

Fig. S1 Cellular patterns of the RAM of the PR and emerged LRP.

- (A) Confocal image of the primary RAM in pSCR::GUS:GFP (endodermis and QC, green) counterstained with PI (cell wall, red). Scale bar = 50 μ m.
- (B) Schematic representation of the cellular pattern of the RAM in the PR created by tracing cell outlines from A. Cell types are colored according to the legend.
- (C) Confocal image of the emerged LRP visualized with a plasma membrane-localized fluorescent marker (WAVE131Y, green). Scale bar = $50 \mu m$.
- (D) Schematic representation of the cellular pattern of the emerged LRP created by tracing cell outlines from C. Cell colors show the putative cell types based on the information from our observations in this study, and previously reported histological study and marker expression patterns (Malamy and Benfey, 1997).

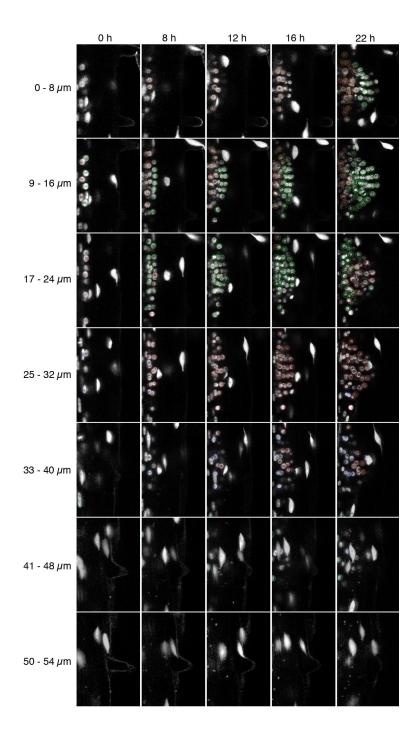


Fig. S2 Original images for 4D nuclei tracking.

Maximum-intensity projections of seven sections at the indicated distance within Z sections at the indicated time points are shown. Circles (4 μ m diameter) drawn in the same colors as in Fig. 2A–F indicate tracked nuclei for 3D representation.

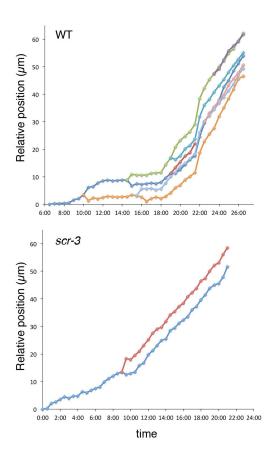


Fig. S3. Relative cell positions of the outer layer-derived cell lineages in the WT and *scr-3* mutant during LRP development.

Cell positions relative to the initial position (stage II) were analysed from the tracking data of Fig. 5.

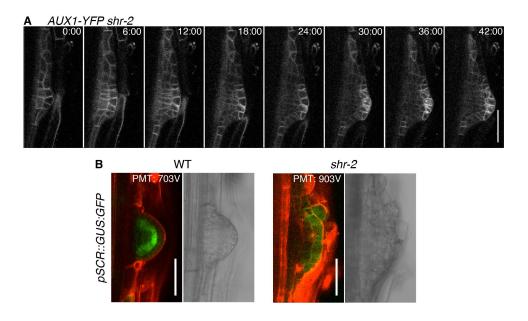


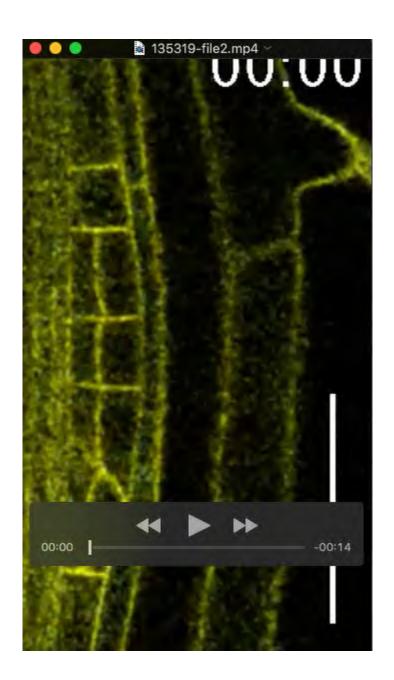
Fig. S4. shr mutation disrupted cell division during LRP development.

- (A) Time-lapse image series of LRP development in AUXI-YFP in the shr-2 background. The elapsed time (h:min) from the start of observation is indicated in each panel. Scale bar = 50 μ m.
- (B) pSCR::GFP:GUS in the shr-2 background. pSCR::GUS:GFP was observed in the second outermost layer of the WT, and the outermost layer of the scr-3 mutant. Because of the large reduction of SCR promoter activity in the shr-2 mutant compared with the WT, we observed the expression using different voltages on a photo multiplier (PMT); 703 V for WT and 903 V for shr-2 with the same laser power (5%). The roots were counterstained with propidium iodide (red). Scale bars = 50 μ m.



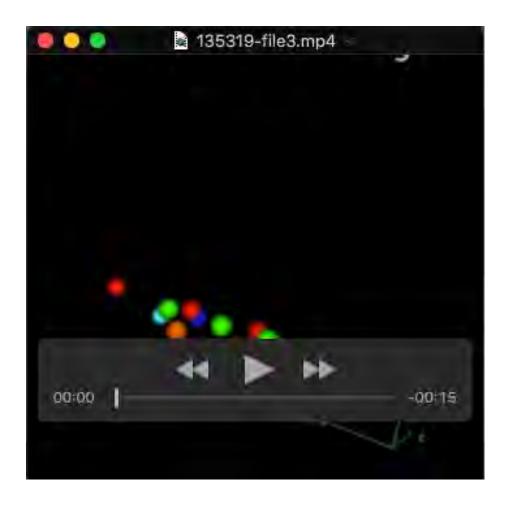
Fig. S5. The scr mutant created mature LRs.

Eleven-day-old seedlings of WT (Col) and scr-3 and shr-2 mutants. Scale bars = 10 mm.



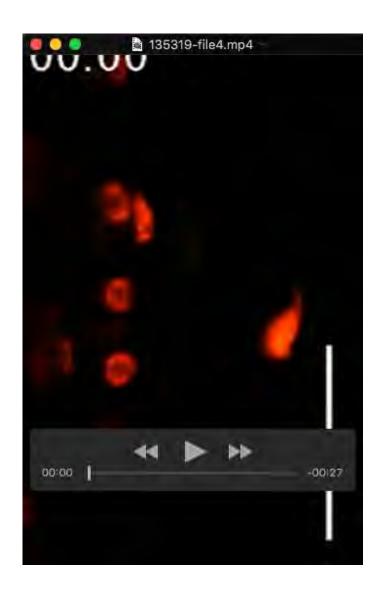
Movie 1. Time-lapse movie of WAVE131Y x QC25::CFP.

Time-lapse movie of LR primordium development visualized using WAVE131Y (plasma membrane, yellow) and QC25::CFP (QC marker, cyan). The elapsed time (h:min) from the start of observation is indicated at the top. Scale bar = $50 \mu m$.



Movie 2. 3D nuclei representation of LRP development.

Nuclei are distinguished by different colors dependent on the initial cell file. The central cell file (green) provides all cells in the medial section, and flanking cell files (red and orange) contribute to the side parts of the LRP. Additional flanking cell files (blue and light blue) only contribute to a small proportion of the LRP.



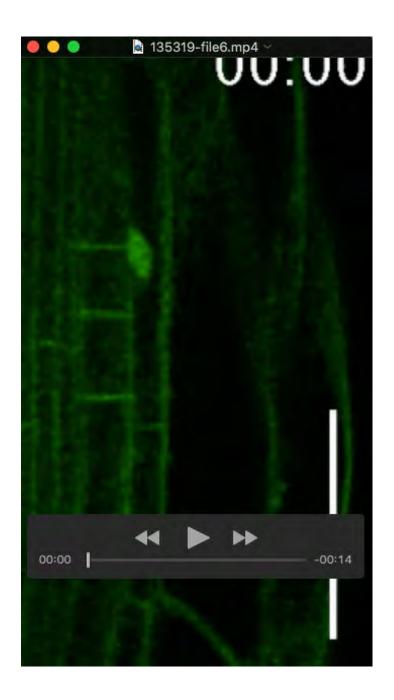
Movie 3. Time-lapse movie of pRPS5a::H2B:tdTomato x pWOX5::n3GFP.

Time-lapse movie of LRP development visualized using pRPS5a::H2B:tdTomato (red) and pWOX5::n3GFP (green). The elapsed time (h:min) from the start of observation is indicated at the top. Scale bar = 50 μ m.



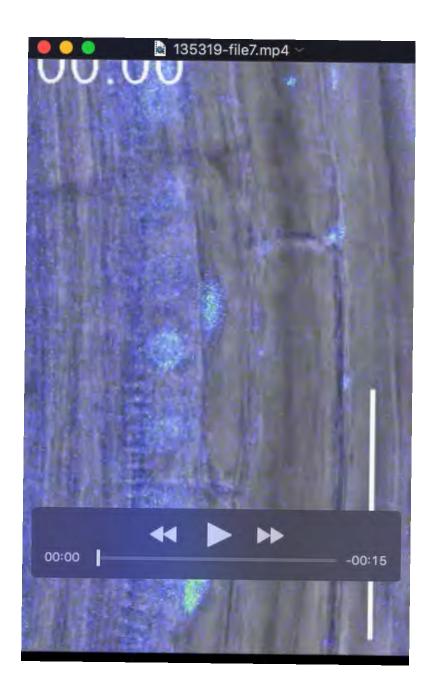
Movie 4. 3D nuclei representation of the QC cell lineage.

QC precursor cells (yellow) were produced by periclinal cell division at the outer layer of stage II, and then acquired QC identity (green). After QC marker expression, the QC underwent longitudinal radial cell division to create four QC cells.



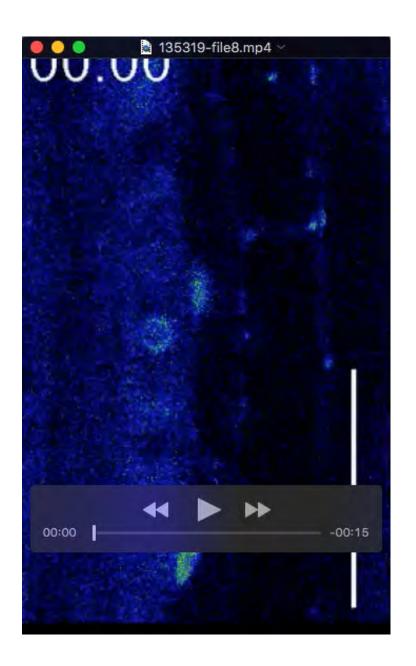
Movie 5. Time-lapse movie of pSCR::GFP:SCR (scr-3) x WAVE131Y.

Time-lapse movie of pSCR::GFP:SCR (nuclei, green) x WAVE131Y (plasma membrane, green) in the scr-3 mutant background. The elapsed time (h:min) from the start of observation is indicated at the top. Scale bar = $50 \mu m$.



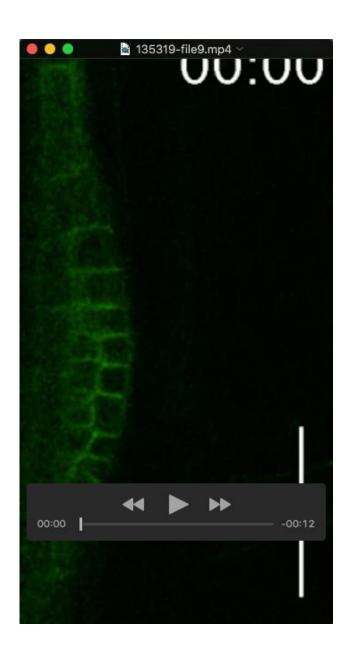
Movie 6. Time-lapse movie of pSHR::SHR:GFP (shr-2) merged with DIC.

The fluorescence intensity of SHR:GFP was described by a rainbow look-up color table. The elapsed time (h:min) from the start of observation is indicated at the top. Scale bar = $50 \mu m$.



Movie 7. Time-lapse movie of pSHR::SHR:GFP (shr-2).

The GFP channel only from Movie 6. Fluorescence intensity of SHR:GFP was described by a rainbow look-up color table. The elapsed time (h:min) from the start of observation is indicated at the top. Scale bar = $50 \, \mu m$.



Movie 8. Time-lapse movie of LRP development in AUX1-YFP scr-3.

The elapsed time (h:min) from the start of observation is indicated at the top. Scale bar = $50 \mu m$.