

Supplementary material:

**Cluster root allocation of white lupin (*Lupinus albus* L.) in soil with
heterogeneous P and water distribution**

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Water availability and distribution

The assessment of soil water availability and distribution in the various treatments of the main experiment was based on water potentials derived from experimental water retention curves (Figure S1) and water content measurements in the plant containers (Figure S2).

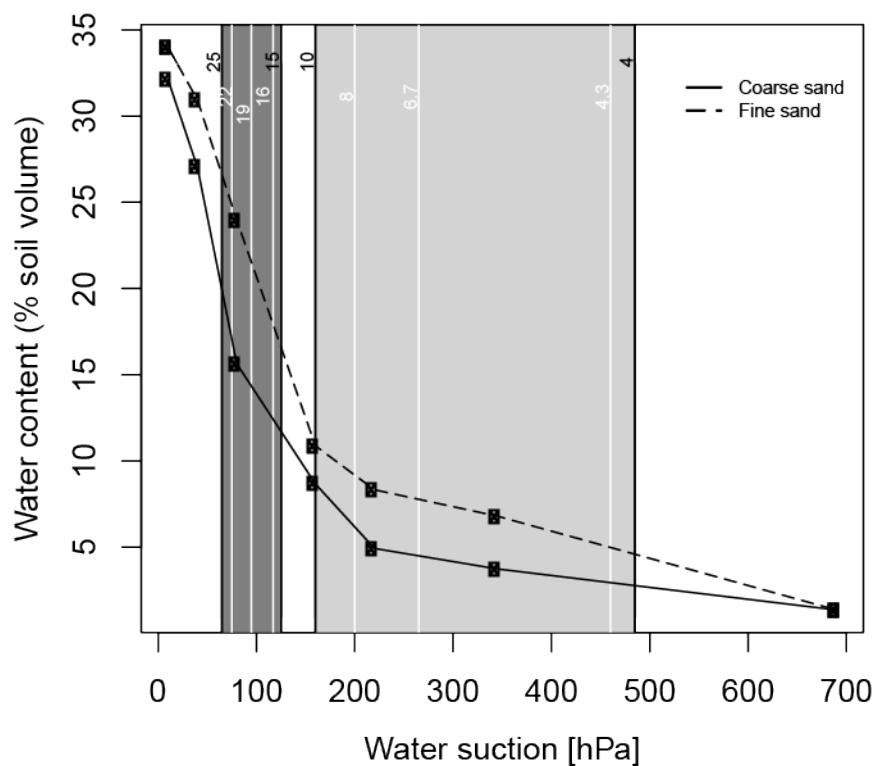


Figure S1: Water retention curves of the two experimental substrates determined by means of the pressure membrane method (Klute, 1986). The vertical lines represent calculated water suctions at medium container depth in the treatments with heterogeneous substrates (as described in the Materials and Methods section). The vertically aligned numbers give the corresponding water contents. The grey areas represent the calculated water suction ranges between maximum and minimum water contents in the high (dark grey) and low (after DAG 12, light grey) water supply treatments.

Water contents and matrix potentials of the fine and coarse sand sections in the heterogeneous treatments were calculated by partitioning the total mass of water in the container between the two sands according to their (interpolated) water retention curves under the assumption of hydraulic equilibrium and negligible non-linearity in the vertical water content gradients resulting from gravity, i.e. by solving the following equation for the matrix (or soil water) potential ψ at medium depth:

$$W(\psi)/V = 1/3 \theta(\psi)_{\text{fine}} + 2/3 \theta(\psi)_{\text{coarse}} \quad (1)$$

where $W(\psi)$ is the total mass of water in the container at matrix potential ψ , V is the bulk volume of the soil, and $\theta(\psi)_{\text{fine}}$ and $\theta(\psi)_{\text{coarse}}$ are the water contents of the fine and the coarse sand at matrix potential ψ , respectively.

In order to determine the dynamics of short-term changes in soil moisture distribution we conducted a supplementary experiment, because it was not possible to do this in the main experiment for logistical reasons,. For this experiment, additional containers with heterogeneous water distribution, but no addition of P, were prepared as described before and treated with either high or low water supply. Plants were grown at ETH for 16 days under the same conditions as described before and then transferred to PSI, where they remained for the entire NR imaging period, which ended on DAG 28. At PSI, the plants were kept at room temperature in the hall of the NEUTRA facility with illumination provided through temporarily installed plant growth lamps (Nurturelite 125 W blue). In the treatments with low water supply, 10 ml of water were added on DAG 24 and 30 ml on DAG 25 In the treatments with high water supply 60 ml of water were supplied on DAG 21. The containers were NR imaged on DAG 17, 22 and 24 and 27 in the treatments with low water supply and on DAG 16, 21 and 22 and 24 in the treatments with high water supply. The NR images were processed as described before. Changes in soil moisture

distribution were visualized by pixel-wise subtraction of an NR image taken at the beginning of an interval from the respective image taken at the end of the interval.

Results of the soil water measurements and supplementary experiment

The averaged soil water contents in the containers of the main experiment fluctuated between 15% and 25% in the treatments with high water supply, with a slight tendency to decrease over time (Figure S2). In the treatments with low water supply, the soil water contents initially ranged between 15 and 17%; after irrigation was discontinued for 10 days after transplanting, they dropped to 8 – 10% within seven days and then gradually decreased to values between 4 and 6 % with the development of the plants.

The soil water potentials inferred from the experimental water contents varied between 60 (equivalent to field capacity) and 120 hPa at high water supply and between 160 and close to 500 hPa in the treatment with low water supply (Figure S1), indicating that water availability was no limitation for plant growth in the former, but became an increasingly limiting factor towards the end of the experiment in the latter treatment.

The water retention curves show that always more than 10% of the bulk volume of both substrates was air-filled porosity, so that we can safely assume that aeration was sufficient at all times at both water supply rates, but in particular at the lower rate (Allmaras *et al.*, 1988; Lipiec and Hakansson, 2000).

Neutron radiography images of plant containers from the side experiment conducted to assess short-term water dynamics are shown in Figure S3. They illustrate that the water content was generally higher in the fine sand than in the coarse sand sections in the heterogeneous soil water treatments. At low water supply it also fluctuated more

strongly over irrigation cycles in the fine sand, in line with the higher specific water capacity of the fine sand in the dry range of the water retention curves. Conversely, water content fluctuations were larger in the sections with coarse than with fine sand over wetting-drying cycles in the treatments with high water supply.

Infiltrating water redistributed much more rapidly in the containers with high than with low water supply rate. Figure S3 shows that no wetting front was visible any more already within one day after watering in both substrates in the treatment with high water supply rate, while the wetting front was still clearly visible more than 2 days after water application in the treatment with low water supply and moved faster in the fine sand than in the coarse sand.

Given that horizontal soil water redistribution was very slow, the larger water content fluctuations in fine sand in the treatments with low water supply indicate that water availability for plants was higher in the fine sand than in the coarse sand in these treatments, whereas the larger fluctuations in coarse sand in the treatments with high water supply do not necessarily mean that the opposite was the case in the treatments with high water supply. The water potential never reached levels known to limit water uptake by roots in the latter treatments, and due to a much higher hydraulic conductivity associated with the higher wetness horizontal fluxes could have much more easily balanced differences in available water between the sections in contrast to the treatments with low water supply.

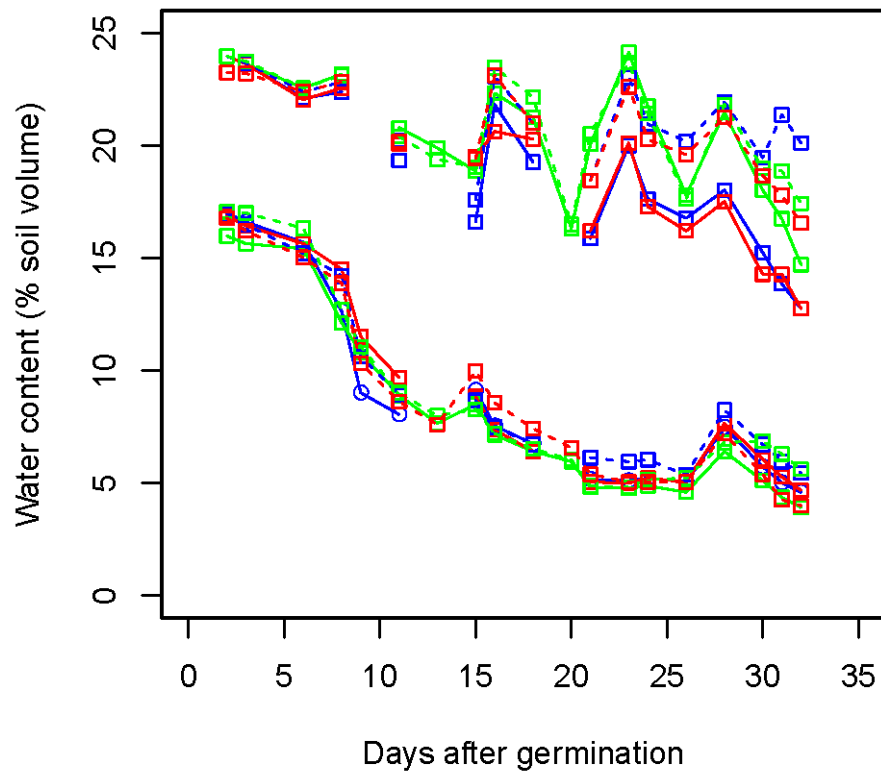


Figure S2: Water content variation over time in containers with heterogeneous (dashed lines) and homogeneous (solid lines) substrates and no P addition (blue lines), homogeneous P fertilization (green lines) or heterogeneous P fertilization (red lines)

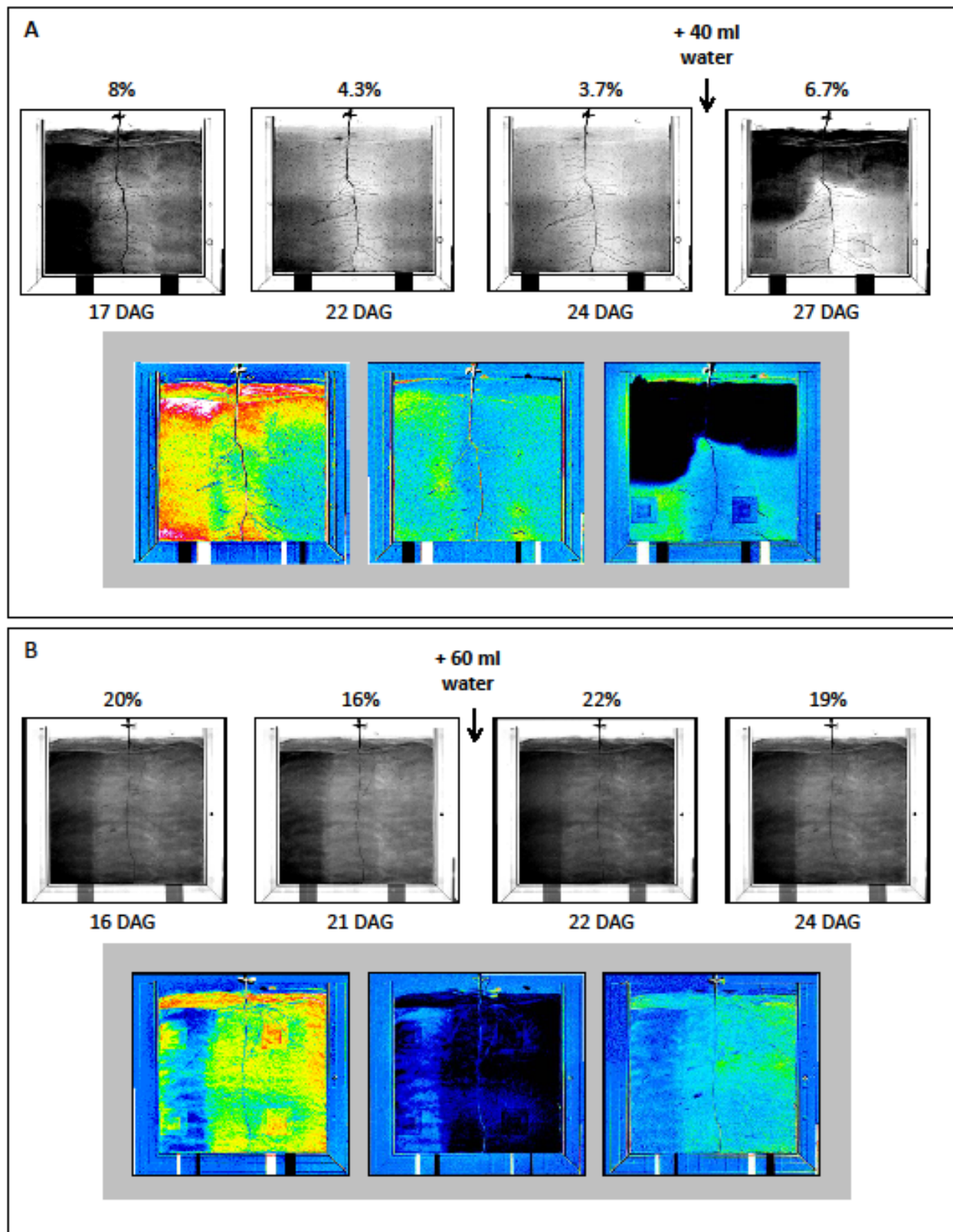


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Figure S3: Neutron radiography (NR) images of soil water distribution at different points in time (grey-value images) and of the changes between these time points (color images) in treatments with heterogeneous substrate at low (A) and high water supply (B). The numbers above the grey value images refer to the volumetric water content (%) at the respective days (DAG: day after germination) given below the images. The colors in the colored images indicate no change (light blue), decrease (green-yellow-red-white) or increase (dark blue) in water content between two subsequent dates.

ANOVA Tables

Table T1: ANOVA results relating to the effects of water distribution (W. Dist.), P fertilization (P) and overall water supply rate (Water) and their interactions on shoot dry weight.

	Chisquare	Degrees of freedom	P value
W.dist	10.40	1	0.001**
P	2.14	2	0.344
Water	81.25	1	0.000***
W.dist x P	0.60	2	0.743
W.dist x Water	1.68	1	0.195
PxWater	3.46	2	0.178
W.dist x Water x P	0.03	2	0.983

Table T2: ANOVA results relating to the effects of water distribution (W. Dist.), P fertilization (P) and overall water supply rate (Water) and their interactions on shoot P concentrations.

	Chisquare	Degrees of freedom	P value
W.dist	0.02	1	0.901
P	36.10	2	0.000***
Water	24.76	1	0.000***
W.dist x P	0.21	2	0.899
W.dist x Water	3.17	1	0.075
PxWater	6.03	2	0.049*
W.dist x Water x P	2.64	2	0.268

Table T3: ANOVA results relating to the effects of water distribution (W. Dist.), P fertilization (P) and overall water supply rate (Water) and their interactions on root length.

	Chisquare	Degrees of freedom	P value
W.dist	2.90	1	0.088
P	2.42	2	0.299
Water	9.61	1	0.002**
W.dist x P	2.98	2	0.226
W.dist x Water	3.16	1	0.076
PxWater	0.13	2	0.937
W.dist x Water x P	1.56	2	0.458

Tables T4: ANOVA results relating to the effects of water distribution (W. Dist.), P fertilization (P) and overall water supply rate (Water) and their interactions on cluster length.

	Chisquare	Degrees of freedom	P value
W.dist	4.20	1	0.041*
P	19.61	2	0.000***
Water	1.32	1	0.251
W.dist x P	0.48	2	0.785
W.dist x Water	0.97	1	0.326
PxWater	2.06	2	0.357
W.dist x Water x P	1.44	2	0.488

Supplementary root data

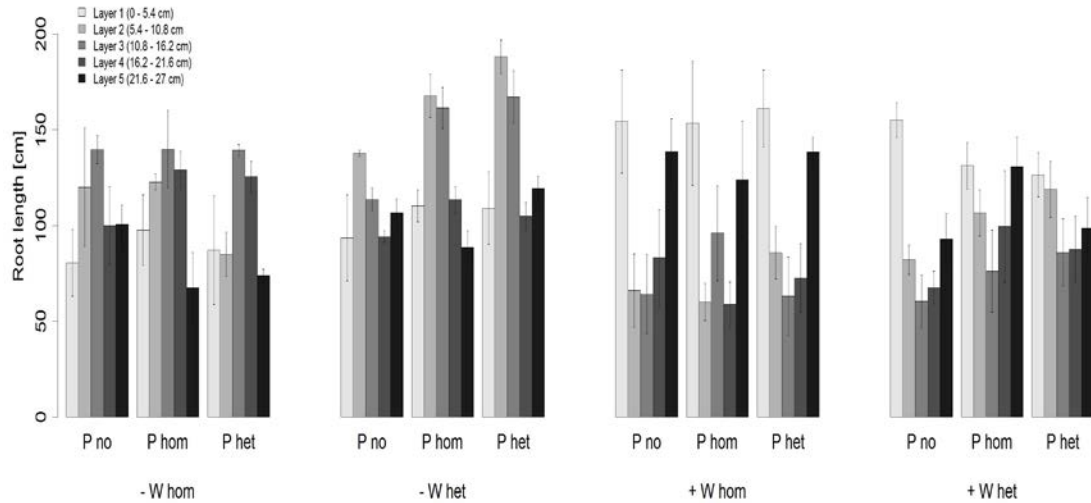


Figure S4: Vertical root length distribution 35 days after germination of white lupin (*Lupinus albus* L.) at low (-) or high overall water supply (+) in homogeneous (W hom) or heterogeneous (W het) substrate and with no additional P supply (P no), homogeneous P fertilization (P hom) or heterogeneous P fertilization (P het). The five bars given for each treatment combination represent the vertical distribution of root length from the top (lightest shading) to the bottom (black) of the containers. The error bars are the standard errors of the mean. At high water supply more root length was produced at the top and the bottom of the containers than at intermediate depths, while the opposite was the case in the treatments with low water supply.

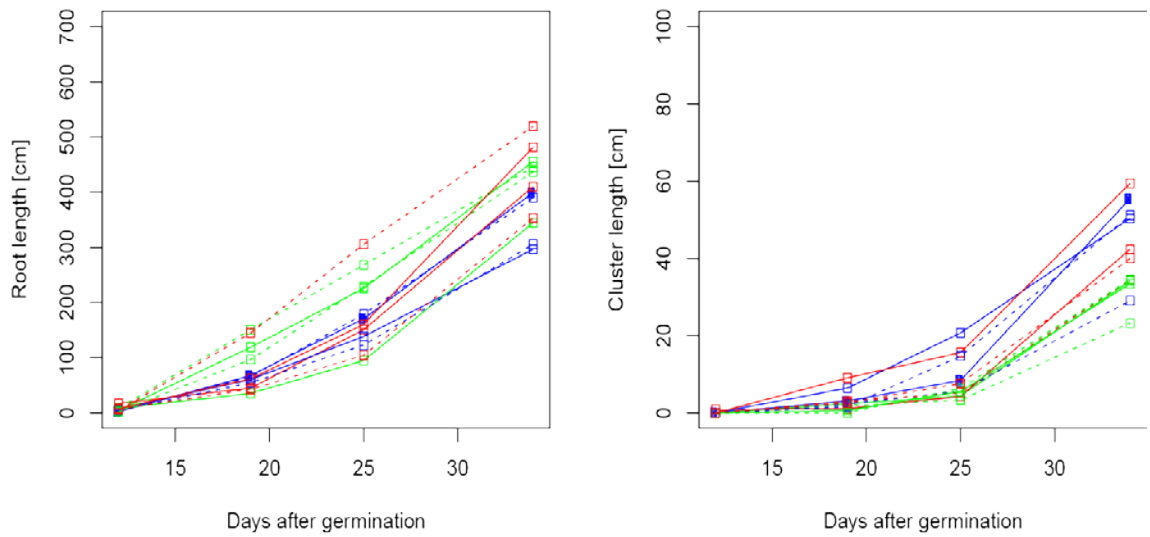


Figure S5: Root length and cluster length production of white lupin (*Lupinus albus* L.) in containers with horizontally heterogeneous (dashed lines) and homogeneous (solid lines) soil water availability and no P addition (blue lines), homogeneous P fertilization (green lines) or heterogeneous P fertilization (red lines). While root length production was almost linear over the experimental growth period, most of the cluster roots were produced between 25 and 35 days (harvest) after germination

Nutrient concentrations in the shoot

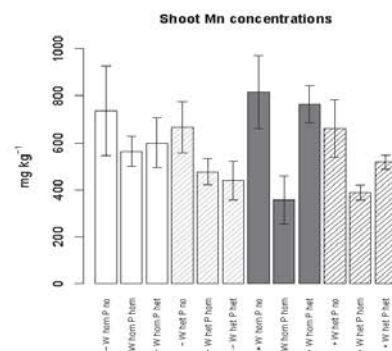
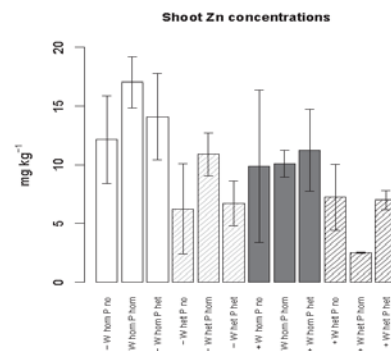
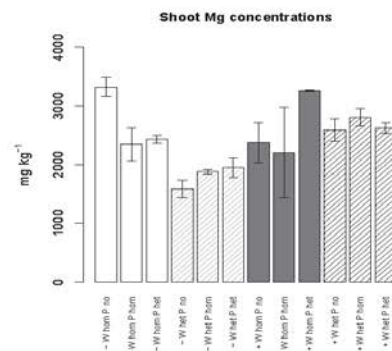
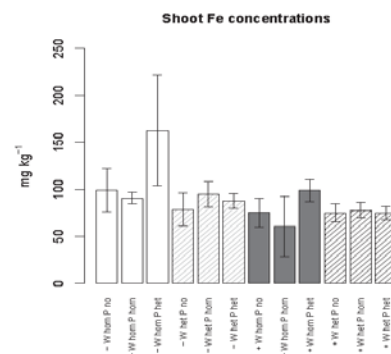
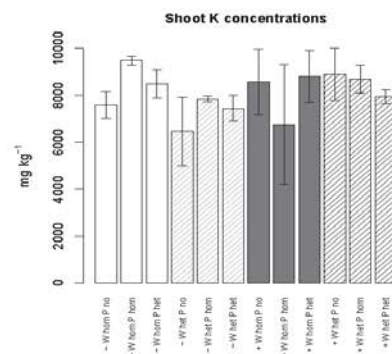
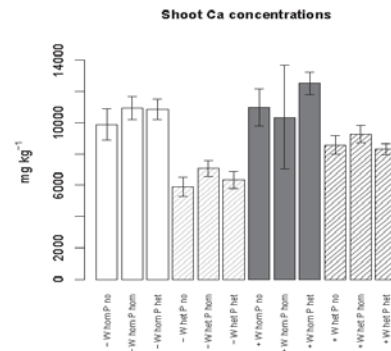
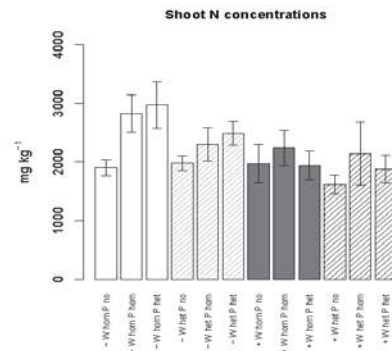


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Figure S6: Shoot element concentrations of white lupin (*Lupinus albus* L.) grown at low (-) or high water supply (+), in homogeneous (W hom) or heterogeneous (W het) substrate with no additional P supply (P no), homogeneous P fertilization (P hom) or heterogeneous P fertilization (P het). Bars represent averages of all plants with herringbone root system. The error bars give the standard errors of the means.