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Supplementary information

Auxin-mediated root branching is determined by the form of available nitrogen

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Auxin-mediated root branching is determined by the form of available nitrogen

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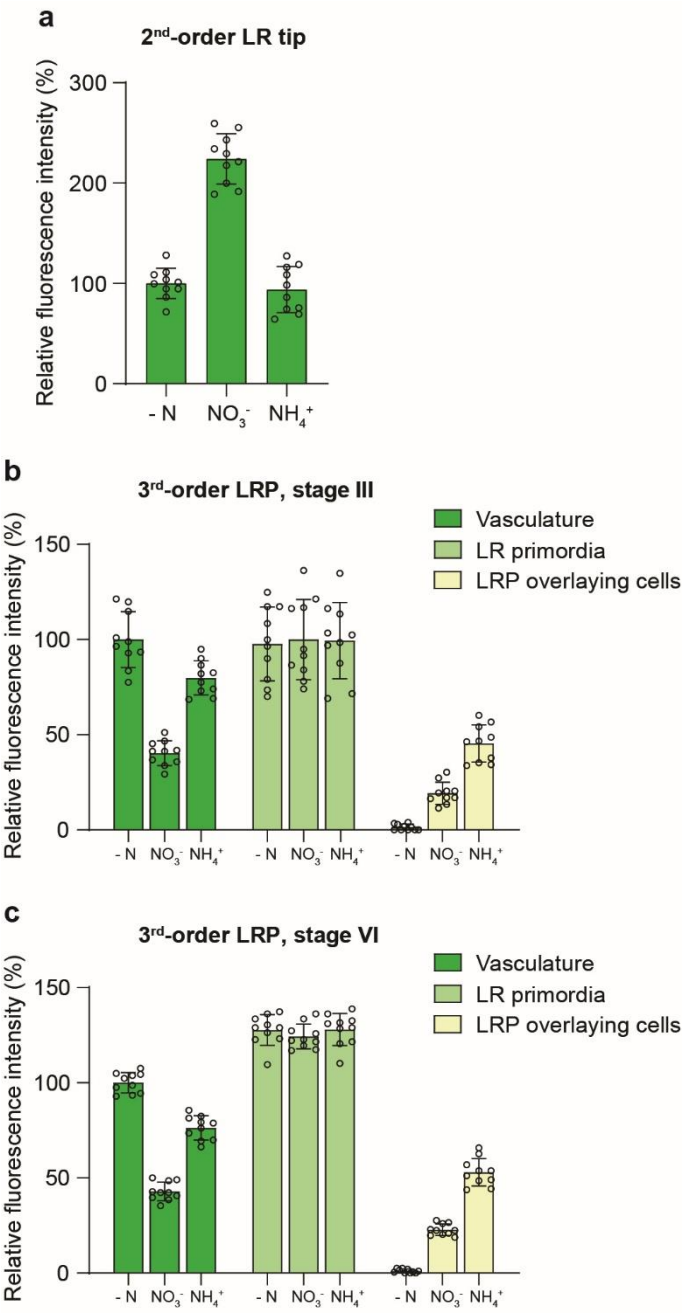
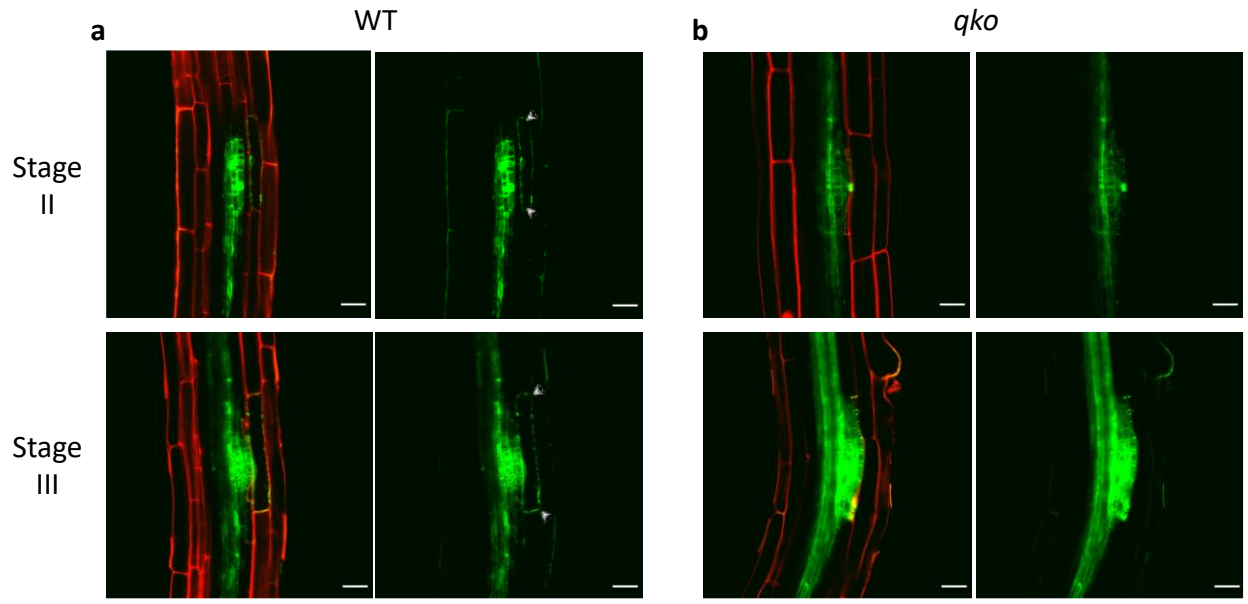
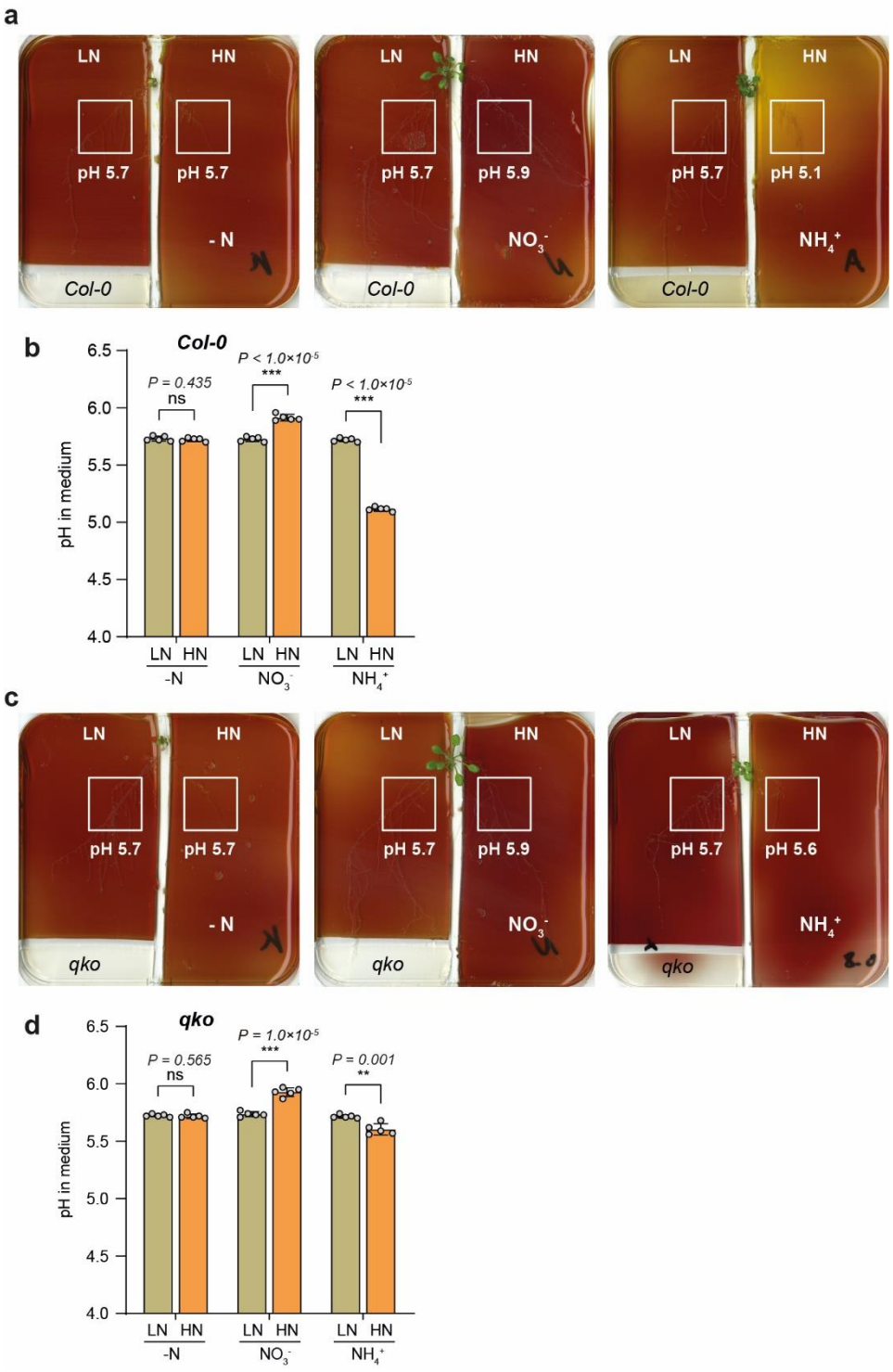


Supplementary Fig. 1: Fluorescence intensity of DR5:GFP in higher-order lateral roots in response to local N supplies.

a-c, Quantitative readout of DR5:GFP fluorescence intensity in roots shown in Fig. 2a-i. GFP fluorescence was measured in 2nd-order lateral root tips (**a**), in 3rd-order lateral root primordia at stage III (**b**), and in 3rd-order lateral root primordia at stage VI (**c**). Bars represent means ± SD, n = 10 independent biological replicates.



Supplementary Fig. 2: Local ammonium supply promotes auxin accumulation in overlaying cells.
a, b, Fluorescence of DR5:GFP in the 3rd-order lateral root primordia in wild-type (WT) (**a**) and *qko* (**b**) plants at developmental stages II and III. Lateral roots were imaged 15 days after these plants were transferred to vertical split agar plates with a 1st-order lateral root exposed to 0.8 mM NH_4Cl on the HN side. White arrows indicate presence of DR5:GFP signal in cortical cells overlaying the 3rd-order lateral root primordia. Representative images from 10 plants per treatment are shown. Scale bars: 20 µm.



Supplementary Fig. 3: Rhizosphere pH changes in response to local N treatments.

a and **c**, Rhizosphere pH of wild-type (WT) (**a**) or *qko* (**c**) plants grown for 15 days on vertical split agar plates supplemented with 0.8 mM KCl (–N), 0.8 mM KNO_3 or 0.8 mM NH_4Cl on the HN side. Colour changes of the pH indicator bromocresol purple represent acidic (yellow) to neutral (brown-purple) pH. **b** and **d**, pH in the agar medium after cultivation of WT (**b**) or *qko* (**d**) plants for 15 days. After removal of plants

from the plates, agar pieces were excised from the HN and LN segment as indicated by 2 x 2 cm white squared areas and dissolved in water to measure pH. Bars represent means ± SD, n = 5 independent biological replicates. Asterisks denote significant differences between HN and LN side at ** $P<0.01$, *** $P<0.001$, ns = not significant, according to two-tailed Student's *t*-test.

Supplementary Table 1. Primers used in this study.

Primer name	Primer sequence
qAUX1-F	TGCTCTGATCAAAGTCTTCTCCT
qAUX1-R	GAAGAGAAGAACCCAGAAATGTG
qLAX3-F	CACCGAGAGTGGTAGGAGGA
qLAX3-R	CTCCAAACCCGAACCCAACT
qUBQ10-F	CTTCGTCAAGACTTTGACCG
qUBQ10-R	CTTCTTAAGCATAACAGAGACGAG
qACTIN2-F	GACCAGCTCTTCCATCGAGAA
qACTIN2-R	CAAACGAGGGCTGGAACAAG