Water uptake patterns of pea and barley responded to drought but not to cropping systems

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Highlights

- Pea and barley shifted to shallower water uptake depths in response to drought.
- No niche differentiation was found between pea and barley in a mixture under drought.
- No differences on changes in uptake depths by drought were found among cropping systems.
- Thus, cropping systems did not compensate drought effects on water uptake patterns.

Abstract

Agricultural production is under threat of water scarcity due to increasingly frequent and severe drought events under climate change. Whether a change in cropping systems can be used as an effective adaptation strategy against drought is still unclear. We investigated how plant water uptake patterns of a field-grown pea-barley (*Pisum sativum* L. and *Hordeum vulgare* L.) mixture, an important fodder intercrop~~crop~~, responded to experimental drought under four cropping systems, i.e., organic intensive tillage, conventional intensive tillage, conventional no-tillage, and organic reduced tillage. Drought was simulated after crop establishment using rain shelters. Proportional contributions to plant water uptake from different soil layers were estimated based on stable water isotopes using Bayesian mixing models. Pea plants always took up proportionally more water from shallower depths than barley plants. Water uptake patterns of neither species were affected by cropping systems. Both species showed similar responses to the drought simulation and increased their proportional water uptake ~~contributions~~ from shallow soil layer (0-20 cm) in all cropping systems. Our results highlight the impact of drought on plant water uptake patterns for two important crop species and suggest that cropping systems might not be as successful as adaptation strategies against drought as previously thought.

Keywords: climate change, conservation tillage, ~~FAST,~~ organic farming, stable water isotope, water uptake depth

1 Introduction

Due to climate change, drought events may occur more frequently and become more severe than at present, and hence water scarcity is worsening in many regions of the world (Schewe *et al.*, 2014; IPCC, 2019). Thus, agriculture is facing increasing pressure to ensure food security under aggravating drought conditions (FAO, 2018; FAO, 2019). Although crop breeding has large potential to enhance agricultural productivity, it should certainly not be seen as the only option. Adaptive crop management to a changing climate is discussed as an additional solution to mitigate yield loss under drought, ~~either~~ potentially by sustaining plant growth, ~~or by~~ enhancing soil water availability, or by promoting mycorrhizal symbiosis (Cochard, 2002; Bot & Benites, 2005; Kundel *et al.*, 2020; Wahdan *et al.*, 2021). Therefore, there is a growing interest in organic farming and conservation tillage (i.e., no tillage or reduced tillage), as these management practices have been shown to be beneficial to soil health and water holding capacity, ecosystem stability, as well as environmental sustainability (e.g., Seitz *et al.*, 2019; Teasdale *et al.*, 2007; Hobbs *et al.*, 2008; Wittwer *et al.*, 2021). However, an evaluation of different cropping systems as a means to support arable crops under drought is still urgently needed (IPCC, 2019).

Understanding plant water relations under drought plays an increasingly important role in promoting sustainable agriculture to secure food production (Penna *et al.*, 2020). Plant water uptake and water use, particularly during critical growing stages, greatly determine physiological processes, survival, and ultimately crop productivity (Boyer & Rao, 1984; Wang *et al.*, 2015). Although many studies reported plant water uptake patterns in response to drought over a broad range of species and environments (e.g., Prechsl *et al.*, 2015; Grossiord *et al.*, 2019; Rasmussen *et al.*, 2020; Ding *et al.*, 2021), only very few focused on arable agriculture (e.g., Zegada-Lizarazu *et al.*, 2006; Borrell *et al.*, 2014; Wu *et al.*, 2018) and none compared arable cropping systems. Moreover, these studies found contrasting responses of crop species to changing environments, illustrating the current gap of knowledge ~~for~~ on plant water relations in cropping systems.

Plant water uptake mainly depends on soil water availability, root properties and distributions, as well as soil-plant interactions (von Freyberg *et al.*, 2020). Soil water availability depends on soil physical characteristics and local climatic conditions. Root systems, including root distribution and functionality, are affected by soil physical and nutritional conditions as well as plant growth stages and species genetics. Soil-root interactions include hydrotropism, root damage caused by drying soil, and soil water redistribution (Caldwell *et al.*, 1998; Whitmore & Whalley, 2009; Dietrich *et al.*, 2017). Furthermore, plant water uptake patterns are highly dynamic and difficult to track. Since the 1960s, stable water isotopes, i.e. oxygen and hydrogen isotopes, have been used in ecohydrology studies (Gonfiantini *et al.*, 1965; Zimmermann *et al.*, 1967), e.g. to assess root water uptake patterns (Rothfuss & Javaux, 2017), to detect foliar water uptake (Berry *et al.*, 2019), as well as to partition evapotranspiration fluxes (Wang *et al.*, 2010). Stable water isotopes have since become a helpful tool to identify plant water uptake sources and quantify source contributions (Dawson & Ehleringer, 1991; Penna *et al.*, 2018). However, studies in agroecosystems have often focussed on grassland species (e.g. Bachmann *et al.*, 2015; Prechsl *et al.*, 2015), much less on crop species as reviewed by Penna *et al.* (2020).

Hence, our experimental field study investigated how different cropping systems, namely organic vs. conventional farming with intensive vs. conservation tillage, affect plant water uptake patterns under drought using stable water isotopes. We focused on a pea-barley (*Pisum sativum* L. and *Hordeum vulgare* L.) mixture, an increasingly popular intercrop for fodder production (Gilliland & Johnston, 1992). We aimed at understanding (1) if pea and barley ~~grown in mixture~~ differ in their water uptake patterns when grown in mixture, (2) how drought affects plant water uptake depths, and (3) if cropping systems affect water uptake depths differently.

2 Materials and Methods

2.1 Research site and experimental setup

The research site is in Rümlang near Zurich (47.26° N, 8.31° E, 489 m a.s.l.), and belongs to the Swiss federal agricultural research station Agroscope. Long-term average annual precipitation at the site is 994 mm, and mean annual air temperature is 9.7 °C (1988 to 2017; MeteoSwiss, 2020). The soil at the research site is a calcareous Cambisol with 23% clay, 34% silt, and 43% sand, and total soil carbon content of 1.6 to 1.8% (Loaiza Puerta *et al.*, 2018). The plant available soil depth is 50-70 cm, and no groundwater is accessible for plants (Kanton Zürich, 2021). Our study used a sub-set of plots in the Farming Systems and Tillage Experiment which began in 2009 with a six-year crop rotation that is typical for Swiss cropping systems (for details see Wittwer *et al.*, 2017). It combines conventional (C) and organic (O) farming with intensive or soil conservation tillage practices. The conventional systems are managed according to the "Proof of Ecological Performance" (PEP) guidelines of the Swiss Federal Office for Agriculture (Swiss Federal Council, 2021), which allows synthetic fertiliser and pesticide applications. The organic systems were managed following the BioSuisse guidelines, prohibiting the use of mineral fertilisers and synthetic plant protection products. Intensive tillage (IT) with a mouldboard plough to 20 cm depth followed by seedbed preparation with a rotary harrow to 5 cm depth was applied in both conventional (C-IT) and organic systems (O-IT). For conservation tillage, direct sowing and no soil management were implemented in the no tillage conventional plots (C-NT) but glyphosate was sprayed before sowing of the main crops for weed control. A disc or rotary harrow, which superficially disturbed the soil for weed control, was used for reduced tillage in organically managed plots (O-RT) to a maximum depth of 10 cm. These four cropping systems were repeated in four blocks following a Latin square design. Cropping system plots had an area of 6 m × 30 m.

In 2018, the same pea (*Pisum sativum* L. cv. 'Alvesta') and barley (*Hordeum vulgare* L. cv. 'Eunova') mixture was sown in all plots on 26 March and harvested on 12 July (108 days). The mixture was composed of 20% and 80% of the recommended sowing densities of pea (90 seeds/m^2) and barley (350 grains/m^2), respectively. The seeds were sown in a mixture with a standard drill-sowing machine. No fertilisation was applied in any of the treatments because the pea plants were expected to fix dinitrogen from the atmosphere.

In order to simulate a future drought scenario (CH2018), portable rain shelters were installed from 22 May to 28 June 2018 (37 days) during the 108-day growing season. This resulted in a 34% ~~of~~ reduction in precipitation from the drought subplots during the growing season in 2018 (from sowing to harvest; Table 1). No irrigation was applied to the control plots during the (unexpected) naturally dry period in June for logistical and rational reasons, i.e., irrigation is unusual for the region and this crop, and the dry period happened during the ripening phase of the crop. The ~~P~~portable, tunnel-shaped rain shelters (metal frames of 3 m × 5 m base area and 2.1 m height at the highest point) were covered with transparent and ultraviolet light-transmissible plastic foil (Gewächshausfolie UV5, 200 µm, Folitec Agrarfolien-Vertrieb, Germany) ~~were installed to simulate a drought period from 22 May to 28 June 2018. Shelters~~ and were open at both ends as well as at both sides and had an opening at the top along the full length. This allowed extensive ventilation and prevented temperature build-up (for technical details see Hofer *et al.*, 2016). Rain running down the foil was collected in PVC half pipes and directed away from the plots (about 2 m). ~~During the drought treatment period, 34% of precipitation during the growing season in 2018 (from sowing to harvest) was excluded from the drought subplots (Table 1).~~ These drought subplots were established in each cropping system (which were in place since 2009) and located directly next to control subplots which received natural precipitation inputs, resulting in a split-plot layout. A total of 16 experimental plots (four cropping systems × four replicates) with 32 subplots (16 plots × two water availability treatments) were used in this study. Our experimental design thus compared replicated drought and control sub-plots in parallel (i.e., at the same time), not after each other (i.e., a temporal replication over multiple years), since in crop rotations, the identical crop cannot be grown on the same field for several years due to soil health issues.

2.2 Climatic data and soil water contents

Precipitation and air temperature data (Table 1; Fig. 1) were obtained from a nearby weather station, Zürich/Kloten (KLO, 47.48° N, 8.54° E, 4.6 km north of the research site, MeteoSwiss, 2020). Soil water content (SWC) was continuously measured and recorded at 10 and 40 cm depths with two replicates per cropping system (EC-5, Decagon Devices Inc., Pullman, WA, USA; factory-calibrated). Data were averaged at 10 min intervals by data loggers (CR1000 and CR216, Campbell Scientific Ltd., Loughborough, UK), then averaged for daily values.

2.3 Plant and soil water samples for stable isotope analysis

Plant and soil samples were collected on 7 May, 25 June, and 11 July 2018, i.e., before the drought treatment (BT), at the end of the treatment (ET), and after the treatment (AT), respectively. Pea was not sampled AT due to progressed senescence. Root crowns were collected for stable isotope analysis of plant xylem water as this part best reflects the mixture of water sources taken up from the soil in herbaceous plants (Barnard *et al.*, 2006; von Freyberg *et al.*, 2020). Four to six individuals were collected and pooled into one sample per species and subplot. Root crowns were cleaned quickly to remove remaining soil and then immediately sealed in air-tight glass tubes (12-ml exetainer, Labco Ltd., Ceredigion, UK). In parallel to the plant sampling, soil samples were collected close to the sampled plants with a soil auger (1 cm diameter). The soil cores were separated into six depth layers – 0-5, 5-10, 10-20, 20-30, 30-40, and 40-60 cm – and then immediately sealed in glass tubes (18 ml, Schott AG, Mitterteich, Germany). All plant and soil samples for stable water isotope analysis were kept in a cool box in the field and then stored at -18 °C before extraction with cryogenic vacuum distillation (Ehleringer & Osmond, 1989). During the extraction, the samples were kept in an 80 °C water bath, extracted under 10^{-2} MPa for 2 h, and the extracted water collected in glass tubes immersed in liquid nitrogen.

2.4 Stable water isotope analyses

The oxygen and hydrogen stable isotope ratios (δ^{18}O and δ^{2}H) of extracted water samples were analysed by coupling a high-temperature elemental analyser (TC/EA, Finnigan MAT, Bremen, Germany) with an isotope ratio mass spectrometer (IRMS, Delta^{plus}XP, Finnigan MAT, Bremen, Germany) via a ConFlo III interface (Finnigan MAT, see Werner *et al.*, 1999) using the high-temperature carbon reduction method~~s~~ described by Gehre *et al.* (2004). All δ^{18}O and δ^{2}H values are expressed relative to the Vienna Standard Mean Ocean Water (VSMOW-SLAP, Craig & Gordon, 1965; Gat, 2010) in parts-per-thousand (or "per mil", ‰; eq. 1):

$$\delta^{18}\mathrm{O} \text{ or } \delta^{2}\mathrm{H} = \frac{R_{\mathrm{SAMPLE}}}{R_{\mathrm{STANDARD}}} - 1 \tag{1}$$

where R is the isotope ratio of the rare isotope to the abundant isotope ($^{18}\mathrm{O}/^{16}\mathrm{O}$ or $^{2}\mathrm{H}/^{1}\mathrm{H}$). The long-term precision of the quality-control standard *IsoLab 1* over the last four years was 0.22‰ for $d^{18}\mathrm{O}$ and 0.59‰ for $d^{2}\mathrm{H}$.

The isotopic composition of precipitation at the global scale shows a linear relationship between the $\delta^{18}\mathrm{O}$ and $\delta^{2}\mathrm{H}$ of meteoric waters (Global Meteoric Water Line, GMWL; Craig, 1961), described by the regression line in a "dual-isotope" $\delta^{18}\mathrm{O}$-$\delta^{2}\mathrm{H}$ plot (eq. 2):

$$\text{GMWL: } \delta^{2}\mathrm{H} = 8.2 \times \delta^{18}\mathrm{O} + 11.7 \tag{2}$$

Similarly, the Local Meteoric Water Line (LMWL) describes the isotopic composition in rainfall for a specific location (Dansgaard, 1964). We fitted the long-term LMWL (1994 to 2017) with monthly mean data from the closest GNIP station (Global Network of Isotopes in Precipitation, Buchs Suhr, 47.37° N, 8.08° E, 34 km from the research site; IAEA, 2020; eq. 3), while the LMWL of 2018 was fitted with data of precipitation samples collected at the research site (after Prechsl *et al.*, 2014; eq. 4) during the growing season and data of 2018 from GNIP Buchs (Fig. S1):

$$\text{long-term LMWL: } \delta^{2}\mathrm{H} = 7.9 \times \delta^{18}\mathrm{O} + 6.4 \tag{3}$$

$$\text{2018 LMWL: } \delta^{2}\mathrm{H} = 8.3 \times \delta^{18}\mathrm{O} + 12.7 \tag{4}$$

2.5 Bayesian mixing model for plant water uptake

Proportional contributions of soil water to plant water uptake (PC) from different depths were estimated using mixing models from the R package ‘simmr’ (Parnell, 2020) within a Bayesian framework based on code by Parnell *et al.* (2013). The $\delta^{18}\mathrm{O}$ or $\delta^{2}\mathrm{H}$ signatures of soil water from the six soil layers were used as sources, and plant xylem water was considered the mixture for modelling in each subplot at different sampling times, i.e., BT, ET, and AT. Missing replicates of soil samples due to sampling difficulties (n = 5 in total) were filled with mean values of the other replicates from the same cropping system and treatment to have balanced model inputs. The model outputs consisted of 10 000 possible combinations of PC from different soil depths from four Markov chain Monte Carlo Bayesian models with at least 300 000 iterations, 50 000 burns, and 100 times of thinning for each chain. The median of the model outputs on PC (MPC) from each soil depth was calculated for each subplot and used for statistical analysis on plant water uptake depths. Compared to the most frequent value of the model outputs, MPCs of all the sources usually sum up closer to 1. To increase clarity of presentation, PC was grouped into three layers, namely shallow (0-20 cm), middle (20-40 cm), and deep (40-60 cm) soil layers for further analyses. The PC values from shallow and middle layers are the sum of PC from soil depths of 0-5, 5-10, and 10-20 cm, and the sum of PC from soil depths of 20-30 and 30-40 cm, respectively. As $\delta^{18}O$ and $\delta^{2}H$ yielded similar results, only the model outputs of $\delta^{18}O$ are described in detail in this paper.

2.6 Data analyses

For data analyses, the whole growing season was divided into three periods based on the drought treatment, namely before the drought treatment (BT; 26 March to 21 May), the drought treatment period itself (22 May to 28 June) which was sampled directly before the removal of shelters on 28 June (termed ET, end of treatment), and after the drought treatment (AT, 29 June to 12 July). All statistical analyses were carried out using R (v3.6.2; R Core Team, 2020). The effects of cropping systems, drought treatment, and species were tested with linear mixed models using the function *lmer()* from the R package ‘lmerTest’ (Kuznetsova *et al.*, 2017). ‘Cropping systems (CS)’, ‘drought treatment (D)’, and ‘blocks’ were three fixed factors (Dixon, 2016), interactive effects between ‘CS’ and ‘D’ with ‘plots’ (accounting for the split-plot design) were considered as random factors. For variables measured on both pea and barley (i.e., stable isotopes of xylem water and MPC for BT and ET), ‘plant species’, ‘CS’, ‘D’, and ‘blocks’ were tested as fixed factors considering interactive effects among ‘plant species’, ‘CS’, and ‘D’ with ‘plots’ and ‘subplots’ as random factors. Diagnostic plots were checked for normality and homoscedasticity of residuals for model assumptions. Differences among cropping systems and between treatments or species were tested by the Tukey HSD (honestly significant difference) test using the function *glht()*, from the R package ‘multcomp’ (Hothorn *et al.*, 2008).

3 Results

3.1 Environmental conditions in drought and control subplots

Air temperatures in 2018 were very high compared to the long-term mean, in particular in May and June, with a daily average air temperature of 15.8 and 18.8 ºC, respectively, while the long-term (1988 to 2017) mean air temperatures in these two months were 13.9 and 17.2 ºC, respectively (Table 1; Fig. 1). Annual precipitation was relatively low (Table 1). While the precipitation in May 2018 (102 mm) was comparable to the long-term mean (1988 to 2017: 105 mm), ~~, with~~ no precipitation fell between 14 June and 2 July 2018 (naturally dry period), resulting in a below-average precipitation in June (40 mm; long-term mean of 102 mm, Table 1), followed by~~and~~ an even more pronounced drought period in July (Fig. 1). ~~Thus, a~~Average daily soil water contents (SWC) in the control subplots ranged from 16% to 29% at 10 cm depth and slightly higher, from 22% to 29%, at 40 cm depth, prior to the rain event on 3 July 2018. After this rain event, SWC increased in all cropping systems at both depths (Fig. 2a, b). Variations in SWC among cropping systems were small, particularly during the naturally dry period ~~drought~~ in June. SWC in drought subplots of all cropping systems decreased continuously during the 37-day drought treatment (22 May to 28 June 2018), averaging to 13% at 10 cm and to 19% at 40 cm soil depth (Fig. 2 c, d). SWC at 10 cm did not show any pronounced differences among cropping systems, while SWC at 40 cm tended to be slightly higher in cropping systems with conservation tillage (O-RT and C-NT) compared to systems with intensive tillage (O-IT and C-IT; Fig. 2b, d).

3.2 Stable isotopes in soil water and plant xylem water

In the dual-isotope space, stable oxygen and hydrogen isotope ratios of soil and plant xylem waters were strongly related with each other ($R^2 = 0.89$ and 0.85, respectively; Fig. S1) and generally fell below the local meteoric water line (LMWL) of 2018, representing evaporation. Stable isotope signatures of xylem water were lower than the LMWL but higher than those of soil water, indicating that xylem water isotope signatures were mixtures of the original source precipitation and the pool of soil water, affected by different degrees of fractionation.

The stable water isotope profiles of soil water showed a characteristic pattern at all times, for all cropping systems and both treatments, with most enriched values in the uppermost soil and increasingly depleted values with increasing soil depth (Table S1; Fig. 3 for δ^{18}O; Fig. S2 for δ^{2}H). The drought treatment showed no significant effects before the treatment (BT) for δ^{18}O nor δ^{2}H (except for δ^{2}H at 20-30 cm; Table 2). In contrast, at the end of the drought treatment (ET), soil water δ^{18}O values from 20-60 cm (20-30, 30-40, and 40-60 cm) as well as δ^{2}H values from all depths were strongly affected by the drought treatment (all $P < 0.05$; Table 2), with more depleted signatures in the drought than in control subplots due to the exclusion of more enriched summer precipitation. Even after the shelters were removed and the treatment had been finished (AT), the drought treatment still significantly affected both δ^{18}O and δ^{2}H of soil water, albeit only in deeper soil depths (30-40 and 40-60 cm for δ^{18}O and 40-60 cm for δ^{2}H; all $P < 0.05$; Table 2). Overall, cropping systems did not significantly affect the stable isotopic signatures in soil water at any time (Table 2).

Pea xylem water was always significantly more enriched in ^{18}O and ^{2}H compared to barley (all $P < 0.001$; Table S2). The δ^{18}O values in xylem water for pea ranged between -8.8‰ and -5.7‰, and significantly lower between -10.1‰ and -5.8‰ for barley (averages per cropping system, treatment, and time; Table 3; Table S2). Similarly, the δ^{2}H values in xylem water for pea ranged between -65.6‰ and -52.1‰, and significantly lower between -74‰ and -47.1‰ for barley (Table 3~~Table 3~~; Table S2~~Table S2~~). Overall, isotopic signatures in xylem water became more enriched in ^{18}O and ^{2}H during the growing season for both pea and barley (Fig. 3, Table S2, Fig. S2). On average, the xylem δ^{18}O for pea was -8.5‰ before the treatment (BT) and -7.2‰ at the end of the treatment (ET), compared to -9.8‰ (BT), -8.8‰ (ET), and -6.3‰ after the treatment (AT) for barley. While average δ^{2}H values for pea were -64.1‰ (BT) and -57.6‰ (ET), δ^{2}H values averaged -72.2‰ (BT), -68.6‰ (ET), and -50.8‰ (AT) for barley (Fig. 3; Table S1~~Table S1~~; Fig. S2~~Fig. S2~~). Since there was a strong relationship between δ^{18}O and δ^{2}H in xylem water (Fig. S1; $R^2 = 0.85$), our analyses are mainly focused on δ^{18}O in the text (but see Table 3~~Table 3~~, Table S2~~Table S2~~, and Fig. S2~~Fig. S2~~ for analyses on δ^{2}H).

For pea, cropping systems did not significantly affect δ^{18}O nor δ^{2}H in xylem water at either time (BT and ET; Table S2~~Table S2~~), while the drought treatment significantly affected the isotopic signatures of ^{18}O only at the end of treatment (ET: $P = 0.022$; no interactions between cropping systems and drought treatment: $P = 0.085$; Table S2~~Table S2~~). ^{18}O in pea xylem water were significantly more enriched in the drought than in the control subplots (on average, δ^{18}O of -6.9‰ and -7.7‰, respectively).

In contrast to pea, cropping systems significantly affected δ^{18}O in barley xylem water (ET: $P = 0.035$; Table S2~~Table S2~~). The drought treatment significantly affected the isotope signatures of both ^{18}O and ^{2}H at the end of treatment (ET: both $P < 0.01$; no interactions between cropping systems and drought treatment; Table S2~~Table S2~~). However, unlike pea, the xylem water of barley showed significantly lower δ^{18}O values in drought than in control subplots for all cropping systems (on average, -9.0‰ and -8.6‰, respectively), although the difference was small (Table S2~~Table S2~~). A similar pattern was also observed for δ^{2}H at the end of treatment (ET), with significantly lower values on average in drought than in control subplots (ET: -71.8‰ and -65.4 ‰, respectively).

3.3 Modelled plant water uptake depths

The outputs of the Bayesian mixing model on the proportional contribution to total plant water uptake (PC) showed highly significantly different behaviours of pea and barley, mirroring some of the differences seen in the xylem water isotopic signatures of these two species (Fig. 4; Fig. 5). Since frequency density distributions provide not only one estimate per soil depth, but a full frequency distribution, the medians were calculated for each soil depth to assist in the analyses (Table S3 for results from δ^{18}O; Table S4 for results from δ^{2}H). As both stable isotope signatures showed similar results, we here focus on results derived from $\delta^{18}O$ only. In addition, we grouped the uptake depths into shallow (0-20 cm as sum of 0-5, 5-10, and 10-20 cm), middle (20-40 cm as sum of 20-30 and 30-40 cm), and deep (the original 40-60 cm) soil layers (Table 4; Table 5). Overall, both species took up water from the entire soil profile studied (0 to 60 cm soil depth), albeit with different proportions depending on species, time (i.e., BT, ET, and AT) and treatment (i.e., control vs. drought; Table 4; Table 5).

For pea, soil water contributions to total plant water uptake decreased with increasing soil depth in both control and drought subplots before (BT) and at the end of the treatment (ET) for all cropping systems (Fig. 4). The median of PC values (MPC) differed significantly among shallow (0-20 cm), middle (20-40 cm), and deep (40-60 cm) layers, averaging 47%, 33%, and 16%, respectively, for both treatments and all cropping systems (BT; Table 5; Fig. 4a, c). At ET, pea plants subjected to drought significantly shifted their water uptake to even higher contributions from the shallow layer (67%) and less uptake from middle (22%) and deep (8%) soil layers compared to BT (Table 5; Fig. 4d; Table S5). Pea plants in control subplots did not display such a significant shift, but remained with average MPC from shallow, middle, and deep soil layers of 52%, 31%, and 14%, respectively (Table 5; Fig. 4b; Table S5). Cropping systems did not significantly affect MPC before (BT) or at the end of (ET) treatment (also no interactions between cropping systems and drought, Table 5; Fig. 4d).

In contrast to pea, barley plants showed very different water uptake patterns before the treatment (BT), with significantly lower PC from the shallow soil layer compared to the middle and deep layers. For barley, MPC values averaged 19%, 44%, and 35% for shallow, middle, and deep soil layers, respectively, for both treatments and all cropping systems (Fig. 5a, d). However, at the end of the treatment (ET), barley plants significantly increased the contributions from the shallow layer in drought subplots, similar to pea (Table 5; Fig. 5e; Table S5), resulting in MPC values of 38%, 41%, and 18% from shallow, middle, and deep soil layers, respectively. The MPC further shifted after the treatment (AT) to values of 62%, 27%, and 10% from shallow, middle, and deep layers, respectively (Fig. 5f). Also in control subplots, barley plants showed the same significant shift from BT to ET, with MPC values at ET of 35%, 34%, and 29% from shallow, middle, and deep layers, respectively (Table 5; Fig. 5b; Table S5), and from ET to AT with MPC values AT of 59%, 29%, and 12% from shallow, middle, and deep layers, respectively (Table 5; Fig. 5c; Table S5). Similar to pea, barley water uptake patterns were not significantly affected by cropping systems (Table 5).

Overall, MPC values from shallow and deep layers for pea and barley were positively correlated (r = 0.64 and 0.55, respectively; Fig. S3). This means when barley took up more water from the shallow layer, so did pea.

Organic as well as reduced/no tillage cropping systems are discussed as adaptation strategies under climate change conditions to ensure arable crop production. Thus, we analysed plant water uptake depths in drought subplots at the end of treatment (ET) more in detail, although cropping systems showed no significant effects on water uptake depths for either species and no interactions occurred between cropping systems and drought treatment (Table 5). Pea plants in both intensive systems (C-IT and O-IT) showed significantly higher (O-IT: 77%) or similar (C-IT: 65%) contributions to total water uptake (as MPC) from the shallow layer (0-20 cm) compared to conservation tillage systems (64% in both C-NT and O-RT; Table 5; Fig. 4d). Conversely, contributions from the middle layer (20-40 cm) for pea at the end of treatment (ET) were only 15% in O-IT compared to 24% in the other three cropping systems (O-RT, C-IT, and C-NT). Differences among cropping systems under drought were even smaller for barley than for pea (Table 5; Fig. 5e). MPC values of barley for uptake from the shallow layer were 47% (C-IT), 39% (O-RT), 31% (O-IT), and 32% (C-NT). Conversely, contributions from the middle layer were the largest in C-NT (47%), followed by O-IT (44%) and O-RT (41%), and lowest in C-IT (34%). The absolute changes in MPC values between before the treatment (BT) and the end of treatment (ET) were not significantly affected by cropping systems for either species, but significantly affected by the drought treatment for pea (for the shallow and middle soil layers; Table S6).

4 Discussion

Root water uptake patterns are often discussed for their important role in plant water relations, but only few studies considered arable crop species (Penna *et al.*, 2020). In addition, most studies on responses of crop root water uptake patterns to drought took place in pots or under controlled conditions (e.g., Zegada-Lizarazu & Iijima, 2004; Araki & Iijima, 2005), so that information on field conditions is particularly scarce, except maize (Ma & Song, 2016), wheat (Ma & Song, 2018), oilseed rape, and barley in monoculture (Wu *et al.*, 2016). Furthermore, studies comparing the role of different cropping systems for crop water uptake are completely lacking. Here, we showed for the first time that root water uptake patterns of field-grown pea and barley in mixture responded to drought but not to different cropping systems. Subjected to a pronounced drought period (37 d without precipitation), both crop species shifted to relying more on shallow soil layer (0-20 cm) for water uptake. This drought response was independent of the cropping system, i.e., organic vs. conventional farming or intensive vs. conservation tillage.

Previous research on root water uptake patterns in crop as well as grassland species showed ambiguous responses to drought. For some species, root water uptake depth was dependent on root distribution during wet periods, but on soil water availability during dry periods (Sprenger *et al.*, 2016). Therefore, utilising more water from deep than from shallow soil layer is typically the anticipated drought response, such as barley in monoculture (Wu *et al.*, 2018), maize (Ma & Song, 2016), wheat, rice, soybean (Zegada-Lizarazu & Iijima, 2004), or chickpea (Purushothaman *et al.*, 2017). However, other studies reported that crop and grassland species do not take up water from deeper depths under drought but even absorb more water from shallow soil layer (e.g., barley in monoculture, maize, pigeon pea, cowpea; Zegada-Lizarazu & Iijima, 2004), or grassland species (Hoekstra *et al.*, 2014; Prechsl *et al.*, 2015; Wu *et al.*, 2016). This is in accordance with our results in which both pea and barley increased their proportional water uptake from shallow layer (0-20 cm) at the end of treatment (ET) in the drought subplots. Although soil water contents (SWC) were still higher at 40 cm than at 10 cm at the end of the treatment (ET; Fig. 2c, d), SWC at 40 cm and 10 cm depths were both very low. Thus, the whole soil profile showed very low water availability at the end of the treatment (ET), and fine root distributions most likely dominated plant water uptake patterns.

Rooting profiles for legumes with increased proportions of deeper roots under drought, e.g., below 23-30 cm, have been reported (Benjamin & Nielsen, 2006; Purushothaman *et al.*, 2017), although different responses in root growth to drought were found among different varieties (Kashiwagi *et al.*, 2006; Kumar *et al.*, 2012; Purushothaman *et al.*, 2017). The architecture of legume root systems is strongly affected by rhizobia, which typically find better living conditions in terms of oxygen and nitrogen concentrations higher up in the soil profile than at greater depths (Concha & Doerner, 2020), also in dry soils. Moreover, barley grown under drought conditions has been reported to develop proportionally more shallow roots (0-20 cm depth) relative to deeper soil depths (Carvalho *et al.*, 2014). Also, studies on grassland plants (both legume and grass species) found increasing root biomass production in shallow soil depths (0-15 cm) in response to drought (e.g., Prechsl *et al.*, 2015). Although we did not investigate root distributions for either crop species, they most likely followed such evolutionary strategies as well during our rather strong, 37-day drought treatment, in addition to recent crop breeding efforts leading to less deep root systems in general (Canadell *et al.*, 1996; Thorup-Kristensen *et al.*, 2020). Moreover, shifting to shallower water uptake depths during drought might actually be beneficial for nutrient acquisition (Querejeta *et al.*, 2021), since not only concentrations of soil water and atmospheric N_2 are higher in the top soil than in the deeper soil, but also litter inputs for N mineralisation. ~~Although we did not investigate root distributions for either crop species, they most likely follow such evolutionary strategies as well, in addition to recent crop breeding efforts leading to less deep root systems in general (Canadell *et al.*, 1996; Thorup-Kristensen *et al.*, 2020).~~ Thus, besides the low soil moisture within the entire soil profile~~s~~, acclimation of the root systems ~~biology clearly~~ most likely also contributed to the shift towards shallower water uptake depths under drought for both pea and barley in this study.

The year 2018 was characterised by low precipitation during our experimental period, when a naturally dry period occurred at the end of our pronounced drought treatment in June (which excluded 34% of the precipitation during the growing season; Table 1). Our treatment compared well with the climate scenarios available for Switzerland, with a 25% reduction of precipitation in 2060, and up to 40% by the end of the century; and an increase of the longest rain-free summer period (June, July, August) from currently 11 days to 20 days (CH2018). The dry period in June ~~which~~ affected pea and barley plants in our control subplots differently (Fig. 6a, b). While pea did not shift its water uptake pattern (Fig. 6a; Table S5), barley grown in the control subplots reacted very similar to the natural ~~11-d~~ dry period (before the ET sampling, 14 to 25 June; Fig. 2) as barley subjected to our drought treatment, namely with a clear shift from deep (40-60 cm) to shallow (0-20 cm) soil layer (Fig. 6b, d; Table S5). However, barley still relied more on water uptake from the deep soil layer during this naturally dry period in the control subplots than under the experimental drought (*P* = 0.017; Table 5). Hence, these different reactions of the two species to the dry period clearly indicated that barley was more susceptible than pea even to a mild water stress. This observation is fully in line with measurements of stem hydraulic traits (i.e., loss of xylem conductance) from the same experiment (Sun *et al.*, 2021). Barley plants lost xylem conductance much earlier than pea plants when xylem water potentials decreased. In addition, legumes like pea can maintain low stomatal conductance to avoid water stress without compromising photosynthesis when growing under conditions with limiting water supply, due to their high foliar N concentrations (Adams *et al.*, 2018). This adds to the hydraulic trait benefits of pea and explains why pea was less affected by the natural dry period. Nevertheless, as shown in our study, if severities and frequencies of droughts increase in the future, one can expect negative consequences not only on the performance of barley, but also of pea (Martin & Jamieson, 1996).

Moreover, the two species growing together in the pea-barley mixture showed distinct niches for root water uptake before drought, with pea relying more on water from shallow (0-20 cm) and barley from deep (40-60 cm) soil layers, in accordance with resource partitioning in the absence of water limitation as observed in intercrops, e.g., pearl millet and cowpea (Zegada-Lizarazu *et al.*, 2006) and in mixed-species grasslands (e.g., Hoekstra *et al.*, 2014). However, the niches became more similar under drought conditions, contradicting ecological theory which postulates more pronounced niche differentiation and less niche overlap under stressful conditions, such as during a drought (see Nippert & Knapp, 2007; Silvertown *et al.*, 2015; Guderle *et al.*, 2018). However, our results were in line with results from biodiversity studies in temperate grasslands (Bachmann *et al.*, 2015; Barry *et al.*, 2020; Hoekstra *et al.*, 2014) which also did not show niche differentiation in response to increased competition or drought~~_ENREF_5~~. Thus, further detailed knowledge on the dynamics of intercrop water uptake patterns is needed to solve this contradiction and to decrease the uncertainty for arable crop production now and under future climate conditions.

As global agriculture has already been considerably compromised by and become increasingly sensitive to climate change (Ortiz-Bobea *et al.*, 2021), farming practices such as organic management and conservation tillage are being discussed widely. They have been shown to improve general soil conditions compared to conventional management and intensive tillage, particularly under drought (Bot & Benites, 2005; Gomiero *et al.*, 2011; Choudhary *et al.*, 2016). For instance, organic management and conservation tillage can increase soil water holding capacity, therefore providing higher water availability than conventional management and intensive tillage (e.g., Colombi *et al.*, 2019; Kundel *et al.*, 2020). In this study, the systems with conservation tillage (C-NT and O-RT) indeed showed slightly higher SWC than systems with intensive tillage (C-IT and O-IT) at 40 cm (Fig. 2d). However, this did not result in any benefit for root water uptake patterns of pea and barley against drought. Water uptake of both species shifted to the shallow layer (0-20 cm) in all cropping systems under drought, without cropping system effects or interactive effects between cropping systems and drought treatment. Thus, any further changes in soil physical characteristics due to the drought treatment among cropping systems did not affect the observed root water uptake patterns. The relatively short period that annual crop species are growing under these conditions might limit the potential benefits from improved soil conditions present in those systems (e.g., Dennert *et al.*, 2018; Loaiza Puerta *et al.*, 2018; Schluter *et al.*, 2018). Although it remains to be seen if the observed behaviour of a pea-barley mixture also holds true for other crop species, our results clearly challenge the potential of cropping management under temperate climate as a tool to adapt arable agriculture to climate change.

5 Conclusions

Water uptake patterns of pea and barley both shifted under drought in all cropping systems and both species relied more on water from the shallow soil layer (0-20 cm) than on water from deeper in the soil profile. This was also the case for organic and reduced/no tillage cropping systems, which are often discussed as beneficial for crop performance, particular under water-limited conditions, and are thus suggested as adaptive~~ed~~ cropping management practices under a future climate. However, in this study, we showed for the first time that cropping systems could not counteract the effects of severe drought ~~effects~~ on plant water uptake patterns for pea and barley grown in mixture. It remains to be seen if this observation also holds true for other, major crops grown under water-limited conditions.

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Author Contribution

NB, AKG, RW, and MH designed the study; QS analysed the data; QS, AKG, and NB wrote the first drafts of the manuscript; all authors discussed the results, revised, and agreed on the final version of the manuscript.

Conflict of Interest

None declared.

Supporting Information

Additional supporting information can be found in the online version of this article.

Table S1 Stable water isotope values (δ^{18}O and δ^{2}H, ‰) of soil in control and drought subplots under different cropping systems.

Table S2 Effects of cropping systems, drought treatment and the interaction on stable isotope data (δ^{18}O and δ^{2}H, ‰) of pea and barley as well as mean ± 1 SE for each species in control and drought subplots under different cropping systems.

Table S3 Effects of cropping systems, drought treatment and the interaction on the median proportional contributions from different soil depths to water uptake of pea and barley as well as mean ± 1 SE of MPC using δ^{18}O data.

Table S4 Effects of cropping systems, drought treatment and the interaction on the median proportional contributions from different soil depths to water uptake of pea and barley as well as mean ± 1 SE MPC using δ^{2}H data.

Table S5 Effects of cropping systems, sampling times and the interaction on the proportional contributions from different soil depths to water uptake of pea and barley simulated from δ^{18}O data in control and drought subplots.

Table S6 Effects of cropping systems, drought treatment and the interaction on absolute changes in median proportional contributions to plant water uptake of pea and barley.

Fig. S1 Dual isotope plot of soil and plant samples from control and drought subplots.

Fig. S2 δ^2H values of soil water from different depths and plant xylem water in each cropping system in 2018.

Fig. S3 Relationships of median proportional contributions to plant water uptake from the shallow and deep soil layers of pea vs. barley.

Data Availability Statement

The data that support the findings of this study will be openly available in the ETH Zurich Repository at https://www.research-collection.ethz.ch/ (DOI: 10.5905/ethz-1002-11290).

Reference

Adams MA, Buchmann N, Sprent J, Buckley TN, Turnbull TL. 2018. Crops, nitrogen, water: Are legumes friend, foe, or misunderstood ally? *Trends in Plant Science* **23**(6): 539-550.

Araki H, Iijima M. 2005. Stable isotope analysis of water extraction from subsoil in upland rice (*Oryza sativa* L.) as affected by drought and soil compaction. *Plant and Soil* **270**(1-2): 147-157.

Bachmann D, Gockele A, Ravenek JM, Roscher C, Strecker T, Weigelt A, Buchmann N. 2015. No evidence of complementary water use along a plant species richness gradient in temperate experimental grasslands. *Plos One* **10**(1): e0116367.

Barnard RL, de Bello F, Gilgen AK, Buchmann N. 2006. The δ^{18}O of root crown water best reflects source water δ^{18}O in different types of herbaceous species. *Rapid Communications in Mass Spectrometry* **20**(24): 3799-3802.

Barry KE, van Ruijven J, Mommer L, Bai YF, Beierkuhnlein C, Buchmann N, de Kroon H, Ebeling A, Eisenhauer N, Guimaraes-Steinicke C, et al. 2020. Limited evidence for spatial resource partitioning across temperate grassland biodiversity experiments. *Ecology* **101**(1): e02905.

Benjamin JG, Nielsen DC. 2006. Water deficit effects on root distribution of soybean, field pea and chickpea. *Field Crops Research* **97**(2-3): 248-253.

Berry ZC, Emery NC, Gotsch SG, Goldsmith GR. 2019. Foliar water uptake: Processes, pathways, and integration into plant water budgets. *Plant Cell and Environment* **42**(2): 410-423.

Borrell AK, van Oosterom EJ, Mullet JE, George-Jaeggli B, Jordan DR, Klein PE, Hammer GL. 2014. Stay-green alleles individually enhance grain yield in sorghum under drought by modifying canopy development and water uptake patterns. *New Phytologist* **203**(3): 817-830.

Bot A, Benites J. 2005. *The importance of soil organic matter: Key to drought-resistant soil and sustained food production*. Food & Agriculture Organization: 35-50.

Boyer JS, Rao IM. 1984. Magnesium and the acclimation of photosynthesis to low leaf water potentials. *Plant Physiology* **74**(1): 161-166.

Caldwell MM, Dawson TE, Richards JH. 1998. Hydraulic lift: Consequences of water efflux from the roots of plants. *Oecologia* **113**(2): 151-161.

Canadell J, Jackson RB, Ehleringer JR, Mooney HA, Sala OE, Schulze ED. 1996. Maximum rooting depth of vegetation types at the global scale. *Oecologia* **108**(4): 583-595.

Carvalho P, Azam-Ali S, Foulkes MJ. 2014. Quantifying relationships between rooting traits and water uptake under drought in Mediterranean barley and durum wheat. *Journal of Integrative Plant Biology* **56**(5): 455-469.

CH2018. 2018. *CH2018 – Climate Scenarios for Switzerland, Technical Report*. National Centre for Climate Services, Zurich: 271.

Choudhary M, Ghasal PC, Kumar S, Yadav R, Singh S, Meena VS, Bisht JK 2016. Conservation agriculture and climate change: an overview. *Conservation Agriculture*: Springer, 1-37.

Cochard H. 2002. Xylem embolism and drought-induced stomatal closure in maize. *Planta* **215**(3): 466-471.

Colombi T, Walder F, Buchi L, Sommer M, Liu KX, Six J, van der Heijden MGA, Charles R, Keller T. 2019. On-farm study reveals positive relationship between gas transport capacity and organic carbon content in arable soil. *Soil* **5**(1): 91-105.

Concha C, Doerner P. 2020. The impact of the rhizobia-legume symbiosis on host root system architecture. *Journal of Experimental Botany* **71**(13): 3902-3921.

Craig H. 1961. Isotopic variations in meteoric waters. *Science* **133**(3465): 1702-1703.

Craig H, Gordon LI 1965. Deuterium and oxygen 18 variation in the ocean and the marine atmosphere. In: Tongiorgi E ed. *Stable Isotopes in Oceanographic Studies and Paleotemperatures*. Pisa, Italy: Consiglio Nazionale delle Ricerche, Laboratorio di Geologia Nucleare, 9-130.

Dansgaard W. 1964. Stable isotopes in precipitation. *Tellus* **16**(4): 436-468.

Dawson TE, Ehleringer JR. 1991. Streamside trees that do not use stream water. *Nature* **350**(6316): 335-337.

Dennert F, Imperiali N, Staub C, Schneider J, Laessle T, Zhang T, Wittwer R, van der Heijden MG, Smits THM, Schlaeppi K, et al. 2018. Conservation tillage and organic farming induce minor variations in

Pseudomonas abundance, their antimicrobial function and soil disease resistance. *FEMS Microbiology Ecology* **94**(8): fiy075.

Dietrich D, Pang L, Kobayashi A, Fozard JA, Boudolf V, Bhosale R, Antoni R, Nguyen T, Hiratsuka S, Fujii N, et al. 2017. Root hydrotropism is controlled via a cortex-specific growth mechanism. *Nature Plants* **3**(6): 17057.

Ding YL, Nie YP, Chen HS, Wang KL, Querejeta JI. 2021. Water uptake depth is coordinated with leaf water potential, water-use efficiency and drought vulnerability in karst vegetation. *New Phytologist* **229**(3): 1339-1353.

Dixon PM. 2016. Should blocks be fixed or random? *Statistics Conference Proceedings, Presentations and Posters* **6**.

Ehleringer JR, Osmond CB. 1989. *Stable isotopes*. Heidelberg, Germany: Chapman & Hall: 281-300.

FAO. 2018. The impact of disasters and crises on agriculture and food security. Rome: Food and Agriculture Organization of the United Nations.

FAO. 2019. Proactive approaches to drought preparedness: Where are we now and where do we go from here? Rome: Food and Agriculture Organization of the United Nations.

Gat JR 2010. The isotopes of hydrogen and oxygen. In: Kwang wei T ed. *Isotope Hydrology: A Study of the Water Cycle*. London, UK: Imperial College Press, 9-21.

Gehre M, Geilmann H, Richter J, Werner RA, Brand WA. 2004. Continuous flow ^{2}H/^{1}H and and ^{18}O/^{16}O analysis of water samples with dual inlet precision. *Rapid Communications in Mass Spectrometry* **18**(22): 2650-2660.

Gilliland TJ, Johnston J. 1992. Barley pea mixtures as cover crops for grass re-seeds. *Grass and Forage Science* **47**(1): 1-7.

Gomiero T, Pimentel D, Paoletti MG. 2011. Environmental impact of different agricultural management practices: Conventional vs. organic agriculture. *Critical Reviews in Plant Sciences* **30**(1-2): 95-124.

Gonfiantini R, Gratziu S, Tongiorgi E. 1965. Oxygen isotopic composition of water in leaves. *Isotopes and Radiation in Soil-Plant Nutrition Studies* **405**: 410.

Grossiord C, Sevanto S, Bonal D, Borrego I, Dawson TE, Ryan M, Wang WZ, McDowell NG. 2019. Prolonged warming and drought modify belowground interactions for water among coexisting plants. *Tree Physiology* **39**(1): 55-63.

Guderle M, Bachmann D, Milcu A, Gockele A, Bechmann M, Fischer C, Roscher C, Landais D, Ravel O, Devidal S, et al. 2018. Dynamic niche partitioning in root water uptake facilitates efficient water use in more diverse grassland plant communities. *Functional Ecology* **32**(1): 214-227.

Hobbs PR, Sayre K, Gupta R. 2008. The role of conservation agriculture in sustainable agriculture. *Philosophical Transactions of the Royal Society B-Biological Sciences* **363**(1491): 543-555.

Hoekstra NJ, Finn JA, Hofer D, Lüscher A. 2014. The effect of drought and interspecific interactions on depth of water uptake in deep- and shallow-rooting grassland species as determined by δ^{18}O natural abundance. *Biogeosciences* **11**(16): 4493-4506.

Hofer D, Suter M, Haughey E, Finn JA, Hoekstra NJ, Buchmann N, Luscher A. 2016. Yield of temperate forage grassland species is either largely resistant or resilient to experimental summer drought. *Journal of Applied Ecology* **53**(4): 1023-1034.

Hothorn T, Bretz F, Westfall P. 2008. Simultaneous inference in general parametric models. *Biometrical Journal* **50**(3): 346-363.

IAEA. 2020. *Global network of isotopes in precipitation*. Vienna, Austria

IPCC. 2019. *Climate change and land. An IPCC special report on climate change, desertification, land degradation, sustainable land management, food security, and greenhouse gas fluxes in terrestrial ecosystems*. In press.

Kanton Zürich. 2021. Soil map of the agricultural areas. Kanton Zürich, Accessed on 25 November 2020, <http://maps.zh.ch/?topic=BoKaZH>

Kashiwagi J, Krishnamurthy L, Crouch JH, Serraj R. 2006. Variability of root length density and its contributions to seed yield in chickpea (*Cicer arietinum* L.) under terminal drought stress. *Field Crops Research* **95**(2-3): 171-181.

Kumar J, Basu PS, Srivastava E, Chaturvedi SK, Nadarajan N, Kumar S. 2012. Phenotyping of traits imparting drought tolerance in lentil. *Crop & Pasture Science* **63**(6): 547-554.

Kundel D, Bodenhausen N, Jorgensen HB, Truu J, Birkhofer K, Hedlund K, Mader P, Fliessbach A. 2020. Effects of simulated drought on biological soil quality, microbial diversity and yields under long-term conventional and organic agriculture. *FEMS Microbiology Ecology* **96**(12): fiaa205.

Kuznetsova A, Brockhoff PB, Christensen RHB. 2017. lmerTest package: Tests in linear mixed effects models. *Journal of Statistical Software* **82**(13): 26.

Loaiza Puerta V, Pereira EIP, Wittwer R, Heijden MVD, Six J. 2018. Improvement of soil structure through organic crop management, conservation tillage and grass-clover ley. *Soil & Tillage Research* **180**: 1-9.

Ma Y, Song XF. 2016. Using stable isotopes to determine seasonal variations in water uptake of summer maize under different fertilization treatments. *Science of the Total Environment* **550**: 471-483.

Ma Y, Song XF. 2018. Seasonal variations in water uptake patterns of winter wheat under different irrigation and fertilization treatments. *Water* **10**(11): 1633.

Martin RJ, Jamieson PD. 1996. Effect of timing and intensity of drought on the growth and yield of field peas (*Pisum sativum* L). *New Zealand Journal of Crop and Horticultural Science* **24**(2): 167-174.

MeteoSwiss. 2020. Swiss Federal Office for Meteorology and Climatology, Accessed on 15 March 2020, <https://www.meteoswiss.admin.ch/home.html?tab=overview>

Nippert JB, Knapp AK. 2007. Linking water uptake with rooting patterns in grassland species. *Oecologia* **153**(2): 261-272.

Ortiz-Bobea A, Ault TR, Carrillo CM, Chambers RG, Lobell DB. 2021. Anthropogenic climate change has slowed global agricultural productivity growth. *Nature Climate Change* **11**(4): 306-312.

Parnell AC. 2020. simmr: A stable isotope mixing model. <https://CRAN.R-project.org/package=simmr>

Parnell AC, Phillips DL, Bearhop S, Semmens BX, Ward EJ, Moore JW, Jackson AL, Grey J, Kelly DJ, Inger R. 2013. Bayesian stable isotope mixing models. *Environmetrics* **24**(6): 387-399.

Penna D, Geris J, Hopp L, Scandellari F. 2020. Water sources for root water uptake: Using stable isotopes of hydrogen and oxygen as a research tool in agricultural and agroforestry systems. *Agriculture Ecosystems & Environment* **291**: ARTN 106790.

Penna D, Hopp L, Scandellari F, Allen ST, Benettin P, Beyer M, Geris J, Klaus J, Marshall JD, Schwendenmann L, et al. 2018. Ideas and perspectives: Tracing terrestrial ecosystem water fluxes using hydrogen and oxygen stable isotopes – challenges and opportunities from an interdisciplinary perspective. *Biogeosciences* **15**(21): 6399-6415.

Prechsl UE, Burri S, Gilgen AK, Kahmen A, Buchmann N. 2015. No shift to a deeper water uptake depth in response to summer drought of two lowland and sub-alpine C_3-grasslands in Switzerland. *Oecologia* **177**(1): 97-111.

Prechsl UE, Gilgen AK, Kahmen A, Buchmann N. 2014. Reliability and quality of water isotope data collected with a lowbudget rain collector. *Rapid Communications in Mass Spectrometry* **28**(8): 879-885.

Purushothaman R, Krishnamurthy L, Upadhyaya HD, Vadez V, Varshney RK. 2017. Genotypic variation in soil water use and root distribution and their implications for drought tolerance in chickpea. *Functional Plant Biology* **44**(2): 235-252.

Querejeta JI, Ren W, Prieto I. 2021. Vertical decoupling of soil nutrients and water under climate warming reduces plant cumulative nutrient uptake, water-use efficiency and productivity. *New Phytologist*, doi: https://doi.org/10.1111/nph.17258.

R Core Team. 2020. R: A language and environment for statistical computing.

Rasmussen CR, Thorup-Kristensen K, Dresboll DB. 2020. Uptake of subsoil water below 2 m fails to alleviate drought response in deep-rooted Chicory (*Cichorium intybus* L.). *Plant and Soil* **446**(1-2): 275-290.

Rothfuss Y, Javaux M. 2017. Reviews and syntheses: Isotopic approaches to quantify root water uptake: A review and comparison of methods. *Biogeosciences* **14**(8): 2199-2224.

Schewe J, Heinke J, Gerten D, Haddeland I, Arnell NW, Clark DB, Dankers R, Eisner S, Fekete BM, Colon-Gonzalez FJ, et al. 2014. Multimodel assessment of water scarcity under climate change. *Proceedings of the National Academy of Sciences of the United States of America* **111**(9): 3245-3250.

Schluter S, Grossmann C, Diel J, Wu GM, Tischer S, Deubel A, Rucknagel J. 2018. Long-term effects of conventional and reduced tillage on soil structure, soil ecological and soil hydraulic properties. *Geoderma* **332**: 10-19.

Seitz S, Goebes P, Puerta VL, Pereira EIP, Wittwer R, Six J, van der Heijden MGA, Scholten T. 2019. Conservation tillage and organic farming reduce soil erosion. *Agronomy for Sustainable Development* **39**(1): 4.

Silvertown J, Araya Y, Gowing D. 2015. Hydrological niches in terrestrial plant communities: A review. *Journal of Ecology* **103**(1): 93-108.

Sprenger M, Leistert H, Gimbel K, Weiler M. 2016. Illuminating hydrological processes at the soil-vegetation-atmosphere interface with water stable isotopes. *Reviews of Geophysics* **54**(3): 674-704.

Sun Q, Gilgen AK, Signarbieux C, Klaus VH, Buchmann N. 2021. Cropping systems alter hydraulic traits of barley but not pea grown in mixture. *Plant, Cell & Environment*, doi: 10.1111/pce.14054.

Swiss Federal Council. 2021. *Verordnung über die Direktzahlungen an die Landwirtschaft (Direktzahlungsverordnung, DZV) vom 23. Oktober 2013 (Stand am 1 Januar 2021) (Swiss council regulation no. 910.13: Ordinance on Direct Payments of 23 October 2013 (as of 1 January 2021)*.

Teasdale JR, Coffman CB, Mangum RW. 2007. Potential long-term benefits of no-tillage and organic cropping systems for grain production and soil improvement. *Agronomy Journal* **99**(5): 1297-1305.

Thorup-Kristensen K, Halberg N, Nicolaisen M, Olesen JE, Crews TE, Hinsinger P, Kirkegaard J, Pierret A, Dresboll DB. 2020. Digging deeper for agricultural resources, the value of deep rooting. *Trends in Plant Science* **25**(4): 406-417.

von Freyberg J, Allen ST, Grossiord C, Dawson TE, Royles J. 2020. Plant and root-zone water isotopes are difficult to measure, explain, and predict: Some practical recommendations for determining plant water sources. *Methods in Ecology and Evolution* **11**(11): 1352-1367.

Wahdan SFM, Reitz T, Heintz-Buschart A, Schadler M, Roscher C, Breitkreuz C, Schnabel B, Purahong W, Buscot F. 2021. Organic agricultural practice enhances arbuscular mycorrhizal symbiosis in correspondence to soil warming and altered precipitation patterns. *Environmental Microbiology* **23**(10): 6163-6176.

Wang LX, Caylor KK, Villegas JC, Barron-Gafford GA, Breshears DD, Huxman TE. 2010. Partitioning evapotranspiration across gradients of woody plant cover: Assessment of a stable isotope technique. *Geophysical Research Letters* **37**(9): 232-256.

Wang LX, Manzoni S, Ravi S, Riveros-Iregui D, Caylor K. 2015. Dynamic interactions of ecohydrological and biogeochemical processes in water-limited systems. *Ecosphere* **6**(8): 133.

Werner RA, Bruch BA, Brand WA. 1999. ConFlo III - An interface for high precision δ^{13}C and δ^{15}N analysis with an extended dynamic range. *Rapid Communications in Mass Spectrometry* **13**(13): 1237-1241.

Whitmore AP, Whalley WR. 2009. Physical effects of soil drying on roots and crop growth. *Journal of Experimental Botany* **60**(10): 2845-2857.

Wittwer RA, Dorn B, Jossi W, van der Heijden MGA. 2017. Cover crops support ecological intensification of arable cropping systems. *Scientific Reports* **7**: ARTN 41911.

Wittwer RA, Bender SF, Hartman K, Hydbom S, Lima RAA, Loaiza V, Nemecek T, Oehl F, Olsson PA, Petchey O, et al. 2021. Organic and conservation agriculture promote ecosystem multifunctionality. *Science Advances* 7(34): eabg6995.

Wu HW, Li J, Zhang CC, He B, Zhang HX, Wu XC, Li XY. 2018. Determining root water uptake of two alpine crops in a rainfed cropland in the Qinghai Lake watershed: first assessment using stable isotopes analysis. *Field Crops Research* **215**: 113-121.

Wu HW, Li XY, Li J, Jiang ZY, Chen HY, Ma YJ, Huang YM. 2016. Differential soil moisture pulse uptake by coexisting plants in an alpine *Achnatherum splendens* grassland community. *Environmental Earth Sciences* **75**(10).

Zegada-Lizarazu W, Iijima M. 2004. Hydrogen stable isotope analysis of water acquisition ability of deep roots and hydraulic lift in sixteen food crop species. *Plant Production Science* **7**(4): 427-434.

Zegada-Lizarazu W, Kanyomeka L, Izumi Y, Iijima M. 2006. Pearl millet developed deep roots and changed water sources by competition with intercropped cowpea in the semiarid environment of northern Namibia. *Plant Production Science* **9**(4): 355-363.

Zimmermann U, Ehhalt D, Münnich KO 1967. Soil-water movement and evapotranspiration: Changes in the isotopic composition of the water. *Isotopes in hydrology. Proceedings of a symposium*.

Table 1 Precipitation and air temperature data from a nearby weather station, Zürich/Kloten (KLO, 47.48° N, 8.54° E, 4.6 km north of the research site, MeteoSwiss, 2020) as well as dates for the growing season (from sowing to harvest) and treatment periods in 2018.

	Date	Total precipitation (mm)	Mean air temperature (°C)
Long-term annual (1988-2017)	1 January to 31 December	994	9.7
Annual (2018)	1 January to 31 December	856	11.2
Long-term May (1988-2017)	1 to 31 May	105	13.9
May 2018	1 to 31 May	102	15.8
Long-term June (1988-2017)	1 to 30 June	102	17.2
June 2018	1 to 30 June	40	18.8
Growing season 2018	26 March to 12 July	231	15.7
Before drought treatment	26 March to 21 May	108	12.7
~~End of~~ During drought treatment	22 May to 28 June	79 (34% of the growing season)	18.7
After drought treatment	29 June to 12 July	44	20.0

Table 2 Effects of cropping systems (CS, df = 3), drought treatment (D, df = 1) and the interaction (CS × D, df = 3) on stable water isotopes (δ^{18}O and δ^{2}H) in different soil depths before the drought treatment on 7 May, at the end of treatment on 25 June, and after the treatment on 11 July (in 2018 tested by linear mixed models (*P* values are given).

Isotope	Depth (cm)	CS	D	CS × D	Blocks
Before drought treatment					
δ^{18}O	0-5	0.580	0.555	0.458	0.788
	5-10	0.119	0.276	0.073	0.367
	10-20	0.489	0.836	0.516	0.459
	20-30	0.201	0.164	0.128	0.069
	30-40	0.135	0.437	0.882	0.311
	40-60	0.960	0.898	0.845	0.404
δ^{2}H	0-5	0.831	0.120	0.423	0.982
	5-10	0.158	0.118	0.056	0.516
	10-20	0.467	0.416	0.574	0.571
	20-30	0.105	**0.026**	0.064	0.181
	30-40	0.089	0.125	0.959	0.308
	40-60	0.560	0.291	0.853	0.436
End of drought treatment					
δ^{18}O	0-5	0.316	0.835	0.253	0.367
	5-10	0.189	0.247	0.766	0.168
	10-20	0.080	0.603	0.920	0.673
	20-30	0.898	**<0.001**	0.852	0.940
	30-40	0.437	**<0.001**	0.651	0.954
	40-60	0.073	**0.008**	0.616	0.594
δ^{2}H	0-5	0.295	**<0.001**	0.168	0.479
	5-10	0.330	**0.005**	0.859	0.215
	10-20	0.091	**0.029**	0.700	0.659
	20-30	0.889	**<0.001**	0.863	0.820
	30-40	0.388	**<0.001**	0.551	0.970
	40-60	0.136	**0.006**	0.469	0.809
After drought treatment					
δ^{18}O	0-5	0.393	0.059	0.848	0.291
	5-10	0.730	0.672	0.111	**0.031**
	10-20	0.538	0.612	0.734	0.993
	20-30	0.933	0.136	0.936	0.944
	30-40	0.881	**0.048**	0.979	0.772
	40-60	0.751	**0.001**	0.560	0.380
δ^{2}H	0-5	0.776	0.056	0.667	0.421
	5-10	0.117	0.958	0.649	0.636
	10-20	0.228	0.887	0.926	0.815
	20-30	0.710	0.104	0.888	0.705
	30-40	0.877	0.050	0.919	0.699
	40-60	0.841	**<0.001**	0.493	0.484

Significant differences are shown in bold ($P < 0.05$).

Table 3 Effects of species (df = 1), cropping systems (CS, df = 3), drought treatment (D, df = 1) and the interaction (species × CS, df = 3; species × D, df = 1; CS × D, df = 3; species x CS × D, df = 3) on stable water isotopes (δ^2H and δ^{18}O) of pea and barley before the drought treatment on 7 May and at the end of treatment on 25 June in 2018 tested by linear mixed models (*P* values are given).

Factor	Before drought treatment		End of drought treatment	
	δ^{18}O	δ^2H	δ^{18}O	δ^2H
Species	**<0.001**	**<0.001**	**<0.001**	**<0.001**
CS	0.251	0.382	**0.038**	0.055
D	0.106	**<0.001**	0.143	**0.001**
Species × CS	0.184	**0.023**	0.312	0.348
Species × D	0.796	0.486	**0.004**	**0.016**
CS × D	0.190	0.117	0.051	0.081
Species × CS × D	0.290	**0.045**	0.120	0.070
Blocks	0.485	0.599	**0.004**	0.162

Significant differences are shown in bold ($P < 0.05$).

Table 4 Effects of species (df = 1), cropping systems (CS, df = 3), drought treatment (D, df = 1) and the interaction (species × CS, df = 3; species × D, df = 1; CS × D, df = 3; species × CS × D, df = 3) on the median proportional contributions from different soil depths to water uptake (MPC) of pea and barley before the drought treatment on 7 May and the end of treatment on 25 June in 2018 tested by linear mixed models (*P* values are given).

Factor	Before drought treatment			End of drought treatment		
	0-20 cm	20-40 cm	40-60 cm	0-20 cm	20-40 cm	40-60 cm
Species	**<0.001**	**0.036**	**<0.001**	**<0.001**	**<0.001**	**<0.001**
CS	0.506	0.555	0.992	0.374	0.440	0.252
D	0.849	0.775	0.629	**0.003**	0.546	**0.004**
Species × CS	0.255	0.865	0.702	0.303	0.799	0.180
Species × D	0.424	0.619	0.336	**0.009**	**0.001**	0.359
CS × D	0.454	0.293	0.098	0.278	0.811	0.141
Species × CS × D	0.404	0.064	0.079	0.201	0.315	0.495
Blocks	0.360	0.667	0.534	**0.008**	0.115	**0.016**

MPC was derived from 10 000 simulations by mixing models using $\delta^{18}O$ data. Proportional contribution from 0-20 cm is the sum from 0-5, 5-10, and 10-20 cm, and 20-40 cm is the sum from 20-30 and 30-40 cm. Significant differences are shown in bold ($P < 0.05$).

Table 5 Median proportional contributions (MPC) from different soil depths to water uptake of pea and barley before the drought treatment on 7 May, at the end of treatment on 25 June, and after the drought treatment on 11 July in 2018 (left) as well as ~~E~~effects of cropping systems (CS, df = 3), drought treatment (D, df = 1) and the interaction (CS × D, df = 3) on MPC ~~as well as the median proportional contributions from different soil depths to water uptake (MPC) of pea and barley before the drought treatment on 7 May, at the end of treatment on 25 June, and after the drought treatment on 11 July in 2018~~ tested by linear mixed models. Means ± 1 SE (MPC) and *P* values are given.

Species	Depth (cm)	MPC~~Mean ± 1 SE~~								*P* value from linear mixed models			
		Control				Drought				CS	D	CS × D	Blocks
		C-IT	C-NT	O-IT	O-RT	C-IT	C-NT	O-IT	O-RT				
Before drought treatment													
	0-20	45±8	46±9	50±6	48±5	47±9AB	54±7B	34±9A	50±4AB	0.823	0.818	0.313	0.780
Pea	20-40	37±6	29±3	32±4	36±3	35±7	27±3	36±8	33±3	0.557	0.834	0.913	0.656
	40-60	16±3	20±8	12±2	13±2	14±3	14±4	26±11	13±1	0.746	0.665	0.216	0.545
	0-20	10±3	26±12	17±9	14±5	25±11AB	30±11B	11±7A	14±6AB	0.302	0.475	0.535	0.058
Barley	20-40	41±16ab	39±9ab	65±16b	22±11a	55±15AB	29±10A	37±21AB	63±13B	0.736	0.707	0.156	0.785
	40-60	49±19ab	31±12ab	15±7a	63±17b	18±6	38±19	49±24	20±8	0.940	0.467	0.100	0.634
End of drought treatment													
	0-20	63±6b	46±13a	48±9ab	51±4ab	65±4AB	64±14A	77±12B	64±7A	0.416	**0.001**	0.170	**0.01**
Pea	20-40	27±5a	36±9b	31±3ab	31±1ab	23±2AB	24±9B	15±8A	24±5B	0.416	**0.003**	0.703	**0.021**
	40-60	8±1a	16±4ab	18±6b	14±5ab	9±1AB	10±4B	6±3A	8±1AB	0.398	**0.008**	0.272	**0.027**
	0-20	43±2	38±11	28±6	30±8	47±7B	32±5A	31±9AB	39±7AB	0.214	0.459	0.488	**0.034**
Barley	20-40	39±3	36±4	32±8	29±6	34±4A	47±4B	44±5AB	41±4AB	0.669	0.065	0.339	0.963
	40-60	15±1a	23±9ab	40±13b	38±13b	15±2	19±3	24±8	17±3	0.207	**0.017**	0.213	**0.028**
After drought treatment													
	0-20	61±9	62±8	56±8	56±8	64±13	55±13	71±8	57±5	0.696	0.546	0.436	**0.001**
Barley	20-40	28±6	25±5	30±5	31±5	25±9	31±9	20±6	31±4	0.664	0.604	0.508	**0.004**
	40-60	10±2	11±3	13±4	12±3	11±4	13±4	7±2	10±1	0.852	0.401	0.225	**<0.001**

MPC was derived from 10 000 simulations by mixing models using δ^{18}O data. Pea plants were already senesced in early July therefore no stable water isotope data are available after the treatment. Proportional contribution from 0-20 cm is the sum from 0-5, 5-10, and 10-20 cm, and 20-40 cm is the sum from 20-30 and 30-40 cm. Mean ± 1 SE for MPC (%) are given for different cropping systems (C-IT for Conventional intensive tillage, C-NT for Conventional no tillage, O-IT for Organic intensive tillage, and O-RT for Organic reduced tillage). Different small and capital letters indicate significant differences among cropping systems in control and drought subplots, respectively, tested with Tukey HSD (honestly significant difference, $P < 0.05$). Significant effects tested with linear mixed models are shown in bold ($P < 0.05$).

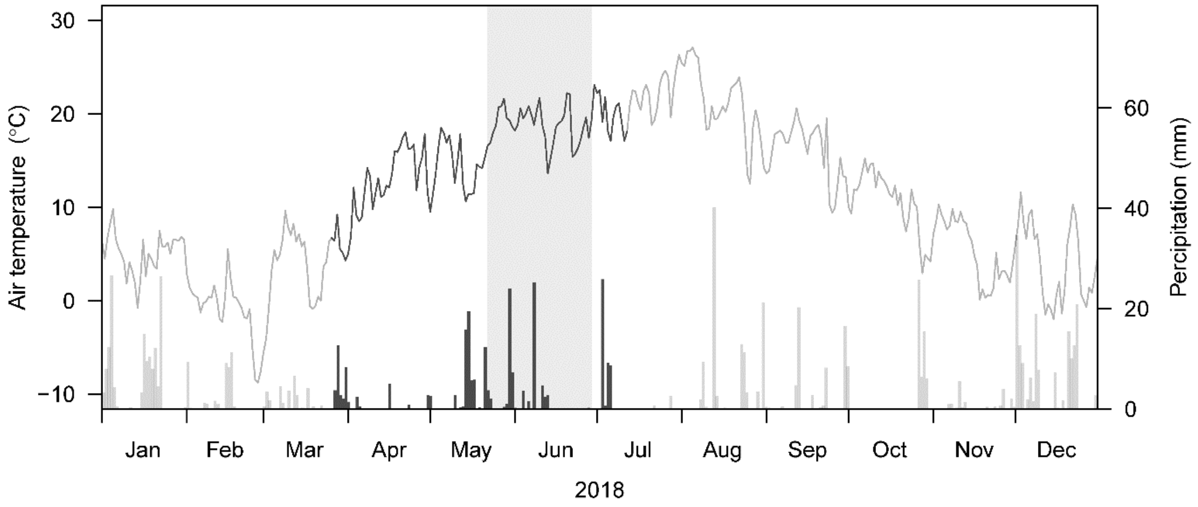

Fig. 1 Daily air temperature and precipitation in 2018. Dark line segments and bars depict the crop growing season from 26 March to 12 July 2018. The shaded area indicates the drought treatment from 22 May to 28 June 2018. Data from the MeteoSwiss station Zürich/Kloten (KLO, 47.48° N, 8.54° E, 4.6 km north of the research site, MeteoSwiss, 2020) are given.

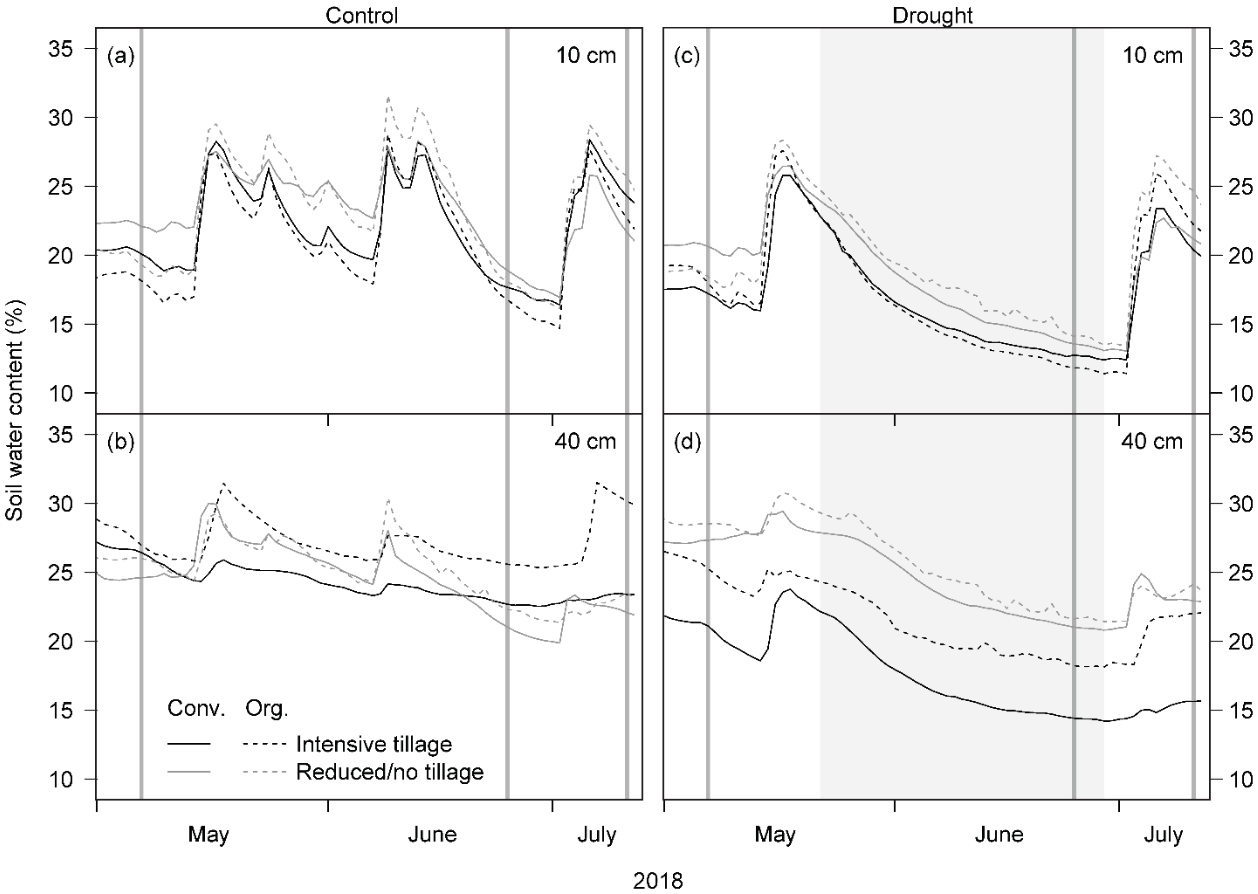

Fig. 2 Daily mean soil water contents at 10 and 40 cm depth in (a, b) control and (c, d) drought subplots under different cropping systems (n = 2 each; Conv. for conventional, Org. for organic). Vertical lines indicate sampling dates for stable water isotopes on 7 May, 25 June, and 11 July 2018. Shaded areas in (c) and (d) represent the drought treatment period from 22 May to 28 June 2018.

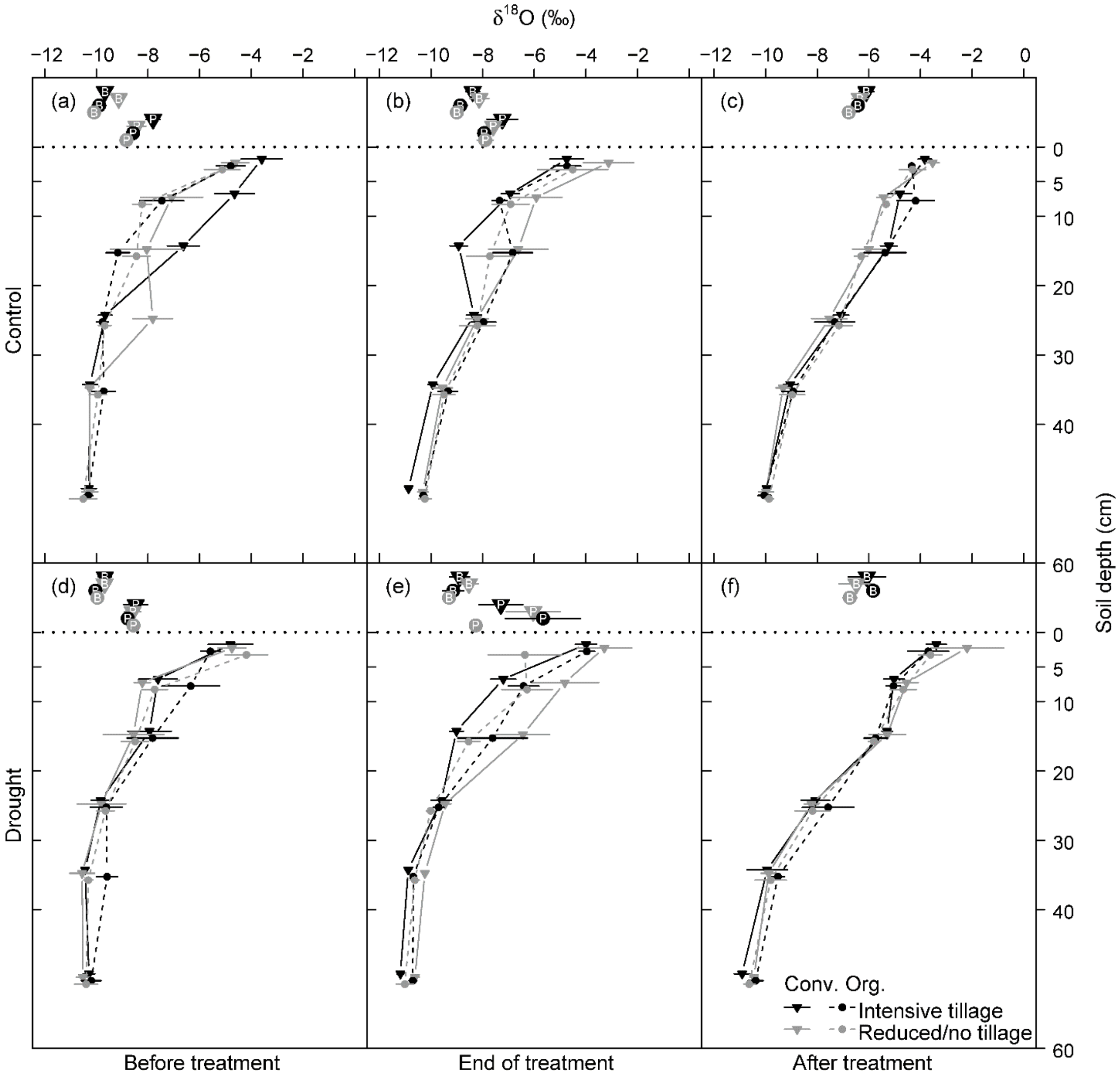

Fig. 3 δ^{18}O values of soil water from different depths and plant xylem water in each cropping system (a, d) before the drought treatment on 7 May, (b, e) at the end of the drought treatment on 25 June, and (c, f) after treatment on 11 July in 2018 (Conv. for conventional, Org. for organic). Horizontal dotted lines separate isotopic composition of soil and plant samples (P for pea, B for barley). Pea plants were already senesced in early July, therefore no stable water isotope data are available after the drought treatment. Means and 1 SE (horizontal bars) are given for each cropping system (n = 3-4).

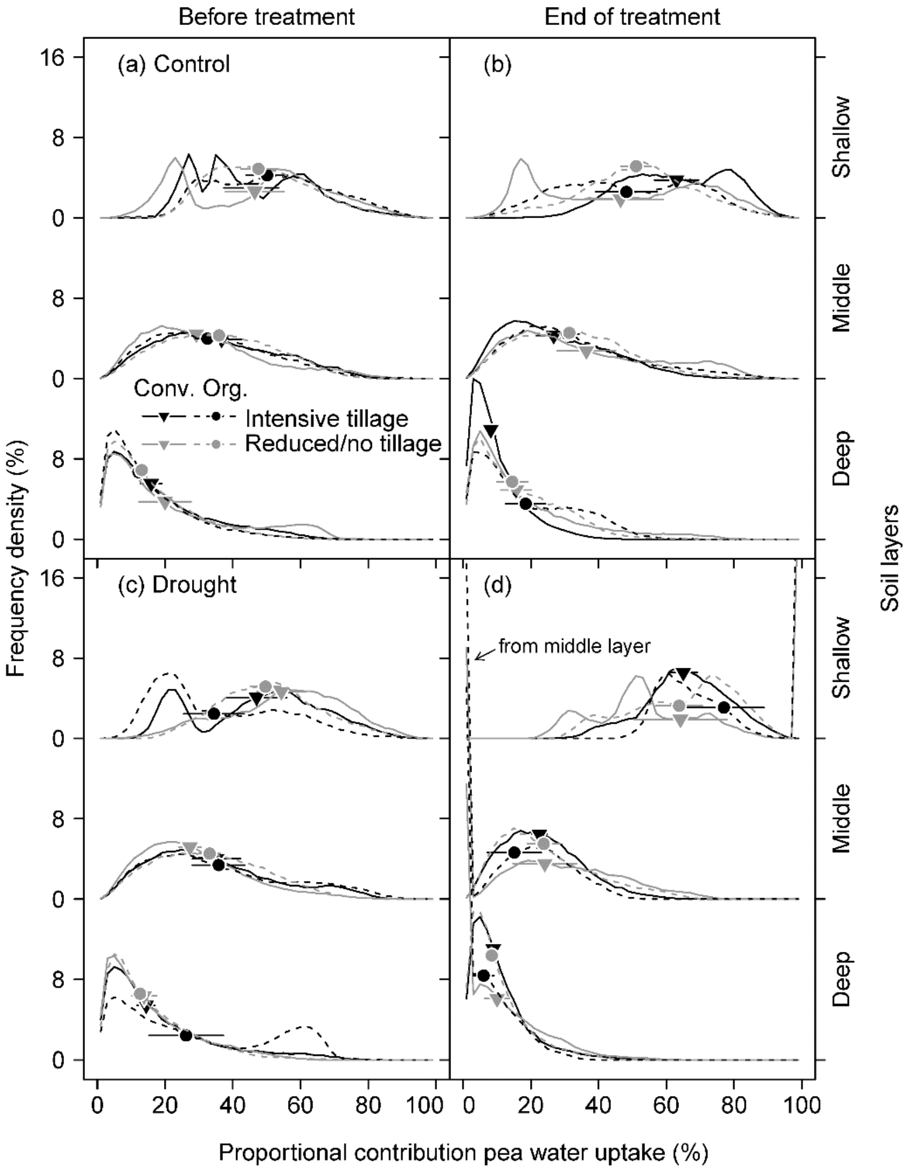

Fig. 4 Frequency density distribution of model outputs on the proportional contribution of soil water to pea water uptake from shallow (0-20 cm, sum of 0-5, 5-10, and 10-20 cm), middle (20-40 cm, sum of 20-30 and 30-40 cm), and deep (40-60 cm) soil layers under different cropping systems (a, b) before the drought treatment on 7 May and (c, d) at the end of treatment on 25 June in 2018. Frequency density was derived from 10 000 simulations at 2% increment of mixing models using $\delta^{18}O$ for each subplot (Conv. for conventional, Org. for organic). Data were pooled for all subplots in each cropping system. Symbols on the curves indicate the median of the model outputs for each soil layer. Means and 1 SE (horizontal bars) of each cropping system are given (n = 3-4).

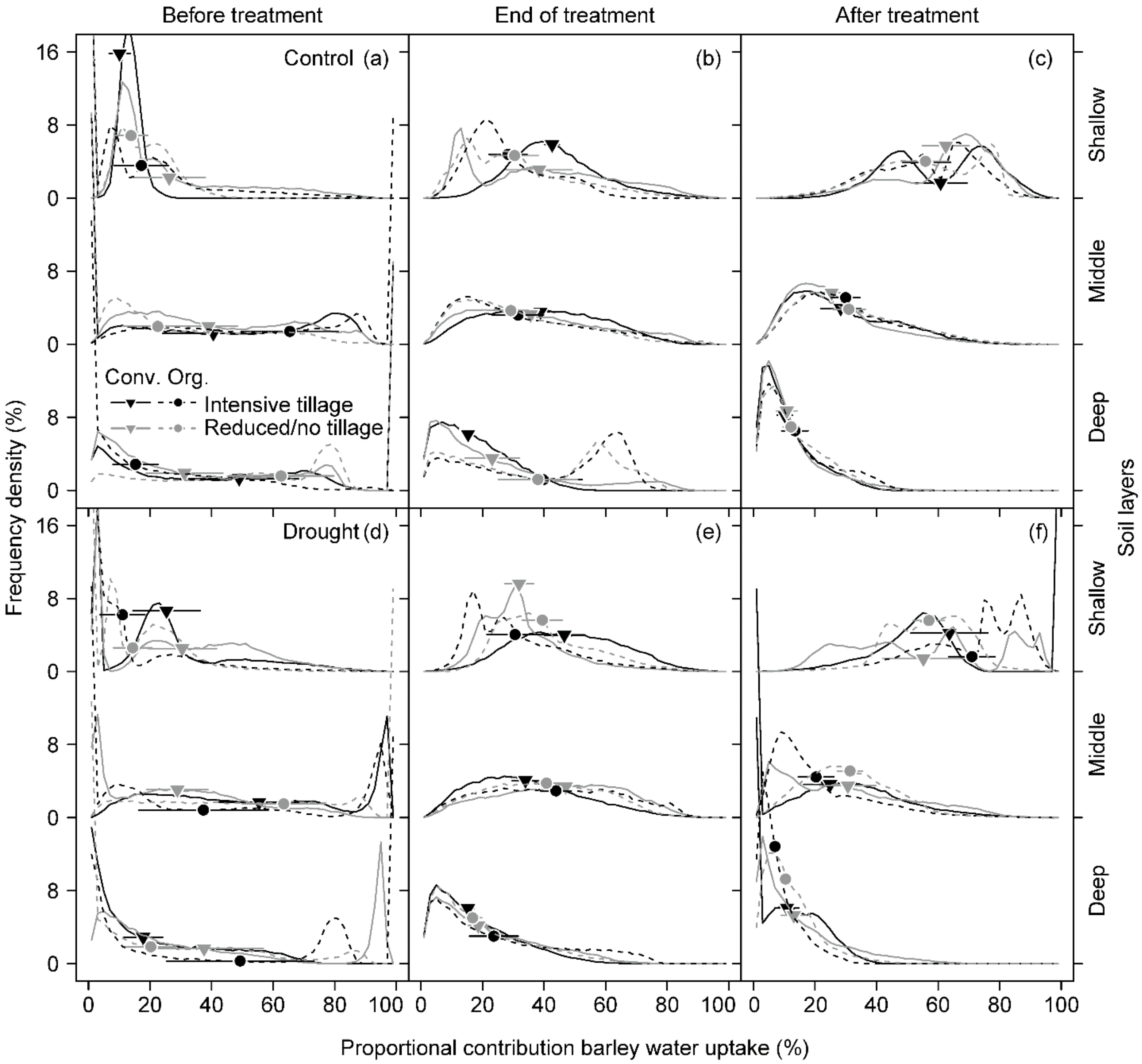

Fig. 5 Frequency density distribution of model outputs on the proportional contribution of soil water to barley water uptake from shallow (0-20 cm, sum of 0-5, 5-10, and 10-20 cm), middle (20-40 cm, sum of 20-30 and 30-40 cm), and deep (40-60 cm) soil layers under different cropping systems (a, b) before the drought treatment on 7 May, (c, d) at the end of treatment on 25 June, and (e, f) after treatment on 11 July in 2018. Frequency density was derived from 10 000 simulations at 2% increment of mixing models using $\delta^{18}O$ for each subplot (Conv. for conventional, Org. for organic). Data were pooled for all subplots in each cropping system. Symbols on the curves indicate the median of the model outputs for each soil layer. Means and 1 SE (horizontal bars) of each cropping system are given (n = 3-4).

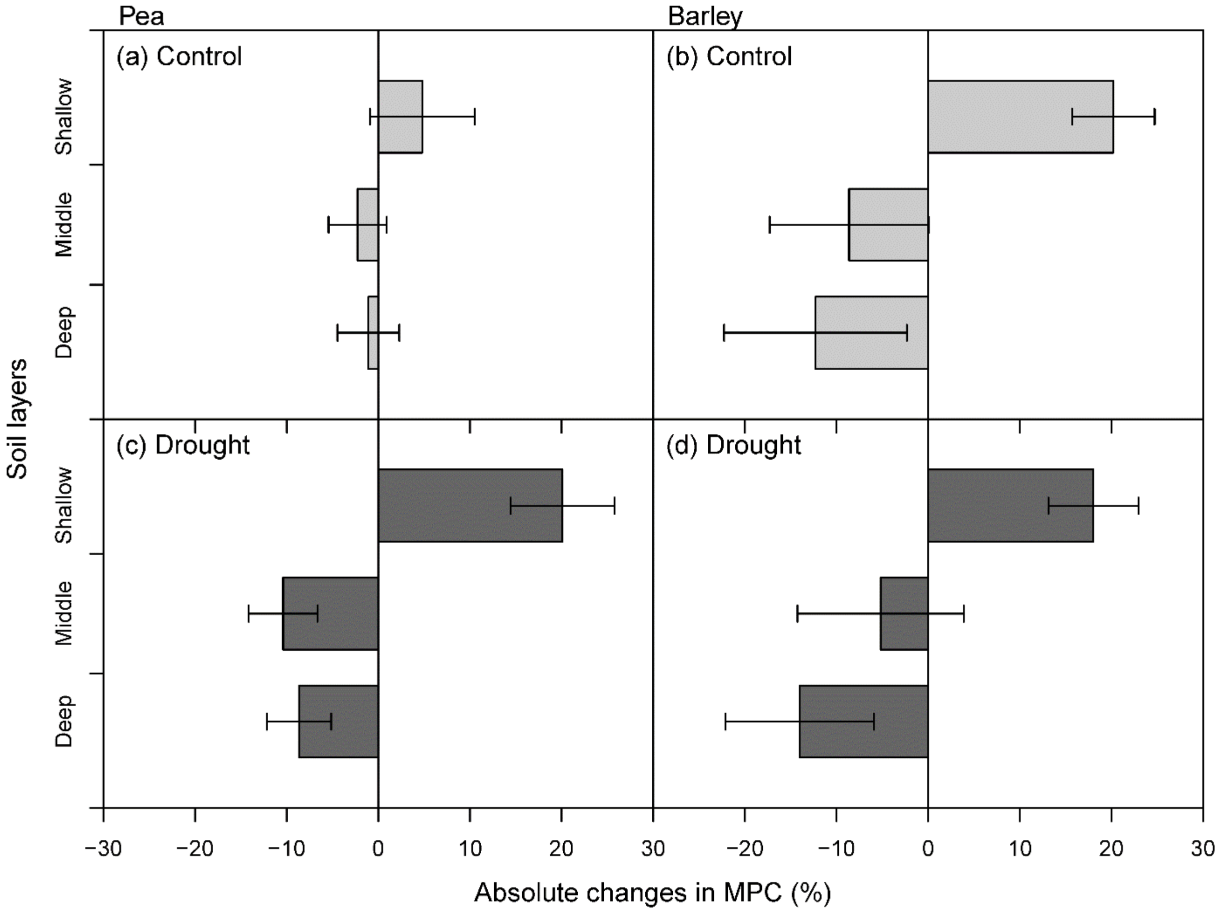

Fig. 6 Absolute changes in median proportional contributions (MPC) to plant water uptake ~~(MPC)~~ of pea (a, c) and barley (b, d), calculated as the difference of MPC at the end (25 June; ET) and before the drought treatment (7 May; BT), from three soil layers in control (a, b) and drought (c, d) subplots in all cropping systems. MPC was derived from 10 000 simulations of mixing models using stable water isotope data. Proportional contribution from the shallow layer is the sum of 0-5, 5-10, and 10-20 cm depths, the middle layer is the sum of 20-30 and 30-40 cm depths, and the deep layer represents 40-60 cm. Means and 1 SE (horizontal lines) are given (n = 14-16).