

# Using water isotopes to analyze water uptake during vegetation succession on abandoned cropland on the Loess Plateau, China



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## ABSTRACT

The Chinese government implemented the largest environmental recovery plan in the 1990s, the Grain for Green Project, on the Loess Plateau to prevent soil erosion. Extensive areas of cultivated land were abandoned and then gradually restored with communities of native vegetation. Little is known, however, about the successional development of these communities and their strategies of water use. We collected soil and root samples from four vegetation communities at different stages of succession (*Artemisia capillaris*, *A. sacrorum*, *Bothriochloa ischaemum* and *Lespedeza davurica*) in the dry and wet seasons of 2015 in the Wangmaogou watershed of the Wuding River. Both the root systems and soil-water contents tended to increase with successional development and fluctuated with changes of the dry and wet seasons. Isotopic analysis indicated that the thawing of winter snow during the dry season in April provided sufficient soil water. The vegetation communities only used the water in the 0–20 cm soil layer during the early successional stage. This range increased to 0–100 cm as the succession developed, with strong seasonal variation; water was accessed from deeper soil during the dry season, and water was accessed from shallower soil during the wet season. Antecedent rainfall, soil-water content and root distribution strongly influenced the use of water in all four vegetation communities. In the process of restoration and succession of vegetation communities, the behavior characteristics and water absorption strategies of the root system are the important theoretical basis for optimizing the selection of species and accelerating the speed of ecological restoration in Chinese Loess Plateau.

## 1. Introduction

Water is the main ecological factor restricting vegetation growth, species richness, primary productivity and grassland stability in arid and semi-arid areas (Bai et al., 2004; Bai et al., 2008; Gao et al., 2011; Heras et al., 2011; Porporato et al., 2004). The absorption of water by the soil/root/stem pathway is a major component of the ecological hydrological process (Chen et al., 2017; Liu et al., 2011; Sprenger et al., 2016; Vargas et al., 2017), and hydrological processes determine the evolution direction and ecological function of the soil/vegetation system (Dawson et al., 2002; Evaristo et al., 2016; Grossiord et al., 2017). Roots are the main agents of water and energy flow between vegetation and soil (Dawson, 1996), and their depth and distribution determine the potential source of soil water for the vegetation (Nie et al., 2011; Yang et al., 2015; Zencich et al., 2002). Studying plant roots and soil water is therefore vital for a comprehensive understanding of the ecological and hydrological processes of vegetation restoration and ecological functions and for evaluating the

characteristics of the use of water resources of entire watersheds.

The stable hydrogen and oxygen isotopes are environmental isotopic elements presenting widely in natural waters. Their relative abundance changes in different water bodies can indicate water circulation processes and plant water use, and thus become an important method widely used in water sources, migration and mixing research (Burgess et al., 2000; Dawson et al., 2002; Meinzer et al., 2001). Grubb (1971) first proposed a mechanism for the stable coexistence of woody and herbaceous plants in arid and semi-arid regions. This two-layer model of water use assumed that plants with different root lengths used different water resources and responded differently to pulses of rain. Plant communities absorb water from different soil depths in the summer and winter. Deep-rooted trees and shrubs rely on the stability of deep soil water, and shallow-rooted plants such as herbs use the unstable soil water near the surface (McCole and Stern, 2007; Ward and Getzin, 2013). Liu et al. (2011) measured the water stable isotopic composition and identify a significant correlation between water use and root distribution of shrubs in a subalpine region. The growth

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characteristics and distribution of root systems strongly affect water use. Drought or continuous changes in precipitation patterns can also differentially affect plant-root activity, which leads to the flexibility of plant water use and the long-term adaptation of plants to drought conditions (Xu and Li, 2006). The use of water by plants varies with season and pulses of rain (Chimner and Cooper, 2004; Guo et al., 2016; Mccole and Stern, 2007).

Stable isotopes have been successfully applied to study water sources of forest cultivation and farmland irrigation (Li et al., 2010; Ma and Song, 2016; Wang et al., 2010a) and in subalpine (Liu et al., 2011), forested (Deng et al., 2014; Jia et al., 2013) and cold areas (Hua et al., 2015; Zhao et al., 2013). The source of water used by plants in vegetation restoration in loessial areas, however, has rarely been analyzed. Studies of farmland converted to natural vegetation in loessial areas have mostly focused on the diversity of the vegetation communities (Qin et al., 2009), soil physical (Xiao and Xue, 2014) or chemical (Xu et al., 2017; Zhao et al., 2017) properties, changes of soil water content (Shi et al., 2019; Wang et al., 2019; Xu et al., 2019) and soil microbial communities (Xiao et al., 2016). Vegetation succession on the Loess Plateau in China has been an important measure for repairing fragile ecosystems and improving soil quality, but plants of different ages have different strategies for absorbing soil water (Dodd et al., 1998). The “Grain for Green Project” implemented by the Chinese government in the 1990s changed the landscape pattern of the annual crops on the Loess Plateau. This change increased the depth of water absorption by plant roots and increased the total amount and storage of soil water, thereby changing the regional hydrological processes (Asbjornsen et al., 2007). The study of water use during vegetation succession in this area however has been neglected and should receive more attention.

We analyzed the characteristics of the root systems of four vegetation communities in different seasons on the plateau. We quantified the contribution of water uptake by plants from various soil depths, investigated the mechanism of the response of roots to changes of soil water and modeled the use of soil water by the vegetation using the MixSIAR model. The results of the present research can provide a scientific basis for the study of key issues such as the mechanism of water use and the ecological water demand on the Loess Plateau.

## 2. Materials and methods

### 2.1. Study area

The study area was in the Wangmaogou watershed of Suide County, Shaanxi Province, China (110°20′26″–110°22′46″E, 37°34′13″–37°36′03″N; 940–1180 m a.s.l.) (Fig. 1). The watershed is the first subregion of the hilly and gully region of the Loess Plateau in the middle reaches of the Yellow River. It is a two-branched depression on the left bank of the Wuding River, with an area of 5.97 km<sup>2</sup>. The Suide County Yellow River Soil and Water Conservation Experimental Station of Wangmaogou has been a site for testing the management of small watersheds since 1950 for improving the status of soil erosion on the plateau. The “Grain for Green Project” has replaced crops on slopes with other vegetation, which is now at various stages of succession.

The watershed has a continental monsoon climate, four distinct seasons, an average temperature of 10.2 °C and an average annual precipitation of 513 mm, concentrated mostly in the wet season (July to October), which accounts for about 73.1% of the annual rainfall. The main soil type is loessial soil, with a cover thickness of about 20–30 m. Sampling slope are all located in the main channel on the left bank of the river on the sunny slope.

### 2.2. Description of the sampling sites

Four plant communities were selected based on succession sequence of vegetation on the Loess Plateau (Cheng, 1999): *Artemisia capillaris*

restored for 2 years, *Artemisia sacrorum* restored for 8 years, *Bothriochloa ischaemum* restored for 15 years and *Lespedeza davurica* restored for 21 years (Table 2). The times of restoration of these four plant communities represent different stages of the succession of natural vegetation in this area (Fig. 2) (Wang et al., 2011).

### 2.3. Data collection

Two small automatic meteorological stations (HOBO, U30, USA) were installed in a ditch of the watershed to record atmospheric temperature and rainfall at 5-min intervals for determining the influence of antecedent rainfall on soil water. The amounts of hydrogen and oxygen isotopes in the rainwater and the antecedent precipitation before sampling are shown in Table 1, and the atmospheric temperature and rainfall during the sampling period are shown in Fig. 3. The average accumulated rainfall from April to June was < 50 mm, April to June was regarded as the dry season and July to October was regarded as the wet season.

Root and soil samples were collected on 26 April, 20 May, 3 June, 21 July, 7 August, 20 September, 19 October and 10 November 2015. Two plots were established for each vegetation community, and 3 quadrats (1 m<sup>2</sup> each) were then established in each plot. The collection points and shrubs were separated by the same distances in the *L. davurica* plots to avoid errors caused by different horizontal distances from the stems. Root samples were collected using a root drill 9 cm in diameter with a barrel length of 10 cm from 0 to 20, 20–40, 40–60, 60–80 and 80–100 cm soil layers (Fig. 4a), based on previous reports of the root systems of the vegetation on the Loess Plateau (Chang et al., 2016b; Li et al., 2004).

Different types of vegetation require different methods for collecting samples of water isotopes. The most representative plants of *A. capillaris*, *A. sacrorum* and *B. ischaemum* were selected in each quadrat. The entire root systems were excavated using a spade and shaken to remove the rhizospheric soil, and the roots and aboveground tissues were separated using scissors. The non-green rhizome-binding fraction (Schwinning, 2008) was selected for collecting plant-water isotopes. Selected branches of *L. davurica* near the ground surface were collected and debarked. All branches of *L. davurica* samples were xylem tissues (Barnard et al., 2010; Wu et al., 2015). The plant-water samples were rapidly loaded into 30-ml sampling bottles, which were sealed with film and immediately transferred to a portable refrigerator (Fig. 4b). Root samples for determining root morphology were collected 0–10 cm from the plants collected for plant-water samples using a soil corer. The roots were separated from the soil and sealed in bags. A subsample of the soil was bottled, sealed and stored as a sample of soil-water isotopes. The samples of plant- and soil-water isotopes were transported to the laboratory and stored at 4 °C until extraction. Finally, the remaining soil was packed into an aluminum box for measuring soil-water content (SWC).

### 2.4. Data processing

The root samples were cleaned and then scanned (EPSON, TWAIN PRO, Japan) in 16 bit grayscale mode at a resolution of 300 DPI (Fig. 4c). The root-system analysis program (WinRHIZO, Regent, Canada) was used to analyze the scans, and root length, surface area and diameter were obtained (Fig. 4d). Root-length density (RLD), the ratio of root length to relative soil volume in each soil layer, was then calculated (Li et al., 2015).

Soil water and plant water were extracted and isotopic experiments were conducted at the Hydrological Cycle Laboratory of Xi'an University of Technology. The soil and plant water were separated by vacuum extraction (LI-2000, LUTL, China) (Jia et al., 2012). The hydrogen and oxygen isotopes were quantified using a liquid water isotope analyzer (DLT-100, LGR, USA). The hydrogen and oxygen isotopic ratios were calculated by the ratio of Vienna standard mean ocean-

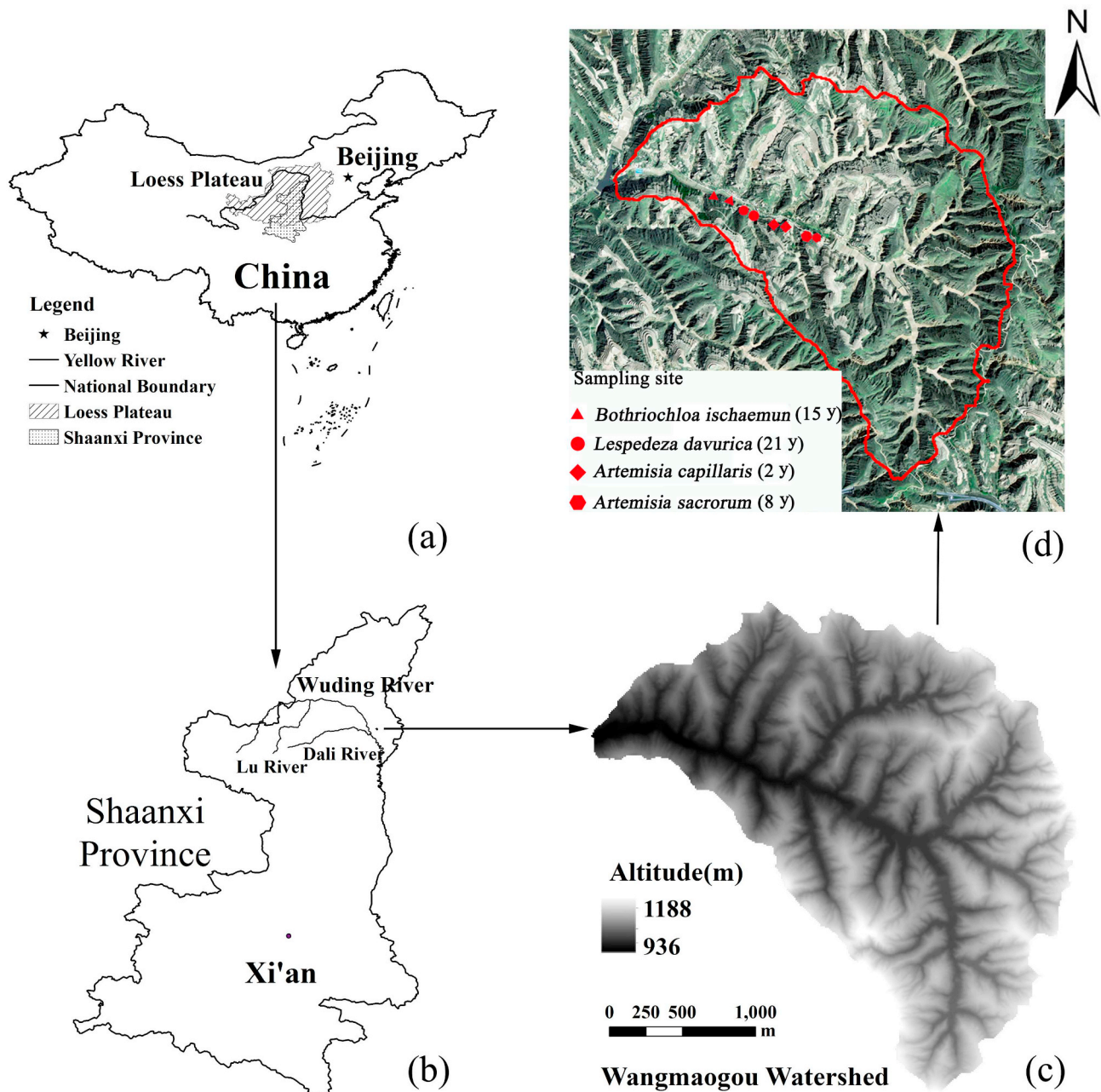


Fig. 1. Study area in Shaanxi province, China (a, b); digital elevation model (c); digital image and sampling sites (d).

water (VSMOW, 0‰), at accuracies of 0.5 and 0.15‰, respectively,  $\delta(\text{‰}) = (R_{\text{sample}} - R_{\text{vsmow}}) / R_{\text{vsmow}} \times 1000$  (1), where  $R_{\text{sample}}$  is the D/H or  $^{18}\text{O}/^{16}\text{O}$  ratio in the water samples, and  $R_{\text{VSMOW}}$  is the D/H or  $^{18}\text{O}/^{16}\text{O}$  ratio of the VSMOW standard water sample. Samples of stable hydrogen and oxygen isotopes in rainwater, snow water, soil water, and plant water were collected from April to November and analyzed (Table 4). A total of 67 rainwater samples, 12 snow-water samples, 706 soil-water samples, and 131 plant-water samples was collected. SWC was calculated by weighing the samples before and after drying at 105 °C,  $\text{SWC}(\%) = (M - M_s) / M_s \times 100$  (2), where  $M$  is soil-water quantity (Kg) and  $M_s$  is soil dry mass (Kg).

## 2.5. The MixSIAR model

The MixSIAR Bayesian mixing model was used to determine sources of soil water used by plant communities. In the experiment, the potential source of water uptake by plant communities was considered to

be soil water at different depths. Groundwater was not considered one of the water resources for plant communities because of the deeply burying (on average 30 m below the sampling site).

In some plant water samples extracted by vacuum distillation, the plant samples evaporate some of the organic volatile substances (insoluble with water, inability to filter) in the process of water distillation. The spectra of methanol-ethanol contaminants with 44 concentration gradients were prepared and calibrated for each contaminated plant water before input to the MixSIAR model. The discrimination values were set to 0 for both  $\delta\text{D}$  and  $\delta^{18}\text{O}$ , mainly because there is no isotope fractionation during plant water uptake.

## 2.6. The local meteoric water line (LMWL) and the global meteoric water line (GMWL)

The LMWL is the linear relationship between  $\delta\text{D}$  and  $\delta^{18}\text{O}$  of rainwater or snow water in a given area and can be compared with the



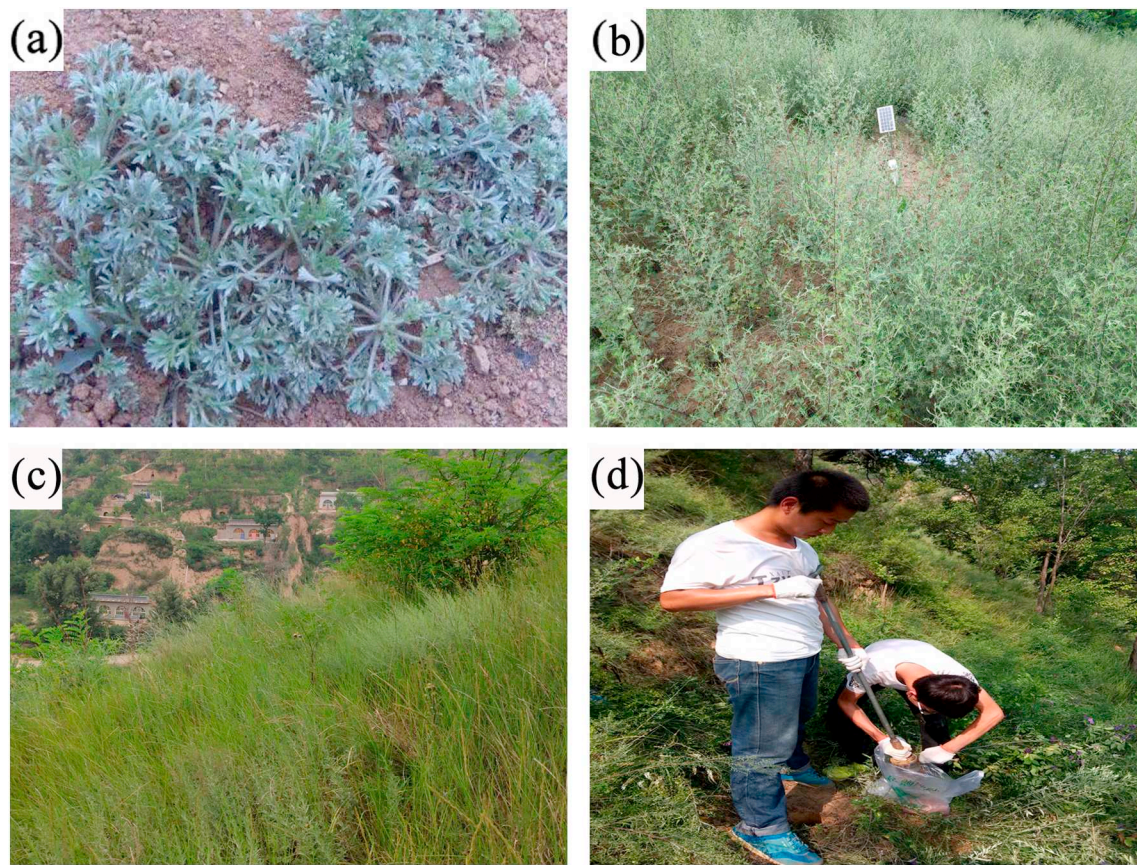


Fig. 2. Sampling site situations, (a) *Artemisia capillaris* (2 y), (b) *Artemisia sacrorum* (8 y), (c) *Bothriochloa ischaemum* (15 y) and (d) *Lespedeza davurica* (21 y).

**Table 1**

Hydrogen and oxygen isotopes of rainwater and precipitation over the study period.

Date	$\delta^{18}\text{O}$					$\delta\text{D}$				
	Antecedent precipitation (mm)	Maximum	Minimum	Average	Standard deviation	Maximum	Minimum	Average	Standard deviation	
15–22 April	0.8	\	\	\	\	\	\	\	\	
23 April					Sampling date					
24 April to 19 May	3.3	\	\	\	\	\	\	\	\	
20 May					Sampling date					
21 May to 2 June	0	\	\	\	\	\	\	\	\	
3 June					Sampling date					
4 June to 20 July	68.6	−8.35	−9.93	−8.95	0.86	−68.64	−69.9	−69.48	0.72	
21 July					Sampling date					
22 July to 6 August	232.2	−7.29	−8.95	−8.06	0.56	−43.85	−62.76	−56.58	6.69	
7 August					Sampling date					
8 August to 19 September	60.6	−3.67	−13.15	−6.8	2.51	−31.47	−97.3	−49.37	17.62	
20 September					Sampling date					
21 September to 18 October	31.6	−7.38	−8.85	−8.23	0.65	−49.17	−58.05	−53.65	3.9	
19 October					Sampling date					
20 October to 9 November	61.0	−4.68	−6.83	−6.06	0.82	−38.55	−48.99	−44.67	3.86	
10 November					Sampling date					

Note: the meaning of “δ” was explained by the formula (1).

global meteoric water line (GMWL,  $\delta\text{D} = 8\delta^{18}\text{O} + 10$ ) to provide an empirical basis for the hydrological cycle of evaporation, condensation, and source of water.

### 3. Results

#### 3.1. Root system and soil-water distribution

The status of root growth in the vegetation communities differed between the stages of succession (Table 3). The RLDs were lower in the

early than the late stages. RLD varied seasonally, e.g. RLD increased in the dry season from April to June, peaked in June at  $20.68 \text{ mm}\cdot\text{cm}^{-3}$ , decreased in the wet season in July and August, reached a minimum of  $3.74 \text{ mm}\cdot\text{cm}^{-3}$  in August and then increased slightly after the wet season. The growth of the root system stagnated at the end of the growing season in October and November. The characteristics of root growth were similar for the 8-y community, but the RLDs increased again in September and November. RLDs in the 15- and 21-y communities were highest in May, at  $17.29$  and  $17.09 \text{ mm}\cdot\text{cm}^{-3}$ , respectively.

RLD varied greatly with depth and season in the 2-y community

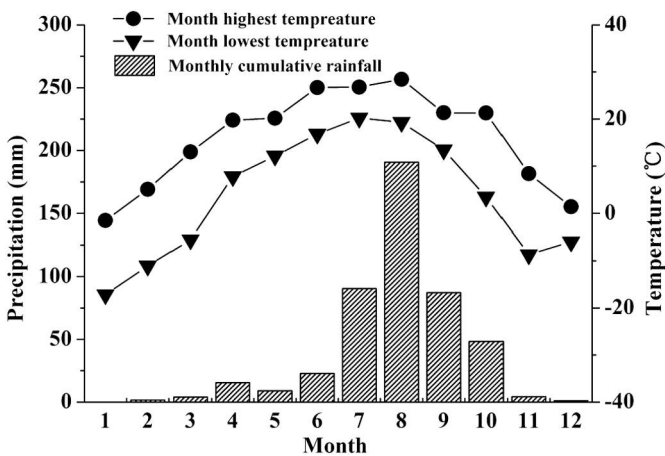


Fig. 3. Distribution of rainfall and temperature in the study area.

(Fig. 5). Roots were mainly distributed in the 0–20 cm layer, and RLD was highest in the 0–20 cm layer in June at  $94.35\text{ mm}\cdot\text{cm}^{-3}$ , which was the highest RLD for the entire study area. RLD was lowest in the 80–100 cm layer in August at  $0.08\text{ mm}\cdot\text{cm}^{-3}$ , which was closely associated with the strategy of root growth and SWC during the wet season (Fig. 6). RLD decreased in the 0–20 cm layer and increased in the 20–100 cm layers in the 8–21-y communities relative to the 2-y communities (Fig. 5). We assumed that the soil layer with > 80% of the total root length was the active layer of the root system, by calculating the percentage of root lengths in each layer. The main root areas of each community were thus 0–20 cm for *A. capillaris* (2 y), 0–35 cm for *A. sacrorum* (8 y), 0–62.5 cm for *B. ischaemum* (15 y) and 0–67.5 cm for *Lespedeza davurica* (21 y), indicating that the root proportions in the deep layers gradually increased with the succession of the vegetation community.

SWC also varied greatly with depth and season (Fig. 6). The 0–20 cm layer of each community was extremely dry, with SWC < 10%, but SWC below 20 cm was sufficient, averaging 20.36% during the dry season in April. The water was due to the thawing of winter ice and snow, and less water was consumed by the vegetation. The SWC of each layer was lowest during the dry season in May and was lowest for all communities in the 0–20 cm layer of the 2-y community at < 5% (Fig. 6). SWC in each layer decreased gradually at the end of the wet season. SWC in the 0–20 cm layer increased slightly during October and November, because rain was abundant, and the plants needed less water at this time, or because root hydraulics were redistributed. SWC tended to gradually increase during the 2–21 years of succession, indicating that the ability of the vegetation communities to store soil water increased with the succession of abandoned farmland.

3.2. Characteristics of isotopic distribution

The isotopic ratios were larger for rainwater than snow water (Table 4).  $\delta^{18}\text{O}$  ranged between  $-13.15$  and  $-1.81\text{‰}$  for rainwater and between  $-19.24$  and  $-10.48\text{‰}$  for snow water.  $\delta\text{D}$  ranged between  $-97.30$  and  $-11.54\text{‰}$  for rainwater and between  $-117.55$  and  $-71.51\text{‰}$  for snow water.  $\delta^{18}\text{O}$  and  $\delta\text{D}$  of the soil-water samples varied with depth, and the mean decreased with depth. Standard deviations also varied consistently, from 3.44 to 0.83 for rainwater and from 17.81 to 4.65 for snow, indicating that the hydrogen and oxygen isotopes at a particular depth began to stabilize over time and that environmental factors greatly affected the soil water near the surface.  $\delta^{18}\text{O}$  was highest in the 0–20 cm layer at 2.88‰. The standard deviations of  $\delta^{18}\text{O}$  and  $\delta\text{D}$  in plant water were within the range of standard deviations for soil-water  $\delta^{18}\text{O}$  and  $\delta\text{D}$ . The standard deviation of  $\delta^{18}\text{O}$  for plant water was 2.36, which was moderate, and the standard deviation of  $\delta\text{D}$  was 18.75, which was large (Table 4).

Table 2  
Status of the experimental plots.

Years of restoration	NOQs	Dominant species	Vegetation type	Altitude (m)	Slope (°)	Coverage (%)	Associated species
2	6	<i>Artemisia capillaris</i>	Semi-shrub, herb	993	29	74	<i>Artemisia scoparia</i> , <i>Sonchus oleraceus</i> , <i>Carduus nutans</i> , <i>Portulaca oleracea</i>
8	6	<i>Artemisia sacrorum</i>	Semi-shrub, herb	1001	28	68	<i>Artemisia capillaris</i> , <i>Poa annua</i> , <i>Cirsium arvense</i> , <i>Lespedeza davurica</i> , <i>Oxytropis bicolor</i>
15	6	<i>Bothriochloa ischaemum</i>	Perennial herb	966	27	76	<i>Poa annua</i> , <i>Lespedeza davurica</i> , <i>Artemisia sacrorum</i> , <i>Ailanthus altissima</i> , <i>Plantago depressa</i>
21	6	<i>Lespedeza davurica</i>	Herbaceous subshrub	971	31	75	<i>Bothriochloa ischaemum</i> , <i>Poa annua</i> , <i>Artemisia sacrorum</i> , <i>Sonchus oleraceus</i> , <i>Carduus nutans</i> , <i>Caragana sinica</i> , <i>Hippophae rhamnoides</i> , <i>Prunus sibirica</i> , <i>Ulmus pumila</i>

Note: NOQs means number of quadrats.

**Table 3**  
Root length density (RLD) means and standard errors ( $\text{mm}\cdot\text{cm}^{-3}$ ).

Month	Years of restoration											
	2			8			15			21		
	Mean	Error	NOFs	Mean	Error	NOFs	Mean	Error	NOFs	Mean	Error	NOFs
April	6.58	4.90	30	11.87	6.47	30	9.46	4.13	30	16.36	4.10	30
May	7.89	5.87	35	11.57	6.91	35	17.29	7.39	35	17.09	5.97	35
June	20.68	18.44	30	16.61	10.99	30	9.09	3.30	30	15.13	4.95	30
July	11.20	8.93	30	10.51	7.28	30	8.07	1.49	30	10.47	2.13	30
August	3.74	2.83	30	4.12	3.01	30	4.85	0.72	60	5.80	1.26	75
September	7.57	5.93	30	9.24	4.40	30	7.43	2.15	30	10.59	2.25	30
October	5.36	3.86	30	4.92	2.31	30	7.18	2.08	30	3.84	1.40	30
November	5.38	4.26	30	12.05	6.69	30	9.81	3.71	30	14.08	4.78	30
Mean		8.55			10.11			9.15			11.67	

Note: NOFs means number of samples.

**Table 4**  
Isotopic ratios of rainwater, snow water, soil water and plant water in the study area.

Water type	Soil layer (cm)	$\delta^{18}\text{O}$ (‰)					$\delta\text{D}$ (‰)				
		Maximum	Minimum	Mean	SD	NOFs	Maximum	Minimum	Mean	SD	NOFs
Rainwater		-1.81	-13.15	-6.49	2.56	67	-11.54	-97.30	-47.35	17.59	67
Snow water		-10.48	-19.24	-12.87	2.73	12	-71.51	-117.55	-86.18	14.47	12
Soil water	0–20	2.88	-13.84	-6.55	3.44	141	-14.03	-95.35	-52.52	17.81	141
	20–40	1.02	-13.26	-7.44	2.83	141	-22.13	-99.25	-60.12	14.70	141
	40–60	0.58	-14.44	-7.93	2.44	142	-21.44	-96.24	-65.14	12.43	142
	60–80	-2.06	-14.33	-8.16	2.20	140	-43.4	-104.53	-65.40	10.80	140
	80–100	-2.77	-14.54	-8.38	2.09	142	-37.66	-119.72	-66.36	10.73	142
Plant water		0.04	-12.12	-6.49	2.36	131	-21.42	-116.06	-61.18	18.75	131

Note: NOFs means number of samples.

**Table 5**  
Variation ranges (%) of soil-water content and contribution in the 0–20 cm layer (dry/wet).

	2 y		8 y		15 y		21 y	
	Mean	NOFs	Mean	NOFs	Mean	NOFs	Mean	NOFs
Soil-water content	5.56/15.01	9/9	7.78/13.97	9/9	6.99/15.62	9/18	5.97/17.09	9/21
Soil-water contribution	52.67/62.32	9/9	9.03/60.32	9/9	8.10/52.06	9/9	3.50/55.84	9/9

Note: NOFs means number of samples.

**Table 6**  
Correlation analysis of influencing factors on shallow sources of water use in the wet season.

Wet season (0–40 cm)	Soil-water contribution	Antecedent rainfall	Soil-water content
Soil-water contribution	1	0.79*	0.81*
Antecedent rainfall	0.79*	1	0.75*
Soil-water content	0.81*	0.75*	1

Note:

\* Indicates a significant correlation at  $P < 0.05$ .

$\delta\text{D}$  and  $\delta^{18}\text{O}$  were correlated with rainfall from June to November, fitting the Local Meteoric Water Line (LMWL:  $\delta\text{D} = 6.57\delta^{18}\text{O} - 4.70$ ;  $R^2 = 0.93$ ,  $n = 67$ ) (Fig. 7). The slope was slightly lower for LMWL than GMWL, and the intercept was much lower than for GMWL, indicating that rainwater had evaporated during rain. The relationship between  $\delta\text{D}$  and  $\delta^{18}\text{O}$  for snow water in January and February in Fig. 5 fitted the Local Meteoric Snow Line (LMSL:  $\delta\text{D} = 5.18\delta^{18}\text{O} - 19.49$ ;  $R^2 = 0.95$ ,  $n = 12$ ) (Fig. 7). The slope and the intercept were lower for LMSL than LMWL, indicating that winter snow evaporated more than

**Table 7**  
Correlation analysis of influencing factors on deep sources of water use in the dry season.

Dry season (40–100 cm)	Soil-water contribution	Antecedent rainfall	Soil-water content
Soil-water contribution	1	-0.79*	0.19
Antecedent rainfall	-0.79*	1	-0.51
Soil-water content	0.19	-0.51	1

Note:

\* Indicates a significant correlation at  $P < 0.05$ .

rainwater. The correlation between  $\delta\text{D}$  and  $\delta^{18}\text{O}$  for soil water in April fitted the equation  $\delta\text{D} = 5.40\delta^{18}\text{O} - 21.70$  ( $R^2 = 0.70$ ,  $n = 48$ ) (Fig. 7). Less rain fell during the dry season in April, but the soil could contain more water below 20 cm depth layer, perhaps due to the melting of snow in winter. A comparison of the snow cover and  $\delta\text{D}$  and  $\delta^{18}\text{O}$  for soil water in April identified a very significant correlation between the stable isotopic ratio of the snow cover and the soil water in April ( $P < 0.01$ ). The slope and intercept of the fitted equation were similar, and  $\delta\text{D}$  and  $\delta^{18}\text{O}$  were larger for soil water than snow water.

The depth at the intersection of the broken and solid lines indicates the main depth of water uptake by plant roots (Fig. 8). This method can approximately indicate the main source of water used by the vegetation. This criterion indicated that the depth of the main water source for the vegetation in the 2-y community throughout the growing season ranged from 0 to 20 cm, except for 50 cm in August (Fig. 8a). The depth of the main water source for the 8-y community was 30–70 cm in the dry season from April to June and 0–20 cm from July to November (Fig. 8b). The depth of the main water source in the 15-y community was 40–80 cm in the dry season from April to June and 0–50 cm from July to November (Fig. 8c). The depth of the main water source in the 21-y community was 30–70 cm in the dry season from April to June and



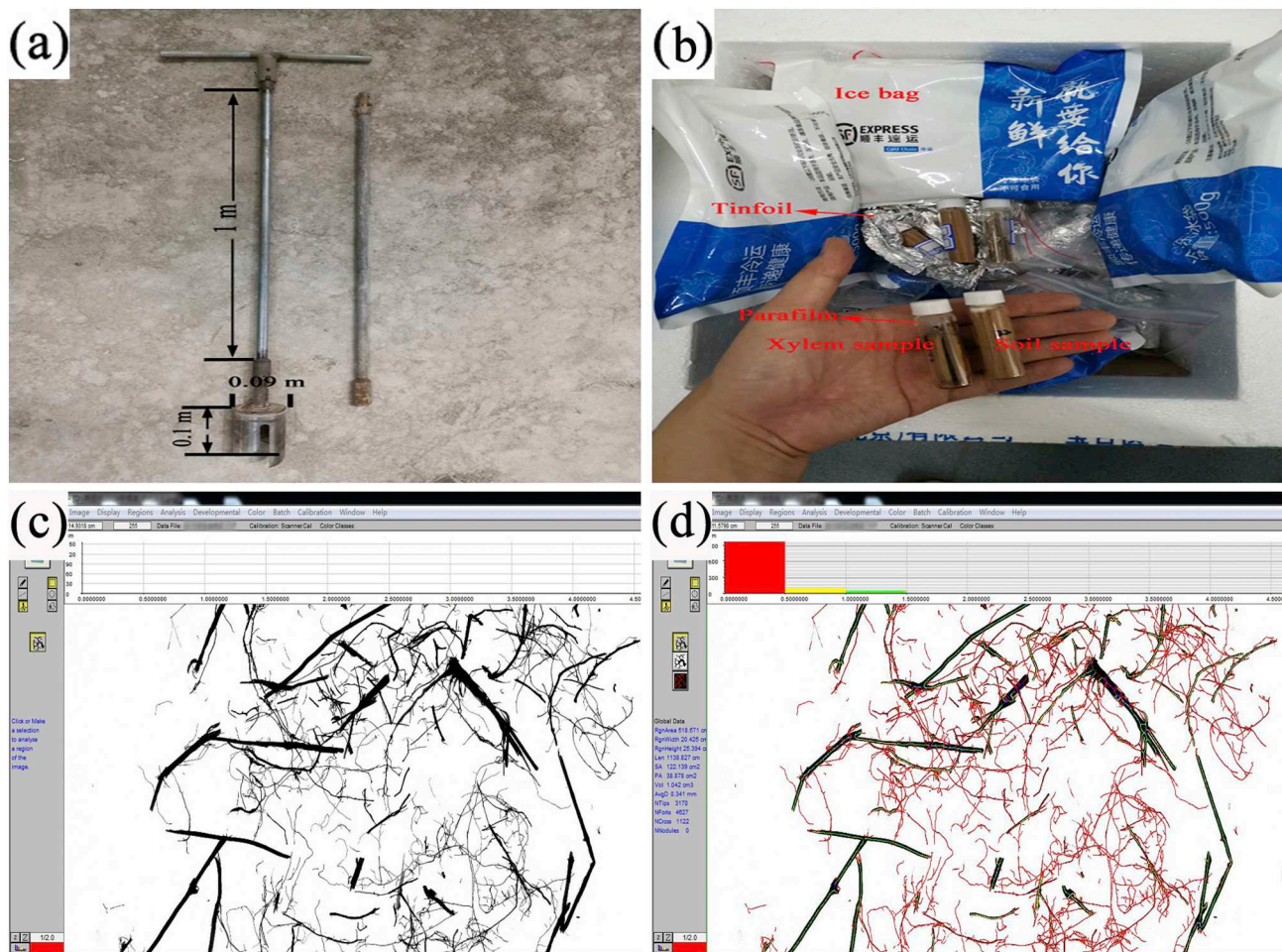


Fig. 4. Related photos of data collection, (a) root drill, (b) storage and transportation of water isotope samples, (c) root scan sample, (d) root system in the analysis process.

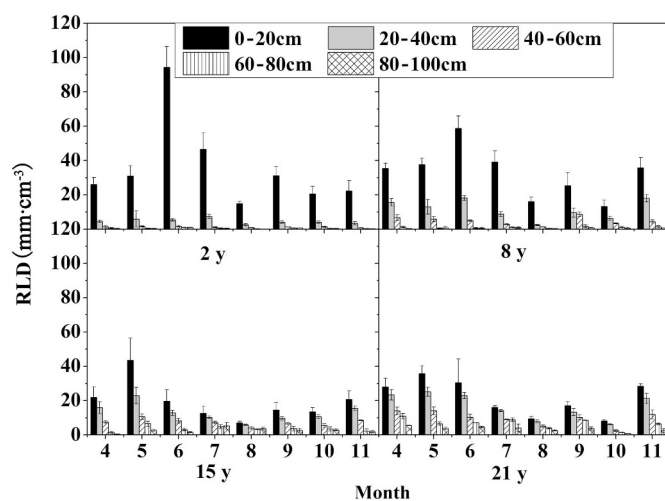


Fig. 5. Vertical distribution of root-length density (RLD) in the vegetation communities.

0–50 cm from July to November. The broken and solid lines had two intersections in April and November. The depths of the main water sources (common intersections) were 30–50 and 20 cm in April and November, respectively (Fig. 8d).

### 3.3. Quantitative analysis of mixing model

Direct contrasts can intuitively identify the depth of the primary water source for plant roots. The specific gravity of water absorption by roots, however, cannot be accurately determined because some figures had multiple intersections. We quantified the contribution of water uptake by plants from different depths using a mixing model to understand the characteristics of soil-water use in different seasons of different vegetation types on the Loess Plateau. The model illustrates the mechanism of response of roots to changes in soil water and the patterns of use of soil-water resources by the plants.

The depth of the main water source was shallowest for the 2-y community (Fig. 9a). Only water from the 0–20 cm layer soil was used from April to November, and the annual average contribution of soil water from this layer was 63.1%. Soil water below 20 cm was used in April, with a contribution of 90.3% (Fig. 9b). The depth of the main water source then shifted downward in May, with more soil water used below 40 cm at a contribution of 72.8%. The depth of water absorption continued to move downward in June, with more soil water used below 60 cm at a contribution of 63.6%. The depth of the main water source returned to 0–20 cm from July to November, with a monthly average contribution of 60.3% (Fig. 9b). Soil water in the 40–80 cm layer was used more frequently from April to June, with a monthly average contribution of 69.2% (Fig. 9c). The monthly contribution of soil water from the 0–20 cm layer was 54% from July to September. There were certain water absorption ratios in each soil layer of 0–100 cm in October, with contributions from the 0–20, 20–40, 40–60, 60–80 and

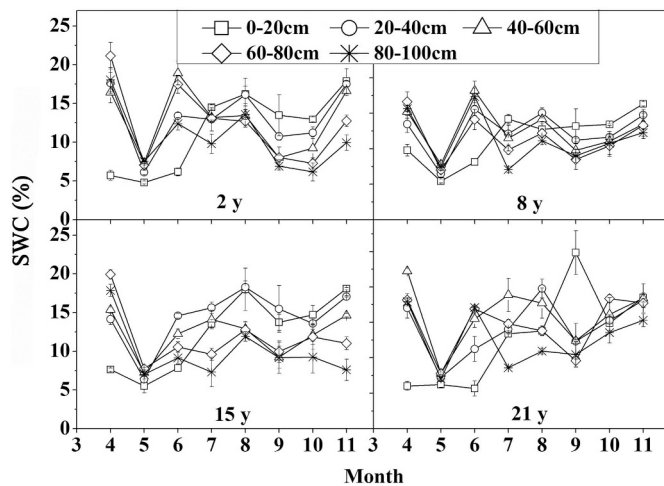


Fig. 6. Seasonal variation of soil-water content in the communities.

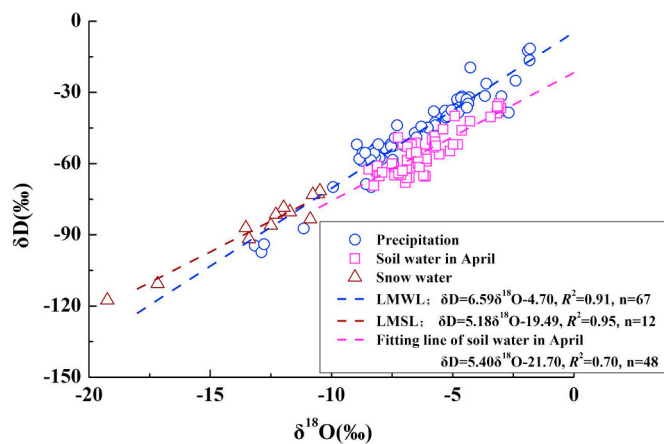


Fig. 7. Relationship between  $\delta D$  and  $\delta^{18}O$  of LMWL, LMSL and soil water in April.

80–100 cm layers of 16.5, 25.5, 20.1, 20.8 and 17.0%, respectively. The depth of the main water source moved upward again to 0–20 cm in November, with a contribution of 81.8% (Fig. 9c). The depth of the main water source was below 20 cm in April, with a soil-water contribution of 96.0% (Fig. 9d). It moved downward to 40 cm in May, with a contribution of 94.6%, and continued to move downward below 60 cm in June, with a contribution of 72.4%. The depth of the main water source then moved upward to the 0–20 cm layer from July to September, with a monthly average contribution of 78.4%. Finally, there were certain water absorption ratios in each soil layer of 0–100 cm from October to November (Fig. 9d).

All four vegetation communities generally used soil water below 20 cm during the dry season in April (Fig. 9). Soil water from melted snow was the main water source for the vegetation (Fig. 7). The depth of the main water source dropped below 40 cm during May and June and moved upward to the 0–40 cm layers during the wet season between July and September. The depth of water absorption gradually extended from the 0–20 cm layer to all layers in October as succession proceeded. The depth of water absorption moved upward again in November to the 0–40 cm layers (Fig. 9).

## 4. Discussion

### 4.1. Effects of root growth and distribution strategies on plant water use

The amount of root system in the dry season is always much more

than that in the wet season on the time scale of plant growth. Plants get more water by proliferating roots in water-poor patches of the soil when the spatial heterogeneity of soil water is evident (Wullschlegel et al., 2001). The lower SWC between May and June allowed the vegetation to allocate more photosynthates to root growth for obtaining scarce soil water (Fig. 5). Soil water in the wet season in July and August was supplied by rainwater (Fig. 3), and the root systems could easily obtain the water needed for growth (Fig. 9). The plants then reduced their allocation of photosynthates to the roots and allocated more to the aboveground tissues. The influence of changes in soil water on root growth from May to August, therefore, indicated the strategies adopted by the communities to adapt to seasonal changes in the soil environment.

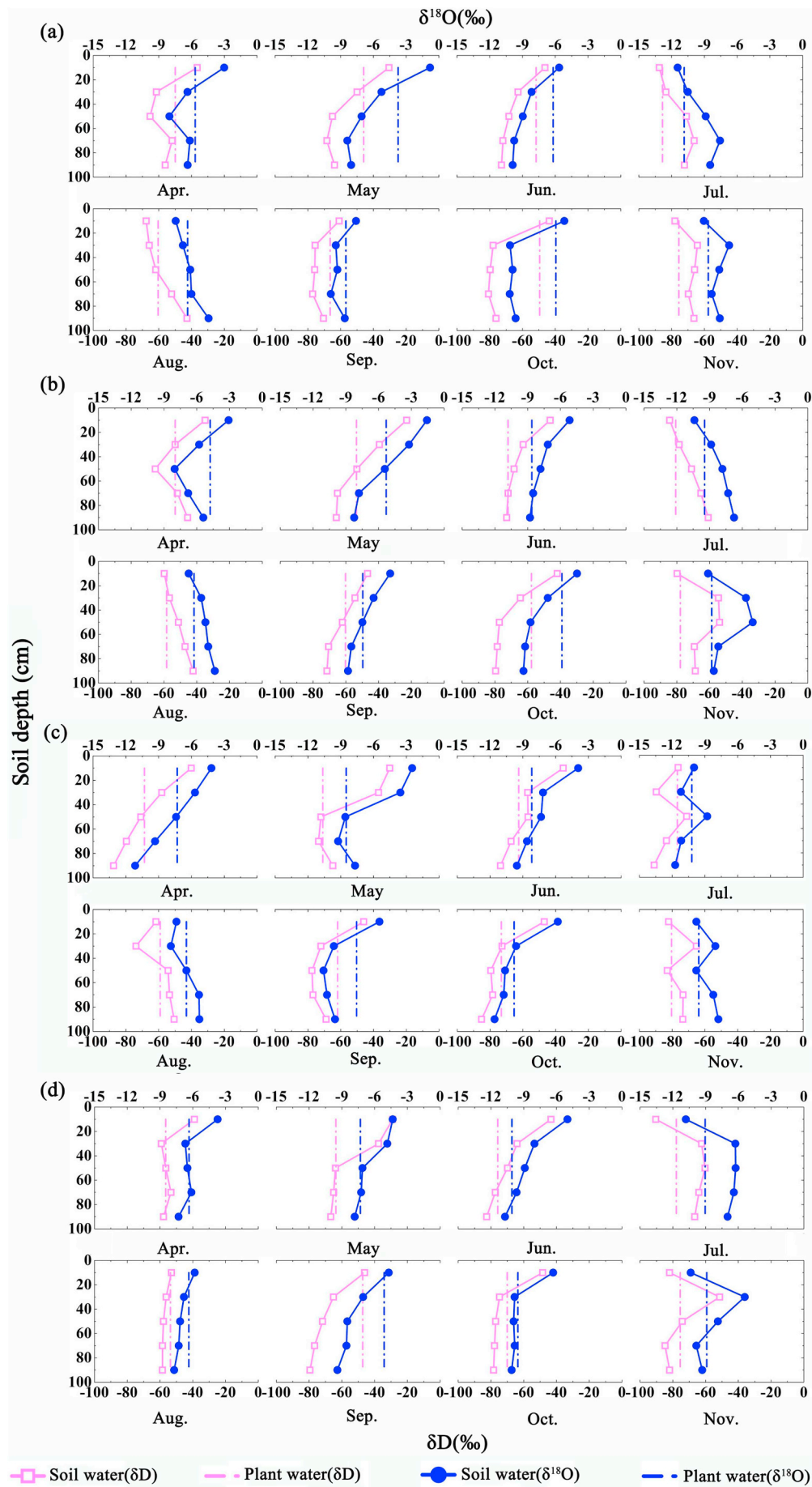
Root length determines the ability of plants to absorb soil water or nutrients (Farrar and Jones, 2000). Root length is more representative than other indicators of the physiological and ecological functions of roots, such as root biomass (Robinson et al., 2003). We, therefore, selected the percentage of root lengths as the internal factor influencing the use of water by plants. We used the percentage of root lengths and soil-water contributions as factors in a correlation analysis to investigate the relationship between root-system characteristics and water use (Fig. 10). Percentage of root length was significantly positively correlated with soil-water contribution in the 2-y community ( $R^2 = 0.86$ ,  $P < 0.01$ ) (Fig. 10). The roots were mainly distributed in the 0–20 cm layer (Fig. 5), accounting for 81.7% of the total root length, the soil-water contributions from the 0–20 cm layer was 63.1% and were consistent with the distribution of root length. They were strongly correlated ( $P < 0.01$ ), with correlation coefficients of 0.93 (2 y), 0.84 (8 y) 0.78 (15 y), 0.67 (21 y). The distribution of roots determines the source of water used by plants, and the distribution of soil water is the key factor affecting root distribution. The distribution of a root system depends mainly on the vertical and horizontal distributions of soil water and the plasticity of root morphology (Du et al., 2010), and roots can detect soil-water gradients and the water characteristics that develop into the moist regions of the soil (Chesson et al., 2004).

The correlation between root length and soil-water contribution gradually decreased as the succession of the plant community developed. The plant roots were sensitive to soil-water responses, indicating good plasticity, in the early stage of succession. This plasticity gradually weakened as the succession developed, with the effect of soil water on root distribution decreasing. Dimorphic root systems may function better in arid and semi-arid areas with shrubs or trees, with lateral roots absorbing shallow soil water in the wet season and taproots absorbing water from deep soil or groundwater in the dry season. Trees and shrubs can thus avoid complete desiccation by reshaping their root morphologies to adapt to seasonal drought (Rossatto et al., 2012). Hao (Hao et al., 2016) reported that species diversity, structure, composition and stability increased, and the use and range of effective resources increased, as succession developed. The depth of water use in our study throughout the growing season was in the ranges of 0–20 cm in the 2-y community, 0–60 cm in the 8-y community, 0–80 cm in the 15-y community and 0–100 cm in the 21-y community. These results indicated that the increase in the effective use of water resources and the range of plants after conversion from farmland on the Loess Plateau were manifested as an increase in the depth of soil-water use (Wang et al., 2017).

### 4.2. Effects of antecedent rainfall and SWC on plant water use

The distribution of precipitation differs between the dry and wet seasons on the Loess Plateau, which leads to seasonal differences of soil-water and root-system distribution. The proportion of water absorbed by plants generally increases as SWC increases (Li et al., 2010). SWC in our study varied greatly in the 0–20 cm layer from the dry to the wet season, increasing from 5.56 to 15.01% for the 2-y community, 7.78 to 13.97% for the 8-y community, 6.99 to 15.62% for the 15-y community





(caption on next page)

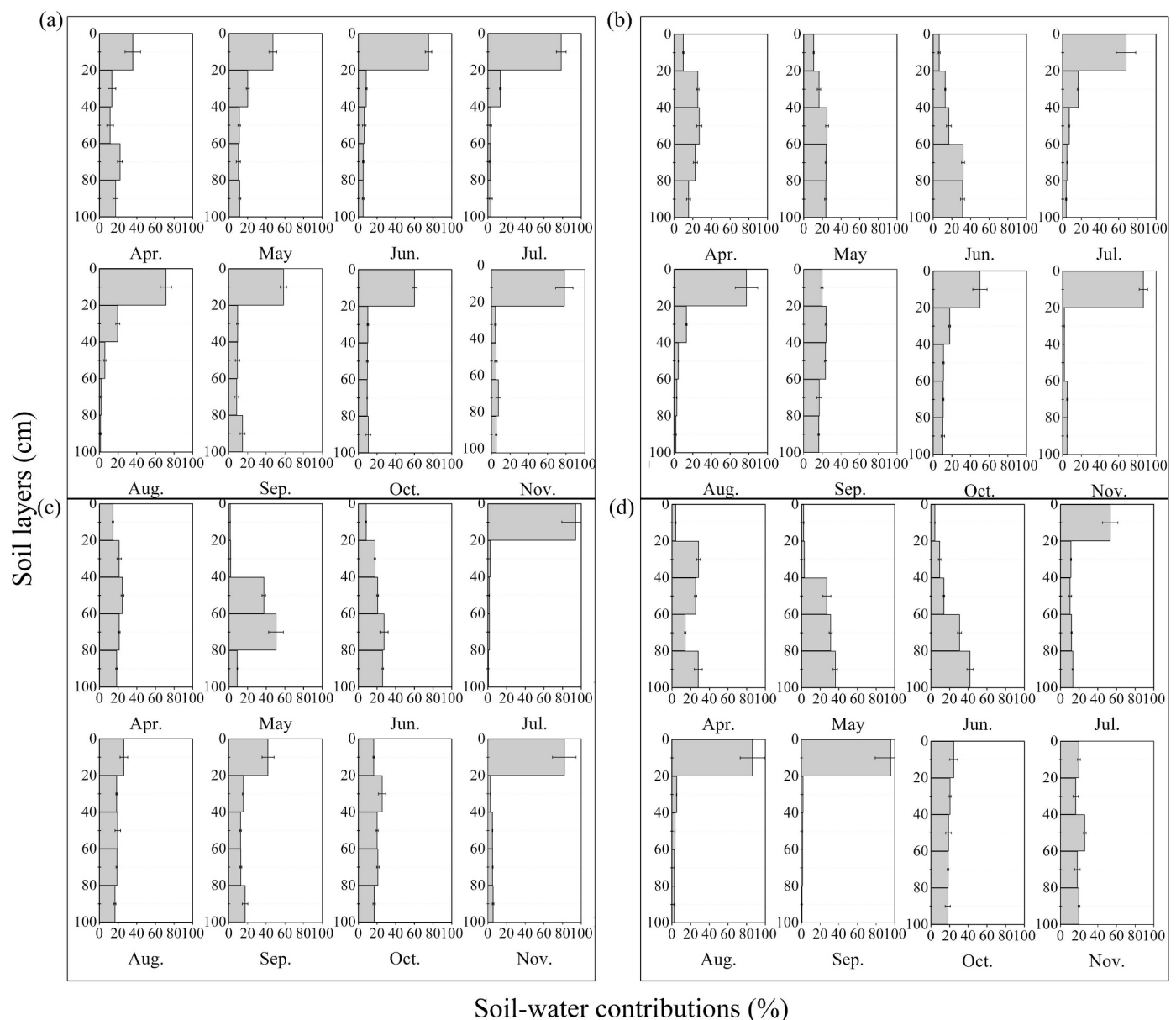
**Fig. 8.** Seasonal variation of  $\delta D$  and  $\delta^{18}O$  of soil and plant water in the profiles for (a) *Artemisia capillaris* (2 y), (b) *Artemisia sacrorum* (8 y), (c) *Bothriochloa ischaemum* (15 y) and (d) *Lespedeza davurica* (21 y).

and 5.97 to 17.09% for the 21-y community, and was similar to the changing trend of the soil-water contribution from this layer (Table 5). The 0–20 cm layer was driest due to the lack of rain, and the plants rapidly integrated more photosynthates into the root system. Thus soil water below 20 cm became the primary source during the dry season. The source of water for vegetation use moved upward to the 0–20 cm layer since the surface 0–20 cm layer is preferentially supplied with soil water in the wet season. Vegetation uses less energy to soak up soil water from the surface layer, absorbing preferentially water stored in the surface layer when it is sufficiently available (Hasselquist and Allen, 2009). The vegetation then allocates fewer resources to the root system and allocates more to the aboveground tissues. Water was more abundant in the 0–80 cm layers since precipitation was unusually frequent in October and November in 2015. The main source of water for the vegetation communities, however, was the 0–20 cm layer, because the

vegetation will soon wither, decreasing the transpiration rate substantially, and root growth will slow sharply, thus greatly reducing the requirement for water, consistent with our results and those of previous studies (Franks et al., 2007; Rose et al., 2003; Zhang et al., 2011).

Antecedent rainfall was also an important factor affecting water use, it was associated with both SWC and water-use contribution. Our results indicated that soil-water sources were divided into shallow layers (0–40 cm) in the wet season and deep layers (40–100 cm) in the dry season. Antecedent rainfall was positively correlated with SWC and water-use contribution in the 0–40 cm layers ( $P < 0.05$ ) (Table 6), indicating that the higher antecedent rainfall, the higher the SWC and soil-water contribution of the 0–40 cm layers during the wet season.

Antecedent rainfall was not significantly correlated with SWC in the 40–100 cm layers ( $P > 0.05$ ) (Table 7). Antecedent rainfall in the dry season did not affect SWC below 40 cm, indicating that rainfall could



**Fig. 9.** Seasonal changes of water uptake by plant roots in the soil profiles for (a) *Artemisia capillaris* (2 y), (b) *Artemisia sacrorum* (8 y), (c) *Bothriochloa ischaemum* (15 y) and (d) *Lespedeza davurica* (21 y).

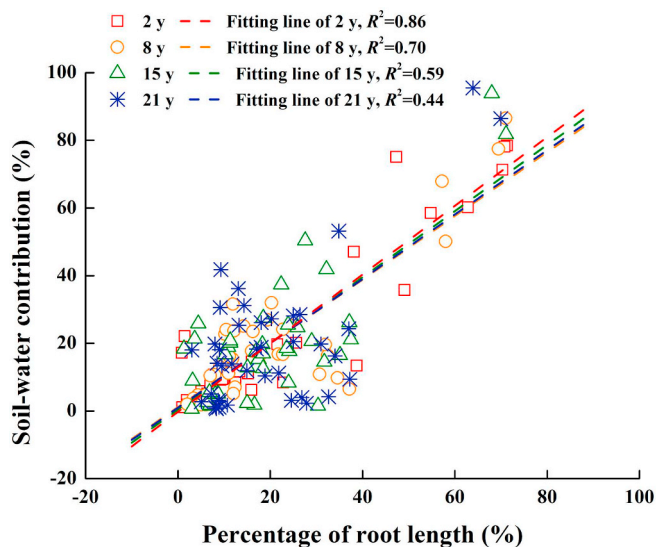


Fig. 10. Linear relationship between root length and water uptake.

not recharge the soil below 40 cm in the dry season. The antecedent rainfall was negatively correlated with the contribution of deep water use ( $P < 0.05$ ), indicating that the higher the antecedent rainfall, the lower the proportion of plants able to absorb the water in deep soil. Plant water in the dry season was sourced mainly from the soil below 40 cm, due to less rainfall, extreme drought and the difficulty in meeting the requirements of plant growth. Thus, the plant roots obtained water from deeper soil. The water in the surface soil, however, was slightly recharged by rain in the dry season. Plants preferentially used the water from the surface layer, and water-use efficiency was lower below 40 cm. The antecedent rainfall in the dry season was therefore negatively correlated with the soil-water contribution, as expected.

#### 4.3. Implications for the relationship between vegetation restoration and soil-water resources

The Loess Plateau has deep soil and a low groundwater level. Soil water from natural precipitation is the only water source of the hillside plants. Vegetation restoration can have negative effects on soil-water resources if water demand is higher than supply. The relationship between vegetation restoration and soil-water resources therefore has many contradictions (Chen et al., 2015; Jian et al., 2015; Wang et al., 2010b). The transformation after the “Grain for Green Project” on a landscape scale on the Loess Plateau has changed the depth of absorption of soil water by hillside plants and has thus changed the original ecological hydrological cycle (Asbjornsen et al., 2007). The depth of soil water absorbed by roots, however, is increasing with the succession of the plant communities. Some trees and shrubs, especially artificially restored forests, may overuse deep soil water (Chang et al., 2016a; Jian et al., 2015). Further study of multiple sampling periods and the interannual changes of water use by plants, and analyzing artificial forests for comparing the advantages and disadvantages of the two restoration methods on the ecological construction on the Chinese Loess Plateau, are needed to resolve these problems. This information will help us to understand the effects of vegetation restoration on regional water resources and the coupling and feedback between ecological vegetation processes and slope hydrological processes.

## 5. Conclusions

We analyzed the dynamic characteristics of roots and soil water, elucidated the isotopic characteristics and quantified the contributions

of plant-water sources on the Chinese Loess Plateau. Plant-root density and SWC increased slightly with the development of vegetation succession. The fitted line of soil-water isotopes in April was similar to that of winter snow, and the isotopic compositions varied more for shallow than deep soil water. The effective depth of the vertical distribution of plant roots gradually increased from 0 to 30 to 0–67.5 cm with the development of vegetation succession, the range of soil water that could be absorbed increased from 0 to 20 to 0–100 cm and varied with season, indicating that water was absorbed from deeper soil in the dry season and from surface soil in the wet season. This seasonal change was affected by plant-root length and antecedent precipitation. Our results are important for vegetation restoration in this arid and semi-arid ecosystem. They provide a scientific basis for the study of the influence of vegetation on the ecological hydrological process of the Wangmaogou watershed and can be used to optimize vegetation collocation in the “Grain for Green Project” on the Chinese Loess Plateau.

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