

product **AS07 204**

FtsH3 + FtsH10 | ATP-dependent zinc metalloprotease FtsH3 + FtsH10 (mitochondrial)

product information

Background	One of the several classes of mitochondrial proteases is membrane bound, ATPdependent FtsH protease. Their function is very important for the control of protein quality and quantity by degradation of unassembled subunits. Other names: AtFtsH3, cell division protease ftsH homolog 3, mitochondrial, AtFtsH10, cell division protease ftsH homolog 10, mitochondrial
Immunogen	<u>KLH</u> -conjugated peptide derived from sequences of <i>Arabidopsis thaliana</i> FtsH3 and FtsH10 with localization to mitochondria <u>Q84WU8</u> , <u>At2g29080</u> and <u>Q8VZI8</u> , <u>At1g07510</u>
Host	Rabbit
Clonality	Polyclonal
Purity	Affinity purified serum in PBS, pH 7.4
Format	Lyophilized in PBS pH 7.4
Quantity	200 µg
Reconstitution	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	Blue Native PAGE (BN-PAGE/SDS-PAGE), Western blot (WB)
Related products	AS11 1789S FtsH2 positive control/quantitation standard AS11 1789 anti-FtsH1-11 ATP-dependent zinc metalloprotease FtsH1-11 AS16 3930 anti-FtsH1 + FtsH5 ATP-dependent zinc metalloprotease FtsH1 + FtsH5 (chloroplastic) AS16 3929 anti-FtsH2 + FtsH8 ATP-dependent zinc metalloprotease FtsH2 + FtsH8 (chloroplastic) AS07 205 anti-FtsH4 ATP-dependent zinc metalloprotease FtsH4 (mitochondrial) AS05 094A anti-FtsH6 ATP-dependent zinc metalloprotease FtsH6 (chloroplastic) AS06 130 anti-FtsH9 ATP-dependent zinc metalloprotease FtsH9 (chloroplastic) AS07 251 anti-FtsH10 ATP-dependent zinc metalloprotease FtsH10 (mitochondrial) Antibodies to other proteins involved in photosynthesis Plant protein extraction buffer Secondary antibodies

Additional information | Blue-native (2D BN/SDS-PAGE) methodology is described in Piechota et al. 2010

Application information

Recommended dilution	1 : 500-1 : 1000 (WB)
Expected apparent MW	80 kDa
Confirmed reactivity	<i>Arabidopsis thaliana</i>
Predicted reactivity	<i>Arabidopsis thaliana</i>
Not reactive in	No confirmed exceptions from predicted reactivity are currently known.
Additional information	
Selected references	Piechota et al. (2010). Identification and characterization of high-molecular-weight complexes formed by m-AAA proteases and prohibitins in mitochondria of <i>Arabidopsis thaliana</i> . In press.

application example

total protein from *Arabidopsis thaliana* mitochondria (20 µg) were separated on 10% acrilamide gel and electrophoresis prepared according to Schägger and von Jagov (Anl. Biochem., 1987, 166:368-379). After running the gel, proteins were transferred to nitrocellulose membrane using wet transfer (0.22% CAPS, pH 11). Transfer was checked by Ponceau S staining. Blot was destained by several quick washings in distilled water and 1 washing in 1X TBS (10 mM T pH 7.5, 150 mM NaCl) (10-15 min.). Blot was blocked by 1.5 hour in 5% milk in TBST (1X TBS, 0,1 20) After blocking blot was washed quickly twice in TBST and incubated 2 hours with primary antibody (dilution 1: 1000 TBST (dilution 1:1000). Washing: two quick washings in TBST and 3 x 10 min. washings in TBST. Then blot was incubated 45-60 min. with a secondary anti-rabbit antibodies conjugated to peroxidase (Sigma, dilution 1:10000) in TBST. Washing: as above. After washing blot was incubated 1-2 min. in ECL solution and exposed to Kodak autoradiography film. Exposure time was 15-60 seconds.

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 (13000rpm, 1 min.).

Courtesy Dr. J. Piechota

