

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

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13 **Highlights**

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

18 **Abstract**

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

34 **Keywords:** climate change, conservation tillage, organic farming, stable water isotope, water  
35 uptake depth

36

## 37 **1 Introduction**

38 Due to climate change, drought events may occur more frequently and become more severe than at  
39 present, and hence water scarcity is worsening in many regions of the world ([Schewe et al., 2014](#);  
40 [IPCC, 2019](#)). Thus, agriculture is facing increasing pressure to ensure food security under  
41 aggravating drought conditions ([FAO, 2018](#); [FAO, 2019](#)). Although crop breeding has large  
42 potential to enhance agricultural productivity, it should certainly not be seen as the only option.  
43 Adaptive crop management to a changing climate is discussed as an additional solution to mitigate  
44 yield loss under drought, potentially by sustaining plant growth, enhancing soil water availability,  
45 or by promoting mycorrhizal symbiosis ([Cochard, 2002](#); [Bot & Benites, 2005](#); [Kundel et al., 2020](#);  
46 [Wahdan et al., 2021](#)). Therefore, there is a growing interest in organic farming and conservation  
47 tillage (i.e., no tillage or reduced tillage), as these management practices have been shown to be  
48 beneficial to soil health and water holding capacity, ecosystem stability, as well as environmental  
49 sustainability (e.g., [Seitz et al., 2019](#); [Teasdale et al., 2007](#); [Hobbs et al., 2008](#); Wittwer et al.,  
50 2021). However, an evaluation of different cropping systems as a means to support arable crops  
51 under drought is still urgently needed ([IPCC, 2019](#)).

52 Understanding plant water relations under drought plays an increasingly important role in  
53 promoting sustainable agriculture to secure food production ([Penna et al., 2020](#)). Plant water uptake  
54 and water use, particularly during critical growing stages, greatly determine physiological  
55 processes, survival, and ultimately crop productivity ([Boyer & Rao, 1984](#); [Wang et al., 2015](#)).

56 Although many studies reported plant water uptake patterns in response to drought over a broad  
57 range of species and environments (e.g., [Prechsl et al., 2015](#); [Grossiord et al., 2019](#); [Rasmussen et](#)  
58 [al., 2020](#); [Ding et al., 2021](#)), only very few focused on arable agriculture (e.g., [Zegada-Lizarazu et](#)  
59 [al., 2006](#); [Borrell et al., 2014](#); [Wu et al., 2018](#)) and none compared arable cropping systems.

60 Moreover, these studies found contrasting responses of crop species to changing environments,  
61 illustrating the current gap of knowledge on plant water relations in cropping systems.

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

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

## 84 **2 Materials and Methods**

### 85 **2.1 Research site and experimental setup**

86 The research site is in Rümlang near Zurich (47.26° N, 8.31° E, 489 m a.s.l.), and belongs to the  
87 Swiss federal agricultural research station Agroscope. Long-term average annual precipitation at the

88 site is 994 mm, and mean annual air temperature is 9.7 °C (1988 to 2017; [MeteoSwiss, 2020](#)). The  
89 soil at the research site is a calcareous Cambisol with 23% clay, 34% silt, and 43% sand, and total  
90 soil carbon content of 1.6 to 1.8% ([Loaiza Puerta et al., 2018](#)). The plant available soil depth is 50-  
91 70 cm, and no groundwater is accessible for plants ([Kanton Zürich, 2021](#)). Our study used a sub-set  
92 of plots in the Farming Systems and Tillage Experiment which began in 2009 with a six-year crop  
93 rotation that is typical for Swiss cropping systems (for details see [Wittwer et al., 2017](#)). It combines  
94 conventional (C) and organic (O) farming with intensive or soil conservation tillage practices. The  
95 conventional systems are managed according to the “Proof of Ecological Performance” (PEP)  
96 guidelines of the Swiss Federal Office for Agriculture ([Swiss Federal Council, 2021](#)), which allows  
97 synthetic fertiliser and pesticide applications. The organic systems were managed following the  
98 BioSuisse guidelines, prohibiting the use of mineral fertilisers and synthetic plant protection  
99 products. Intensive tillage (IT) with a mouldboard plough to 20 cm depth followed by seedbed  
100 preparation with a rotary harrow to 5 cm depth was applied in both conventional (C-IT) and organic  
101 systems (O-IT). For conservation tillage, direct sowing and no soil management were implemented  
102 in the no tillage conventional plots (C-NT) but glyphosate was sprayed before sowing of the main  
103 crops for weed control. A disc or rotary harrow, which superficially disturbed the soil for weed  
104 control, was used for reduced tillage in organically managed plots (O-RT) to a maximum depth of  
105 10 cm. These four cropping systems were repeated in four blocks following a Latin square design.  
106 Cropping system plots had an area of 6 m × 30 m.

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

113 In order to simulate a future drought scenario (CH2018), portable rain shelters were installed from  
114 22 May to 28 June 2018 (37 days) during the 108-day growing season. This resulted in a 34%  
115 reduction in precipitation from the drought subplots during the growing season in 2018 (from  
116 sowing to harvest; Table 1). No irrigation was applied to the control plots during the (unexpected)  
117 naturally dry period in June for logistical and rational reasons, i.e., irrigation is unusual for the  
118 region and this crop, and the dry period happened during the ripening phase of the crop. The  
119 portable, tunnel-shaped rain shelters (metal frames of 3 m × 5 m base area and 2.1 m height at the  
120 highest point) were covered with transparent and ultraviolet light-transmissible plastic foil  
121 (Gewächshausfolie UV5, 200 µm, Folitec Agrarfolien-Vertrieb, Germany) and were open at both  
122 ends as well as at both sides and had an opening at the top along the full length. This allowed  
123 extensive ventilation and prevented temperature build-up (for technical details see [Hofer et al.,](#)  
124 [2016](#)). Rain running down the foil was collected in PVC half pipes and directed away from the plots  
125 (about 2 m). These drought subplots were established in each cropping system (which were in  
126 place since 2009) and located directly next to control subplots which received natural precipitation  
127 inputs, resulting in a split-plot layout. A total of 16 experimental plots (four cropping systems ×  
128 four replicates) with 32 subplots (16 plots × two water availability treatments) were used in this  
129 study. Our experimental design thus compared replicated drought and control sub-plots in parallel  
130 (i.e., at the same time), not after each other (i.e., a temporal replication over multiple years), since in  
131 crop rotations, the identical crop cannot be grown on the same field for several years due to soil  
132 health issues.

## 133 **2.2 Climatic data and soil water contents**

134 Precipitation and air temperature data (Table 1; Fig. 1) were obtained from a nearby weather  
135 station, Zürich/Kloten (KLO, 47.48° N, 8.54° E, 4.6 km north of the research site, [MeteoSwiss,](#)  
136 [2020](#)). Soil water content (SWC) was continuously measured and recorded at 10 and 40 cm depths  
137 with two replicates per cropping system (EC-5, Decagon Devices Inc., Pullman, WA, USA; factory-

138 calibrated). Data were averaged at 10 min intervals by data loggers (CR1000 and CR216, Campbell  
139 Scientific Ltd., Loughborough, UK), then averaged for daily values.

### 140 **2.3 Plant and soil water samples for stable isotope analysis**

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

### 156 **2.4 Stable water isotope analyses**

157 The oxygen and hydrogen stable isotope ratios ( $\delta^{18}\text{O}$  and  $\delta^2\text{H}$ ) of extracted water samples were  
158 analysed by coupling a high-temperature elemental analyser (TC/EA, Finnigan MAT, Bremen,  
159 Germany) with an isotope ratio mass spectrometer (IRMS, Delta<sup>plus</sup>XP, Finnigan MAT, Bremen,  
160 Germany) via a ConFlo III interface (Finnigan MAT, see [Werner \*et al.\*, 1999](#)) using the high-  
161 temperature carbon reduction method described by [Gehre \*et al.\* \(2004\)](#). All  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values are  
162 expressed relative to the Vienna Standard Mean Ocean Water (VSMOW-SLAP, [Craig & Gordon,](#)  
163 [1965](#); [Gat, 2010](#)) in parts-per-thousand (or "per mil", ‰; eq. 1):

$$\delta^{18}\text{O} \text{ or } \delta^2\text{H} = \frac{R_{\text{SAMPLE}}}{R_{\text{STANDARD}}} - 1 \quad (1)$$

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

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

$$\text{GMWL: } \delta^2\text{H} = 8.2 \times \delta^{18}\text{O} + 11.7 \quad (2)$$

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

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

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

## 176 **2.5 Bayesian mixing model for plant water uptake**

177 Proportional contributions of soil water to plant water uptake (PC) from different depths were  
 178 estimated using mixing models from the R package ‘simmr’ ([Parnell, 2020](#)) within a Bayesian  
 179 framework based on code by [Parnell et al. \(2013\)](#). The  $\delta^{18}\text{O}$  or  $\delta^2\text{H}$  signatures of soil water from the  
 180 six soil layers were used as sources, and plant xylem water was considered the mixture for  
 181 modelling in each subplot at different sampling times, i.e., BT, ET, and AT. Missing replicates of  
 182 soil samples due to sampling difficulties ( $n = 5$  in total) were filled with mean values of the other  
 183 replicates from the same cropping system and treatment to have balanced model inputs. The model

184 outputs consisted of 10 000 possible combinations of PC from different soil depths from four  
185 Markov chain Monte Carlo Bayesian models with at least 300 000 iterations, 50 000 burns, and 100  
186 times of thinning for each chain. The median of the model outputs on PC (MPC) from each soil  
187 depth was calculated for each subplot and used for statistical analysis on plant water uptake depths.  
188 Compared to the most frequent value of the model outputs, MPCs of all the sources usually sum up  
189 closer to 1. To increase clarity of presentation, PC was grouped into three layers, namely shallow  
190 (0-20 cm), middle (20-40 cm), and deep (40-60 cm) soil layers for further analyses. The PC values  
191 from shallow and middle layers are the sum of PC from soil depths of 0-5, 5-10, and 10-20 cm, and  
192 the sum of PC from soil depths of 20-30 and 30-40 cm, respectively. As  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  yielded  
193 similar results, only the model outputs of  $\delta^{18}\text{O}$  are described in detail in this paper.

## 194 **2.6 Data analyses**

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

209 by the Tukey HSD (honestly significant difference) test using the function *glht()*, from the R  
210 package ‘multcomp’ ([Hothorn et al., 2008](#)).

## 211 **3 Results**

### 212 **3.1 Environmental conditions in drought and control subplots**

213 Air temperatures in 2018 were very high compared to the long-term mean, in particular in May and  
214 June, with a daily average air temperature of 15.8 and 18.8 °C, respectively, while the long-term  
215 (1988 to 2017) mean air temperatures in these two months were 13.9 and 17.2 °C, respectively  
216 (Table 1; Fig. 1). Annual precipitation was relatively low (Table 1). While the precipitation in May  
217 2018 (102 mm) was comparable to the long-term mean (1988 to 2017: 105 mm), no precipitation  
218 fell between 14 June and 2 July 2018 (naturally dry period), resulting in a below-average  
219 precipitation in June (40 mm; long-term mean of 102 mm, Table 1), followed by an even more  
220 pronounced drought period in July (Fig. 1). Average daily soil water contents (SWC) in the control  
221 subplots ranged from 16% to 29% at 10 cm depth and slightly higher, from 22% to 29%, at 40 cm  
222 depth, prior to the rain event on 3 July 2018. After this rain event, SWC increased in all cropping  
223 systems at both depths (Fig. 2a, b). Variations in SWC among cropping systems were small,  
224 particularly during the naturally dry period in June. SWC in drought subplots of all cropping  
225 systems decreased continuously during the 37-day drought treatment (22 May to 28 June 2018),  
226 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  
227 any pronounced differences among cropping systems, while SWC at 40 cm tended to be slightly  
228 higher in cropping systems with conservation tillage (O-RT and C-NT) compared to systems with  
229 intensive tillage (O-IT and C-IT; Fig. 2b, d).

### 230 **3.2 Stable isotopes in soil water and plant xylem water**

231 In the dual-isotope space, stable oxygen and hydrogen isotope ratios of soil and plant xylem waters  
232 were strongly related with each other ( $R^2 = 0.89$  and  $0.85$ , respectively; Fig. S1) and generally fell  
233 below the local meteoric water line (LMWL) of 2018, representing evaporation. Stable isotope  
234 signatures of xylem water were lower than the LMWL but higher than those of soil water,

235 indicating that xylem water isotope signatures were mixtures of the original source precipitation and  
236 the pool of soil water, affected by different degrees of fractionation.

237 The stable water isotope profiles of soil water showed a characteristic pattern at all times, for all  
238 cropping systems and both treatments, with most enriched values in the uppermost soil and  
239 increasingly depleted values with increasing soil depth (Table S1; Fig. 3 for  $\delta^{18}\text{O}$ ; Fig. S2 for  $\delta^2\text{H}$ ).

240 The drought treatment showed no significant effects before the treatment (BT) for  $\delta^{18}\text{O}$  nor  $\delta^2\text{H}$   
241 (except for  $\delta^2\text{H}$  at 20-30 cm; Table 2). In contrast, at the end of the drought treatment (ET), soil  
242 water  $\delta^{18}\text{O}$  values from 20-60 cm (20-30, 30-40, and 40-60 cm) as well as  $\delta^2\text{H}$  values from all  
243 depths were strongly affected by the drought treatment (all  $P < 0.05$ ; Table 2), with more depleted  
244 signatures in the drought than in control subplots due to the exclusion of more enriched summer  
245 precipitation. Even after the shelters were removed and the treatment had been finished (AT), the  
246 drought treatment still significantly affected both  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  of soil water, albeit only in deeper  
247 soil depths (30-40 and 40-60 cm for  $\delta^{18}\text{O}$  and 40-60 cm for  $\delta^2\text{H}$ ; all  $P < 0.05$ ; Table 2). Overall,  
248 cropping systems did not significantly affect the stable isotopic signatures in soil water at any time  
249 (Table 2).

250 Pea xylem water was always significantly more enriched in  $^{18}\text{O}$  and  $^2\text{H}$  compared to barley (all  $P <$   
251  $0.001$ ; Table S2). The  $\delta^{18}\text{O}$  values in xylem water for pea ranged between  $-8.8\text{‰}$  and  $-5.7\text{‰}$ , and  
252 significantly lower between  $-10.1\text{‰}$  and  $-5.8\text{‰}$  for barley (averages per cropping system, treatment,  
253 and time; Table 3; Table S2). Similarly, the  $\delta^2\text{H}$  values in xylem water for pea ranged  
254 between  $-65.6\text{‰}$  and  $-52.1\text{‰}$ , and significantly lower between  $-74\text{‰}$  and  $-47.1\text{‰}$  for barley (Table  
255 3; Table S2). Overall, isotopic signatures in xylem water became more enriched in  $^{18}\text{O}$  and  $^2\text{H}$   
256 during the growing season for both pea and barley (Fig. 3, Table S2, Fig. S2). On average, the  
257 xylem  $\delta^{18}\text{O}$  for pea was  $-8.5\text{‰}$  before the treatment (BT) and  $-7.2\text{‰}$  at the end of the treatment  
258 (ET), compared to  $-9.8\text{‰}$  (BT),  $-8.8\text{‰}$  (ET), and  $-6.3\text{‰}$  after the treatment (AT) for barley. While  
259 average  $\delta^2\text{H}$  values for pea were  $-64.1\text{‰}$  (BT) and  $-57.6\text{‰}$  (ET),  $\delta^2\text{H}$  values averaged  $-72.2\text{‰}$   
260 (BT),  $-68.6\text{‰}$  (ET), and  $-50.8\text{‰}$  (AT) for barley (Fig. 3; Table S1; Fig. S2). Since there was a

261 strong relationship between  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  in xylem water (Fig. S1;  $R^2 = 0.85$ ), our analyses are  
262 mainly focused on  $\delta^{18}\text{O}$  in the text (but see Table 3, Table S2, and Fig. S2 for analyses on  $\delta^2\text{H}$ ).  
263 For pea, cropping systems did not significantly affect  $\delta^{18}\text{O}$  nor  $\delta^2\text{H}$  in xylem water at either time  
264 (BT and ET; Table S2), while the drought treatment significantly affected the isotopic signatures of  
265  $^{18}\text{O}$  only at the end of treatment (ET:  $P = 0.022$ ; no interactions between cropping systems and  
266 drought treatment:  $P = 0.085$ ; Table S2).  $^{18}\text{O}$  in pea xylem water were significantly more enriched  
267 in the drought than in the control subplots (on average,  $\delta^{18}\text{O}$  of  $-6.9\text{‰}$  and  $-7.7\text{‰}$ , respectively).  
268 In contrast to pea, cropping systems significantly affected  $\delta^{18}\text{O}$  in barley xylem water (ET:  $P =$   
269  $0.035$ ; Table S2). The drought treatment significantly affected the isotope signatures of both  $^{18}\text{O}$   
270 and  $^2\text{H}$  at the end of treatment (ET: both  $P < 0.01$ ; no interactions between cropping systems and  
271 drought treatment; Table S2). However, unlike pea, the xylem water of barley showed significantly  
272 lower  $\delta^{18}\text{O}$  values in drought than in control subplots for all cropping systems (on average,  $-9.0\text{‰}$   
273 and  $-8.6\text{‰}$ , respectively), although the difference was small (Table S2). A similar pattern was also  
274 observed for  $\delta^2\text{H}$  at the end of treatment (ET), with significantly lower values on average in drought  
275 than in control subplots (ET:  $-71.8\text{‰}$  and  $-65.4\text{‰}$ , respectively).

### 276 **3.3 Modelled plant water uptake depths**

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

287 with different proportions depending on species, time (i.e., BT, ET, and AT) and treatment (i.e.,  
288 control vs. drought; Table 4; Table 5).

289 For pea, soil water contributions to total plant water uptake decreased with increasing soil depth in  
290 both control and drought subplots before (BT) and at the end of the treatment (ET) for all cropping  
291 systems (Fig. 4). The median of PC values (MPC) differed significantly among shallow (0-20 cm),  
292 middle (20-40 cm), and deep (40-60 cm) layers, averaging 47%, 33%, and 16%, respectively, for  
293 both treatments and all cropping systems (BT; Table 5; Fig. 4a, c). At ET, pea plants subjected to  
294 drought significantly shifted their water uptake to even higher contributions from the shallow layer  
295 (67%) and less uptake from middle (22%) and deep (8%) soil layers compared to BT (Table 5; Fig.  
296 4d; Table S5). Pea plants in control subplots did not display such a significant shift, but remained  
297 with average MPC from shallow, middle, and deep soil layers of 52%, 31%, and 14%, respectively  
298 (Table 5; Fig. 4b; Table S5). Cropping systems did not significantly affect MPC before (BT) or at  
299 the end of (ET) treatment (also no interactions between cropping systems and drought, Table 5; Fig.  
300 4d).

301 In contrast to pea, barley plants showed very different water uptake patterns before the treatment  
302 (BT), with significantly lower PC from the shallow soil layer compared to the middle and deep  
303 layers. For barley, MPC values averaged 19%, 44%, and 35% for shallow, middle, and deep soil  
304 layers, respectively, for both treatments and all cropping systems (Fig. 5a, d). However, at the end  
305 of the treatment (ET), barley plants significantly increased the contributions from the shallow layer  
306 in drought subplots, similar to pea (Table 5; Fig. 5e; Table S5), resulting in MPC values of 38%,  
307 41%, and 18% from shallow, middle, and deep soil layers, respectively. The MPC further shifted  
308 after the treatment (AT) to values of 62%, 27%, and 10% from shallow, middle, and deep layers,  
309 respectively (Fig. 5f). Also in control subplots, barley plants showed the same significant shift from  
310 BT to ET, with MPC values at ET of 35%, 34%, and 29% from shallow, middle, and deep layers,  
311 respectively (Table 5; Fig. 5b; Table S5), and from ET to AT with MPC values AT of 59%, 29%,

312 and 12% from shallow, middle, and deep layers, respectively (Table 5; Fig. 5c; Table S5). Similar  
313 to pea, barley water uptake patterns were not significantly affected by cropping systems (Table 5).  
314 Overall, MPC values from shallow and deep layers for pea and barley were positively correlated ( $r$   
315 = 0.64 and 0.55, respectively; Fig. S3). This means when barley took up more water from the  
316 shallow layer, so did pea.

317 Organic as well as reduced/no tillage cropping systems are discussed as adaptation strategies under  
318 climate change conditions to ensure arable crop production. Thus, we analysed plant water uptake  
319 depths in drought subplots at the end of treatment (ET) more in detail, although cropping systems  
320 showed no significant effects on water uptake depths for either species and no interactions occurred  
321 between cropping systems and drought treatment (Table 5). Pea plants in both intensive systems (C-  
322 IT and O-IT) showed significantly higher (O-IT: 77%) or similar (C-IT: 65%) contributions to total  
323 water uptake (as MPC) from the shallow layer (0-20 cm) compared to conservation tillage systems  
324 (64% in both C-NT and O-RT; Table 5; Fig. 4d). Conversely, contributions from the middle layer  
325 (20-40 cm) for pea at the end of treatment (ET) were only 15% in O-IT compared to 24% in the  
326 other three cropping systems (O-RT, C-IT, and C-NT). Differences among cropping systems under  
327 drought were even smaller for barley than for pea (Table 5; Fig. 5e). MPC values of barley for  
328 uptake from the shallow layer were 47% (C-IT), 39% (O-RT), 31% (O-IT), and 32% (C-NT).  
329 Conversely, contributions from the middle layer were the largest in C-NT (47%), followed by O-IT  
330 (44%) and O-RT (41%), and lowest in C-IT (34%). The absolute changes in MPC values between  
331 before the treatment (BT) and the end of treatment (ET) were not significantly affected by cropping  
332 systems for either species, but significantly affected by the drought treatment for pea (for the  
333 shallow and middle soil layers; Table S6).

#### 334 **4 Discussion**

335 Root water uptake patterns are often discussed for their important role in plant water relations, but  
336 only few studies considered arable crop species ([Penna et al., 2020](#)). In addition, most studies on  
337 responses of crop root water uptake patterns to drought took place in pots or under controlled

338 conditions (e.g., [Zegada-Lizarazu & Iijima, 2004](#); [Araki & Iijima, 2005](#)), so that information on  
339 field conditions is particularly scarce, except maize ([Ma & Song, 2016](#)), wheat ([Ma & Song, 2018](#)),  
340 oilseed rape, and barley in monoculture ([Wu et al., 2016](#)). Furthermore, studies comparing the role  
341 of different cropping systems for crop water uptake are completely lacking. Here, we showed for  
342 the first time that root water uptake patterns of field-grown pea and barley in mixture responded to  
343 drought but not to different cropping systems. Subjected to a pronounced drought period (37 d  
344 without precipitation), both crop species shifted to relying more on shallow soil layer (0-20 cm) for  
345 water uptake. This drought response was independent of the cropping system, i.e., organic vs.  
346 conventional farming or intensive vs. conservation tillage.

347 Previous research on root water uptake patterns in crop as well as grassland species showed  
348 ambiguous responses to drought. For some species, root water uptake depth was dependent on root  
349 distribution during wet periods, but on soil water availability during dry periods ([Sprengrer et al.,](#)  
350 [2016](#)). Therefore, utilising more water from deep than from shallow soil layer is typically the  
351 anticipated drought response, such as barley in monoculture ([Wu et al., 2018](#)), maize ([Ma & Song,](#)  
352 [2016](#)), wheat, rice, soybean ([Zegada-Lizarazu & Iijima, 2004](#)), or chickpea ([Purushothaman et al.,](#)  
353 [2017](#)). However, other studies reported that crop and grassland species do not take up water from  
354 deeper depths under drought but even absorb more water from shallow soil layer (e.g., barley in  
355 monoculture, maize, pigeon pea, cowpea; [Zegada-Lizarazu & Iijima, 2004](#)), or grassland species  
356 ([Hoekstra et al., 2014](#); [Prechsl et al., 2015](#); [Wu et al., 2016](#)). This is in accordance with our results  
357 in which both pea and barley increased their proportional water uptake from shallow layer (0-20  
358 cm) at the end of treatment (ET) in the drought subplots. Although soil water contents (SWC) were  
359 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  
360 cm depths were both very low. Thus, the whole soil profile showed very low water availability at  
361 the end of the treatment (ET), and fine root distributions most likely dominated plant water uptake  
362 patterns.

363 Rooting profiles for legumes with increased proportions of deeper roots under drought, e.g., below  
364 23-30 cm, have been reported ([Benjamin & Nielsen, 2006](#); [Purushothaman \*et al.\*, 2017](#)), although  
365 different responses in root growth to drought were found among different varieties ([Kashiwagi \*et\*](#)  
366 [al., 2006](#); [Kumar \*et al.\*, 2012](#); [Purushothaman \*et al.\*, 2017](#)). The architecture of legume root systems  
367 is strongly affected by rhizobia, which typically find better living conditions in terms of oxygen and  
368 nitrogen concentrations higher up in the soil profile than at greater depths ([Concha & Doerner,](#)  
369 [2020](#)), also in dry soils. Moreover, barley grown under drought conditions has been reported to  
370 develop proportionally more shallow roots (0-20 cm depth) relative to deeper soil depths ([Carvalho](#)  
371 [et al., 2014](#)). Also, studies on grassland plants (both legume and grass species) found increasing  
372 root biomass production in shallow soil depths (0-15 cm) in response to drought (e.g., [Prechsl \*et al.\*,](#)  
373 [2015](#)). Although we did not investigate root distributions for either crop species, they most likely  
374 followed such evolutionary strategies as well during our rather strong, 37-day drought treatment, in  
375 addition to recent crop breeding efforts leading to less deep root systems in general ([Canadell \*et al.\*,](#)  
376 [1996](#); [Thorup-Kristensen \*et al.\*, 2020](#)). Moreover, shifting to shallower water uptake depths during  
377 drought might actually be beneficial for nutrient acquisition ([Querejeta \*et al.\*, 2021](#)), since not only  
378 concentrations of soil water and atmospheric N<sub>2</sub> are higher in the top soil than in the deeper soil, but  
379 also litter inputs for N mineralisation. Thus, besides the low soil moisture within the entire soil  
380 profile, acclimation of the root systems most likely also contributed to the shift towards shallower  
381 water uptake depths under drought for both pea and barley in this study.

382 The year 2018 was characterised by low precipitation during our experimental period, when a  
383 naturally dry period occurred at the end of our pronounced drought treatment in June (which  
384 excluded 34% of the precipitation during the growing season; Table 1). Our treatment compared  
385 well with the climate scenarios available for Switzerland, with a 25% reduction of precipitation in  
386 2060, and up to 40% by the end of the century; and an increase of the longest rain-free summer  
387 period (June, July, August) from currently 11 days to 20 days (CH2018). The dry period in June  
388 affected pea and barley plants in our control subplots differently (Fig. 6a, b). While pea did not shift

389 its water uptake pattern (Fig. 6a; Table S5), barley grown in the control subplots reacted very  
390 similar to the natural dry period (before the ET sampling, 14 to 25 June; Fig. 2) as barley subjected  
391 to our drought treatment, namely with a clear shift from deep (40-60 cm) to shallow (0-20 cm) soil  
392 layer (Fig. 6b, d; Table S5). However, barley still relied more on water uptake from the deep soil  
393 layer during this naturally dry period in the control subplots than under the experimental drought ( $P$   
394 = 0.017; Table 5). Hence, these different reactions of the two species to the dry period clearly  
395 indicated that barley was more susceptible than pea even to a mild water stress. This observation is  
396 fully in line with measurements of stem hydraulic traits (i.e., loss of xylem conductance) from the  
397 same experiment ([Sun et al., 2021](#)). Barley plants lost xylem conductance much earlier than pea  
398 plants when xylem water potentials decreased. In addition, legumes like pea can maintain low  
399 stomatal conductance to avoid water stress without compromising photosynthesis when growing  
400 under conditions with limiting water supply, due to their high foliar N concentrations ([Adams et al.,](#)  
401 [2018](#)). This adds to the hydraulic trait benefits of pea and explains why pea was less affected by the  
402 natural dry period. Nevertheless, as shown in our study, if severities and frequencies of droughts  
403 increase in the future, one can expect negative consequences not only on the performance of barley,  
404 but also of pea ([Martin & Jamieson, 1996](#)).

405 Moreover, the two species growing together in the pea-barley mixture showed distinct niches for  
406 root water uptake before drought, with pea relying more on water from shallow (0-20 cm) and  
407 barley from deep (40-60 cm) soil layers, in accordance with resource partitioning in the absence of  
408 water limitation as observed in intercrops, e.g., pearl millet and cowpea ([Zegada-Lizarazu et al.,](#)  
409 [2006](#)) and in mixed-species grasslands (e.g., [Hoekstra et al., 2014](#)). However, the niches became  
410 more similar under drought conditions, contradicting ecological theory which postulates more  
411 pronounced niche differentiation and less niche overlap under stressful conditions, such as during a  
412 drought (see [Nippert & Knapp, 2007](#); [Silvertown et al., 2015](#); [Guderle et al., 2018](#)). However, our  
413 results were in line with results from biodiversity studies in temperate grasslands ([Bachmann et al.,](#)  
414 [2015](#); [Barry et al., 2020](#); [Hoekstra et al., 2014](#)) which also did not show niche differentiation in

415 response to increased competition or drought. Thus, further detailed knowledge on the dynamics of  
416 intercrop water uptake patterns is needed to solve this contradiction and to decrease the uncertainty  
417 for arable crop production now and under future climate conditions.

418 As global agriculture has already been considerably compromised by and become increasingly  
419 sensitive to climate change ([Ortiz-Bobea et al., 2021](#)), farming practices such as organic  
420 management and conservation tillage are being discussed widely. They have been shown to  
421 improve general soil conditions compared to conventional management and intensive tillage,  
422 particularly under drought ([Bot & Benites, 2005](#); [Gomiero et al., 2011](#); [Choudhary et al., 2016](#)). For  
423 instance, organic management and conservation tillage can increase soil water holding capacity,  
424 therefore providing higher water availability than conventional management and intensive tillage  
425 (e.g., [Colombi et al., 2019](#); [Kundel et al., 2020](#)). In this study, the systems with conservation tillage  
426 (C-NT and O-RT) indeed showed slightly higher SWC than systems with intensive tillage (C-IT  
427 and O-IT) at 40 cm (Fig. 2d). However, this did not result in any benefit for root water uptake  
428 patterns of pea and barley against drought. Water uptake of both species shifted to the shallow layer  
429 (0-20 cm) in all cropping systems under drought, without cropping system effects or interactive  
430 effects between cropping systems and drought treatment. Thus, any further changes in soil physical  
431 characteristics due to the drought treatment among cropping systems did not affect the observed  
432 root water uptake patterns. The relatively short period that annual crop species are growing under  
433 these conditions might limit the potential benefits from improved soil conditions present in those  
434 systems (e.g., [Dennert et al., 2018](#); [Loaiza Puerta et al., 2018](#); [Schluter et al., 2018](#)). Although it  
435 remains to be seen if the observed behaviour of a pea-barley mixture also holds true for other crop  
436 species, our results clearly challenge the potential of cropping management under temperate climate  
437 as a tool to adapt arable agriculture to climate change.

## 438 **5 Conclusions**

439 Water uptake patterns of pea and barley both shifted under drought in all cropping systems and both  
440 species relied more on water from the shallow soil layer (0-20 cm) than on water from deeper in the

441 soil profile. This was also the case for organic and reduced/no tillage cropping systems, which are  
442 often discussed as beneficial for crop performance, particular under water-limited conditions, and  
443 are thus suggested as adaptive cropping management practices under a future climate. However, in  
444 this study, we showed for the first time that cropping systems could not counteract the effects of  
445 severe drought on plant water uptake patterns for pea and barley grown in mixture. It remains to be  
446 seen if this observation also holds true for other, major crops grown under water-limited conditions.

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#### 457 **Author Contribution**

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

#### 461 **Conflict of Interest**

462 None declared.

#### 463 **Supporting Information**

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

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

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

470 Table S3 Effects of cropping systems, drought treatment and the interaction on the median  
471 proportional contributions from different soil depths to water uptake of pea and barley as well as  
472 mean  $\pm$  1 SE of MPC using  $\delta^{18}\text{O}$  data.

473 Table S4 Effects of cropping systems, drought treatment and the interaction on the median  
474 proportional contributions from different soil depths to water uptake of pea and barley as well as  
475 mean  $\pm$  1 SE MPC using  $\delta^2\text{H}$  data.

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

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

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

482 Fig. S2  $\delta^2\text{H}$  values of soil water from different depths and plant xylem water in each cropping  
483 system in 2018.

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

#### 486 **Data Availability Statement**

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

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- 688

689 Table 1 Precipitation and air temperature data from a nearby weather station, Zürich/Kloten (KLO,  
690 47.48° N, 8.54° E, 4.6 km north of the research site, [MeteoSwiss, 2020](#)) as well as dates for the  
691 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
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

692

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

Isotope	Depth (cm)	CS	D	CS × D	Blocks
Before drought treatment					
$\delta^{18}\text{O}$	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
$\delta^2\text{H}$	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	<b>0.026</b>	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					
$\delta^{18}\text{O}$	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	<b>&lt;0.001</b>	0.852	0.940
	30-40	0.437	<b>&lt;0.001</b>	0.651	0.954
	40-60	0.073	<b>0.008</b>	0.616	0.594
$\delta^2\text{H}$	0-5	0.295	<b>&lt;0.001</b>	0.168	0.479
	5-10	0.330	<b>0.005</b>	0.859	0.215
	10-20	0.091	<b>0.029</b>	0.700	0.659
	20-30	0.889	<b>&lt;0.001</b>	0.863	0.820
	30-40	0.388	<b>&lt;0.001</b>	0.551	0.970
	40-60	0.136	<b>0.006</b>	0.469	0.809
After drought treatment					
$\delta^{18}\text{O}$	0-5	0.393	0.059	0.848	0.291
	5-10	0.730	0.672	0.111	<b>0.031</b>
	10-20	0.538	0.612	0.734	0.993
	20-30	0.933	0.136	0.936	0.944
	30-40	0.881	<b>0.048</b>	0.979	0.772
	40-60	0.751	<b>0.001</b>	0.560	0.380
$\delta^2\text{H}$	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	<b>&lt;0.001</b>	0.493	0.484

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

698 Table 3 Effects of species (df = 1), cropping systems (CS, df = 3), drought treatment (D, df = 1) and  
 699 the interaction (species × CS, df = 3; species × D, df = 1; CS × D, df = 3; species × CS × D, df = 3)  
 700 on stable water isotopes ( $\delta^2\text{H}$  and  $\delta^{18}\text{O}$ ) of pea and barley before the drought treatment on 7 May  
 701 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	
	$\delta^{18}\text{O}$	$\delta^2\text{H}$	$\delta^{18}\text{O}$	$\delta^2\text{H}$
Species	<b>&lt;0.001</b>	<b>&lt;0.001</b>	<b>&lt;0.001</b>	<b>&lt;0.001</b>
CS	0.251	0.382	<b>0.038</b>	0.055
D	0.106	<b>&lt;0.001</b>	0.143	<b>0.001</b>
Species × CS	0.184	<b>0.023</b>	0.312	0.348
Species × D	0.796	0.486	<b>0.004</b>	<b>0.016</b>
CS × D	0.190	0.117	0.051	0.081
Species × CS × D	0.290	<b>0.045</b>	0.120	0.070
Blocks	0.485	0.599	<b>0.004</b>	0.162

702

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

704 Table 4 Effects of species (df = 1), cropping systems (CS, df = 3), drought treatment (D, df = 1) and  
 705 the interaction (species × CS, df = 3; species × D, df = 1; CS × D, df = 3; species × CS × D, df = 3)  
 706 on the median proportional contributions from different soil depths to water uptake (MPC) of pea  
 707 and barley before the drought treatment on 7 May and the end of treatment on 25 June in 2018  
 708 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	<b>&lt;0.001</b>	<b>0.036</b>	<b>&lt;0.001</b>	<b>&lt;0.001</b>	<b>&lt;0.001</b>	<b>&lt;0.001</b>
CS	0.506	0.555	0.992	0.374	0.440	0.252
D	0.849	0.775	0.629	<b>0.003</b>	0.546	<b>0.004</b>
Species × CS	0.255	0.865	0.702	0.303	0.799	0.180
Species × D	0.424	0.619	0.336	<b>0.009</b>	<b>0.001</b>	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	<b>0.008</b>	0.115	<b>0.016</b>

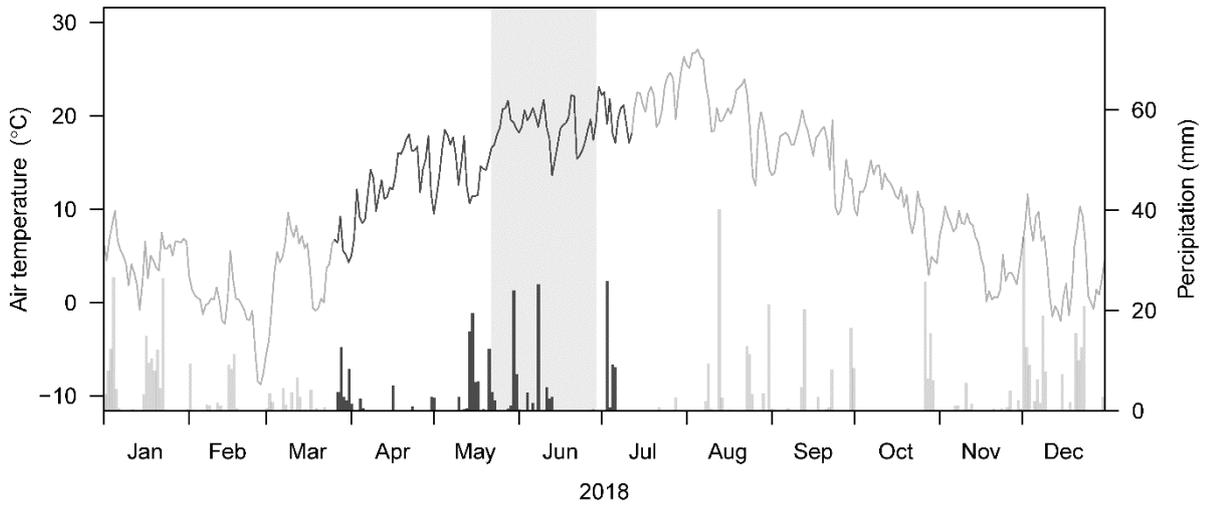
709

710 MPC was derived from 10 000 simulations by mixing models using  $\delta^{18}\text{O}$  data. Proportional  
 711 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  
 712 20-30 and 30-40 cm. Significant differences are shown in bold (*P* < 0.05).

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

Species (cm)	Depth		MPC								<i>P</i> 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														
Pea	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	
	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	
Barley	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	
	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														
Pea	0-20	63±6b	46±13a	48±9ab	51±4ab	65±4AB	64±14A	77±12B	64±7A	0.416	<b>0.001</b>	0.170	<b>0.01</b>	
	20-40	27±5a	36±9b	31±3ab	31±1ab	23±2AB	24±9B	15±8A	24±5B	0.416	<b>0.003</b>	0.703	<b>0.021</b>	
	40-60	8±1a	16±4ab	18±6b	14±5ab	9±1AB	10±4B	6±3A	8±1AB	0.398	<b>0.008</b>	0.272	<b>0.027</b>	
Barley	0-20	43±2	38±11	28±6	30±8	47±7B	32±5A	31±9AB	39±7AB	0.214	0.459	0.488	<b>0.034</b>	
	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	<b>0.017</b>	0.213	<b>0.028</b>	
After drought treatment														
Barley	0-20	61±9	62±8	56±8	56±8	64±13	55±13	71±8	57±5	0.696	0.546	0.436	<b>0.001</b>	
	20-40	28±6	25±5	30±5	31±5	25±9	31±9	20±6	31±4	0.664	0.604	0.508	<b>0.004</b>	
	40-60	10±2	11±3	13±4	12±3	11±4	13±4	7±2	10±1	0.852	0.401	0.225	<b>&lt;0.001</b>	

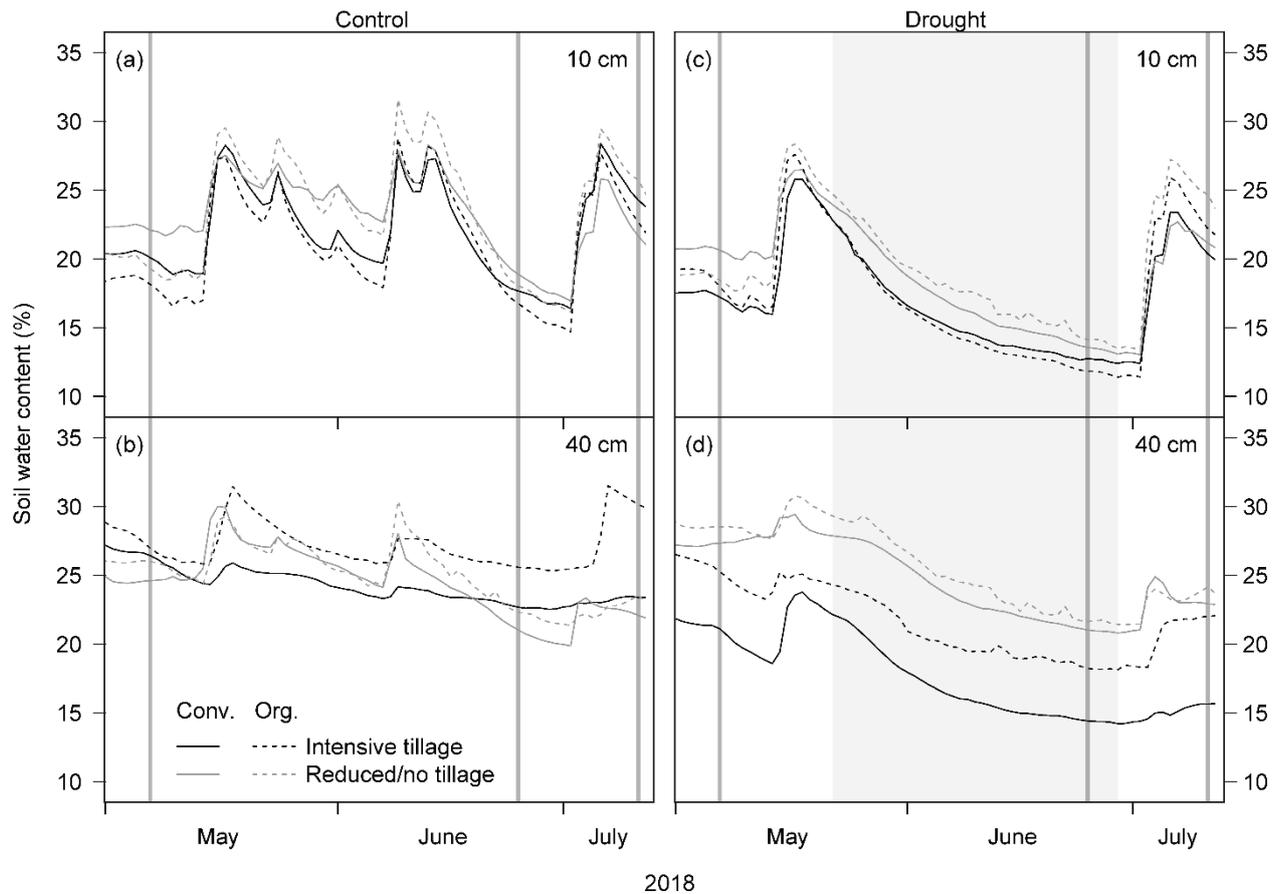
718  
 719 MPC was derived from 10 000 simulations by mixing models using  $\delta^{18}\text{O}$  data. Pea plants were  
 720 already senesced in early July therefore no stable water isotope data are available after the  
 721 treatment. Proportional contribution from 0-20 cm is the sum from 0-5, 5-10, and 10-20 cm, and 20-  
 722 40 cm is the sum from 20-30 and 30-40 cm. Mean ± 1 SE for MPC (%) are given for different  
 723 cropping systems (C-IT for Conventional intensive tillage, C-NT for Conventional no tillage, O-IT  
 724 for Organic intensive tillage, and O-RT for Organic reduced tillage). Different small and capital  
 725 letters indicate significant differences among cropping systems in control and drought subplots,  
 726 respectively, tested with Tukey HSD (honestly significant difference, *P* < 0.05). Significant effects  
 727 tested with linear mixed models are shown in bold (*P* < 0.05).



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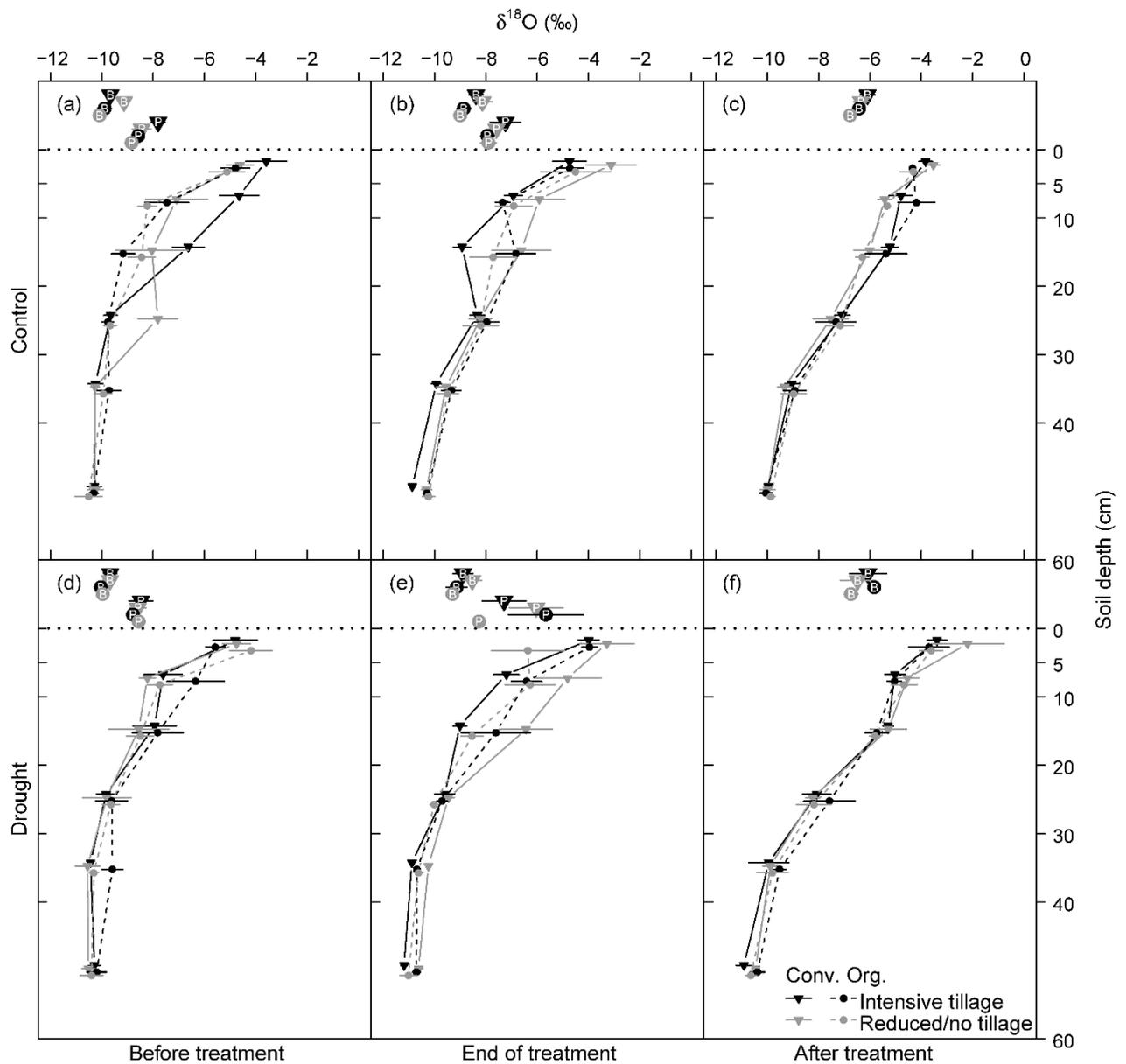
729 Fig. 1 Daily air temperature and precipitation in 2018. Dark line segments and bars depict the crop  
 730 growing season from 26 March to 12 July 2018. The shaded area indicates the drought treatment  
 731 from 22 May to 28 June 2018. Data from the MeteoSwiss station Zürich/Kloten (KLO, 47.48° N,  
 732 8.54° E, 4.6 km north of the research site, [MeteoSwiss, 2020](#)) are given.

733

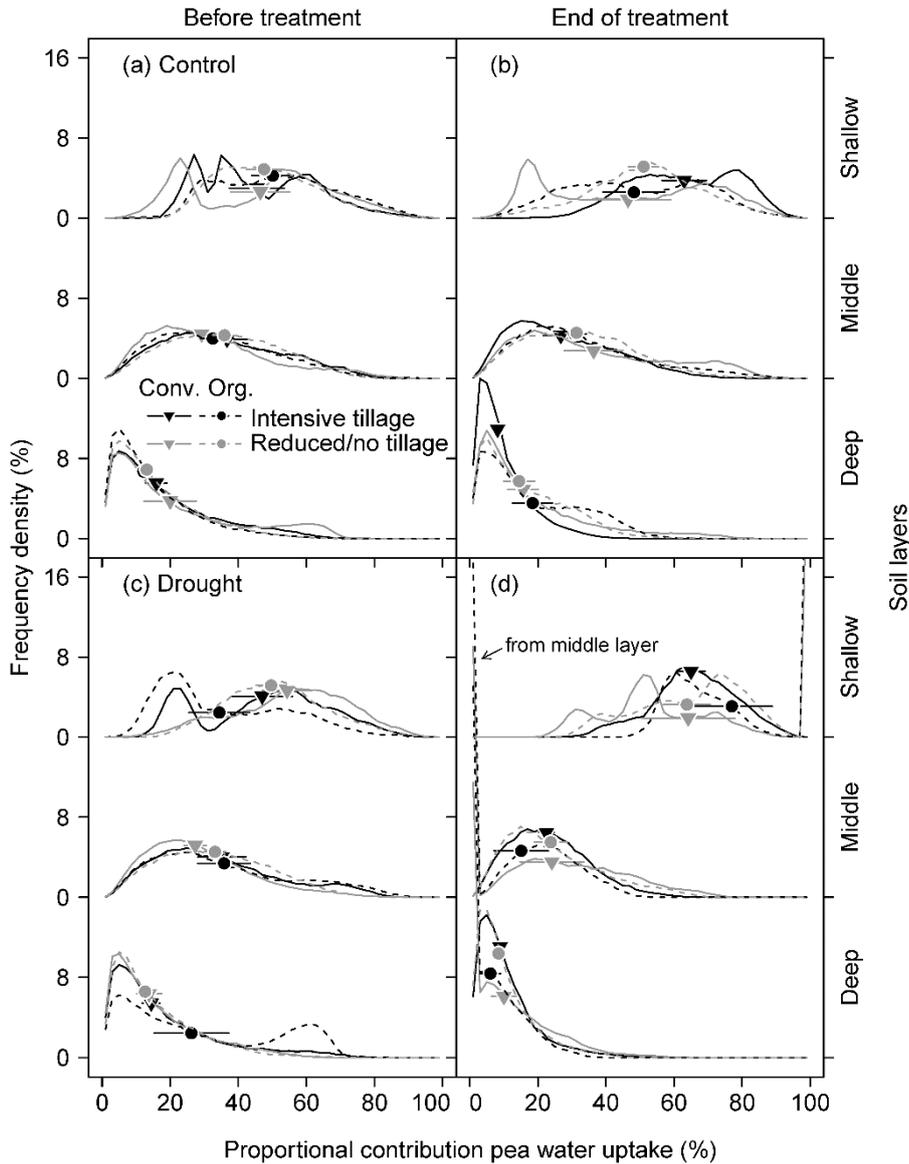


734

735 Fig. 2 Daily mean soil water contents at 10 and 40 cm depth in (a, b) control and (c, d) drought  
 736 subplots under different cropping systems (n = 2 each; Conv. for conventional, Org. for organic).  
 737 Vertical lines indicate sampling dates for stable water isotopes on 7 May, 25 June, and 11 July  
 738 2018. Shaded areas in (c) and (d) represent the drought treatment period from 22 May to 28 June  
 739 2018.



740  
 741 Fig. 3  $\delta^{18}\text{O}$  values of soil water from different depths and plant xylem water in each cropping  
 742 system (a, d) before the drought treatment on 7 May, (b, e) at the end of the drought treatment on 25  
 743 June, and (c, f) after treatment on 11 July in 2018 (Conv. for conventional, Org. for organic).  
 744 Horizontal dotted lines separate isotopic composition of soil and plant samples (P for pea, B for  
 745 barley). Pea plants were already senesced in early July, therefore no stable water isotope data are  
 746 available after the drought treatment. Means and 1 SE (horizontal bars) are given for each cropping  
 747 system (n = 3-4).

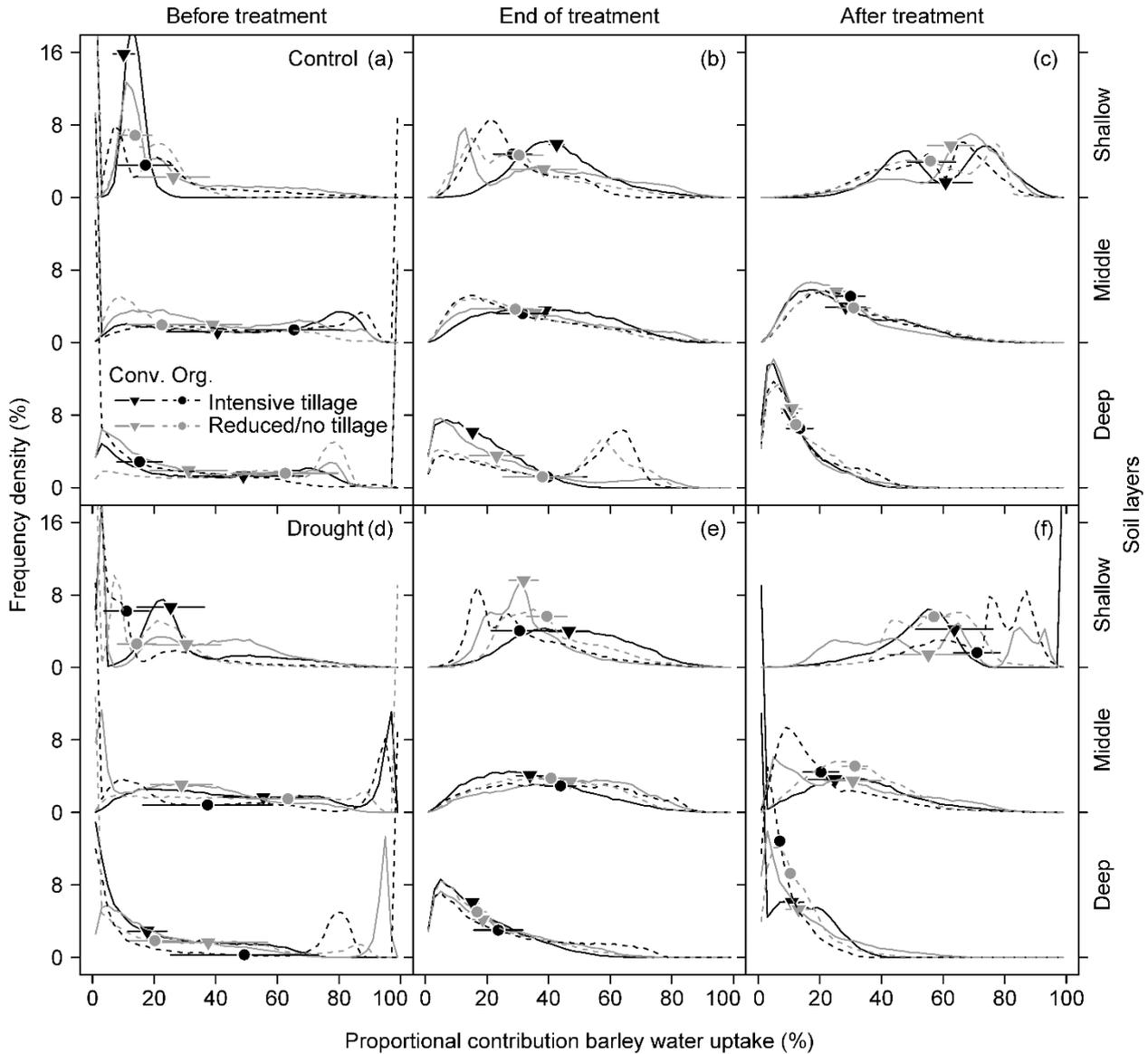


748

749 Fig. 4 Frequency density distribution of model outputs on the proportional contribution of soil water  
 750 to pea water uptake from shallow (0-20 cm, sum of 0-5, 5-10, and 10-20 cm), middle (20-40 cm,  
 751 sum of 20-30 and 30-40 cm), and deep (40-60 cm) soil layers under different cropping systems (a,  
 752 b) before the drought treatment on 7 May and (c, d) at the end of treatment on 25 June in 2018.

753 Frequency density was derived from 10 000 simulations at 2% increment of mixing models using  
 754  $\delta^{18}\text{O}$  for each subplot (Conv. for conventional, Org. for organic). Data were pooled for all subplots  
 755 in each cropping system. Symbols on the curves indicate the median of the model outputs for each  
 756 soil layer. Means and 1 SE (horizontal bars) of each cropping system are given (n = 3-4).

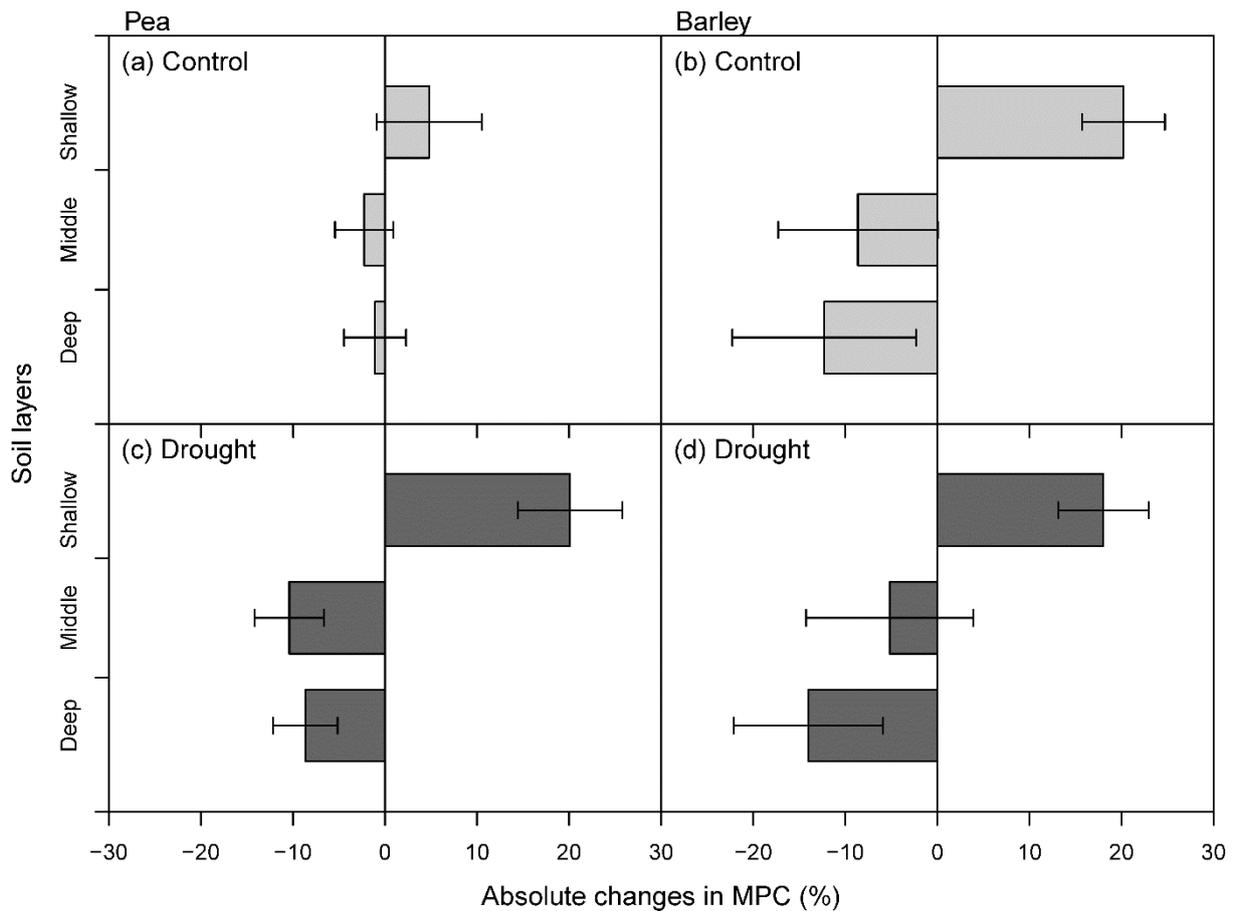
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758

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

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770 Fig. 6 Absolute changes in median proportional contributions (MPC) to plant water uptake of pea  
 771 (a, c) and barley (b, d), calculated as the difference of MPC at the end (25 June; ET) and before the  
 772 drought treatment (7 May; BT), from three soil layers in control (a, b) and drought (c, d) subplots in  
 773 all cropping systems. MPC was derived from 10 000 simulations of mixing models using stable  
 774 water isotope data. Proportional contribution from the shallow layer is the sum of 0-5, 5-10, and 10-  
 775 20 cm depths, the middle layer is the sum of 20-30 and 30-40 cm depths, and the deep layer  
 776 represents 40-60 cm. Means and 1 SE (horizontal lines) are given (n = 14-16).

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