

product **AS11 1747**
DHAR2 | Dehydroascorbate Reductase 2

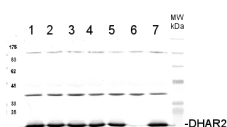
product information

background	DHAR2 (Dehydroascorbate Reductase 2) the protein is induced by jasmonic acid and oxidative chemical stresses and is a key component of the ascorbate recycling system. Involved in redox homeostasis under biotic and abiotic inducers. Localized in cytoplasm. Synonymes: chloride intracellular channel homolog 2, CLIC homolog 2, glutathione-dependent dehydroascorbate reductase 2, DHAR2, CytDHAR, GSH-dependent dehydroascorbate reductase 2.
immunogen	<u>KLH</u> -conjugated synthetic peptide derived from known DHAR1 sequence of <i>Arabidopsis thaliana</i> <u>Q9FRL8</u> , <u>At1g75270</u>
antibody format	rabbit polyclonal affinity purified serum lyophilized
quantity	200 µg for reconstitution add 200 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)
related products	<u>AS11 1746</u> Anti-DHAR1, rabbit antibodies
additional information	to be added when available

application information

recommended dilution	1 : 5000 with standard ECL (WB)
expected apparent MW	23.6 23.4 kDa
confirmed reactivity	<i>Arabidopsis thaliana</i>
predicted reactivity	dicots including: <i>Ricinus communis</i> , trees: <i>Populus trichocarpa</i>
not reactive in	no confirmed exceptions from predicted reactivity are currently known
additional information	to be added when available
selected references	<u>Grefen</u> et al. (2009). The determination of protein-protein interactions by the mating-based split-ubiquitin system (mbSUS). <i>Methods Mol Biol</i> 479:217-233.

application example



1cm² of a leaf from *Arabidopsis thaliana* Col-0 (1) and or t-DNA insertion lines dhar1-1 (2), dhar1-2 (3), dhar1-3 (4), dhar2-1 (5), dhar2-2 (6), dhar1-3 EOS-DHAR1 (7), was extracted using 200µl Lyse&Load-Buffer (Grefen et al. 2009). 10 µl were separated on a 15% SDS-PAGE and blotted 1h to PVDF (using Bjerrum Buffer in a semidry blot). Blots were blocked with 5% Milk in 1xTBS-Tween20 (1%) for 1h at room temperature (RT) with agitation. Blot was incubated in the primary antibody at a dilution of 1:5000 (in 5% Milk 1xTBS-Tween20 (1%) + 0.01 % NaN₃) ON at 4°C with agitation. The antibody solution was decanted and the blot was washed 3 times for 10 minutes with 1x TBS-Tween20 at RT with agitation. Blot was incubated in secondary antibody BioRad

anti-rabbit IgG AP-conjugate (#170-6518) diluted to 1:2000 in 5% Milk 1xTBS-Tween20 (1%) + 0.01 % NaN₃ for 1h at RT with agitation. The blot was washed as above, equilibrated in staining buffer (100mM Tris-HCl, 100mM NaCl, 5mM MgCl₂, see Grefen et al. 2009) and developed for 5-15 min. with staining solution (Nitro blue tetrazolium chloride (NBT) and 5-bromo-4-chloro-3-indoylphosphate-p-toluidin (BCIP) in staining buffer).

Courtesy Dr. Christopher Grefen, UK