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**Product no AS20 4408**

# Anti-Delta-VPE | Vacuolar-processing enzyme delta-isozyme

## Product information

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|---|---|
| **Immunogen** | Recombinant, His6-tagged, delta-VPE from *Arabidopsis thaliana,* UniProt: Q9LJX8, TAIR: At3g20210 |
| **Host** | Rabbit |
| **Clonality** | Polyclonal |
| **Purity** | Total IgG. Protein A purified in PBS, 50% glycerol. Filter sterilized. |
| **Format** | Liquid at 2 mg/ml. |
| **Quantity** | 100 µg |
| **Storage** | Store at -20°C; once reconstituted make aliquots to avoid repeated freeze-thaw cycles. 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

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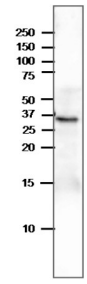
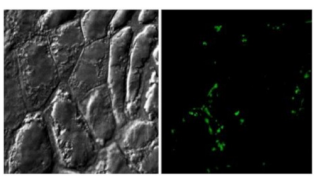
| **Recommended dilution** | 1: 500 (IF), 1: 5000 (IL), 1: 5000 (WB) |
| **Expected \| apparent MW** | 52 kDa (inactive precursor) \| 37-38 kDa (mature, active form) |
| **Confirmed reactivity** | *Arabidopsis thaliana* |
| **Predicted reactivity** | *Camelina sativa, Capsella rubella, Eutrema salsugineum, Raphanus sativus* Species of your interest not listed? Contact us |
| **Not reactive in** | No confirmed exceptions from predicted reactivity are currently known |
| **Additional information** | Subcellular localisation is seed specific and restricted to developing seeds at 7 days after anthesis, Delta-VPE is detected at lower levels in flowers siliques, specifically in seed coats |
| **Selected references** | Kunieda et al. (2013). Spatiotemporal secretion of PEROXIDASE36 is required for seed coat mucilage extrusion in Arabidopsis. Plant Cell . 2013 Apr;25(4):1355-67.doi: 10.1105/tpc.113.110072. (Western blot, Arabidopsis thaliana)<br>Kunieda et al. (2008). NAC family proteins NARS1/NAC2 and NARS2/NAM in the outer integument regulate embryogenesis in Arabidopsis. Plant Cell. 2008 Oct;20(10):2631-42.doi: 10.1105/tpc.108.060160. (Western blot, Arabidopsis thaliana)<br>Nakaune et al. (2005). A vacuolar processing enzyme, deltaVPE, is involved in seed coat formation at the early stage of seed development. Plant Cell. 2005 Mar;17(3):876-87. doi: 10.1105/tpc.104.026872. (Immunofluorescence, Immunolocalisation by electron microscopy, Western blot, Arabidopsis thaliana) |




*Arabidopsis thaliana* maturing siliques was freshly extracted to a crude extract with 2x SDS-sample buffer (+ 2ME) for SDS-PAGE and denatured with 4X SDS buffer at 95°C for 5 min. were separated on 15-20 % SDS-PAGE and blotted to PVDF membrane in wet system. Blot was blocked with 3 % skim milk/TBS-T, 1h/RT with agitation. Blot was incubated in the primary antibody at a dilution of 1: 5000 in TBS-T for 1h/RT. The antibody solution was decanted and the blot was washed 4 times for 10 min in TBS-T at RT with agitation. Blot was incubated in matching secondary antibody (anti-rabbit IgG horse radish peroxidase conjugated) diluted to 1:10 000 in for 1h/RT with agitation. The blot was washed as above and developed with a chemiluminescent detection reagent, following manufacture's recommendations.



Left panel: differential Interference Contrast Image
Right panel: Immunofluorescent staining
Material: developing seeds of *Arabidopsis thaliana*
Fixation: 7.2% (w/v) formaldehyde, 0.1% (v/v) Nonidet P-40, 10% (v/v) dimethyl sulfoxide, and 50 mM Na-phosphate buffer, pH 7.2 for 40 minutes.
Wash, 2x: TBS-T for 5 min, incubated in TBS-T containing 5% (w/v) Cellulase Onozuka R-10 and 2% (w/v) Pectolyase Y-23 for 20 min at 30°C
Was: 2x with TBS-T
Blocking: 30 min. in 2 % BSA in TBS-T
Primary antibody: 1: 500, 40 min.
Wash: 3x5 min. with TBS-T
Secondary antibodies: goat anti-rabbit AlexaFluor 488 conjugated
Wash: 3x 5 min. TBS-T and mounted