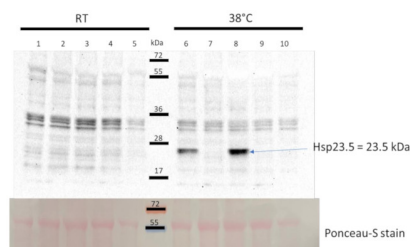


Product no **AS21 4524****Anti-HSP23.5 | Heat shock protein 23.5 (mitochondrial)****Product information**

<b>Immunogen</b>	Truncated recombinant HSP23.5 of <i>Arabidopsis thaliana</i> , UniProt: <a href="#">Q9FGM9-1</a> , TAIR: <a href="#">AT5G51440</a>
<b>Host</b>	Rabbit
<b>Clonality</b>	Polyclonal
<b>Purity</b>	Immunogen affinity purified serum, in PBS pH 7.4.
<b>Format</b>	Lyophilized
<b>Quantity</b>	50 µg
<b>Reconstitution</b>	For reconstitution add 50 µl, of sterile water.
<b>Storage</b>	Store lyophilized/reconstituted 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**

<b>Recommended dilution</b>	1 : 1000 (WB)
<b>Expected   apparent MW</b>	23.5 kDa
<b>Confirmed reactivity</b>	<i>Arabidopsis thaliana</i>
<b>Predicted reactivity</b>	Species of your interest not listed? <a href="#">Contact us</a>
<b>Not reactive in</b>	No confirmed exceptions from predicted reactivity are currently known
<b>Selected references</b>	To be added when available, antibody available in December 2021.

**Samples:**

- 1- 20 µg of *Arabidopsis thaliana* Col-0 whole protein extract
- 2- 20 µg of an *Arabidopsis thaliana* Hsp23.5 null mutant whole protein extract
- 3- 20 µg of an *Arabidopsis thaliana* Hsp23.6 null mutant whole protein extract
- 4- 20 µg of an *Arabidopsis thaliana* Hsp23.5 and Hsp23.6 double null mutant whole protein extract
- 5- 20 µg of an *Arabidopsis thaliana* Hsp23.5, 23.6 and 26.5 triple null mutant whole protein extract

Protein for lanes 1-5 was extracted from plants kept at RT

- 6- 20 µg of *Arabidopsis thaliana* Col-0 whole protein extract
- 7- 20 µg of an *Arabidopsis thaliana* Hsp23.5 null mutant whole protein extract
- 8- 20 µg of an *Arabidopsis thaliana* Hsp23.6 null mutant whole protein extract
- 9 - 20 µg of an *Arabidopsis thaliana* Hsp23.5 and Hsp23.6 double null mutant whole protein extract
- 10 - 20 µg of an *Arabidopsis thaliana* Hsp23.5, 23.6 and 26.5 triple null mutant whole protein extract

Protein for lanes 6-10 was extracted from plants heat stressed at 38°C for 1 hour, left to recover at RT for 3 hours and then flash frozen.

20 µg/well of total protein extracted from 10 day old *Arabidopsis thaliana* seedlings (WT and mutant, flash frozen in liquid nitrogen and stored at -85°C) with 60 mM Tris-HCl pH 6.8, 60 mM DTT, 2% SDS, 1.5% Sucrose and 0.05% bromophenol blue and denatured with 1X SDS Loading Dye (10% glycerol, 0.002% bromophenol blue, 0.5% SDS, 50 mM Tris-HCl pH 6.8) at 95°C for 5 min were separated on 4- 20% SDS-PAGE (nUView Tris-Glycine Precast Gel, NuSep) and blotted 1h to nitrocellulose (0.45 µm pore size) using semi-dry transfer. Blot was blocked with 5% milk in TBS-T for 15 minutes, blocking solution was replaced and blocked for another 15 minutes for 30 minutes total with agitation. The blot was incubated in the primary antibody at a dilution of 1:1000 for ~6 hours at 4°C with agitation. The antibody solution was decanted, and the blot was

rinsed briefly twice, then washed for 10 minutes, a process repeated three times in total for 30 minutes of washes with TBS-T at RT with agitation. Blot was incubated in Goat Anti- rabbit IgG (horse radish peroxidase conjugated) diluted 1:5000 in TBS-T with agitation. The blot was washed as above and developed for 2 minutes in homemade ECL solution (1.25 mM Luminol, 0.1 M Tris pH 8.5, 0.2 mM p-coumaric acid, 1 mL of solution mixed with 3  $\mu$ L of 3% hydrogen peroxide just before development). Exposure time was 11 minutes in a G:Box iChemi XT (Syngene).

Courtesy of Fabian Suri-Payer, Vierling Lab, Department of Biochemistry & Molecular Biology University of Massachusetts Amherst, USA