

Supplementary Data

SITPR1, a tomato tetratricopeptide repeat protein, interacts with the ethylene receptors NR and LeETR1, modulating ethylene and auxin responses and development

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Running title:

SITPR1, ethylene signalling and development

Supplementary Material

Figure S1. Interaction assay of SITPR1 with the ethylene receptors.

A. Receptors and SITPR1 constructs used for yeast two-hybrid assays (TMD = transmembrane domain; DB = LexA DNA binding domain; AD = activation domain).

B. Verification of bait protein synthesis in yeast by western blot using anti-LexA antibody. 1. DB-LeETR1; 2. DB-LeETR2; 3. DB-NR; 4. DB-LeETR4; 5. DB-LeETR5; 6. DB-LeETR6; 7. Bait vector pEG202 containing LexA DNA binding domain.

C. Activation assay of *LacZ* reporter by interaction of SITPR1 and the receptors: 1, LeETR1/SITPR1; 2, LeETR2/SITPR1; 3, NR/SITPR1; 4, LeETR4/SITPR1; 5 LeETR5/SITPR1; 6, LeETR6/SITPR1; P, positive control pSH17-4; N, Negative control pRFHM1. Gala: galactose.

Figure S2. Adventitious root formation on cuttings from wild type and SITPR1 transgenic plants with and without IBA treatment. Cuttings about 8 cm in length from the side shoots of wild type and the transgenic plants were dipped in talcum powder containing 0 or 1,000 $\mu\text{g/g}$ IBA. Ten cuttings were taken from plants of each line and propagated in 5 cm pots containing perlite and maintained in the glasshouse in high humidity. After three weeks, the total number of root initials for each cutting was examined and photographed. Experiments were repeated twice.

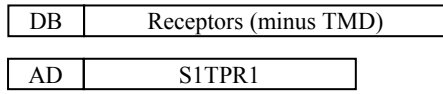
Figure S3. Co-localization of the SITPR1-GFP fusion with the red-fluorescent membrane stain FM 4-64. *Arabidopsis* roots (A-C) and hypocotyls (D-F) expressing SITPR1-GFP were treated with the red-fluorescent membrane dye FM 4-64 (Invitrogen) and examined using a Leica TCS SP2 confocal scanning microscope. (A and D) SITPR1-GFP. (B and E) FM 4-64 dye. (C and F) Bright-field image. Scale bar = 75 μm (root) and 31 μm (hypocotyl).

Table S1. Adventitious root formation on cuttings from wild type and transgenic line treated with 0 or 1000 $\mu\text{g/g}$ IBA

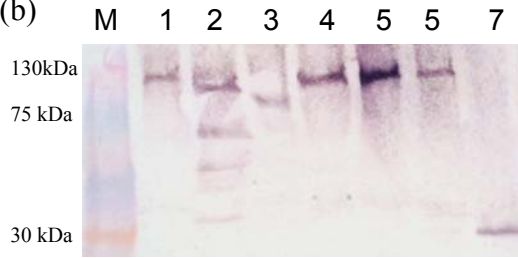
Lines	0			1000 $\mu\text{g/g}$		
	Rooted cuttings	Total root no	%	Rooted cuttings	Total root no	%
Wild type	10	15 \pm 5	100	10	100.5 \pm 19.5	100
3278A	9	8 \pm 8	53	10	47 \pm 30	47
3286A	5	2.6 \pm 4.4	17	9	19.5 \pm 19.5	19
3272A	5	2.2 \pm 5	15	10	17.3 \pm 15.7	17
3273A	5	3.7 \pm 7	25	9	30 \pm 37	30

Data represent the mean of two independent experiments.

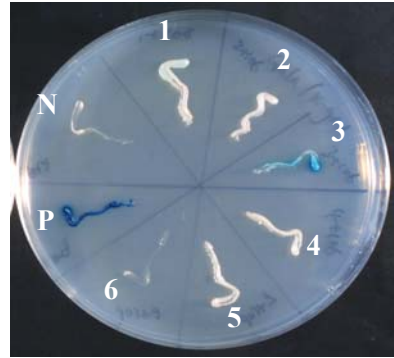
(a)



(b)



(c)



gala/x-gal

Figure S1

0 IBA

1000 $\mu\text{g/g}$ IBA



Figure S2

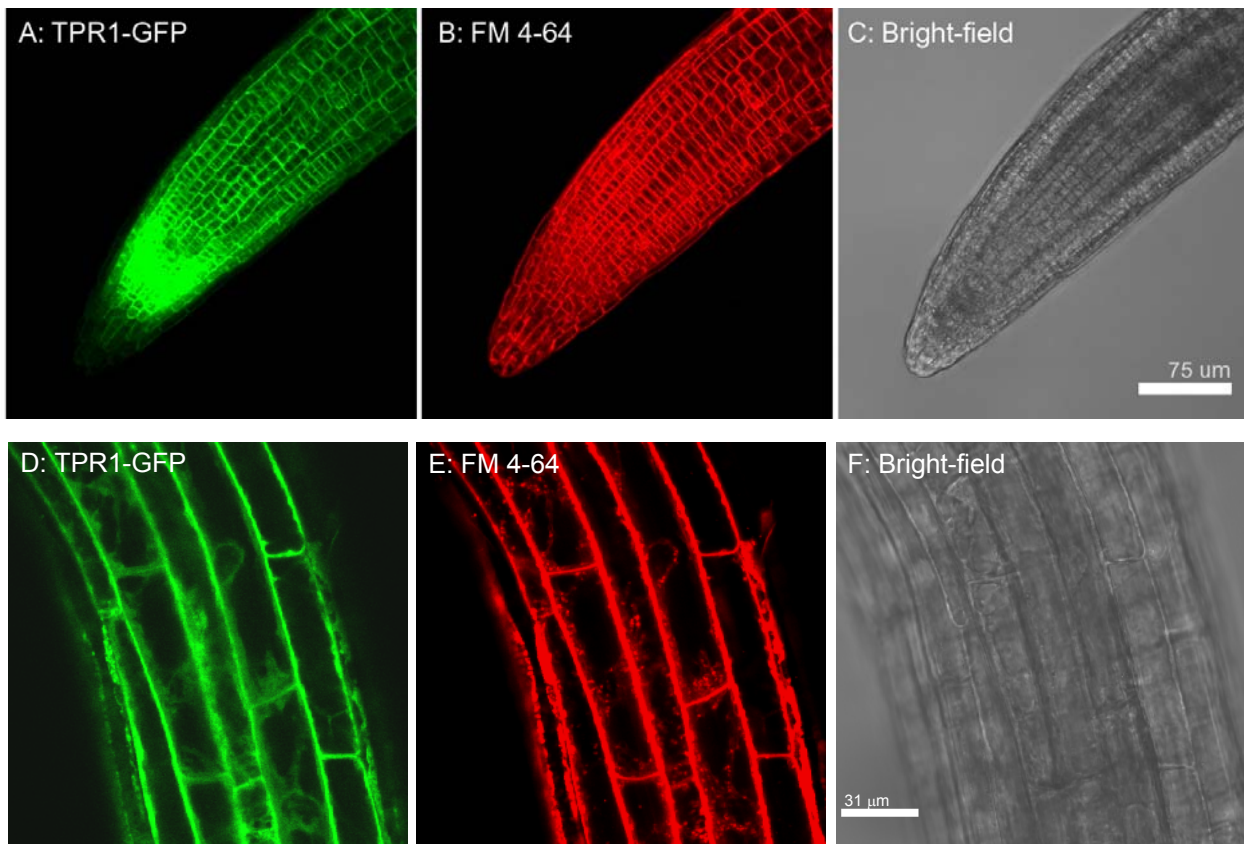


Figure S3