Supporting Information

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Fig. S1. rp/4d and TirT mutants reduced Bodipy dye signal with or without NAA treatment. (A–C) Bodipy dye staining pattern in root transition zone of 7-dold seedlings from Col (A), rp/4d (B), or TirT (C) after treatment with DMSO (0.1%) for 6 h. (Scale bars, 10 μ m.) (D–F) Bodipy dye staining pattern in root transition zone of 7-d-old seedlings from Col (D), rp/4d (E), or TirT (F) after treatment with 100 nM NAA for 6 h. (Scale bars, 10 μ m.) (G) Quantification of signal intensity of Bodipy dye in Col, rp/4d or TirT mutants after DMSO or NAA treatment. Images from A–F were taken from a single scan in the middle section of the transition region in the root. For G, 300 cells from 20 seedlings were used for data quantification. Error bars represent SD.



Fig. 52. Time-course of Nile red staining pattern after NAA induction. (A–D) Nile red staining pattern in root transition zone after treatment by DMSO or 100 nM NAA treatment for 2 h (B), 5 h (C), or 24 h (D), respectively. For all of the images, the *Left* is nonpolar lipid, *Center* is polar lipid, and *Right* is a merged image from the other two. Images represent data taken from at least three independent experiments. (Scale bars, 10 μ m.)



Fig. S3. Nile red staining pattern after treatment with different hormones. (*A–F*) Nile red staining pattern in root transition zone after treatment by DMSO, NAA (*B*), 2-NAA (*C*), 6-BA (*D*), JA (*E*), or ABA (*F*), respectively. For all of the images, the *Left* is nonpolar lipid, *Center* is polar lipid, and *Right* is merged image from the above two. Images represent data taken from at least three independent experiments. (Scale bars, 10 µm.)

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Fig. 54. Phenotype comparison between T-DNA insertion mutant and Col wild-type seedlings. (*A*) Phenotypes of 10-d-old seedlings of Col and T-DNA mutants. (Scale bars, 1 cm.) (*B*) Nile red staining of 7-d-old seedlings of Col and T-DNA mutants. (Scale bars, 10 μm.) Images represent data taken from three independent experiments.

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Fig. 55. rp/4d mutants displayed hypersensitivity to sphingolipid and sterol inhibitors but not fatty acid inhibitor. (*A–E*) Growth phenotypes of 7-d-old seedlings of Col and rp/4d mutants on medium with DMSO (0.1%) (*A*), 500 nM FB1 (*B*), 50 μ M FEN (*C*), 200 nM flufenacet (*D*), or 5 μ M auxinole (*E*). (Scale bars, 1 cm.) (*F*) Quantification of root inhibition rate of Col and rp/4d mutants grown on medium with different lipid inhibitors or auxinole compared with on medium with DMSO. **P* < 0.05, ***P* < 0.001. One-hundred roots of each genotype were used for quantification. Error bars represent SD. (*G–K*) quantification of root deviation angel from vertical position in Col and rp/4d mutants when grown on medium with DMSO (0.1%) (*G*), 500 nM FB1 (*H*), 50 μ M FEN (*I*), 200 nM flufenacet (*J*), or 5 μ M auxinole (*K*). The *x* axis represents the range of different deviation angels. The *y* axis represents the percentage of roots fall into different deviation angel ranges;120 roots of each genotype were used for quantification.



Fig. S6. Lipid inhibitors reduce NAA induced lipid accumulation as revealed by Nile red staining. (A) Nile red staining of root transition zone in 7-d-old seedlings after treatment with DMSO (0.1%), FB1 (5 μ M), PDMP (1 μ M), FEN (100 μ M), flufenacet (1 μ M), or auxinole (5 μ M) alone or together with 100 nM NAA for 6 h. (*B*) Bodipy dye staining of root transition zone in 7-d-old seedlings after treatment with DMSO (0.1%), FB1 (5 μ M), FEN (100 μ M), flufenacet (1 μ M), or auxinole (5 μ M), PDMP (1 μ M), FEN (100 μ M), flufenacet (1 μ M), or auxinole (10 μ M) alone or together with 100 nM NAA for 6 h. Images represent data taken from three independent experiments. (Scale bars, 10 μ m.)

Dataset S1. List of genes down-regulated in polysome-bound mRNA but not in total mRNA according to RNA sequencing result

Dataset S1

Dataset S2. List of genes in lipid metabolism pathway that were tested by qRT-PCR

Dataset S2

The genes labeled with red letters were not consistent with RNA sequencing data and the ones labeled with green letters were chosen to analyze the phenotypes of T-DNA insertion mutants.

Dataset S3. List of genes in vesicular trafficking pathways which were validated by qRT-PCR

Dataset S3

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Dataset S4. Primers used in qRT-PCR

Dataset S4

Dataset S5. Amount of lipid in each head group class in 7-d-old seedlings of Col and rpl4d mutants

Dataset S5

Dataset S6. List of T-DNA insertion mutants used in the report

Dataset S6