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This product is for research use only (not for diagnostic or therapeutic use)

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## Product no AS11 1780 Anti-IRT1 | Iron regulated transporter 1

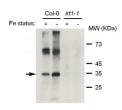
#### **Product information**

Immunogen	KLH-conjugated synthetic peptide derived from Arabidopsis thaliana IRT1 sequence, Q38856, At4g19690
Host	Rabbit
Clonality	Polyclonal
Purity	Immunogen affinity purified serum in PBS pH 7.4.
Format	Lyophilized
Quantity	50 μg
Reconstitution	For reconstitution add 50 µl of sterile water
Storage	Store lyophilized/reconstituted at -20°C; once reconstituted make aliquots to avoid repeated freeze-thaw cycles, This antibody can be stored in a solution containg 50 % glycerol, final concentration, 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**

Recommended dilution	1 : 5000 (WB)
Expected   apparent MW	36.7 kDa
Confirmed reactivity	Arabidopsis thaliana
Predicted reactivity	Brassica napus, Brassica oleracea, Camelina sativa, Capsella rubella, Noccaea caerulescens, Thlaspi cerulescens
	Species of your interest not listed? Contact us
Not reactive in	Horderum vulagre, Solanum lycopersicum, Tagetes erecta
Selected references	<u>Cao</u> et al. (2024). Spatial IMA1 regulation restricts root iron acquisition on MAMP perception. Nature . 2024 Jan;625(7996):750-759. <u>Gautam</u> et al. (2021) IRONMAN Tunes Responses to Iron Deficiency in Concert with Environmental pH. bioRxiv 2021.02.16.431461; doi: https://doi.org/10.1101/2021.02.16.431461 <u>Ivanov</u> et al. (2014). SORTING NEXIN1 Is Required for Modulating the Trafficking and Stability of the Arabidopsis IRON-REGULATED TRANSPORTER1. Plant Cell. 2014 Mar 4. <u>Selote</u> et al. (2014). Iron-binding E3 ligase mediates iron response in plants by targeting bHLH transcription factors. Plant Physiol. 2014 Dec 1. pii: pp.114.250837.

### **Application example**



**5 μg of total protein** from Arabidopsis thaliana wild type (Col-0) and IRT1 mutant (irt1-1) extracted with SDG buffer (62 mM Tris-HCL pH 8.6, 2.5 % SDS, 2 % dithiothreitol, 10 % glycerol) were separated on 15 % **SDS-PAGE** and blotted 1h to **nitrocellulose**. Blots were blocked with 5 % milk in TBST for 1h at room temperature (RT) with agitation. Blot was incubated in the primary antibody at a dilution of 1: 5 000 for 1h at RT with agitation in TBST with 2.5 % milk. The antibody solution was decanted and the blot was rinsed briefly three times, then washed once for 10 min in TBST at RT with agitation. Blot was incubated in secondary antibody (anti-rabbit IgG horse radish peroxidase conjugated, from Agrisera, <u>AS09</u> 602) diluted to 1:10 000 in for 1h at RT with agitation. The blot was washed as above and developed for 5 min with ECL according to the manufacturers instructions. Exposure time was 3 seconds.

Iron-sufficient medium contained 50 µM Fe, +Fe condition, iron-deficient medium 0 µM Fe, -Fe condition.

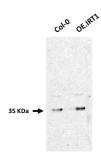
Courtesy Dr. Petra Bauer and Dr. Rumen Ivanov, Saarland University, Germany



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60 μg of total protein from *Arabidopsis thaliana* wild type (Col- 0) and IRT1 over-expressor(OE.IRT1) extracted with Celytic buffer (C2360 Sigmaaldrich) were separated on 15 % SDS- PAGE and blotted 30 min. to PVDF turbo-blot membrane. Blots were blocked with 5 % milk in TBST for 1h at room temperature (RT) with agitation. Blot was incubated in the primary antibody at a dilution of 1: 5 000 for Over night at 4°C with agitation in TBST with 2.5 % milk. The antibody solution was decanted and the blot was rinsed briefly three times, then washed once for 5 min in TBST at RT with agitation. Blot was incubated in secondary antibody (AS09 602, anti-rabbit IgG horse radish peroxidase conjugated, from Agrisera ) diluted to 1:10 000 in for 1h at RT with agitation. The blot was washed as above and developed for 15 min with chemiluminescent detection reagent: AgriseraECL Bright. Exposure time was 1 minute. Hoagland medium contained 50 μM Fe.

Courtesty Haitham Ayeb, Louvain Institute of Biomolecular Science and Technology (LIBST), Belgium