Supplementary information for article: Root growth dynamics and allocation as a response to rapid and local changes in soil moisture

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S1 Sensitivity analysis of software parameters

In order to ensure that the measurements obtained from the software were consistent and were not influenced by the parameter threshold selection, noise cutoff and max gap (two main parameters used by the software to construct the root system) were varied within a given range. The choice of different values of max gap in NMRooting (from 2 till 7) led to visually similar

- 5 curves (Fig. S1.1a). Different values of noise cutoff (0.04 to 0.12) also led to visually similar curves except for the value of 0.04 (Fig. S1.1b). So, aside from this value, the parameter choice did not obviously influence the trend of root length detection. The 3D image of the root system produced by the software when the noise cutoff level was set to 0.04 was not realistic (Fig. S2.2a). It overestimated root length by even recognizing roots outside the system. The image quality and realism increased significantly once the noise cutoff was increased to 0.06 (Fig. S2.2b). This further supports our parameter selection for the
- 10 image analysis.



Figure S1.1. Growth rate in L2 of a root system under (a) a range of values of maximum gap and (b) a range of values of noise cutoff.



Figure S2.1. Approach used to determine the start of Phase 2. The lines correspond to the VWC in Layer 2 of three T1 plants (receiving pulses in L2 during Phase 1 and pulses in L1 during Phase 2). Phase 2 started when the VWC dropped below 7.5% in L2 after Phase 1 (it took 2 days in Plant 3 and three days in Plants 10 and 14).



Figure S2.2. 3-dimensional representation of the same root system using two different levels of noise cutoff. (a) Root system representation generated using a noise cutoff of 0.04. The root detection is erroneous and the signal has many artifacts. (b) Root system representation generated using a noise cutoff of 0.06. The root detection is considered realistic.



Figure S2.3. Higher presence of intertwined and clustered lateral roots in L2 compared to L1. Roots in L1 are easier to distinguish and to track all the way to the tip. This is the reason why individual root selection was not performed in L2, as such selection would have been difficult and prone to errors.



Figure S2.4. Medians of scaled growth rates for each T1 plant and data used to answer Question 1. Only data points obtained on the first day of the pulse series and 48 hours later were considered, regardless of the phase.



Figure S2.5. Medians of scaled growth rates for each T2 plant and data used to answer Question 1.



Figure S2.6. Medians of the scaled growth rates for each control plant (where VWC was kept at 15% in both L1 and L2 during both Phase 1 and 2).



Figure S2.7. Violin plots of growth rates during the three experimental phases in T1, T2 and controls. Panels on the left refer to L1 (50 data points in each violin), and on the right to L2 (10 data points in each violin). (a)-(b): T1; (c)-(d): T2; (e)-(f): controls. The thick black line inside each violin indicates the range between the 25^{th} and 75^{th} quartiles, while the white circles mark the median values.



Figure S2.7. Refer to previous caption.