

product **AS10 699**
AOX | alternative oxidase

product information

background	Alternative oxidases (AOX) are quinol oxidases located in the inner mitochondrial membrane of plants. They function as terminal oxidases in the alternate electron transport pathway, oxidizing ubiquinone to reduce oxygen to water.
immunogen	alternative oxidase purified from <i>Sauromatum guttatum</i> (the voodoo lily)
antibody format	mouse monoclonal IgG1 serum liquid
quantity	1,5 ml
storage	store at -20°C; for long term storage at store at -80°C ; 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), immunoprecipitation (IP), antibody column
additional information	antibody is supplied in cell culture medium with 1mM sodium azide as preservative Protocol for plant mitochondria isolation can be found here.

application information

recommended dilution	1 : 100 with standard ECL (WB, IP)
expected apparent MW	36-40 36-40 for <i>Arabidopsis thaliana</i>
confirmed reactivity	<i>Arabidopsis thaliana</i> , <i>Glycine max</i> (AOX1,2,3), <i>Sauromatum guttatum</i>
predicted reactivity	dicots including: <i>Nicotiana tabacum</i> , monocots including: <i>Oryza sativa</i> , diatoms, fungus <i>Aspergillus sp.</i> , <i>Neurospora crassa</i> , yeast, protozoa: <i>Trypanosoma sp.</i>
not reactive in	no confirmed exceptions from predicted reactivity known in the moment
additional information	Antibody is recognizing native and denatured AOX
selected references	Costa-de-Oliveira et al. (2012). An alternative respiratory pathway on <i>Candida krusei</i> : implications on susceptibility profile and oxidative stress. FEMSyeat Res. January 23 (ahead of print). Elthon et al. (1989). Monoclonal antibodies to the alternative oxidase of higher plant mitochondria. Plant Physiol. 89:1311-1317.

application example

25 µg of *Arabidopsis thaliana* mitochondrial wild type fraction (**1**) mitochondrial fraction from a mutant with increased AOX level (**2**), total wild type leaf extract (**3**), total leaf extract from AOX overproducing mutant (**4**) were separated on 10% gel and blotted on **nitrocellulose** membrane using wet transfer (0.22% CAPS, pH 11). Filters were blocked (1.5h) in 5% milk in TBST (1X TBS, 0,1% Tween 20), incubated with 1: 100 anti-AOX monoclonal antibodies (2h in TBST) followed by 1 h incubation with 1: 10 000 Agrisera secondary anti-mouse HRP-coupled antibodies ([AS09 627](#)) and visualized with standard ECL on Kodak autoradiography film for 15-60 s. Mitochondria were isolated as described by [Urantowka](#) et al. (Plant Mol Biol, 2005, 59:239-52). Mitochondrial pellets were suspended in 1X Laemmli buffer (5% beta-mercaptoetanol, 3.7% glycerol, 1.1% SDS, 23 mM Tris- HCl pH 6.8, 0.01% bromophenol blue), heated (95°C, 5 min.) and centrifuged (13 000rpm, 1 min.). Leaf extracts were prepared as described by [Martinez-Garcia](#) et al. (Plant J., 1999, 20:251-7).

Courtesy Dr. Janusz Piechota, Wrocław University, Poland

