

product **AS09 577A**

V-ATPase | epsilon subunit of tonoplast H⁺ATPase (affinity purified)

product information

background	Plant vacuole V-ATPase is responsible for energization of transport of ions and metabolites, and acts as well 'house-keeping' and as a stress response enzyme. V-ATPase is a multi-subunit enzyme composed of a membrane sector and a cytosolic catalytic sector. It is related to the FoF1 ATP synthase. Alternative protein names: Vacuolar proton pump subunit E, Protein EMBRYO DEFECTIVE 2448
immunogen	<u>KLH</u> -conjugated synthetic peptide chosen from subunit E of plant V-ATPase including <i>Arabidopsis thaliana</i> <u>At4g11150</u> . Peptide is conserved in vacuolar H ⁺ -ATPase subunit E, isoform 1 to 3 (VHA-E1).
antibody format	goat polyclonal affinity purified serum lyophilized
quantity	200 µg for reconstitution add 100 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.
tested applications	western blot, (WB)
additional information	to be added when available

application information

recommended dilution	1 : 1 000 with alkaline phosphatase or 1: 3000 with ECL (WB)
expected apparent MW	26 31 kDa (<i>Arabidopsis thaliana</i>)
confirmed reactivity	dicots including: <i>Arabidopsis thaliana</i> , <i>Nicotiana tabacum</i> , <i>Solanum lycopersicum</i> , monocots including: <i>Avena strigosa</i>
predicted reactivity	dicots including: <i>Lycopersicum esculentum</i> , <i>Mesembryanthemum sp.</i> , <i>Petunia sp.</i> , <i>Phaseolus sp.</i> , <i>Pteris vittata</i> (fern), <i>Ricinus communis</i> , <i>Vitis vinifera</i> and monocots including <i>Hordeum vulgare</i> , <i>Oryza sativa</i> , <i>Zea mays</i> , algae, <i>Physcomitrella patens</i> , <i>Chlamydomonas reinhardtii</i> , <i>Thellungiella sp.</i> , bull frog, chicken, bovine, <i>Drosophila melanogaster</i> , human, mouse, rat
not reactive in	no confirmed exceptions from predicted reactivity known in the moment
additional information	V-ATPase is very sensitive for the redox of the SDS buffer. We recommend using at least 50-100 mM DTT freshly prepared before handling the sample.

2 hours incubation with primary antibody is recommended over over night incubation which can contribute to increased background.

selected references to be added when available. Antibody released in November 2009

application example

6 µg of total SDS-extracted protein from *Avena strigosa* roots (R) and leaves (L) , were separated on NuPage LDS-PAGE 4-12% gradient acrylamide gel (Invitrogen) and blotted 1h to nitrocellulose. Filters were blocked 1h with 5% low-fat milk powder in TBS and probed with anti-V-ATPase antibodies (AS09 577 , 1:2000, 1h) and secondary anti-goat (1:5000, 1 h) antibody (Alexa 647) in TBS containing 5% low fat milk powder. Antibody incubations were followed by washings in TBS-T (containing 0.05% Tween-20, 0.1% Triton X-100) . All steps were performed at RT with agitation. Blots were scanned with a Typhoon scanner.

Courtesy Dr. Sam Mugford (JIC)

