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product AS10 685

ADH | alcohol dehydrogenase (hypoxia marker)

product information

background	Alcohol dehydrogenase **(ADH)** is an enzyme playing a crucial role in the fermentative metabolism in plants subjected to low oxygen stress. It is known to be synthesized preferentially under low oxygen conditions.
immunogen	KLH-conjugated peptide derived from available ADH sequences including *Arabidopsis thaliana* P06525
antibody format	rabbit polyclonal serum lyophilized
quantity	100 µl 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: 3000 with standard ECL (WB)
expected | apparent MW	42 | 42 kDa (*Arabidopsis thaliana*)
confirmed reactivity	*Arabidopsis thaliana, Oryza sativa*
predicted reactivity	dicots including: *Brassica napus, Glycine max*, *Pisum sativum, Solanum tuberosum, Sorghum bicolor, Ricinus communis, Vitis vinifera,* monocots including: *Hordeum vulgare, Oryza sativa, Zea mays*, trees: *Picea sitchensis, Populus trichocarpa*,
not reactive in	Allyl alcohol dehydrogenase of *Nicotiana tabacum*, accession 75206691
additional information	to be added when available
selected references	to be added when available, antibody released in September 2010

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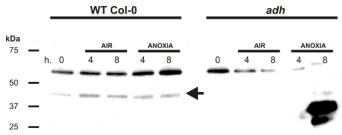
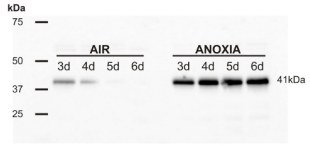
application example



20 µg of total protein from *Arabidopsis thaliana* seedlings (0-4-8 hours of anoxic treatment with aerobic control) of WT Col-0 and adh mutant extracted with an SDS Extraction Buffer (60mM Tris-HCl pH 8.0, 2% SDS, 1,5% Sucrose) were separated on XT CRITERION 10%Bis-Tris (BioRad) SDS-PAGE and blotted 1h to PVDF. Blot was blocked immediately in milk in TBS-T for 1h at room temperature (RT) with agitation. Blot was incubated in the anti-ADH antibodies at a diluition of 1: 3000 in milk in TBS-T for 3h at RT with agitation. Blot was incubated in secondary antibody (goat anti-rabbit IgG HRP conjugated from Agrisera, AS09 602) diluited 1:20 000 in milk in TBS-T for 50 min at RT and then washed as above and developed for 2 min with standard ECL. Images of the blot were obtained using BioSpectrum AC Imaging System (UVP). Exposure time was 10 min The arrow indicates ADH (42kDa, as expected) . There is a cross reacting band in *Arabidopsis thaliana* between 50-70 kDa. *The large band in the right corner of the membrane is likely a staining artefact.*



20 µg of total protein from *Orysa sativa* coleoptiles (3-4-5-6 days of germination under aerobic and anoxic conditions) extracted with an SDS Extraction Buffer (60mM Tris-HCl pH 8.0, 2% SDS, 1,5% Sucrose) were separated on XT CRITERION 10% Bis-Tris (BioRad) SDS-PAGE and blotted 1h to PVDF. The blot was blocked immediately in milk in TBS-T for 1h at room temperature (RT) with agitation. Blot was incubated in the anti-ADH antibodies at a diluition of 1: 3000 in milk in TBS-T for over night with agitation. Blot was incubated in secondary antibody (goat anti-rabbit IgG HRP conjugated from Agrisera, AS09 602) diluted 1:20 000 in milk in TBS-T for 50 min at RT and then washed as above and developed for 2 min with standard ECL. Images of the blot were obtained using BioSpectrum AC Imaging System (UVP). Exposure time was 10 min. The band corresponds to ADH (41 kDa).
Courtesy Dr. Eleonora Paparelli,Scuola Superiore Sant'Anna, Italy