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Product no AS08 343A

Cyt c | Cytochrome c

Product information

Immunogen	KLH-conjugated synthetic peptide derived from *Arabidopsis thaliana* cytochrome c protein sequence, UniProt:D7KMK0 (C-1) D7LY03 (C-2), TAIR: At1g22840 (Cytc1) and At4g10040 (Cytc2)
Host	Rabbit
Clonality	Polyclonal
Purity	Immunogen 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 at -20°C; once reconstituted make aliquots to avoid repeated freeze-thaw cycles and Store at -80°C. 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.

Application information

Recommended dilution	1: 100 (IL), 1 : 5000 (WB)
Expected | apparent MW	12.5 | 14 kDa (for *Arabidopsis thaliana)*
Confirmed reactivity	*Arabidopsis thaliana, Brassica oleracea, Glycine max, Pisum sativum, Zea mays*
Predicted reactivity	cytc1 and cytc2 from following species: *A. theoprasi, Brassica napus, Brassica oleracea, Cannabis sativa, C. maxima, Chlamydomonas reinhardtii (*peptide target partially conserved*), Lupinus luteus, Medicago truncatula, Nicotiana tabacum, Oryza sativa, Ostreococcus (*peptide target partially conserved*), P. aurea, Physcomitrium patens, Ricinus communis, S. nigra, Solanum lycopersivum, Vitis vinifera.* Species of your interest not listed? Contact us
Not reactive in	*Arabidopsis thaliana* CytC6, *Chinesecuscuta* sp.
Additional information	https://www.agrisera.com/en/artiklar/goat-anti-rabbit-igg-hl-ap-conjugated-.htmlThe presence of cytochrome c in the cysotol is a marker of PCD (programmed cell death)
Selected references	Soria et al. (2024).Functional resilience: An active oxidative phosphorylation system prevails amid foreign proteins in holoparasitic plants. Current Plant Biology Volume 37, March 2024, 100322. Canal et al. (2024).Cytochrome c levels affect the TOR pathway to regulate growth and metabolism under energy-deficient conditions. New Phytol. 2024 Mar;241(5):2039-2058. Guo et al. (2021) The pentatricopeptide repeat protein GEND1 is required for root development and high temperature tolerance in Arabidopsis thaliana,Biochemical and Biophysical Research Communications,Volume 578,2021,Pages 63-69,ISSN 0006-291X,https://doi.org/10.1016/j.bbrc.2021.09.022.(https://www.sciencedirect.com/science/article/pii/S0006291X21013164) Wang et al. (2020) Rerouting of ribosomal proteins into splicing in plant organelles. BioRxiv, DOI: 10.1101/2020.03.03.974766 . Dai et al. (2020). Pentatricopeptide repeat protein DEK46 is required for multi-sites mitochondrial RNA editing and maize seed development. J Exp Bot. 2020 Jul 25;eraa348.doi: 10.1093/jxb/eraa348. Waltz et al. (2019). Small is big in Arabidopsis mitochondrial ribosome. Nat Plants. 2019 Jan;5(1):106-117. doi: 10.1038/s41477-018-0339-y. Doronina et al. (2019). Structural and Functional Features of the Wheat Embryo Sac's Antipodal Cells during Differentiation. Russ J Dev Biol 50, 194–208. (immunolocalization)

Application example

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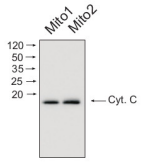
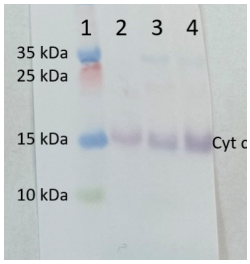


Mitochondrial proteins (15 ug) from *Arabidopsis thaliana* mitochondria was separated on 16% acrilamide gel and electrophoresis prepared according to Schägger and von Jagov (Anl. Biochem., 1987, 166:368-379). After running the gel, proteins were transferred to PVDF membrane using wet transfer (Roti®-Blot 2, Roth). Transfer was checked by Ponceau S staining. Blot was destained by several quick washings in distilled water and 1 washing in 1X TBS (10 mM T pH 7.5, 150 mM NaCl) (10-15 min.).Blot was blocked by 1.5 hour in 5% milk in TBST (1X TBS, 0,1% Tween 20) After blocking blot was washed quickly twice in TBST and incubated 2 hours with primary antibody (dilution 1: 1000) in TBST. Washing: two quick washings in TBST and 3 x 10 min. washings in TBST. Then blot was incubated 45-60 min. with a secondary anti-rabbit antibodies conjugated to peroxidase (Agrisera AB, dilution 1:10 000, AS09 602) in TBST. Washing: as above. After washing blot was incubated 1-2 min. in chemiluminescent detection reagent. Chemiluminescence was detected by BioSpectrum® Imaging System (UVP). Exposure time was 5 seconds.

Courtesy Dr. Janusz Piechota, Wrocław University, Poland



Lane:

1 - MW marker

2 - 10 ug of *Arabidopsis thaliana* whole leaf extract

3 - 10 ug of *Zea mays* whole leaf extract

4- 10 ug of *Solanum lycopersicum* whole leaf extract

10 µg/well of total protein extracted from *Arabidopsis thaliana*, *Zea mays* and *Solanum lycopersicum*, total cell extract, stored at -80°C. Exact buffer components were 1xPEB (AS08 300) with protease inhibitor (0.1 mg/ml for Pefabloc SC, Roche) and denatured with Invitrogen LDS sample buffer (4X) at 70°C/5 min. Samples were separated on Invitrogen NuPage Bis-Tris 4-12% SDS-PAGE and blotted for 1 h to Invitrogen PVDF (pore size of 0.2 um), using wet transfer. Blot was blocked with 5% milk in TBS-T for 1h/RT with agitation. Blot was incubated in the primary antibody at a dilution of 1: 500 for 1h/RT with TBS-T Blocking. 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 matching secondary antibody (anti-rabbit IgG ALP conjugated, **AS09 607** lot 2302) diluted to 1: 1 000 in TBS-T Blocking for 0,5h/RT with agitation. The blot was washed as above and developed with **AS19 BCIP-NBT-PLUS** lot 09269221 for 3min. As soon as the desired band was detectable, the membrane was washed in generous amounts of deionized water before placing the membrane on Whatman paper to dry. Image was captured after 1 h.

Courtesy Agrisera