal protein fraction (P100/M)
6: pmr4-1/cals12 – microsomal protein fraction (P100/M)

Total proteins were isolated from 60 *Arabidopsis thaliana* seedlings (10-day-old) of Col-0 (wild- type; lanes 1, 3, 5) and pmr4-1/cals12 null mutant [ref 1] (lanes 2, 4, 6). Using differential centrifugation, the total protein fractions (lanes 1, 2) were separated into soluble proteins (S100; lanes 3, 4) and microsomal proteins (M/P100; lanes 5, 6) as previously published by our lab [ref 2]. Proteins were denatured at 65°C for 5 min. 30 µg of proteins were separated on an 8 % SDS-PAGE and transferred for 70 min at 55V using a tank transfer system to nitrocellulose membrane. Blots were blocked with 1x PBS + 0.1 % Tween 20 (PBS-T) + 5% milk for 1 h at room temperature (RT) with agitation. To test primary -CalS12/PMR4 by Agrisera (AS21 4567), different dilutions of the antibody were used as indicated in the figure. Membrane portions probed with -CNX1/2 (AS12 2365, membrane ER) and -MPK6 [ref 1] served as loading controls. Blots were incubated with the primary antibodies overnight at 4°C with agitation in 1x PBS-T + 5% milk. The primary antibody solutions were decanted, and the blots were washed 4 times (6-8 minutes each) in 1x

PBS-T at RT with agitation prior to incubation with secondary antibody Goat anti Rabbit IgG (H&L) –HRP conjugated (AS09 602-trial) diluted to 1 : 20,000 in 1x PBS-T + 5% milk for 2 h at RT with agitation. The blots were washed as above and developed for 4 min with chemiluminescent detection reagent ECL Bright (AS16 ECL-N). Exposure time to X-ray films was indicated in figure.

Courtesy of Kelly Mason and Antje Heese; University of Missouri, Div. Biochemistry, IPG (USA)

References

[1] Mason K, Ekanayake G, Heese A (2020). Staining and automated image quantification of callose in Arabidopsis cotyledons and leaves. Methods Cell Biology: Plant Cell Biology 160:181-199.
[2] LaMontagne, E.D., Collins, C.A., Peck, S.C. and Heese, A. 2016. Isolation of microsomal membrane proteins from Arabidopsis thaliana. Curr. Protoc. Plant Biol. 1:217-234. doi: 10.1002/cppb.20020