Crops grown in mixtures show niche partitioning in spatial water uptake

Anja Schmutz[1] and Christian Schöb[1,2]

[1]Department of Environmental Systems Science, ETH Zurich, Universitätstrasse 2, 8092 Zurich, Switzerland

[2]current address: University Rey Juan Carlos, c/ Tulipán s/n, 28933 Móstoles, Madrid, Spain

Correspondence

Anja Schmutz

Email: anja.schmutz@usys.ethz.ch

Abstract

1. More diverse plant communities generally produce more biomass than monocultures. This benefit of plant diversity is supposed to stem from resource complementarity of species in mixtures. Different plant species might use the resources spatially, temporally, or chemically in different ways. Along the same lines, for agricultural production crop mixtures outperform monocultures. Complementarity in water use of crop species in mixtures could result in higher yield. Therefore, complementarity in water use could be relevant for crop production in the future when water for agriculture becomes increasingly limited.
2. Here we used the stable isotopes of water and a Bayesian model to investigate the spatial water uptake patterns of six different crops species and how these patterns change upon increasing crop diversity. In addition, we calculated niche overlaps of water uptake and compared them among the different diversity levels.
3. The spatial water uptake pattern differed among crop species. The effect of crop diversity had a minor effect on water uptake but varied strongly depending on the crop species. Niche overlap in water uptake was highest in monocultures and decreased strongly in mixtures. Additionally, we show that increased competition intensity caused stronger changes in water uptake patterns.
4. *Synthesis*. We found evidence for niche partitioning of water uptake in crop mixtures which might lead to a more efficient use of the limited resource water in intercropping systems. Intercropping can therefore increase and stabilise crop yields under forthcoming drier climatic conditions.

Introduction

Water is essential for plant growth and is also a limiting factor for crop production especially in dry regions. To fully exploit different water sources available on agricultural land, mixed cropping (i.e. the simultaneous cultivation of two and more crops in the same field) might be a solution. In fact, mixed cropping has already shown to increase water-use efficiency compared to monocropping practice (Morris and Garrity, 1993). Furthermore, mixed cropping systems show stable or even enhance crop yield even under drought stress (Renwick et al., 2020). Yield benefits in mixed cropping systems can be explained through complementary resource use where distinct species use resources temporally and/or spatially different (Barry et al., 2019).

Plant water uptake is influenced by many physiological and (abiotic and biotic) environmental factors. Physiological characteristics include plant root architecture and distribution (Ferchaud et al., 2015; Y. Liu et al., 2011; Tron et al., 2015), plant life cycle (i.e. annuals vs perennials) (Ehleringer et al., 1991) and temporal dynamics such as development stage (Ma and Song, 2018; Wang et al., 2017). Abiotic factors such as soil water content (Y. Liu et al., 2011), temperature and precipitation history (Nippert and Knapp, 2007; Querejeta et al., 2021) and topography (Nippert and Knapp, 2007) can also influence plant water uptake. Furthermore, biotic factors such as plant diversity (O'Keefe et al., 2019; Schwendenmann et al., 2015) and co-existing plants (Moreno Gutiérrez et al., 2012) have also been shown to affect plant water uptake.

To study spatial plant water uptake, the natural abundance of the stable isotopes in plant and soil water (Allison et al., 1983; Barnes and Allison, 1983; Dalton, 1988) and the subsequent analysis with mixing models (Wang et al., 2019) can be used. Most of

the studies exploring spatial plant water uptake using this method were conducted in natural systems (Beyer et al., 2018; Y. Liu et al., 2011; Querejeta et al., 2021), in grasslands (O'Keefe et al., 2019) or in forests (Schwendenmann et al., 2015). Some studies also used crops such as wheat (Liu et al., 2021, 2020; Ma and Song, 2018; Yang et al., 2018), barley and rapeseed (Wu et al., 2018), and maize (Asbjornsen et al., 2007; Ma and Song, 2016; Wang et al., 2010). Some studies linked also diversity with plant water uptake in grassland and tree species, respectively, and found complementarity in water use of co-occurring species (O'Keefe et al., 2019; Schwendenmann et al., 2015). But we are not aware of any study linking water uptake of crops to crop diversity in intercrop systems.

Thus, this study aimed to investigate the spatial water uptake of different crop species and to examine if there were changes in spatial water uptake with increasing crop diversity and in specific crop combinations. A field experiment was conducted where six different crops were grown in different diversities (one, two or three species grown together). The natural abundance of stable isotopes in plant and soil water and the mixing model MixSIAR (Stock et al., 2020) was used to calculate the proportion of water from different soil layers taken up by the plants. Furthermore, the niche overlap between pairs of species were calculated. We hypothesised that different crop species differ in their proportions of water taken up from different soil layers, the plants show plasticity in water uptake and shift to different water sources depending on the neighbour crop identity and species diversity.

Material and methods

Study site

The field experiment was conducted near Zürich, Switzerland (coordinates 47°26′19.917″N 8°29′58.930″E and 455 m a.s.l.). The field site is located in a

temperate climate with annual mean temperature of 9.3 °C and annual mean perception around 1000 mm (norm period 1981-2010, MeteoSchweiz). During the field experiment, temperature ranged from 0°C in the beginning of May up to 36°C at the end of June (Fig. 1). During June 2019, the hottest days ever were recorded, and June was the second hottest June since 1864. Furthermore, June was a dry month with only two-thirds of usual rainfall during this month (Fig. 1).

The soil at the study site was a brown earth which is profound and with normal water-permeability. The soil consists of 56% sand, 18% clay, 25% silt, 3% hummus, 8% gravel and 2% stones.

Seed material

In this experiment, six crop species were used, namely spring barley (*Hordeum vulgare* var. Atrika), spring wheat (*Triticum aestivum* var. Fiorina), faba bean (*Vicia faba* var. Fanfare), pea (*Pisum sativum* var. Astronaute), linseed (*Linum usitatissimum* var. Marquise) and rapeseed (*Brassica napus* subsp. *napus* var. Campino). The seeds were purchased from a local retailer (UFA Samen) and are local varieties commonly cultivated in Switzerland. These crop species can be grouped into three functional groups: cereals including wheat and barley, legumes containing faba bean and pea and (oilseed) herbs with linseed and rapeseed.

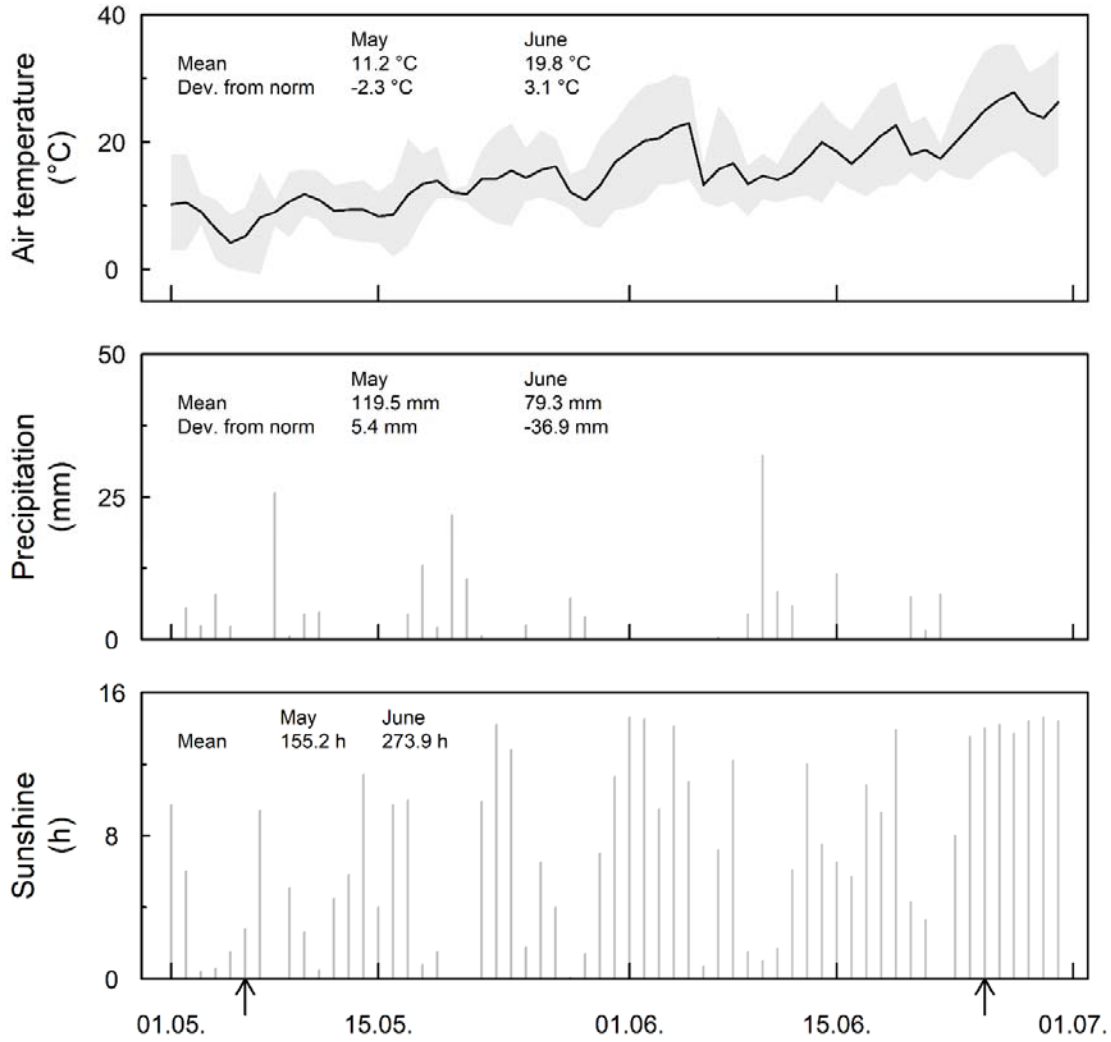

Fig. 1. Air temperature (day mean ± day minima and maxima), precipitation and sunshine duration between May and June 2019. Arrows indicate sowing (6 May 2019) and the sampling event (25 June 2019). For temperature, the daily mean, minima and maxima are shown. For perception and sunshine duration, the day sum is drawn. The monthly mean of the norm period (1981-2010) and the deviation from the norm period of the months May and June 2019 are also indicated. The Data is from the MeteoSchweiz weather station Affoltern (approx. 2 km away from our field site).

Experimental design

The six crop species were grown in plant communities as monocultures and mixtures with either two or three species, respectively. For the 2-species mixtures, all possible combinations among the crop species were cultivated. As for the 3-species mixtures, all possible combinations among the functional groups were grown together (Table S1). In total 29 different plant communities were grown. The 29 community plots

together with three control plots with no plants were arranged in blocks of 4 x 8 plots. This complete set was replicated three times following a randomised-complete block design. Additionally, all crop species were grown as single plants. These single plants experienced no above- and belowground interaction during the whole cropping season. Thus, these plots were weeded regularly to prevent any growth of weeds which could interact with the crops. To exclude any above- and belowground interaction with other crops, all single plants were grown in one separate block which contained five replicates of each crop, randomly allocated within the block. All plots measured 0.5 × 0.5 m and were separated by a metal fence to a depth of approx. 30 cm.

Sowing was conducted by hand on the 6 May 2019 with the sowing density recommended by the seed retailer (210 kg ha^{-1} for wheat, 180 kg ha^{-1} for barley, 250 kg ha^{-1} for faba bean, 275 kg ha^{-1} for pea, 60 kg ha^{-1} for linseed and 6 kg ha^{-1} for rapeseed). The sowing ratio (%) was 50:50 and 33:33:33 for 2-species and 3-species mixtures, respectively, except for rapeseed which had 20% in both 2-species and 3-species mixtures (Table S1).

Water sampling

Sampling for natural ^{18}O and ^{2}H abundance took place on 25 June 2019. In four blocks (three with the community plots and one with single plants), two control plots per block were randomly selected for soil core sampling. Soil cores were taken with a core sampler and the soil was collected in the following depths: 0, 5, 10, 15, 20, 30, 50 and 75 cm.

For the examination of source water, water extracted from the root crown has already shown to match the source water best (Barnard et al., 2006). Hence, for the community plots the root crown of one random individual per species and plot was collected. For linseed, three individuals per plot were pooled, because one individual

did not contain enough water for extraction. For the single plants, three plants per crop were randomly chosen for sampling. The soil and plant samples were collected in 12 ml glass vials, closed with a chlorobutyl rubber septum cap (Exetainer©, Labco Limited) and stored at -20°C until further processing.

Isotope analysis

Water from samples was extracted following the Cryogenic vacuum method (Dalton, 1988). Subsequently, extracted water was analysed for hydrogen and oxygen isotopic composition (δ^2H and $\delta^{18}O$) following the protocol by Werner and Brand (2001). The results were normalised to VSMOW (Coplen, 1988) and are expressed in the δ-notation (Eq. 1).

$$\delta\ ^2H\ [‰] = \left(\frac{\left(\frac{^{18}O}{^{16}O}\right)_{sample}}{\left(\frac{^{18}O}{^{16}O}\right)_{standard}} - 1 \right) * 1000 \qquad \textbf{(Eq. 1)}$$

Biomass sampling

During the water sampling, the shoot from the sampled plants were collected in paper bags and dried at 80°C for at least 48h. Subsequently, the dry biomass was weighed.

Data analysis

The complete analysis was carried out in R version 4.0.2 (R Core Team, 2020).

For the relations between $\delta^{18}O$ and δ^2H in soil water (Fig. 2a) a linear regression model was conducted. For the $\delta^{18}O$ and δ^2H along the soil profile second-order polynomial regressions were computed (Figs. 2b and 2c).

To quantify the proportion of water uptake (PWU) by the different plants grown in the different diversities, the Bayesian isotope mixing model MixSIAR (Stock et al., 2018) was used. This model allows to calculate the proportion of sources (here the soil water from the specific soil depth) of unknown mixtures (here the sampled plant xylem water). In advance, the measured $\delta^{18}O$ and δ^2H values from the specific soil

depths were combined into the following soil layers: shallow (0-10 cm), middle (15-30 cm) and deep (>50 cm). This spatial stratification of the source allows better model inference and analysis (Phillips et al., 2005; Stock et al., 2018). Four models were computed. First, the global PWU was calculated in model 1 (Fig. S1). Model 2 shows the PWU in the different diversity levels (Fig. S1). Model 3 describes the PWU from the six crops in the different diversity levels (fixed factor) with the replicates as random factor (Fig. 3). Model 4 was run for each replicate (specific species composition as fixed factor, no random factor). This model describes how the water uptake differs when specific species are grown together (Fig. S2). Each model was run with a chain length of 300'000, burn-in of 200'000, thinning of 100 and with three chains ("long" run in MixSIAR).

Furthermore, as a measure of the intensity of plant-plant interactions, the relative interaction index (RII) with the biomass of the plant when grown in community (B_c; monoculture, 2- or 3-species mixture, respectively) and when grown without any other plant (B_s; single plants) was calculated (Armas et al., 2004) (Eq. 2, Fig. 4).

$$RII = \frac{(B_c - B_s)}{(B_c + B_s)} \qquad \textbf{(Eq. 2)}$$

The RII gives information about how the biomass changes upon interaction with neighbours, RII<0 indicates competition (negative interaction) whereas RII>0 indicates facilitation (positive interaction). Deviations of RII from zero were tested with a one-sample t-test. To test for effects of diversity on RII within species, a generalised linear model (GLM) with beta distribution and subsequent post-hoc (estimated marginal means) were calculated. RII was previously transformed to fit a beta distribution (Eq. 3).

$$RII_t = \frac{(RII + 1)}{2} \qquad \textbf{(Eq. 3)}$$

For the relationship between RII and mean PWU, a GLM with beta distribution was computed with the mean PWU as response variable and the RII, soil layer diversity (monoculture, 2- and 3-species mixture) and their interaction as explanatory variables (Fig, S3, Table 2). This relationship gives information of how spatial water uptake changes upon interaction intensity, crop diversity and soil depth.

Finally, niche overlap in water uptake was calculated from the overlap of the posterior distribution estimated in model 3 (Fig. S4). For this, the overall of the kernel density estimates between all the possible species combinations was calculated with the R package *overlapping* (Pastore, 2018). The use of overlapping estimates of two or more kernel densities has been suggested for calculating niche overlap rather than only compare means or use the geometrical overlap of kernel densities (Pastore, 2018; Pastore and Calcagnì, 2019; Swanson et al., 2015). Subsequently, a hierarchical cluster analysis (*hclust* function in R) was applied to the overlapping values (Fig. 5). This analysis reveals patterns in niche overlaps in the different diversity levels, soil layers and in the specific species combinations.

Results

Isotopic signature of soil water

The $\delta^{18}O$ and $\delta^{2}H$ values from water extracted in the specific soil depths were correlated ($P < 0.001$) and showed $\delta^{18}O$ values between -1‰ and -11‰ and $\delta^{2}H$ values between -27‰ and -77‰ (Fig. 2a). When looking at the isotopic signature along the soil profile the following pattern is visible: The topsoil is isotopically enriched, and the isotopic signatures of $\delta^{18}O$ and $\delta^{2}H$ decrease with increasing soil depth ($P < 0.001$ for both $\delta^{18}O$ and $\delta^{2}H$) (Figs. 2b, 2c).

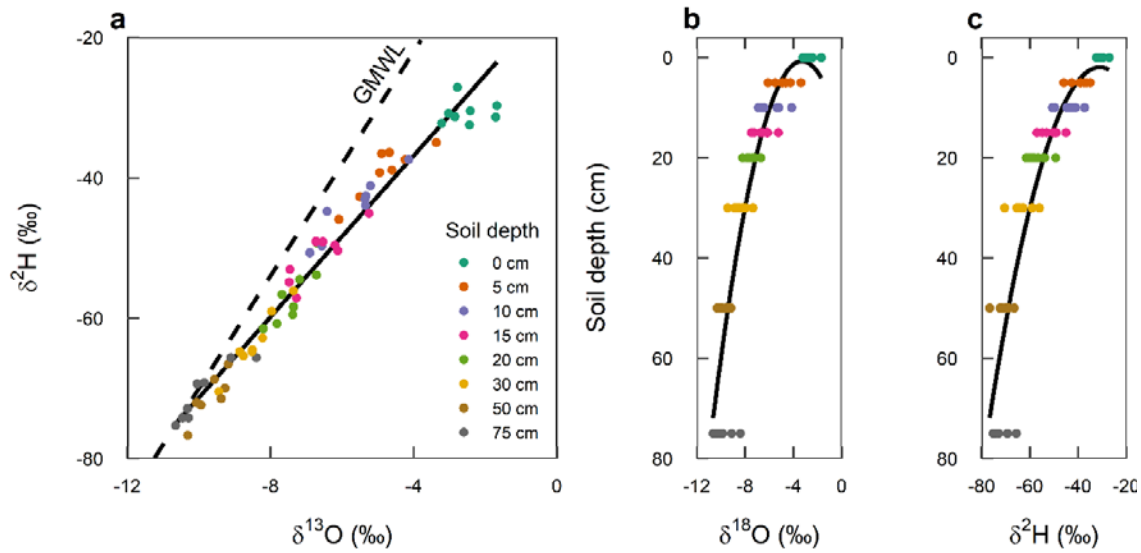

Fig. 2. Isotopic signature of $\delta^{18}O$ and $\delta^{2}H$ (‰) from water extracted at specific soil depths. (**a**) Relationship between $\delta^{18}O$ and $\delta^{2}H$. Water extracted from the soil depth are coloured accordingly (points in different colours). The regression line of the soil water is shown (solid line; y = -13.8 + 5.7x; P < 0.001; R^2 = 0.97; N=64). The global meteoric water line is also shown (GMWL; dashed line; y = 10 + 8x). (**b**) $\delta^{2}H$ values along the soil profile. Shown is the regression line (y = -26 + 165 - 48x^2; P < 0.001; R^2 = 0.82) and the individual samples per soil depth (coloured points; N=8). (**c**) $\delta^{18}O$ values long the soil profile. Shown is the regression line (y = -26 + 162x - 65x^2; P < 0.001; R^2 = 0.85) and the individual samples per soil depth (coloured points; N=8).

Proportion of water uptake in the different soil layers

In our experiment, almost 75% of water was taken up in the shallow soil layer (0-10 cm) (Fig. S1a; model 1). This pattern did not change in the different diversity levels but across all crop species (Fig. S1b; model 2). Nevertheless, differences in the mean PWU among crop species grown in the diversity levels among crop species were visible (Fig. 3; model 3). In general, the deep soil layer (>50 cm) was not important for any species and in any diversity level. The highest amount of water taken up in this layer was 9% of total water by rapeseed. The importance of the shallow and middle (0-10 cm and 15-30 cm, respectively) soil layers differs among crop species and between the diversity levels (Fig. 3). For the cereals, barley and wheat, the shallow and middle soil layers were both important sources of water. Cereals grown as single plants used water from shallow and middle soil layer almost equally (around 50% of water uptake in these layers). For wheat, the proportion did

not change when grown in a community (monoculture) and with increasing diversity (2- and 3-species mixtures), whereas in barley the middle soil layer became less important in community and with increasing diversity. For both legumes, faba bean and pea, most of the water was taken up in the shallow soil layer (up to 90% of the total amount of water). The proportion did not change among the diversity levels. For linseed, shallow and middle soil layer were both an important source of water (approx. 60-70% and 25-35%, respectively). On the other hand, rapeseed showed a clear shift from shallow and middle soil layer (approx. 60% and 30-35%, respectively), when grown as single plant or in monocultures, to middle soil layer as the main water source (approx. 70%) in mixtures.

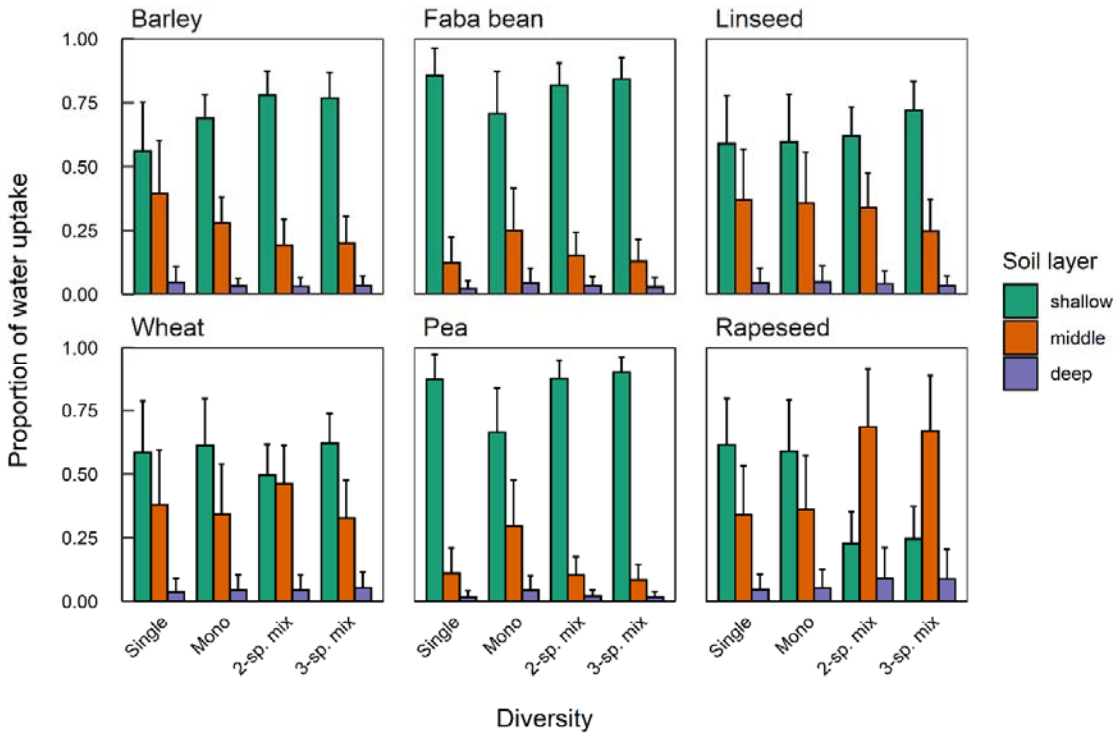

Fig. 3. Proportion of water uptake from the different soil layers by the six crops barley, wheat, faba bean, pea, linseed, and rapeseed grown as single plant, in monoculture and in 2- and 3-species mixtures (mean + standard deviation of the mean). The proportions were calculated with the mixing model MixSIAR (model 3; species*diversity as fixed factor, species composition as random factor).

Competition intensity

Across almost all species and diversity levels, RII was significantly smaller than zero (Fig. 4). Only faba bean and linseed grown in monoculture showed no reduction of biomass upon interaction. RII differed among species (P<0.001, Table 1). Rapeseed showed the highest reduction of biomass upon interaction with neighbours (approx. -0.85), closely followed by the two cereals barley and wheat (approx. -0.7 and -0.6., respectively). Across all crop species, diversity had no effect on RII (Table 1). Only linseed responded to diversity with a lower RII when grown in mixtures compared to when grown in monoculture (Fig. 4).

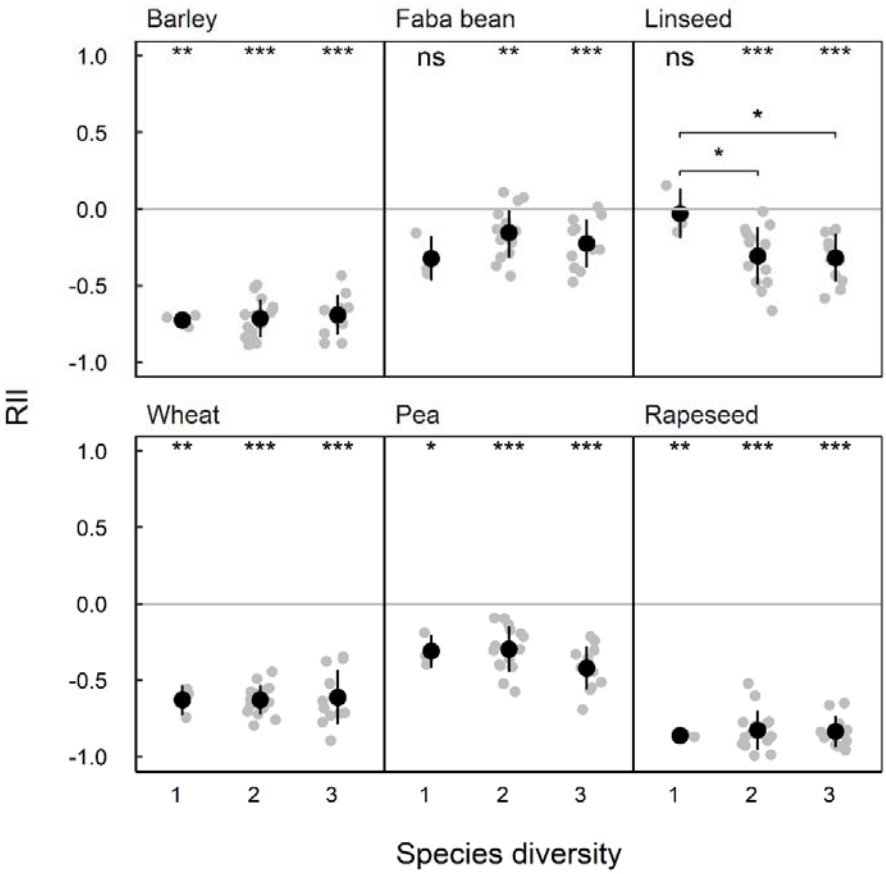

Fig. 4. Relative interaction index (RII) calculated as the relative change in biomass between individuals grown in community (monocultures and 2- and 3-species mixtures) and as single plant without neighbours. Drawn is the mean ± the standard deviation of the mean (black points and lines) and the individual data points (grey points). Significant differences to zero of the RII are shown at the top of the panels (one-sample t-test). Significant differences among species diversity within species are shown with brackets (least-squares means). Asterisks correspond with * p<=0.05, ** p<=0.01 and *** p<=0.001.

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Table 1. Type III-analysis of variance (ANOVA) of the generalised linear model with the transposed relative interaction index (RII) as response variable and species (barley, wheat, faba bean, pea, linseed and rapeseed) and diversity (monoculture, 2- and 3-species mixture) and their interaction as explanatory variable. Df: degrees of freedom of term, Chisq: Wald chi-squared, P-value: error probability. P-values in bold are significant with α=0.05. N=177.

	Df	**Chisq**	**P-value**
species	5	252.34	**<0.001**
diversity	2	0.88	0.643
species × diversity	10	13.28	0.209

Relationship between RII and mean PWU

To investigate if patterns in water uptake change upon interaction intensity, the relationship between mean PWU and RII was calculated. The GLM and subsequent type-III ANOVA indicated significant effects of RII × soil layer × diversity ($p<0.001$; Table 2). This implies that the mean PWU is correlated with RII, but the correlation depends on the soil layer and on plant diversity. In all soil layers, mean PWU and RII were related in the mixtures but not in the monocultures (Fig. S3). In the two- and three-species mixtures the correlation is positive in the shallow soil layer and negative in the two deeper soil layers. In other words, more intense competition led to a lower proportion of water uptake in the shallow soil layer, whereas in the other soil layers, more intense competition led to a higher proportion of water uptake. This indicates that upon more intense competition, plants use more water from deeper soil.

Table 2. type-III analysis of variance (ANOVA) from the generalised linear model with mean proportion of water uptake as response variable (estimated in model 4) and the relative interaction index (RII), soil layer (shallow, middle and deep) and diversity (monoculture, 2- and 3-species mixture) and their interaction as explanatory variables. Df: degrees of freedom of term, Chisq: Wald chi-squared, P-value: error probability. P-values in bold are significant with α=0.05. N=513.

	Df	Chisq	P-value
RII	1	0.00	0.978
soil layer	2	49.61	**<0.001**
diversity	2	0.32	0.854
RII × soil layer	2	0.01	0.997
RII × diversity	2	0.19	0.908
soil layer × diversity	4	18.84	**<0.001**
RII × soil layer × diversity	4	14.35	**0.006**

Niche overlap

The cluster analysis revealed three distinct clusters of PWU among the different diversity levels and soil layers (Fig. 6, x-axis): the first cluster with the monocultures independent of soil layer (D1) and the deep soil layer independent of the diversity level (L3), the second cluster with the shallow and middle soil layer (L1 and L2) in single plants (Ds) and the third cluster with the shallow and middle soil layer (L1 and L2) in 2- and 3-species mixtures (D2 and D3). The first group including all monocultures was characterised with high overlap values (≥ 0.5). In the second cluster including the single plants, overlap values depended strongly on the hypothetical species combination. When the two legumes (faba bean and pea) would be combined with the other species, overlap values would be low (<0.3) whereas these values would be higher among the other species (>0.8). The third cluster including all the mixtures shows low to very low overlap values (<0.5, with some exceptions). This indicates that niche overlap is lowest in the two mixtures. Further, niche overlap is also less in mixtures than we would expect from the hypothetical species combinations of isolated (single) plants.

The clustering of the species combinations (y-axis) revealed two clear clusters (Fig. 6). These two clusters mainly differ in their overlap value estimated for the two mixtures (D2 and D3) in the two upper soil layers (L1 and L2). The cluster with higher

overlap values included all mixtures with barley or faba bean, except when they are combined with rapeseed and faba bean-wheat, and the combination wheat-linseed. Overlap values of rapeseed with any other species were low (<0.29), as are the combinations of wheat with faba bean and pea, and pea-linseed. To summarise, niche overlap in water uptake is lowest in the mixtures – especially when the combination includes rapeseed or wheat with legume.

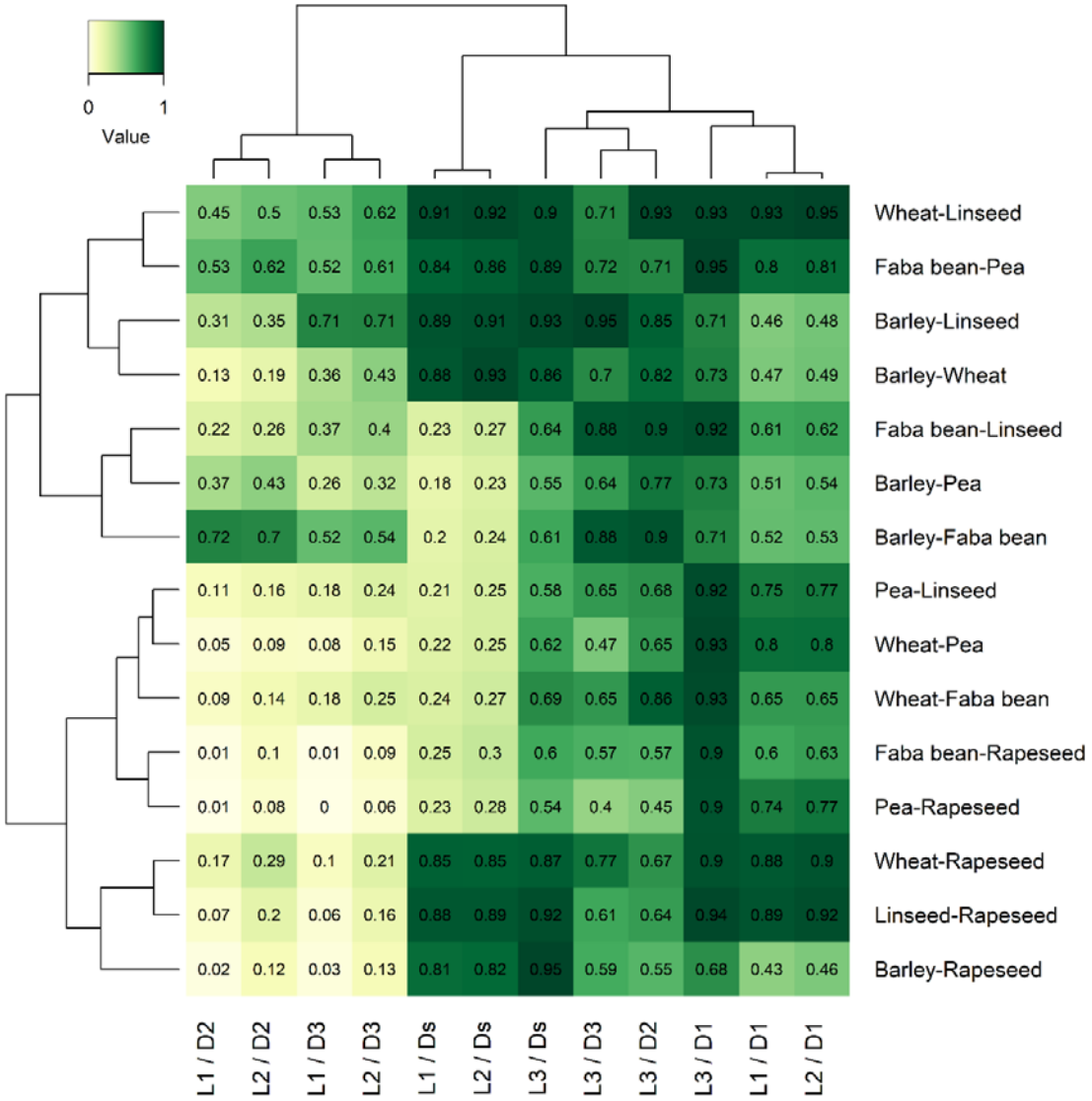

Fig. 5. Heatmap of the estimated overlaps from the posterior densities estimated by model 3. Overlaps were calculated between all the possible combinations of species and for the interactions of soil layer (L; 1=shallow, 2=middle, 3=deep) and the diversity (D; s=single plant, 1=monoculture, 2=2-species mixture, 3=3-species mixture). Overlap values range from zero to one and are also indicated in the corresponding squares. Histograms show hierarchical clustering (*hclust* function in R).

Discussion

The methods applied, which included stable isotopes of water and subsequent application of mixing models, allowed to investigate the spatial water uptake of six different crop species grown in monocultures and mixtures and to assess niche partitioning between species pairs.

General patterns in spatial water uptake and the relationship to root distribution

The shallow (0-10 cm) and middle (15-30 cm) soil layer were the most relevant water sources for the inspected crop species. These findings are in accordance to the plant water uptake globally in the temperate climate zone (Amin et al., 2019). Nevertheless, spatial water uptake depends strongly on plant growth stage (Ma and Song, 2018, 2016; Wang et al., 2017; Wu et al., 2018). These studies show that crop species used a big proportion from shallow soil (0-20 cm) in early stages and shift to deeper soil layers just before flowering. In our study, the crops were in the stage of heading (wheat and barley), flowering (faba bean), and early fruit development (pea, linseed and rapeseed). During these stages, the roots are expected to be fully developed (Weaver, 1926). Plant roots therefore explore the greatest possible soil area and depth.

In fact, it is suggested that water uptake and root distribution are tightly coupled (Gardner, 1964). However, results from field studies are controversial with evidences of both positive and negative relationships between PWU and root length density (Guderle et al., 2018; Liu et al., 2021; Ma and Song, 2018). The six different crop species indeed have distinct rooting patterns (Fan et al., 2016; Kutschera et al., 2009). For instance, rapeseed shows a higher root length density compared to other oilseed and pulse crops (among others linseed and pea) (L. Liu et al., 2011), whereas faba bean has in general a shallow root system (Li et al., 2006). These distinct rooting patterns are reflected in the observed differences in spatial water uptake of the crop species.

Crop diversity and its effect on competition and plasticity in water uptake

In our system, competition (i.e. RII) was very strong. Almost all species showed a reduction of biomass when grown in communities. However, literature suggests that

more diverse communities show reduced competition compared to monocultures due to niche differentiation (Loreau and Hector, 2001). In our system, the effect of crop diversity on competition intensity was limited, and even showed the opposite direction (stronger competition with increasing crop diversity as seen in linseed).

Competition also affected water uptake patterns of the investigated crop species. Upon more intensive competition, the crops used less water from shallow soil and more from deeper soil. Hence, plant roots seem to escape into deeper soil layers when competition is strong, but only when grown in mixture. These effects suggest plasticity in spatial water uptake in mixtures, but not in monocultures, which is supported by other studies (Wang and Callaway, 2021). From the six crop species investigated, only rapeseed and barley showed plasticity in spatial water uptake in response to crop diversity. Rapeseed showed a clear shift and switched from shallow to deeper soil layer in more diverse cultures (2- and 3-species mixtures). In contrast, barley used more water from the shallow soil layer with increasing crop diversity. The other crop species showed almost no response to crop diversity. This implies that plasticity in the six crop species investigated is limited.

In general, literature suggests that plant diversity influences spatial water uptake for instance in tree and grassland communities (Guderle et al., 2018; O'Keefe et al., 2019; Schwendenmann et al., 2015). Furthermore, plant root plasticity in response to neighbouring plants can be high (Callaway et al., 2003) and plant roots segregate spatially to avoid competition (Schenk et al., 1999). Even though plants are suggested to be highly plastic in general (Sultan, 1987), domestication of crops might have reduced the plasticity, for example in water use (Matesanz and Milla, 2018). Nevertheless, this plasticity might be relevant for crop production (Nicotra and Davidson, 2010). It is proposed that future breeding programs should emphasise on adaptive plasticity of crop species (Brooker et al., 2022; Milla et al., 2017) such as

root plasticity (Schneider and Lynch, 2020) especially in more variable environments and due to climate change (Matesanz et al., 2010; Nicotra et al., 2010). Furthermore, higher plasticity might also be favourable in mixtures, particularly for the complementary use of resources (Zhu et al., 2015).

Complementarity in spatial water uptake and niche partitioning

It is suggested that resource complementarity is stronger in more diverse communities (Barry et al., 2019). This is mainly due to species-specific differences in niche occupation. Without interaction, plant species occupy a specific resource niche (fundamental niche) (Vandermeer, 1972). Upon interaction with other plant species, these fundamental niches can shift (realised niche) (Vandermeer, 1989). In our experiment, the plants without any interaction (single plants) represent the fundamental niche in spatial water uptake. These fundamental niches do not change much when the crop species are grown in monoculture. However, when the crops are grown in mixtures, these niches shift substantially. Consequently, niche overlaps between crop pairs were greatly reduced in mixtures. This finding suggest niche partitioning of spatial water uptake in more diverse crop communities, similar to previous findings from semi-natural grasslands (Guderle et al., 2018).

Furthermore, we found that niche partitioning depended on the soil depth and on the specific hypothetical species composition. Niche overlaps were smaller in the first 30 cm of the soil and increased in the deeper soil layer, but only in the mixtures. The deep soil layer (50-75 cm) was a relatively unimportant water source across the six crop species. These findings are in accordance with results from grassland communities, which showed that increased species diversity can lead to a higher water uptake especially in 20-30 cm soil (but also up to 60 cm soil depth) (Guderle et al., 2018). Additionally, niche partitioning was higher in mixtures that included rapeseed and wheat. Especially the combination of rapeseed with one of the

legumes showed almost no overlap in spatial water uptake. The combination of crop species with spatially contrasting water uptake patterns can facilitate efficient use of available water in the field, in particular when water is limited.

Conclusion and further research

In this study we demonstrate that the investigated crop species used water spatially different.

We further show that plasticity of spatial water uptake in response to plant diversity is limited since only two out of six crop species responded to higher plant diversity. Moreover, niche overlap was greatly reduced in mixtures compared to monocultures. Apart from diversity, also soil depth and specific species composition influenced niche overlap of pairs of crops.

It would now be interesting to link niche partitioning in water use to yield in agricultural systems. Additionally, investigation of drought resilience upon niche partitioning would be relevant especially in rainfed agricultural areas and where drought events might become more frequent due to climate change (IPCC, 2014). In this context, investigation of the temporal dynamic of niche partitioning and water use might be interesting in irrigated agricultural areas to improve efficient application of irrigation water.

Author contributions

AS planned and conducted the experiment, analysed the data, and wrote the manuscript. CS gave inputs to the experimental design, data analysis and the manuscript.

Data Availability Statement

The data will be published via an open-access repository zenodo or similar.

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