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product AS09 468

V-ATPase, c | vacuolar H+-ATPase, subunit c (16 kDa)

product information

background	**V-ATPase c subunit** is located in vacuole and is involved in ATP synthesis cuopled proton transport. This protein is coded by ATVHA-C3 gene. Alternative names: AT4g34720/T4L20_300
immunogen	KLH-conjugated synthetic peptide derived from *Arabidopsis thaliana* V-ATPase subunit c,Q6IDA4
antibody format	rabbit polyclonal, serum,
quantity	100 µl
storage	store at -20°C; 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 material adhering to the cap or sides of the tubes.
tested applications	ELISA (ELISA), western blot (WB)
additional information	**Antibody will only work properly on purified vacuolar membranes** 0.1 % sodium azide is added as preservative. For antibody re-suspending information check the tube lable. Antibodies will detect target protein in a few µg of a crude preparation loaded per well. If purified preparations of vacuolar membranes are used, one µg load per well should be sufficient. Subunit c is one of most hydrophobic proteins (can be dissolved in organic solvent such as a mixture of chloroform/methanol solution). It is prone to aggregation even in the presence of SDS. **Therefore, before loading on the gel membrane fractions should be incubated in buffer containing 2 % SDS at 60° or 70°C for 10 min or at 25°C for 30 min.** Protocol of isolation of plant vacuolar membranes can be found here.

application information

recommended dilution	1: 8000 (ELISA), 1: 2000 with standard ECL (WB)
expected | apparent MW	16 | 16 kDa (*Arabidopsis thaliana*)
confirmed reactivity	*Arabidopsis thaliana, Raphanus sativus*
predicted reactivity	dictos including: *Cucumis sativus, Gossypium mexicanum, Phaseolus aureus, Rcicinus communis*, monocots including: *Oryza sativa, Triticum aestivum, Zea mays*, trees: *Picea sitchensis, Populus trichocarpa*

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not reactive in	no confirmed exceptions from predicted reactivity known in the moment
additional information	Protein or membrane sample should be treated at 70°C for 10 min before loading on the gel. Diluted antibody solution can be used 2 to 3 times within one month if it contains 0.1 % sodium azide as preservative and is stored at -20ºC to -80ºC. Manufactured by Operon Biotechnologies.
selected references	Yoshihiro et al. (2006) Immunochemical analysis of aquaporin isoforms in Arabidopsis suspension-cultured cells. Cells. Biosci.Biotechnol. Biochem. 70: 980-987.

application example

1 µg, 5 and 10 µg of vacuolar membrane fraction/lane from Raphanus sativus were separated on 12 % **SDS-PAGE** and blotted 1h to PVDF membrane (40 min. at 10 V using BioRad semidry transfer). Filters were blocked 1h with 5 % low-fat **milk powder** in TBS-T (0.05% Triton X.100). Membranes were washed 5 times with TBS-T, each time in a fresh polystyrene box and probed with anti-V-ATPase c subunit antibodies (AS09 468, **1:1000**, 1h) and secondary anti-rabbit (**1:2000,** 1 h). All steps were performed in RT with agitation.