Water sources of major plant species along a strong climatic gradient in the inland Heihe River Basin

Liangju Zhao [a, b*], Cong Xie [a], Xiaohong Liu [c], Ninglian Wang [a], Zhang Yu [c], Xiying Dong [a], Lixin Wang [d*]

[a] Shaanxi Key Laboratory of Earth Surface System and Environmental Carrying Capacity, College of Urban and Environmental Sciences, Northwest University, Xi'an 710069, China

[b] State Key Laboratory of Continental Dynamics, Northwest University, Xi'an 710069, China

[c] School of Geography and Tourism, Shaanxi Normal University, Xi'an 710119, China

[d] Department of Earth Sciences, Indiana University–Purdue University Indianapolis (IUPUI), Indianapolis, IN 46202, USA

Running title: Plant water use strategy along climate gradient

***Corresponding author**

Dr. Liangju Zhao

College of Urban and Environmental Sciences

Northwest University

Xi'an, Shann'xi, 710069, China

E-mail: lzb.ac.cn

Cell phone: 15934832330

Dr. Lixin Wang

Department of Earth Sciences

Indiana University-Purdue University Indianapolis

Indianapolis, IN, 46202, USA

E-mail: lxwang@iupui.edu

Office phone number: 317-274-7764

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ABSTRACT

Aim Knowledge on vegetation water sources is crucial to understand the ecohydrological processes and ecological management of arid and semi-arid ecosystems. The identification and quantification of plant water uptake from precipitation, soil and groundwater remain challenging along large climatic gradient.

*Method*s Stable oxygen isotope compositions of xylem water, soil water and groundwater were analyzed to assess seasonal and spatial patterns of water uptake of 11 major plant species along the Heihe River Basin.

Conclusions In the upper reaches, soil water recharged by precipitation was the main plant water source, and plants extracted water from the shallow soil water in wet season while used more deep soil water in dry season. In the middle reaches of desert-oasis ecotone, the water sources of shrubs shifted between soil moisture and groundwater depending on variations of precipitation and groundwater level, while shrubs at Gobi relied on deep soil water and shallow soil water after rainfall. In the lower reaches, the driest part of the region, groundwater and deep soil water were main water sources for the riparian plants. Groundwater was stable water source for shrubs growing on the planted shrubland, and soil water was stable water sources for shrubs growing at Gobi. Our results also revealed that water use strategies of the same species were plastic under different groundwater level and precipitation. This study identified water use patterns of different plant species along a climatic gradient and provided scientific implication for water management of different ecosystems of the arid and semi-arid ecosystems.

Keywords Groundwater · Heihe River Basin · oxygen isotope · hydrogen isotope · plant water source · soil water

Introduction

Water is one of the most important factors affecting plant survival and growth in arid and semiarid regions, where evaporative demand exceeds precipitation and water resources are scarce (Reynolds et al., 2007; Wang et al., 2010; Wang et al., 2012). Depending on their belowground rooting system and habitat, different plants may use different water supplies such as precipitation/snow (winter), groundwater, river and soil water (both shallow and deep). Plants can also shift their water supply depending on seasonality as plants often depend on access to deep and moist soil layers to withstand heat waves and droughts (e.g., Eggemeyer et al., 2009; Rossatto et al., 2012; Schwinning et al., 2005). Therefore, understanding on different water sources in water-limited areas is important to maintain the structure and function of these largely diverse arid and semi-arid ecosystems covering forest, grasslands, shrublands, mountain meadows, desert-oasis ecotones, riparian forest and Gobi of the Heihe River Basin.

Studies have shown that different water sources often have different hydrogen and oxygen isotopic compositions (δ^2H, $\delta^{18}O$). Therefore, the isotopic composition of plant tissue water can be an effective proxy to determine plant potential water sources (Ehleringer et al., 1998; Lanning et al., 2020), which can be different along a climatic gradient. To understand different plant water sources and areas with effective water uptake, we used the Heihe River Basin, the second largest inland water basin in northwestern China. The area covers semi-arid region in the upper reaches, arid region in the middle reaches, and extremely arid region in the lower reaches. We examined the stable isotope composition of different water pools that support different plant species especially in the lower and middle reaches, where water has become a limiting factor for vegetative growth. Water uptake patterns of *Tamarix ramosissima* (Sun et al., 2016), *Haloxylon ammodendron* (Zhou et al., 2017) in the middle reaches, of

Populus euphratica, *Tamarix ramosissima*, *Sophora alopecuroides*, *Sonchus oleraceus* and *Herba Taraxaci* (Zhao et al., 2008; Ruan et al., 2014) and *Populus euphratica*, *Taramrix chinensis* and *Reaumuria soongorica* (Fu et al., 2014) in the lower reaches have been studied. These plants may have variations in their belowground rooting strategies, which are critical for plant water access. To our knowledge, at basin scale, there have been no systematic studies on seasonal and annual variations of water uptake patterns across strong climate gradient.

Based on stable oxygen and hydrogen isotopic composition ($\delta^{18}O$ and $\delta^{2}H$) of precipitation, soil water and shallow groundwater of the Heihe River Basin, we investigated the recharge sources of shallow groundwater and soil water of these ecosystems. In addition, we used variations of $\delta^{18}O$ of soil water, groundwater and plant xylem water in 11 different plant species in 15 sites, including trees, shrubs and grasses to reveal their water sources and areas with effective water uptake along a climatic gradient of the Heihe River Basin (acronyms are given in Table 1). The information on the mechanisms of plant water use and the strategies of adapting to arid environments will be useful in selecting the adaptive species when restoring and rebuilding degraded desert ecosystems and maintaining their stability.

Table 1 here

Materials and Methods

Study sites

The study took place at the upper reaches (Qilian Mountains), the middle reaches (Linze) and the lower reaches (Ejina) with distinct climatic conditions within the Heihe River Basin, northwestern China (Table 2 and Fig. 1).

Fig. 1 + Table 2 here

We used Pailugou and Yeniugou to represent middle mountains and alpine region of the Qilian Mountains. In Yeniugou, the long-term (1959-2000) mean annual precipitation is 401.4 mm, 80% of it occurs between June and September. The annual mean temperature is about -3.1°C, with the lowest monthly mean temperature being recorded in January (-17.2°C) and the highest monthly mean temperature in July (9.2ºC). The temperature is above 0°C from May to September. Meanwhile, the highest and the lowest temperature in Pailugou, which is located in the middle of the Qilian Mountains, are 12.2°C (July) and -12.9°C (January), respectively. The area has a mean annual temperature of 0.7°C. Annual precipitation averages 369.2 mm. Both areas have similar ecosystem types consisting of mountain grasslands, mountain meadows, high mountain meadows, swamp meadows and forests.

Due to the absence of long-term climatic data in Linze (middle reaches), we used climatic data in Zhangye, which is about 60 km from Linze. The long-term (1951- 2012) mean annual temperature is about 7.3°C, with a mean January temperature of -9.8°C and a mean July temperature of 21.8°C. Mean annual precipitation is 129.9 $mm \cdot year^{-1}$, with 73.7% of the rainfall occurring between June and September. The main ecosystem types are planted oasis, desert-oasis ecotone and Gobi Desert in the middle reaches.

In the lower reaches (Ejina), the long-term (1960-2012) mean annual temperature is 8.9 °C, with a mean January temperature of -11.5 °C and a mean July temperature of 27.0 °C. Mean annual precipitation is 34.9 $mm \cdot year^{-1}$, with 74.3 % of the rainfall occurring between June and September. Ejina is considered one of the driest regions in China. The main ecosystem types are riparian forest, planted shrubland and Gobi in the lower reaches.

Field sampling

Between 2007 and 2012, annual field sampling was conducted during the growing seasons in each region. In the upper reaches, sampling was conducted in June 2009 and 2011, August 2007, 2009 and 2012 and September 2011. In the middle reaches, sampling was in June 2010 and August 2012, and in June 2007 and 2010, August 2008, 2009, and August 2012 in the lower reaches of the Heihe River Basin. The detailed sampling information is shown in Table 2.

Ten different ecosystems along the Heihe River Basin were selected (Fig. 1; Table 2). Nine sites were selected at mountain grassland (U1), swamp meadow (U2 and U4), mountain meadow (U3) and Qinghai spruce forest (from U5 to U9-9) at the upper reaches; four sites were selected at desert-oasis ecotone (from M1-10 to M3-12) and Gobi (M4-10 and M4-12) at the middle reaches; and six sites were selected at riparian forest (from L1-07 to L4-08), planted shrubland (L5-10 and L5-12) and Gobi (L6-10 and L6-12:) at the lower reaches, respectively (Fig. 1; Table 2).

Plant sampling

In the upper reaches: the dominated plants were *Stipa capillata* at U1 site, *Polygonum viviparum* at U2 site, *Stipa purpurea* at U3 site and *Stipa capillata* at U4 site. The root samples of *Stipa capillata*, *Polygonum viviparum*, and *Stipa purpurea* were taken from above sites. In the Qinghai spruce forest, the dominated plants were Qinghai spruce and *Potentilla fruticosa* at U5 site, Qinghai spruce at U6, U7 site and U8-12 site, Qinghai spruce and *Stipa capillata* at U8-11, and *Qinghai spruce*, *Potentilla fruticosa* and *Polygonum viviparum* at U9-6 and U9-9. The root samples of *Stipa capillata* and stem samples of Qinghai spruce and *Potentilla fruticosa* were taken from the above sites. At U7 site, samples were taken from 5 pm on July 31 to 10 pm on August 2 2009 with 2 h interval for Qinghai spruce stem. At U9-6 site, samples were taken from 6 am on June 23 to 8 am on June 24 and from 6 am to 8 pm on June 25, 2011 with 1 h interval for Qinghai spruce and *Potentilla fruticosa* stem and with 2 h interval

for *Polygonum viviparum* root. At U9-9 site, samples were taken from 6 am to 10 pm on September 2 and from 8 am on September 6 to 5 pm 8 2011 with 1h interval for Qinghai spruce and *Potentilla fruticosa* stem and with 2h interval for *Polygonum viviparum* root. At U8-11 site, plant samples were taken from 6 am on June 27 to 6 pm on June 28 2011 with 1h interval for Qinghai spruce stem and with 2h interval for *Stipa capillata* root (Table 2).

In the middle reaches: The dominated plants were *Tamarix ramosissima* at M1-10 and M1-12, *Haloxylon ammodendron* at M2-10, M2-12, and M3-12 at the desert-oasis ecotone. At Gobi, the dominated plants were *Reaumuria soongorica* and *Nitraria tangutorum* at M4-10 and *Reaumuria soongorica* at M4-12. The stem samples *Tamarix ramosissima*, *Haloxylon ammodendron*, *Reaumuria soongorica* and *Nitraria tangutorum* were taken from above sites. Especially, in M1-10 and M2-10 sites, stem samples of *Tamarix ramosissima* and *Haloxylon ammodendron* were taken from 6 am on June 15 to 6 am on June 16 2010 with 2h interval. In M4-10, stem samples of *Reaumuria soongorica* and *Nitraria tangutorum* were taken from 6 am on June 18 to 6 am on June 19 2010 with 2 h interval (Table 2).

In the lower reaches: The dominated plants were *Populus euphratica* and *Sophora alopecuroides* at L1-07, L1-09 and L1-10, *Populus euphratica* at L1-12, *Tamarix ramosissima* at L2-10 and L2-12, and *Populus euphratica*, *Tamarix ramosissima* and *Sophora alopecuroides* at L3-08 and L4-08 in the riparian forest. The dominated plant was *Haloxylon ammodendron* at L5-10 and L5-12 at the planted shrubland, and the dominated plant was *Reaumuria soongorica* at L6-10 and L6-12 at Gobi. The root samples of *Sophora alopecuroides* and stem samples of *Populus euphratica*, *Tamarix ramosissima*, *Haloxylon ammodendron* and *Reaumuria soongorica* were taken from above sites. At L9-09 site, stem samples of *Populus euphratica* and root samples of *Sophora alopecuroides* from August 6 6am to

August 9 2pm 2009 with 2h interval were taken with three replicates. In both L1-10 and L2-10, stem samples of *Populus euphratica* and *Tamarix ramosissima* were taken from 6 am on June 21 to 6 am on June 22 2010 with 2h interval, and root samples of *Sophora alopecuroides* were also taken with three replicates. In the L5-10, stem samples of *Haloxylon ammodendron* were taken from 6 am on June 23 to 6 am on June 24 2010 with 2h interval. At Gobi (L6-10), stem samples of *Reaumuria soongorica* were taken from 6 am on June 26 to 10 am on June 27 2010 with 2h interval. In both L3-08 and L4-08, root samples of *Sophora alopecuroides* and stem samples of *Populus euphratica* and *Tamarix ramosissima* were taken from 5 am to 9 pm on August 20 2008 with 2h interval (Table 2).

For plant samples, two bottles with 8 ml root samples from 10 to 15 herbaceous plants, 4 to 6 shrub plants and 3 to 4 woody plants around one soil profile were selected to extract water and measure δ^2H and $\delta^{18}O$. The sampling date, species and plant parts are listed in Table 2.

Soil and groundwater sampling

In the upper reaches: In June 2009, soil samples in 5, 10, 30 and 50 cm deep at U1 and U2, in 5, 10 and 20 cm deep at U3 and U4, and in 10, 30 and 70 cm at U6 were taken in June 2009. At U5, soil samples in 5, 10, 15, 20, 40 and 60 cm deep, and at U7, in 5 cm and from 10 to 60 cm with 10 cm increment were taken in August 2007 and 2009, respectively. At U8-11, soil samples in 3 and 5 cm, as well as from 10 to 60 cm with 10 cm increment, followed by 80, 100 and 120 cm were collected in June 2011. At U8-12, soil samples in 5 cm and from 10 to 160 cm with 20 cm increment were collected in August 2012. At both L9-6 and L9-9 sites, soil samples in 3, 5, 10, 15, 20, 40, 60, 80 and 90 cm of soil profile were collected in June and September 2011.

In the middle reaches: In the desert-oasis ecotone, soil samples in 10, 40, 70, 100, and 130 cm,

and from 160 to 220 cm with 20 cm increment in the soil profile were taken at M1-10 in June 2010. In August 2012, soil samples in 5, 10, and from 20 to 300 cm with 20 cm increment in the soil profile were taken at M1-12. At M2-10, soil samples in 10, 40, 60, 70, 100, 150 and 200 cm were taken in June 2010. In August 2012, soil samples in 5 and 10 cm, and from 20 to 300 cm with 20 cm increment at M2-12, and in 5 and 10 cm, and from 20 to 260 cm with 20 cm interval at M2-13 were taken, respectively. At Gobi site, soil samples in 10, 20, 40, and 50 cm, and from 80 to 200 cm with 20 cm increment at M4-10 were taken in June 2010. At M4-12 site, soil samples in 10, 15, 20, 25, from 30 to 80 cm with 10 cm interval, 100, 110, and from 120 to 200 cm with 20 cm increment were taken in August 2012.

In the lower reaches: At the riparian forest of the lower reaches of the Heihe River Basin, at L1-07, soil samples in 20 to 160 cm deep with 20 cm increment and saturated layer in the soil profile were taken in June 2007. Groundwater was also sampled and the depth of groundwater table at this site was 160 cm. At L1-09, groundwater and soil samples of 5, 8, 10, 30 cm, and from 40 to 160 cm with 20 cm interval in the soil profile were taken in August 2009. At this site, the depth of groundwater table was almost 160 cm. At L1-10, groundwater table depth was almost 180 cm and soil samples were taken from 20 to 180 cm deep with 20 cm increment in June 2010. At L1-12, groundwater at nearly 200 cm deep was sampled, followed by soil sample collection from the following depths: 5, 10 cm, and from 20 to 200 cm with 20 cm interval in the soil profile in August 2012. At L2-10 and L2-12 sites, groundwater (200 cm depth) and soil samples from 20 to 200 cm with 20 cm interval and in 5, 10 cm, and from 20 to 160 cm with 20 cm interval were taken in June 2010 and August 2012, respectively. At L3-08 and L4-08, soil samples of 3, 5 cm, and from 20 to 240 cm with 20 cm interval in the soil profile were taken in August 2008. At these sites, the groundwater table was deeper than 5.0 m. At the planted

shrubland site (L5-10), soil samples from 20, 40, 50, 60, 80, 100, 140, 170, 200, 230 and 250 cm deep were taken in June 2010. Soil samples were taken from 20 to 160 cm with a 20 cm interval, in addition to surface soils (5 and 10 cm) and deep soils (165, 180, 200 and 220 cm) in August 2012 (L5-12). Groundwater tables were 250 and 220 cm at L5-10 and L5-12, respectively; samples of groundwater were taken simultaneously with the soil samples. At the Gobi sites (L6-10), soil samples were taken from 20 to 160 cm deep with 20 cm interval, as well as from 175, 180, 185, 200, 220 and 255 cm soil layers in June 2010. Soil samples from 5 and 10 cm, and from 20 to 300 cm with 20 cm interval were taken in August 2012 (L6-12). Groundwater table was deeper than 5.0 m in both L6-10 and L6-12. All soil samples were put in glass containers and were sealed immediately with Parafilm. To measure soil gravimetric water content (w/w %), 20 ml of soil samples from all soil layers with three glass bottles were used. In addition, two glass bottles, each contained 8 ml soil sample, were used to measure δ^2H and $\delta^{18}O$ in every soil profile by extracting the water.

Precipitation sampling

Precipitation samples were collected at Yeniugou (P1: 3320 m a.s.l.), Hulugou (P2: 3020 m a.s.l.), and Pailugou (P3: 2700 m a.s.l.) in the upper reaches, at Zhangye (P4: 1483 m a.s.l.) in the middle reaches, and at Ejina (P5: 920 m a.s.l.) in the lower reaches (Fig. 1a). At Yeniugou, samples were collected for individual events from June 2008 to September 2009. At Hulugou, single-event precipitation samples were collected from July to September 2009 and May to October 2014. At Pailugou, precipitation was sampled once per two hours during two precipitation periods in July and August 2009, and single-event samples were collected from September to November 2008 and June 2011 to October 2014. At Ejina, single-event precipitation samples were collected from January 2007

to December 2010. Stable isotope composition of previous years precipitation (1986-2003) at Zhangye (Fig. 1a P4) were obtained from the GNIP database (http://nds121.iaea.org/wiser) (Zhao et al., 2012). To prevent evaporation of the sampled water, rain samples for each precipitation event were collected and immediately transferred to fill air-tight 8 ml or 20 ml plastic bottles (Brand CNW, Germany). The solid samples (snow and hail) were collected and then melted in low-density polyethylene zip-lock bags at room temperature before being sealed into plastic bottles. We used new low-density polyethylene bags for each sample. All samples were stored at 6 to 8°C prior to analysis.

Water extraction for isotope analyses

All samples were processed at the Key Laboratory of Ecohydrology of the Inland River Basin, Northwest Institute of Eco-Environment and Resources, Chinese Academy of Sciences. Water was extracted from root, stem and soil with cryogenic vacuum distillation (Ehleringer et al., 2000, West et al., 2006). Samples in extraction vials were heated to 100°C and evaporated water was trapped in U-tubes, submerged in liquid nitrogen. We have done extensive water extraction tests in the laboratory including using species from other regions that do not show any significant difference between source water and xylem water after the extraction. West et al., (2006) estimated a minimum extraction time of 60-75 min for woody stems, 40 min for clay soils, 30 min for sandy soils, and 20 to 30 min for leaves during vacuum distillation to obtain an unfractionated water sample. In our study, extraction was performed under a vacuum of 0.03 hPa for at least two hours in order to ensure an unfractionated water sample (West et al., 2006). The extracted water samples were sealed with Parafilm, placed in a bath and allowed to thaw. The liquid water was then transferred to a 2 ml vial for $\delta^{18}O$ and $\delta^{2}H$ analysis.

Measurement of soil water content

Gravimetric water content of each soil sample was measured by weighing the soil sample, then heating the sample for 24 h at 105°C. The samples were then cooled in a desiccator and the dry soil was weighted.

Isotope analysis

The $\delta^{18}O$ and $\delta^{2}H$ values of the water samples were measured using Isoprime isotope ratio mass spectrometer (Isoprime Ltd, UK) coupled to a Euro EA3000 element analyzer at Heihe Key Laboratory of Ecohydrology and River Basin Science, Northwest Institute of Eco-Environment and Resources. To avoid any memory effect associated with continuous-flow methods, measurements of each sample were repeated five times, and the first values were discarded. The accuracy was better than ±1.0‰ for $\delta^{2}H$ and ±0.2‰ for $\delta^{18}O$. The $\delta^{18}O$ and $\delta^{2}H$ were calibrated using two international standard materials (V-SMOW and GISP or SLAP) and one working standard. The $\delta^{18}O$ and $\delta^{2}H$ values are expressed in ‰ on a V-SMOW–SLAP scale. This method is a mass-based method of analysis, and trace amounts of contaminants are unlikely to have a large effect on the isotopic value of a water sample measured by IRMS due to the relatively small mass contribution that they make to the total amount of ^{1}H, ^{2}H, ^{16}O and ^{18}O isotopes in the sample (West *et al.*, 2010).

Data analysis

The Bayesian isotope mixing model (MixSIAR) was used to determine the uptake fractions of water sources (Parnell et al., 2010), and the software package MixSIAR (Stock and Semmens, 2013) was used for the analysis of source water contributions to the plant isotopic composition. MixSIAR is a flexible framework to create mixing models based on the Bayesian theory (Bowen et al., 2018;

Erhardt and Bedrick, 2013; Moore and Semmens, 2008; Parnell et al., 2010, 2013), and it is available to download from the packages section of the Comprehensive R Archive Network site (CRAN)-http://cran.r-project.org/.

One of the main principles of the isotope tracing methodology is the assumption that isotope fractionation during root water uptake does not occur (Allison et al., 1983; Dawson and Ehleringer, 1991; Ehleringer and Dawson, 1992; White et al., 1985). If it is true, the $\delta^{18}O$ and $\delta^{2}H$ of xylem water should always be within the range of values of all water sources. However, xylem water $\delta^{18}O$ could always be interpreted as a mixture of deep and shallow soil waters, but the $\delta^{2}H$ of xylem water was sometimes more depleted than the considered water sources (Barbeta et al., 2019). Vargas et al. (2017) showed that *P. americana* plants discriminated against hydrogen isotopes about 10 times more than oxygen isotopes during water uptake. Brooks et al. (2010) and Oerter and Bowen (2019) reported that $\delta^{18}O$–$\delta^{2}H$ plots of xylem water occupy the $\delta^{18}O$–$\delta^{2}H$ space well below the soil water line, suggestive of deuterium fractionation processes during root water uptake. Other previous studies also found that the isotopic compositions of xylem water are relatively depleted compared to those of the considered sources (De Deurwaerder et al., 2018; Ellsworth and Williams, 2007; Evaristo et al., 2017; Geris et al., 2017; Oerter and Bowen, 2019; Oerter et al., 2014; Wang et al., 2017; Zhao et al., 2016). If such fractionation processes are not considered, the estimation of plant water sources may be inaccurate. Evaristo et al. (2017) showed that erroneous results could be obtained when a simple mass balance approach using only hydrogen isotopes was implemented, but they also concluded that results were less sensitive to deuterium fractionation when both deuterium and oxygen isotopes were combined within a Bayesian inference approach. Therefore, we selected typical sites characteristic of deuterium fractionation (M1-10, L2-12, L5-10 and L3-08) and deuterium non-fractionation (U7, U9-9, M4-12

and L6-12) to calculate the water source contributions using $\delta^{18}O$ alone, $\delta^{2}H$ alone and both $\delta^{18}O$ and $\delta^{2}H$ by the Bayesian isotope mixing model. We find remarkedly differences among the three methods if deuterium fractionation occurs, especially at L5-10, which contributions of groundwater to HA were 70.0% for $\delta^{18}O$, 0.2% for $\delta^{2}H$ and 49.9% for both $\delta^{18}O$ and $\delta^{2}H$ methods, respectively (Table S1). We also compared the results with deuterium fractionation using $\delta^{18}O$ alone, $\delta^{2}H$ alone and both $\delta^{18}O$ and $\delta^{2}H$ between the Iso-Source model (Phillips and Gregg, 2003) and the Bayesian isotope mixing model (Table S2), and found similar results using $\delta^{18}O$ alone by both approaches in the calculated sites. Therefore, in this study, similar to previous studies, we assumed that oxygen isotope fractionation does not occur during plant uptake water, and we used the Bayesian isotope mixing model to quantify the relative contribution of water sources for different plant species based on $\delta^{18}O$ data alone. In addition, in our study, when $\delta^{18}O$ of plant xylem water was not within the range of values of all water sources, we took 100% as the contributions of their nearest water sources such as U1, U2, M1-12 and L3-08 (Table 5). The most probable sources of water uptake were estimated by comparing the $\delta^{18}O$ of stem water with soil water and groundwater. Precipitation and river water were not considered as precipitation is low and all sites are far away from the main river.

Results

Seasonal precipitation and soil water content

Fig. 2+Table 3 here

Mean annual precipitation of the upper reaches, middle reaches and lower reaches are 404.1, 129.9, and 34.9 mm, respectively (Fig. 1b). In order to indicate the plant water use strategy responses to precipitation, the precipitation of two months before sampling date was used (Fig. S1). During our

study periods, corresponding to precipitation along the basin scale and in the lower reaches with water supply from the middle reaches, the profile mean soil water content varied greatly from the upper reaches to the lower reaches. The profile mean soil water content were 34.2%, 2.0% and 8.2%, and varied from 17.0±2.3 (U3) to 64.5%±3.9 (U4), from 1.6±0.4 (M3-12) to 2.3%±0.6 (M1-10), and from 6.6±5.9 (L1-12) to 12.4%±7.5 (L2-10) in the upper, middle and lower reaches, respectively (Table 3). Soil water content (SWC) of the profiles at each study site also varied greatly (Fig. 2a-2g). The SWC of the profiles in the upper reaches (Fig. 2a-2b) and middle reaches (Fig. 2c-2e) were relatively stable. However, the riparian forest, planted shrubland and Gobi in the lower reaches have steeper SWC gradients than those of the upper and middle reaches, and the water table is overlain by uniformly dry soil in the lower reaches (Fig. 2f-2g).

Isotopic compositions of different water pools

Fig. 3 + 4 and Table 4 here

The $\delta^{18}O$ and $\delta^{2}H$ in event-based precipitation varied from -33.3 to 13.1‰ and -253.4 to 113.0‰ at three mountainous sites of the upper reaches, and from -25.3 to 4.9‰ and -217.8 to 36.4‰ at Ejina of the lower reaches, respectively (Fig. S2). The slopes and intercepts of the local meteoric water lines (LMWLs) were 7.883 and 14.270, 7.013 and -2.871, and 7.731 and -6.948, respectively at the upper, the middle and the lower reaches (Fig. S2).

The isotopic composition of soil water exhibited both rainfall and groundwater effects and varied greatly along the strong climatic gradient in the inland Heihe River Basin (Fig. 3 and 4; Table 4). In general, the $\delta^{18}O/\delta^{2}H$ values of soil water were most negative in the upper reaches (-6.6±2.5‰/-48.8±12.9‰) associated with greater precipitation amount, and were negative at the riparian site (-3.6±3.4‰/-39.3±11.1‰) and the planted shrubland (-4.4±3.6‰/-55.3±7.2‰) in the lower reaches

associated with shallower groundwater level. They were more positive at the desert-oasis ecotone (-0.5±4.2‰/-37.4±16.3‰) and the Gobi (1.8±1.9‰/-30.3±8.7‰) in the middle reaches, and the Gobi (0.4±2.8‰/-45.5±5.7‰) in the lower reaches (Fig. 3 and 4; Table 4). In addition, except for 2012 (a precipitation event occurred just before sampling), the $\delta^{18}O$ values of soil water in the upper soil layers were higher than those of the lower layers due to evaporation (Fig. 4). The slope and intercept of the soil water evaporation lines (SWELs) decreased from the upper to lower reaches (except 2009 in the lower reaches), and were lower than those of their corresponding local meteoric water lines (LMWLs) (Fig. S2; Fig. 3). During the study period, the $\delta^{18}O/\delta^{2}H$ value of groundwater was more negative in the middle reaches (-7.8±0.2‰/-49.7±0.5‰) than that of the lower reaches (-6.7±1.0‰/-47.7±9.7‰) (Fig. 3 and 4; Table 4), while the groundwater exhibited relatively more steady isotope values in the middle reaches than those of the lower reaches, especially at planted shrubland and Gobi (Fig. 3; Table 4). Different from soil water variations, the $\delta^{18}O/\delta^{2}H$ values of xylem water varied with their potential water sources (Fig. 3 and 4; Table 4). For example, the more negative $\delta^{18}O/\delta^{2}H$ values of xylem water were found at sites in the middle (-7.5±0.3‰/-60.9±4.1‰ for *Tamarix ramosissima*, and -7.4‰/-67.8‰ for *Haloxylon ammodendron*) and the lower reaches (-7.6±0.3‰/-69.2±2.4‰ at planted shrubland, and -5.4‰±1.3‰/-50.5±5.2‰ at riparian forest), where groundwater is available to plants. And the more negative $\delta^{18}O/\delta^{2}H$ values of xylem water were also found in the upper reaches (-6.1±1.9‰/-48.1±11.6‰ at Qinghai spruce forest, and -3.5±1.9‰/-48.9±8.4‰ at alpine grassland meadow region) (Fig. 3 and 4; Table 4) affected by precipitation. In addition, the $\delta^{18}O/\delta^{2}H$ values of xylem water of tree, shrub and herbaceous plants became progressively more positive at Qinghai spruce forest in the upper reaches and riparian forest in the lower reaches (Fig. 3 and 4; Table 4). The more positive $\delta^{18}O/\delta^{2}H$ values of xylem water were found at sites in the middle (-0.6±0.6‰/-33.4±5.1‰ for

Haloxylon ammodendron at desert-oasis ecotone, and 0.8±0.8‰/-33.5±1.8‰ at Gobi) and lower reaches (-2.5±0.2‰/-49.1±5.5‰ at Gobi) (Fig. 3 and 4; Table 4), where soil water is likely the main water source to plants.

Contributions of potential water sources along the climatic gradient

Figure 5 and Table 5 here

The contributions of potential water sources to different plant species varied along the strong climatic gradient and different soil water environments (Fig. S1; Fig. 2, 4 and 5; Table 5). In the upper reaches, the water use patterns of plant species were varied and controlled by precipitation. For example, grasses and herbaceous plants used water chiefly from the top 10 cm of the soil profile throughout the year, and more than 70% water sources came from the top 5 cm of the soil profile during wet season and at high soil water content sites. Shrubs such as *Potentilla fruticosa* also used surface soil water during wet season (0-5 cm) and used shallow soil water during dry season (up to 15 cm). Qinghai spruce used deeper water sources, chiefly down to 40/60 cm in wet season and deeper (up to 120 cm) in dry season (Fig. S1a; Fig. 2a-b; Fig. 4a-b and 5a; Table 5).

In the middle reaches, main water sources were deep soil water/groundwater/precipitation, and their contributions varied with precipitation (Fig. S1b; Fig. 2c-e, 4c-d and 5b; Table 5). At the desert-oasis ecotone, groundwater was the main water source for *Tamarix ramosissima*, which were not affected by precipitation. For *Haloxylon ammodendron*, when groundwater was available, it was completely dependent on it. However, when groundwater was too deep, deep soil water was the main water sources for it, and the contributions varied with precipitation. Similarly, *Reaumuria soongorica* and *Nitraria tangutorum* used deeper layer soil water at Gobi with low precipitation (Fig. S1b; Fig. 2c-e, 4c-d and 5b; Table 5).

In the lower reaches, in the extremely arid region such as Ejina, where the annual precipitation is about 39 mm, the main and stable water sources of plants were shallow groundwater/deep soil water recharged by groundwater, which were not affected by precipitation (Fig. 2f-i, 4e-g and 5c-d; Table 5). At the riparian forest, groundwater and their corresponding saturated layer soil water were the main water source to *Populus euphratica*. Soil water was the stable water source of herbaceous plant such as *Sophora alopecuroides*. For shrub such as *Tamarix ramosissima*, deep soil water was its main water source, and it also used groundwater. In addition, water sources of *Populus euphratica*, *Tamarix ramosissima* and *Sophora alopecuroides* differed remarkably at the same site such as L3-08 and L4-08. *Populus euphratica* mainly depended on groundwater, *Tamarix ramosissima* depended on deep soil water and groundwater, and *Sophora alopecuroides* depended on soil water (Fig. 4e; Fig. 5c; Table 5). For the planted shrubs of the planted shrubland, groundwater and deep soil water recharged from groundwater were main water sources for *Haloxylon ammodendron* (Fig. 4f; Fig. 5d; Table 5). For shrubs at Gobi, deep soil water recharged from groundwater was the stable water source of *Reaumuria soongorica* (Fig. 4g; Fig. 5d; Table 5).

Discussion

Isotopic patterns of different water pools

For precipitation, the slopes of the LMWLs of the upper (7.883), the middle (7.013) and the lower reaches (7.731) (Fig. S2) were slightly lower than that of the GMWL (8), and the intercepts of the upper reaches (14.270) was higher than that of the GMWL (10), while of the middle (-2.871) and lower reaches (-6.948) were very low (Fig. S2). Our results indicated that the local climatic factors (e.g., strong moisture recycling, re-evaporation of raindrops during precipitation and seasonality of

precipitation) affected the precipitation isotope ratios along the Heihe River Basin, and stronger evaporation occurred at the middle and lower reaches (Fig. 1; Fig. S2) (Mook, 2000; Zhao et al., 2019).

For soil water isotopes, the mean $\delta^{18}O$ and $\delta^{2}H$ values varied significantly (Fig. 3; Table 4). These soil $\delta^{18}O$ and $\delta^{2}H$ variations revealed the complex affecting factors on isotopic discrimination under different environments, for example, precipitation infiltration and evaporation at the upper reaches, evaporation, groundwater recharge and precipitation infiltration at the middle reaches, and evaporation, groundwater recharge and surface water delivery at the lower reaches (Cheng et al. 2014; Hu et al. 2015; Vereecken et al. 2016; Zhang et al. 2018). The decreases of slopes and intercepts of SWELs from the upper to the lower reaches (Fig. 3) revealed the significant decrease of relative humidity from the upper to the lower reaches of the Heihe River Baisn (Fig. 1), and these variations can be explained by the increase in the effective thickness of the vapor transport layer (Barnes and Allison, 1988) and the stronger soil isotopic kinetic effect from the upper to the lower reaches (Cooper et al. 1991). The slopes and intercepts of SWELs were significantly lower than their corresponding LMWLs, which revealed strong evaporation effect on soil water, and these evaporation effect increased gradually from the upper to lower reaches (Fig. 1). For most sampling dates, the profile of soil water $\delta^{18}O$ was characterized by more positive isotopic values in shallow soil layers and more negative values in deeper and saturated soil layers, and these profile variation ranges increased gradually from the upper to the lower reaches (Fig. 3 and 4). However, at U8-12, U9-6 and U9-9 of the upper reaches, at M2-12 and M4-12 in the middle reaches and at L5-12 and L6-12 in the lower reaches, an inverse curve was found with depleted values in shallow soil layers, and this inverse pattern could be explained by infiltration of precipitation characterized by negative isotopic values (Newman et al., 1997).

For xylem water isotopes, significant differences were found among the mean $\delta^{18}O/\delta^{2}H$ values of

plant xylem water along the Heihe River Basin (Fig. 3 and 4; Table 4). These results revealed that there were very complex water sources for different plants along the Heihe River Basin. The mean δ^2H of groundwater were much higher than those of xylem water of *Populus euphratica*, *Tamarix ramosissima* and *Haloxylon ammodendron*, suggesting possible deuterium fractionation occurred between xylem sap/stem tissue water and their water source for these three species (Brooks et al., 2010; De Deurwaerder et al., 2018; Evaristo et al., 2017; Geris et al., 2017; Oerter and Bowen, 2019; Wang et al., 2017; Zhao et al., 2016).

Biplots of δ^2H and $\delta^{18}O$: In the upper reaches, the plots of δ^2H-$\delta^{18}O$ of shallow soil water and xylem water were relatively far away from its corresponding LMWL. This trend was particularly obvious for δ^2H and $\delta^{18}O$ of xylem water, suggesting that: (i) plant water source originated from soil water, (ii) soil water came from the local precipitation, and (iii) strong soil evaporation was occurred in the shallow soil layer in the upper reaches (Fig. 3a). In the middle reaches, the plots of δ^2H and $\delta^{18}O$ of xylem water in desert-oasis ecotone and Gobi suggested that plant water source of these sites came mainly from shallow groundwater and soil water (Fig. 3b). At the riparian forest and planted shrubland of the lower reaches, the plots of δ^2H and $\delta^{18}O$ of xylem water and soil water were near the shallow groundwater and river water, indicating that soil water and xylem water were from groundwater recharged from river (Fig. 3c). The δ^2H-$\delta^{18}O$ plots of soil water and xylem water were far away from the plots of river water and groundwater in Gobi, indicating the strong evaporation occurred at these regions, and soil water was the main water source for *Reaumuria soongorica* (Fig. 3d).

Contributions of potential water sources along the climatic gradient

In the upper reaches: Surface soil water (up to 5 cm) and shallow soil water (up to 10/15 cm) were the main water sources for herbaceous plants and shrubs, and the herbaceous plants and shrubs

preferentially used 0-5 cm soil water during wet season or under well-watered conditions. During dry season or under well-stressed conditions, the herbaceous plants shifted to use 0-10 cm soil water, and shrubs shifted to 15 cm soil water (Fig. S1a; Fig. 2a; Fig. 4a; Fig. 5a; Table 5). These results indicated that as herbaceous plants and shrubs usually have shallow rooting system, the alpine steppe meadow zone in the upper reaches of the Heihe River Basin where these herbaceous plants dominate is prone to degradation due to decreasing precipitation, and these results also highlighted the importance of precipitation as the main controlling factor of water use patterns for shrubs in the upper reaches of the Heihe River Basin. Trees such as Qinghai Spruce appeared to be acquiring water preferentially from the upper 40/60 cm of the soil profile during wet season. However, Qinghai spruce seemed to be tapping water mostly from greater depths during dry season, and the depths reached 120 cm (Fig. S1a; Fig. 2b; Fig. 4b; Fig. 5a; Table 5). Our results indicated that the controlling factor of water uptake was precipitation and water use patterns of different plant species varied with precipitation and species in the upper reaches.

As climate becomes drier from the upper to the middle reaches, we observed that groundwater or deep soil moisture contributed majority of the water needs of shrubs (Fig. 4c-d; Fig. 5b; Table 5). Some species exhibited strong plasticity in water uptake sources (Fig. 4c-d; Fig. 5b; Table 5). For example, groundwater was only water source for *Tamarix ramosissima*. *Haloxylon ammodendron* preferentially access to groundwater even under high precipitation if groundwater is its stable water source. However, contributions of 0-40cm soil water to *Haloxylon ammodendron* increased dramatically after large precipitation when it cannot use groundwater (Fig. S1b; Fig. 1c; Fig. 4c; Fig. 5b; Table 5), revealing that water use strategy of *Haloxylon ammodendron* was controlled by groundwater level, deep soil water and precipitation (Fig. 4d; Fig. 5; Table 45). Deep soil water and precipitation were the main

water source for *Reaumuria soongorica* and *Nitraria tangutorum*, and their water use strategy was controlled by both deep soil water and precipitation (Fig. S1b; Fig. 4d; Fig. 5b; Table 5).

In the extremely arid region (i.e., Ejina) of the lower reaches, the main and stable water sources of plants were shallow groundwater and deep soil water recharged by groundwater (Fig. 4e-g; Fig. 5c-d; Table 5). For trees: shallow groundwater and statured soil water layer in the riparian forest were the main water sources for *Populus euphratica* (Fig. 4e; Fig. 5c; Table 5). These results were consistent with the results of Pettit et al. (2018) who reported that the dominant tree species *Eucalyptus camaldulensis* (river red gum) growing at riparian of Maules Creek is capable of utilizing groundwater even to depths >10 m. At L1-10, the main water source of *Populus euphratica* came from deep soil water (71.1%) and groundwater (21.1%), relating to water delivery from the middle reaches to the lower reaches (Table 5; Cheng et al. 2014; Hu et al. 2015; Zhang et al. 2018). For herbaceous plants: soil water was the stable water source of herbaceous plant such as *Sophora alopecuroides*, although the contributions of soil water varied with soil water content (Fig. 2; Fig. 4e; Fig. 5c; Table 5). For shrub such as *Tamarix ramosissima*, except at L2-10, deep soil water was its main water source, and it also used groundwater (Fig. 4e; Fig. 5c; Table 5). In addition, as aridity and groundwater depth increased, we found that coexisting plant species adopted different water use strategies in extremely water-limited environments. For example, water sources of *Populus euphratica*, *Tamarix ramosissima* and *Sophora alopecuroides* differed remarkably, and their main water sources were groundwater for *Populus euphratica*, deep soil water and groundwater for *Tamarix ramosissima*, and soil water for *Sophora alopecuroides* with groundwater level > 10.0 m, (Table 5). Our results were consistent with the findings of several other studies, which demonstrated that coexisting plant species would adopt different plasticity in water use strategies in water-limited environments (West et al., 2007; Eggemeyer

et al., 2009). For the planted shrubland: deep soil water recharged from groundwater and groundwater were main water sources of *Haloxylon ammodendron* (Fig. 4f; Fig. 5d; Table 5). For Gobi: due to the extremely low precipitation, pulses of high precipitation (e.g., 12.1 mm precipitation in July 2012) did not affect the water sources for the shrubs. Deep soil water recharged from groundwater below 160 cm was the stable water source of *Reaumuria soongorica* at Gobi (Fig. 4g; Fig. 5d; Table 5). Therefore, in the lower reaches, the riparian forest and the planted shrubland relied primarily on groundwater and deep soil moisture to survive. Deep soil water recharged from groundwater below 220 cm was a stable water source of *Reaumuria soongorica*. The maintenance of groundwater level has a vital role in maintaining the stability of oasis in the lower reaches of the Heihe River Basin (Fig. 5c-d; Table 5).

The species-specific water use strategy adaptations

In our study, *Tamarix ramosissima*, *Haloxylon ammodendron* and *Reaumuria soongorica* appear in both the middle and lower reaches. In the middle reaches, groundwater was the only water source for *Tamarix ramosissima* under low soil water conditions, and its water use patterns did not respond to precipitation (Fig. 2c, Fig. 4c; Fig. 5b; Table 5). However, its water use patterns showed great flexibility in the lower reaches. It used soil water when soil water content was high, and then used deep soil water and groundwater when it can get groundwater (Fig. 2g; Fig. 4e; Fig. 5c; Table 5). These results indicated that *Tamarix ramosissima* increase its adaptive capacity through changing its water use mode under extremely arid environment in the lower reaches. For *Haloxylon ammodendron*, it used soil water when it cannot access groundwater and contributions of shallow soil water varied depending on precipitation. Its main water source shifted to groundwater when it can get groundwater

in the middle reaches (Fig. 2d; Fig. 4c; Fig. 5b; Table 5). In the lower reaches, groundwater and deep soil water recharged from groundwater were its only water sources even under relatively high water soil content (>10% at about 140 cm) at L5-10 (Fig. 2h; Fig. 4f; Fig. 5d; Table 5). These results indicated that in the lower reaches, groundwater was the main water source for *Haloxylon ammodendron* and maintaining a suitable groundwater level is very important. For *Reaumuria soongorica*, soil water was its main water source in both middle and lower reaches. However, its water use patterns varied remarkably. In the middle reaches, *Reaumuria soongorica* used shallow soil water when there is a large precipitation (45.0% water from 0 to 80 cm at M4-12; 9.6% water from 0 to 80 cm at M4-10 (Fig. 2e; Fig. 4d; Fig. 5b; Table 5), and the contributions of below 120 cm soil water to *Reaumuria soongorica* were 80.2% at M4-10 (6.7 mm) and 36.7% at M4-12 (32.8 mm) (Table 5). In the lower reaches, main water sources of *Reaumuria soongorica* were from below 220 cm soil water recharged from groundwater, and its water use patterns did not respond to precipitation (Fig. 2i; Fig. 4g; Fig. 5d; Table 5). These results also highlighted that in the lower reaches, groundwater was the main water source for *Reaumuria soongorica* ecosystem at Gobi.

Summary and implications

Our study suggested that there were significantly different water sources for various plants under different climatic conditions in the Heihe River Basin, northwestern China. In the upper reaches, when precipitation is ample, it recharges soil water, which then becomes the main plant water source. Plants used shallow soil water during wet season, and used deeper soil water during dry season. Water uptake patterns thus vary inter-annually following seasonal fluctuations in precipitation and soil water. As climate becomes drier in the middle reaches, plants relied on groundwater/deeper soil water sources,

although precipitation still had contributions to a certain degree. Some variations occurred with species and ecosystem types. At the driest part of the Heihe River Basin, water use strategy was not affected by precipitation. Groundwater and deep soil water recharged by groundwater were potential water sources for different plants in the riparian forest, the planted shrubland and Gobi. The maintenance of groundwater level has vital role in maintaining the stability of oasis in the lower reaches of the Heihe River Basin. Lastly, there are emerging evidence that there are potential deuterium fractionation during plant water uptake (Brooks et al., 2010; De Deurwaerder et al., 2018; Evaristo et al., 2017; Geris et al., 2017; Oerter and Bowen, 2019; Wang et al., 2017; Zhao et al., 2016), our extensive field observation added critical information on this aspect.

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Table 1 The acronyms of hydrogen and oxygen isotopic parameters as well as plants with their functional types of the species.

Table 2 The sampling species, sampling plant components and sampling dates in different regions of the Heihe River Basin.

Table 3 The mean soil water content (%) of soil profile at the different study sites in the Heihe River Basin. The detailed information is shown in the Table 2.

Table 4 Variations of $\delta^{18}O$ and $\delta^{2}H$ (‰) in plant xylem water and their potential water source of the Heihe River Basin.

Table 5 Contributions of possible water sources to different plants (%) of different ecosystem types based on oxygen isotopes. The acronyms of plants as well as hydrogen and oxygen isotopic parameters are shown in the Table 1. The UR, MR and LR indicate the upper, middle and lower reaches, respectively. The SW and GW indicate soil water and groundwater. The U1, U2, U3 and U4 indicate mountain grassland, mountain meadow, high mountain meadow and swamp meadow in the upper reaches, respectively, in the upper reaches. U5, U6, U7, U8-11, U8-12, U9-6 and U9-9 indicate Qinghai Spruce forest in the upper reaches. M1-10, M1-12, M2-10, M2-12 and M3-12 indicate desert-oasis ecotone, and M4-10 and M4-12 indicate Gobi in the middle reaches. L1-07, L1-09, L1-10, L1-12, L2-10, L2-12, L3-08 and L4-08 indicate riparian forest in the lower reaches. L5-10 and L5-12 indicate planted shrubland, and L6-10 and L6-12 indicate Gobi in the lower reaches. Because $\delta^{18}O$ of plant xylem water was not within the range of values of all water sources, we took 100% as the contributions of their nearest water sources of U1, U2, M1-12 and L3-08.

Table 1 The acronyms of hydrogen and oxygen isotopic parameters as well as plants with their functional types of the species.

Acronym	The full name	Plant species		
		Acronym	The full name	Functional type of the species
UR	The upper reaches of the HRB	QS	Qinghai Spruce	Evergreen coniferous tree
MR	The middle reaches of the HRB	PE	*Populus euphratica*	Deciduous broadleaf tree
LR	The lower reaches of the HRB	HA	*Haloxylon ammodendron*	Deciduous broadleaf shrub
HRB	The Heihe River Basin	NT	*Nitraria tangutorum*	Deciduous broadleaf shrub
Qilian Mt.	Qilian Mountain	PF	*Potentilla fruticosa*	Deciduous broadleaf shrub
QSF	Qinghai spruce forest	RS	*Reaumuria soongorica*	Deciduous shrub
MG	Mountain grassland	TR	*Tamarix ramosissima*	Deciduous lanceolate leaf shrub
MM	Mountain meadow	PV	*Polygonum viviparum*	Polygonaceae perennial herb
SM	Swamp meadow	SA	*Sophora alopecuroides*	Leguminous perennial herb
DO	Desert-oasis ecotone	SC	*Stipa capillata*	Gramineous perennial herb
GB	Gobi	SP	*Stipa purpurea Griseb*	Gramineous perennial herb
RF	Riparian forest			
ASF	Artificial shrubbery forest			
GMWL	Global meteoric water line			
LMWL	Local meteoric water line			
$\delta^{18}O$	Oxygen isotope ratio			
$\delta^{2}H$	Hydrogen isotope ratio			

Table 2 The sampling species, sampling plant components and sampling dates in different regions of the Heihe River Basin.

Study region	Ecosystem type	Study time	Locations ID	Altitude (m)	Longitude	Latitude	Plant species	Groundwater level
The UR	Mountain grassland	2009/6/7	U1	2774	99.9	38.8	SC root	Spring
	Swamp meadow	2009/6/7	U2	3040	99.9	38.8	PV root	
	Mountain meadow	2009/6/8	U3	3476	99.5	38.6	SP root	
	Swamp meadow	2009/6/8	U4	3732	99.6	38.6	SC root	
	Qinghai spruce forest	2007/8/21	U5	2594	100.3	38.6	QS and PF stem	
		2009/6/7	U6	2654	99.6	38.8	QS stem	
		2009/7/31-8/2	U7	2774	100.3	38.5	QS stem	
		2011/6/27-28	U8-11	2780	100.3	38.6	QS stem and SC root	
		2012/8/1	U8-12				QS stem	
		2011/6/23-25	U9-6	2900	100.3	38.5	QS, PF and PV	
		2011/9/2-8	U9-9					
The MR	Desert-oasis ecotone	2010/6/15-16	M1-10	1386	100.1	39.4	TR stem	About 5.0 m
		2012/8/3	M1-12					
		2010/6/15-16	M2-10	1386	100.1	39.4	HA stem	
		2012/8/3	M2-12					
		2012/8/4	M3-12	1440	/	/	HA stem	
	Gobi	2010/6/18-19	M4-10	1413	100.1	39.4	RS and NT stem	>10.0 m
		2012/8/5	M4-12				RS stem	
The LR	Riparian forest	2007/6/19	L1-07	930	101.2	42	PE stem and SA root	1.6 m
		2009/8/6-9	L1-09					1.8 m
		2010/6/21-22	L1-10					2.0 m
		2012/8/8	L1-12				PE stem	2.0 m
		2010/6/21-22	L2-10	930	101.2	42	TR stem	2.0 m
		2012/8/8	L2-12					2.0 m
		2008/8/20	L3-08	920	101.1	42	PE, TR and SA	> 10.0 m
		2008/8/20	L4-08	921	101.1	42		
	Planted shrubland	2010/6/23-24	L5-10	910	101	41.9	HA stem	2.5 m

	2012/8/9	L5-12					2.2 m
Gobi	2010/6/26-27	L6-10	906	101.1	42.3	RS stem	> 5.0 m
	2012/8/11	L6-12					

Table 3 The profile mean soil water content (%) at the different study sites in the Heihe River Basin. The detailed information is shown in the Table 2.

The upper reaches	U1	U2	U3	U4	U6	U7	U8-11	U8-12	U9-6	U9-9	Mean
Profile mean soil water content (%)	24.9	59.6	17.0	64.5	30.6	34.7	23.8	26.3	35.6	25.4	34.2
Standard deviation	4.3	9.8	2.3	3.9	6.7	8.2	4.3	4.0	7.4	4.0	5.5
The middle reaches	M1-10	M1-12	M2-10	M2-12	M3-12	M4-10	M4-12				Mean
Profile mean soil water content (%)	2.3	2.3	1.9	2.0	1.6	1.4	2.2				2.0
Standard deviation	0.6	0.3	0.4	0.4	0.4	0.3	0.7				0.4
The lower reaches	L1-09	L1-10	L1-12	L2-10	L2-12	L5-10	L5-12	L6-10	L6-12		Mean
Profile mean soil water content (%)	9.1	9.7	6.6	12.4	8.8	4.7	6.6	7.1	9.2		8.2
Standard deviation	7.2	7.8	5.9	7.5	8.0	3.5	5.3	9.5	9.7		7.2

Table 4 Variations of $\delta^{18}O$ and $\delta^{2}H$ (‰) in plant xylem water and their potential water source of the Heihe River Basin.

The upper reaches		Soil water		Plant		Xylem water			Groundwater	
Alpine grassland meadow region		$\delta^{18}O$ (‰)	$\delta^{2}H$ (‰)			$\delta^{18}O$ (‰)	$\delta^{2}H$ (‰)		$\delta^{18}O$ (‰)	$\delta^{2}H$ (‰)
	Mean±SD	-5.3±2.7	-45.9±10.1	Herbaceous plants	Mean±SD	-3.5±1.9	-48.9±8.4	Mean±SD	/	/
	Max	0.2	-30.7		Max	-1.4	-29.4	Max	/	/
	Min	-8.8	-62.3		Min	-6.8	-59.7	Min	/	/
Qinghai spruce forest	Mean±SD	-7.0±2.3	-49.8±13.6	Qinghai Spruce	Mean±SD	-6.9±1.5	-50.8±10.4	Mean±SD	/	/
	Max	-1.4	-18.4		Max	-4.3	-37.3	Max	/	/
	Min	-11.7	-84.7		Min	-9.7	-67.3	Min	/	/
				Potentilla fruticosa	Mean±SD	-5.9±1.7	-47.5±13.2			
					Max	-4.2	-36.2			
					Min	-7.6	-62.0			
				Herbaceous plants	Mean±SD	-4.7±2.7	-43.2±15.3			
					Max	-1.7	-29.4			
					Min	-6.8	-59.7			
The middle reaches										
Desert-oasis ecotone	Mean±SD	-0.5±4.3	-37.4±16.5	*Tamarix ramosissima*	Mean±SD	-7.5±0.3	-60.9±4.1	Mean±SD	-7.8±0.2	-49.5±0.5
	Max	11.7	5.0		Max	-7.3	-58.0	Max	-7.6	-49.1
	Min	-7.4	-70.1		Min	-7.7	-63.8	Min	-8.0	-50.2
				Haloxylon ammodendron	Mean±SD	-2.1±4.7	-44.9±20.2			
					Max	1.0	-29.9			
					Min	-7.4	-67.8			
	Mean±SD	1.8±1.9	-30.3±8.7	*Reaumuria soongorica*	Mean±SD	0.8±0.8	-33.5±1.8	Mean±SD	-8.0	-50.2
Gobi	Max	5.9	-12.5	*Nitraria tangutorum*	Max	1.9	-31.0	Max	/	/
	Min	-0.6	-43.4		Min	0.1	-35.0	Min	/	/
The lower reaches										
	Mean±SD	-3.6±3.4	-39.3±11.1	*Populus euphratica*	Mean±SD	-6.2±1.1	-50.7±4.5	Mean±SD	-6.2±1.0	-42.0±4.9
Riparian forest	Max	6.1	-3.6		Max	-4.7	-46.0	Max	-4.5	-36.1
	Min	-7.3	-56.1		Min	-7.9	-58.7	Min	-7.6	-50.2
				Tamarix ramosissima	Mean±SD	-5.5±0.9	-52.2±7.4			
					Max	-4.3	-42.6			
					Min	-6.5	-60.0			

				Sophora alopecuroides	Mean±SD	-4.4±1.4	-48.7±5.1			
					Max	-2.6	-45.6			
					Min	-5.7	-56.3			
Planted shrubland	Mean±SD	-4.4±3.7	-55.3±7.5	*Haloxylon ammodendron*	Mean±SD	-7.6±0.3	-69.2±2.4	Mean±SD	-7.7±0.1	-63.0±1.9
	Max	4.7	-41.6		Max	-7.3	-67.5	Max	-7.6	-61.7
	Min	-7.8	-66.1		Min	-7.8	-70.9	Min	-7.7	-64.4
Gobi	Mean±SD	0.4±2.9	-45.5±5.9	*Reaumuria soongorica*	Mean±SD	-2.5±0.2	-49.1±5.5	Mean±SD	-7.5±0.1	-55.4±3.5
	Max	7.2	-36.0		Max	-2.3	-45.2	Max	-7.4	-52.9
	Min	-3.3	-56.9		Min	-2.7	-53.0	Min	-7.6	-57.9

Table 5 Contributions of possible water sources to different plants (%) of different ecosystem types based on oxygen isotopes. The acronyms of plants as well as hydrogen and oxygen isotopic parameters are shown in the Table 1. The UR, MR and LR indicate the upper, middle and lower reaches, respectively. The SW and GW indicate soil water and groundwater. The U1, U2, U3 and U4 indicate mountain grassland, mountain meadow, high mountain meadow and swamp meadow in the upper reaches, respectively, in the upper reaches. U5, U6, U7, U8-11, U8-12, U9-6 and U9-9 indicate Qinghai Spruce forest in the upper reaches. M1-10, M1-12, M2-10, M2-12 and M3-12 indicate desert-oasis ecotone, and M4-10 and M4-12 indicate Gobi in the middle reaches. L1-07, L1-09, L1-10, L1-12, L2-10, L2-12, L3-08 and L4-08 indicate riparian forest in the lower reaches. L5-10 and L5-12 indicate planted shrubland, and L6-10 and L6-12 indicate Gobi in the lower reaches. Because $\delta^{18}O$ of plant xylem water was not within the range of values of all water sources, we took 100% as the contributions of their nearest water sources of U1, U2, M1-12 and L3-08.

Study region	Ecosystem type	Locations ID		Plant species	Water sources and their contribution ratios to plant (Mean ± SD (%))					Groundwater level
The UR	Mountain grassland	U1	Water sources		5cm SW	10cm SW	30cm SW	50cm SW		Spring
			Contribution percentages	SC	**100**	/	/	/		
	Mountain meadow	U2	Water sources		5cm SW	10cm SW	30cm SW	50cm SW		
			Contribution percentages	PV	**100**	/	/	/		
	High mountain meadow	U3	Water sources		5cm SW	10cm SW	20cm SW			
			Contribution percentages	SP	30.2±17.3	41.3±23.3	28.5±17.1			
	Swamp meadow	U4	Water sources		5cm SW	10cm SW	20cm SW			
			Contribution percentages	SC	84.3±5.8	9.3±5.8	6.4±3.6			
	Qinghai Spruce forest	U5	Water sources		5cm SW	10cm SW	20cm SW	40cm SW	60cm SW	
			Contribution percentages	QS	14.1±9.6	20.0±15.7	22.8±17.0	22.9±17.4	20.2±14.5	
			Contributio percentages	PF	61.5±12.4	11.5±10.3	8.7±6.5	10.0±7.7	8.3±5.9	

		U6	Water sources		10cm SW	30cm SW	70cm SW					
			Contribution percentages	QS	20.2±12.1	36.3±22.3	43.5±22.5					
		U7	Water sources		5cm SW	10cm SW	20cm SW	30cm SW	40cm SW	50cm SW	60cm SW	
			Contribution percentages	QS	63.5±5.6	8.4±7.5	5.3±3.8	5.1±3.6	6.5±5.2	5.4±3.9	5.7±4.2	
		U8-11	Water sources		3cm SW	5cm SW	10cm SW	20~40cm SW	50~60cm SW	80cm SW	100~120cm SW	
			Contribution percentages	QS	9.9±6.5	11.1±7.6	11.4±8.1	18.1±15.5	17.6±14.4	16.5±12.3	15.4±10.8	
			Contribution percentages	SC	62.8±17.1	21.0±16.3	16.2±11.2	/	/	/	/	
		U8-12	Water sources		5cm SW	10-40cm SW	40-60cm SW	60-100cm SW	100-120cm SW	120-140-160cm SW		
			Contribution percentages	QS	21.8±17.3	20.9±12.2	21.6±17.4	14.5±11.3	11.9±8.8	9.3±6.2		
		U9-6	Water sources		3~5cm SW	5~10cm SW	10~15cm SW	15~20cm SW	20~60cm SW	60~90cm SW		
			Contribution percentages	QS	8.2±5.6	8.3±5.6	15.8±13.6	26.2±19.9	28.4±15.0	13.2±10.0		
			Contribution percentages	PF	31.0±18.3	31.1±19.9	15.8±11.7	12.0±7.8	10.1±6.2			
			Contribution percentages	PV	38.8±22.0	37.9±25.1	13.3±9.2	10.1±6.4				
		U9-9	Water sources		3~5cm SW	5~10cm SW	10~20cm SW	20~40cm SW	40-60cm SW	80~90cm SW		
			Contribution percentages	QS	27.3±15.4	19.9±16.2	9.9±7.2	21.9±18.9	11.7±8.9	9.4±6.8		
			Contribution percentages	PF	64.7±17.8	12.5±17.1	4.9±3.7	7.5±6.3	5.4±4.1	5.0±3.6		
			Contribution percentages	PV	69.7±20.7	21.7±20.5	8.6±5.4	/				
The MR	Desert-oasis ecotone	M1-10	Water sources	TR	10cm SW	40-70cm SW	100cm SW	130-160cm SW	180-200cm SW	220cm SW	GW	About 5.0 m
			Contribution percentages		1.1±0.7	1.6±1.0	1.8±1.3	2.6±2.0	3.3±3.0	8.3±16.7	81.3±16.3	
		M1-12	Water sources		5-20cm SW	40-60cm SW	80-160cm SW	180cm SW	200-220cm SW	240-280cm SW	300cmSW	GW
			Contribution percentages		/	/	/	/	/	/	/	**100**
		M2-10	Water sources	HA	10cm SW	40cm SW	60cm SW	70cm SW	100cm SW	160cm SW	200cm SW	
			Contribution percentages		6.1±4.3	8.5±6.4	15.9±14.6	9.2±7.5	18.8±17.0	20.3±17.9	21.2±12.1	
		M2-12	Water sources		5cm SW	10cm SW	20-40cm SW	60-140cm SW	160-220cm SW	240-280cm SW	300cm SW	
			Contribution percentages		11.1±8.1	16.3±13.9	10.4±7.3	16.1±13.6	17.9±15.4	15.9±11.8	12.3±7.6	
		M3-12	Water sources		5-10cm SW	20-40cm SW	60-120cm SW	140-220cm SW	240-260cm SW	GW		

			Contribution percentages		0.7±0.4	0.8±0.5	1.0±0.07	1.3±1.0	2.1±2.0	94.1±2.4			
	Gobi	M4-10	Water sources		10cm SW	20-80cm SW	100cm SW	120cm SW	140cm SW	160-200cm SW			
			Contribution percentages	RS	3.4±2.1	6.2±4.8	10.2±10.9	18.6±19.6	23.7±22.6	37.9±23.9			
			Contribution percentages	NT	4.2±2.5	7.8±5.8	12.5±11.8	21.6±19.6	24.8±21.2	29.1±20.3			>10.0 m
		M4-12	Water sources		10-15cm SW	20-40cm SW	50-70cm SW	80cm SW	100-120cm SW	120-160cm SW	180-200cm SW		
			Contribution percentages	RS	12.2±9.9	7.8±5.3	10.6±7.9	14.4±13.0	18.2±15.3	18.3±14.6	18.4±12.9		
The LR	Riparian forest	L1-07	Water sources						80-100cm SW		GW		
			Contribution percentages	PE							93	Zhao et al., 2008	1.6 m
			Contribution percentages	TR							90		
			Contribution percentages	SA					97				
		L1-09	Water sources		5cm SW	8cm SW	10-30cm SW	40-60cm SW	80-120cm SW	140-160cm SW	GW		
			Contribution percentages	PE	2.4±1.6	2.6±1.8	3.8±3.0	5.1±6.7	11.0±17.3	42.0±31.0	33.2±32.0		1.8 m
			Contribution percentages	SA	11.3±7.1	12.7±8.1	24.5±18.8	25.6±18.4	25.9±16.1				
		L1-10	Water sources		20cm SW	40cm SW	60-80cm SW	100-140cm SW	160-180cm SW	GW			
			Contribution percentages	PE	7.7±3.2	15.4±10.6	20.2±16.4	16.7±12.8	18.8±14.9	21.1±17.2			
			Contribution percentages	SA	26.4±6.1	31.8±20.1	22.1±14.5	19.6±12.7					
		L1-12	Water sources		5-10cm SW	20cm SW	40-100cm SW	120cm SW	140-160cm SW	180-200cm SW	GW		
			Contribution percentages	PE	2.8±1.8	4.0±3.0	7.9±9.2	35.2±25.1	16.7±19.8	22.5±23.6	11.0±11.1		
		L2-10	Water sources	TR	20cm SW	40cm SW	60cm SW	80-100cm SW					
			Contribution percentages		9.3±3.5	17.9±10.9	37.1±22.0	35.7±21.2					
		L2-12	Water sources		5-10cm SW	20cm SW	40cm SW	60-80cm SW	100-140cm SW	160cm SW	GW		
			Contribution percentages		3.4±22.0	4.5±3.1	6.8±6.6	13.5±14.8	23.0±21.6	31.5±22.4	17.2±18.3		2.0 m
		L3-08	Water sources		0-5cm SW	5-20cm SW	40-60cm SW	80-120cm SW	140-180cm SW	200-240cm SW	GW		
			Contribution percentages	PE	/	/	/	/	/	/	**100**		
			Contribution percentages	TR	2.5±1.4	3.1±2.0	5.4±4.1	9.1±10.0	18.2±20.7	24.0±24.6	37.7±26.2		
			Contribution percentages	SA	3.6±2.1	4.7±3.0	8.7±6.9	18.3±19.1	28.4±23.9	36.4±24.8			
		L4-08	Water sources		10cm SW	20cm SW	40-60cm SW	100cm SW	120-200cm SW	220-240cm SW	GW		
			Contribution percentages	PE	2.2±1.4	3.8±2.7	4.7±3.7	5.6±4.6	6.6±6.1	33.8±28.8	43.3±30.2		
			Contribution percentages	TR	5.2±2.9	9.4±6.8	12.6±10.5	17.3±15.5	18.7±16.3	18.4±14.0	18.4±13.5		
			Contribution percentages	SA	11.0±4.9	19.7±14.4	20.2±16.8	17.6±13.8	16.5±12.8	14.9±11.2			>10.0 m

Planted shrubland	L5-10	Water sources	HA	20cm SW	40-60cm SW	80-100cm SW	140-160cm SW	170-200cm SW	230-250cm SW	GW		2.5 m
		Contribution percentages		1.5±1.0	1.9±1.4	2.3±1.7	3.1±2.7	3.8±3.4	17.5±27.7	70.0±25.9		
	L5-12	Water sources		5cm SW	10cm SW	20-40cm SW	60-100cm SW	120-140cm SW	165cm SW	180-220cm SW	GW	2.2 m
		Contribution percentages		2.7±2.0	5.0±4.5	4.2±3.5	7.3±9.1	6.9±6.9	35.7±24.7	27.8±27.3	10.4±13.3	
Gobi	L6-10	Water sources	RS	140cm SW	160cm SW	175-180cm SW	185cm SW	200cm SW	220cm SW	255cm SW		
		Contribution percentages		4.0±2.9	4.3±3.4	5.6±4.8	5.0±4.1	5.4±4.7	7.7±10.1	68.1±11.8		
	L6-12	Water sources		5-20cm SW	40-100cm SW	120cm SW	140-160cm SW	200cm SW	220-240cm SW	260-280cm SW		> 5.0 m
		Contribution percentages		2.9±2.1	2.2±1.4	2.7±1.8	3.3±2.5	3.8±3.0	18.4±26.1	66.6±25.0		

Figure captions

Fig. 1 Sampling sites (a), environmental conditions of the upper reaches (b), the middle reaches (c), and the lower reaches (e) of the Heihe River Basin.

Fig. 2 Soil water content (%) of the mountain grassland zone (a) and the Qinghai spruce forest (b) of the upper reaches (UR), the desert-oasis ecotone (c) and the Gobi (d) of the middle reaches (MR), and the riparian forest (f and g), the planted shrubland (h) and the Gobi (i) of the lower reaches (LR) of the Heihe River Basin.

Fig. 3 Relationships of δ^2H and $\delta^{18}O$ of soil water, xylem water, river water and shallow groundwater of the upper reaches (the UR) (a), the middle reaches (the MR) (b) and the lower reaches (the LR) in the riparian forest (c) and in the planted shrubland and the Gobi (d) of the Heihe River Basin. The acronyms of plants as well as the LMWL are shown in the Table 1.

Fig. 4 The $\delta^{18}O$ in soil water and groundwater at the profile and plant stem water of the mountain grassland zone (a) and the Qinghai spruce forest (b) of the upper reaches (UR**),** the desert-oasis ecotone (c) and the Gobi (d) of the middle reaches (MR**),** and the riparian forest (e), the planted shrubland (f) and the Gobi (g) of the lower reaches (LR) of the Heihe River Basin. The acronyms of plants as well as the LMWL are shown in the Table 1.

Fig. 5 The summary results of the water sources of different plants and their controlling factors along the climatic gradient of the Heihe River Basin. Figure 5a, b, c and d indicate the Qinghai spruce forest, the mountain grassland, the mountain meadow and the swamp meadow in the upper reaches (a), the desert-oasis ecotone (red dotted box) and the Gobi (green dotted box) in the middle reaches (b), the riparian forest (c), and the planted shrubland (blue dotted box) and Gobi (grey dotted box) (d) in the lower reaches, respectively. GW, SW and SSW indicate the groundwater, soil water and saturated soil water, respectively. The TR, HA, RS, PE and SA indicate *Tamarix ramosissima*, *Haloxylon ammodendron*, *Reaumuria soongoric*a, *Populus euphratica*, and *Sophora alopecuroides*, respectively.

Fig. 1

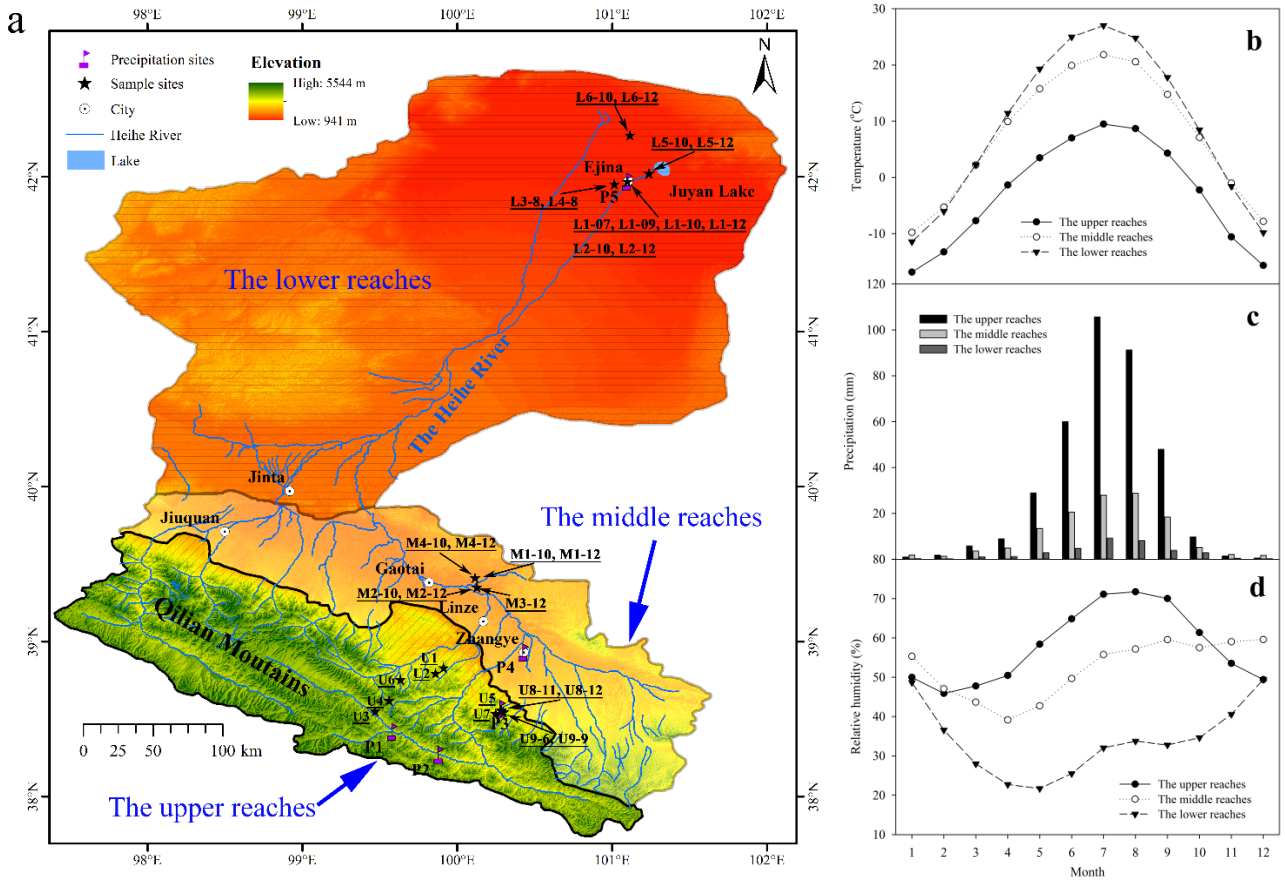
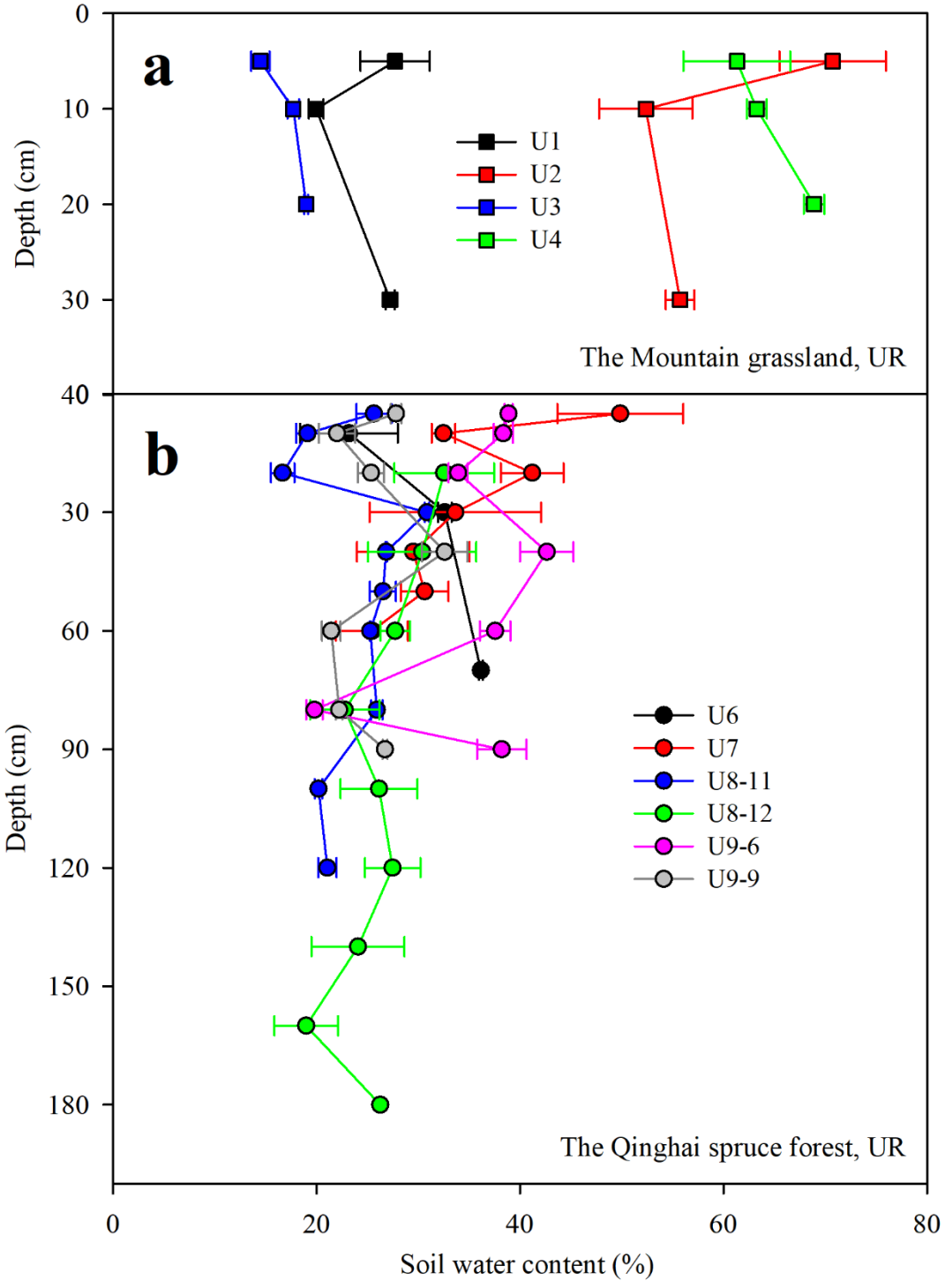
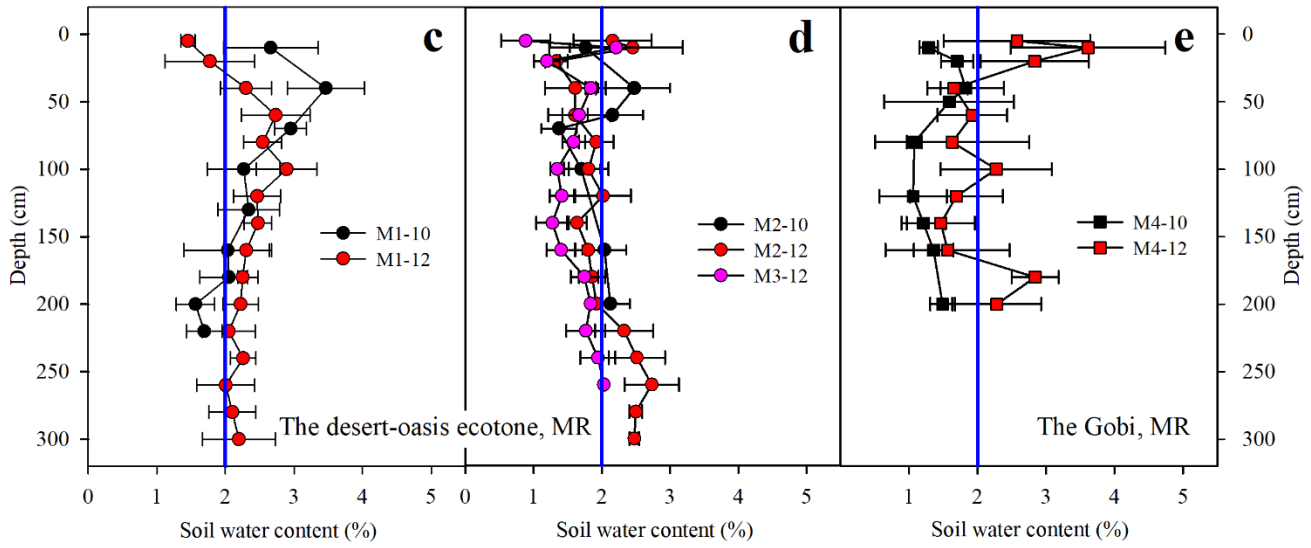
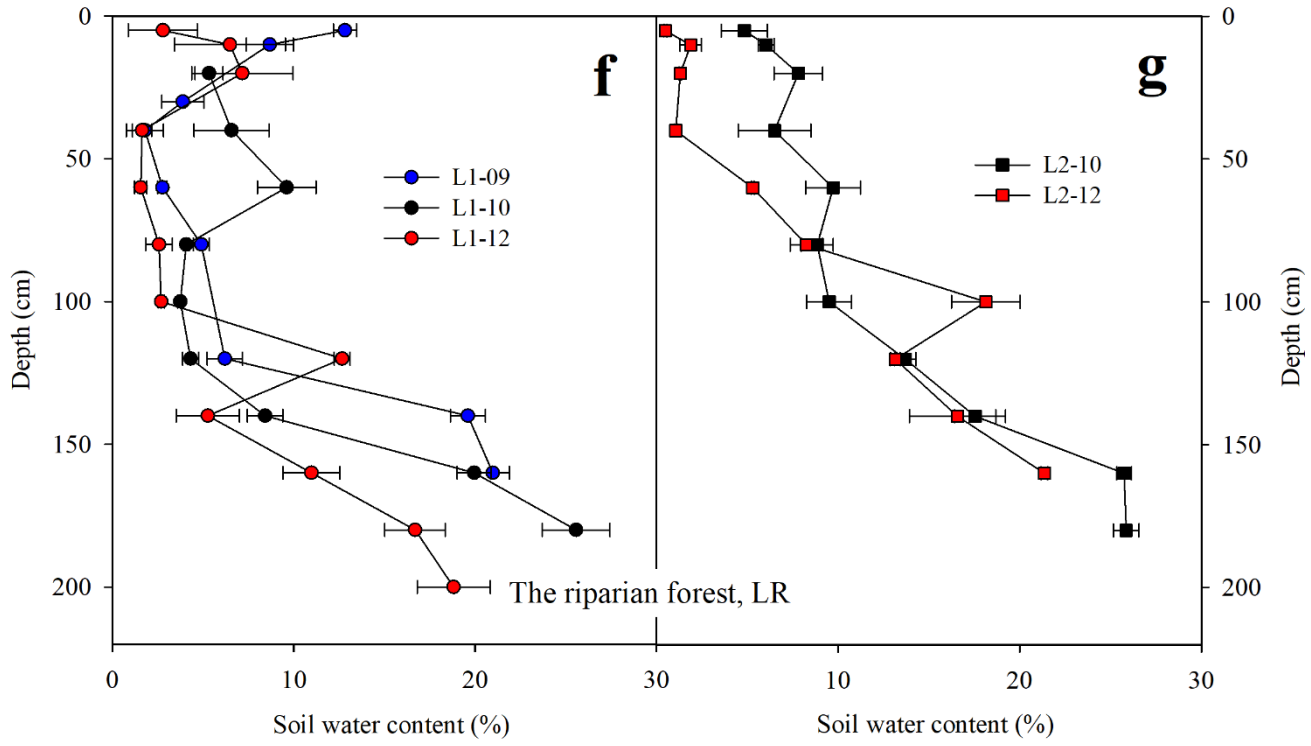

a
Precipitation sites
Sample sites
City
Heihe River
Lake
Elevation
High: 5544 m
Low: 941 m
N
98°E
99°E
100°E
101°E
102°E
42°N
41°N
40°N
39°N
38°N
L6-10, L6-12
L5-10, L5-12
Ejina
Juyan Lake
L3-8, L4-8
P5
L1-07, L1-09, L1-10, L1-12
L2-10, L2-12
The lower reaches
The Heihe River
Jinta
Jiuquan
The middle reaches
M4-10, M4-12
M1-10, M1-12
Gaotai
M2-10, M2-12
Linze
M3-12
Zhangye
Qilian Moutains
P4
U1
U2
U6
U4
U5
U7
U8
U3
P3
U8-11, U8-12
U9-6, U9-9
P1
P2
0 25 50 100 km
The upper reaches
b
Temperature (°C)
The upper reaches
The middle reaches
The lower reaches
c
Precipitation (mm)
The upper reaches
The middle reaches
The lower reaches
d
Relative humidity (%)
The upper reaches
The middle reaches
The lower reaches
1 2 3 4 5 6 7 8 9 10 11 12
Month

Fig. 2


a
b
U1
U2
U3
U4
U6
U7
U8-11
U8-12
U9-6
U9-9
The Mountain grassland, UR
The Qinghai spruce forest, UR
Depth (cm)
Depth (cm)
Soil water content (%)
0
10
20
30
40
60
90
120
150
180
0
20
40
60
80


c
d
e
M1-10
M1-12
M2-10
M2-12
M3-12
M4-10
M4-12
The desert-oasis ecotone, MR
The Gobi, MR
Depth (cm)
Soil water content (%)
0
50
100
150
200
250
300
1
2
3
4
5


f
g
L1-09
L1-10
L1-12
L2-10
L2-12
The riparian forest, LR
Depth (cm)
Soil water content (%)
0
50
100
150
200
10
20
30

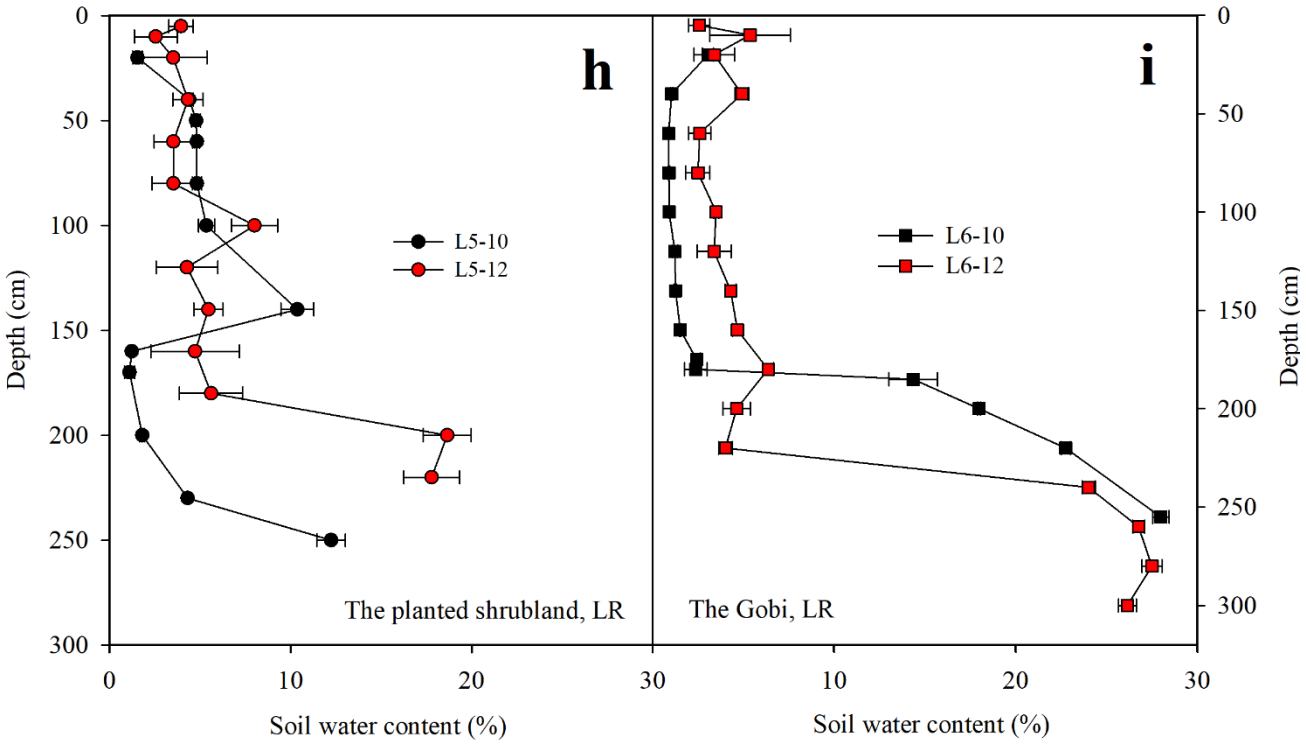
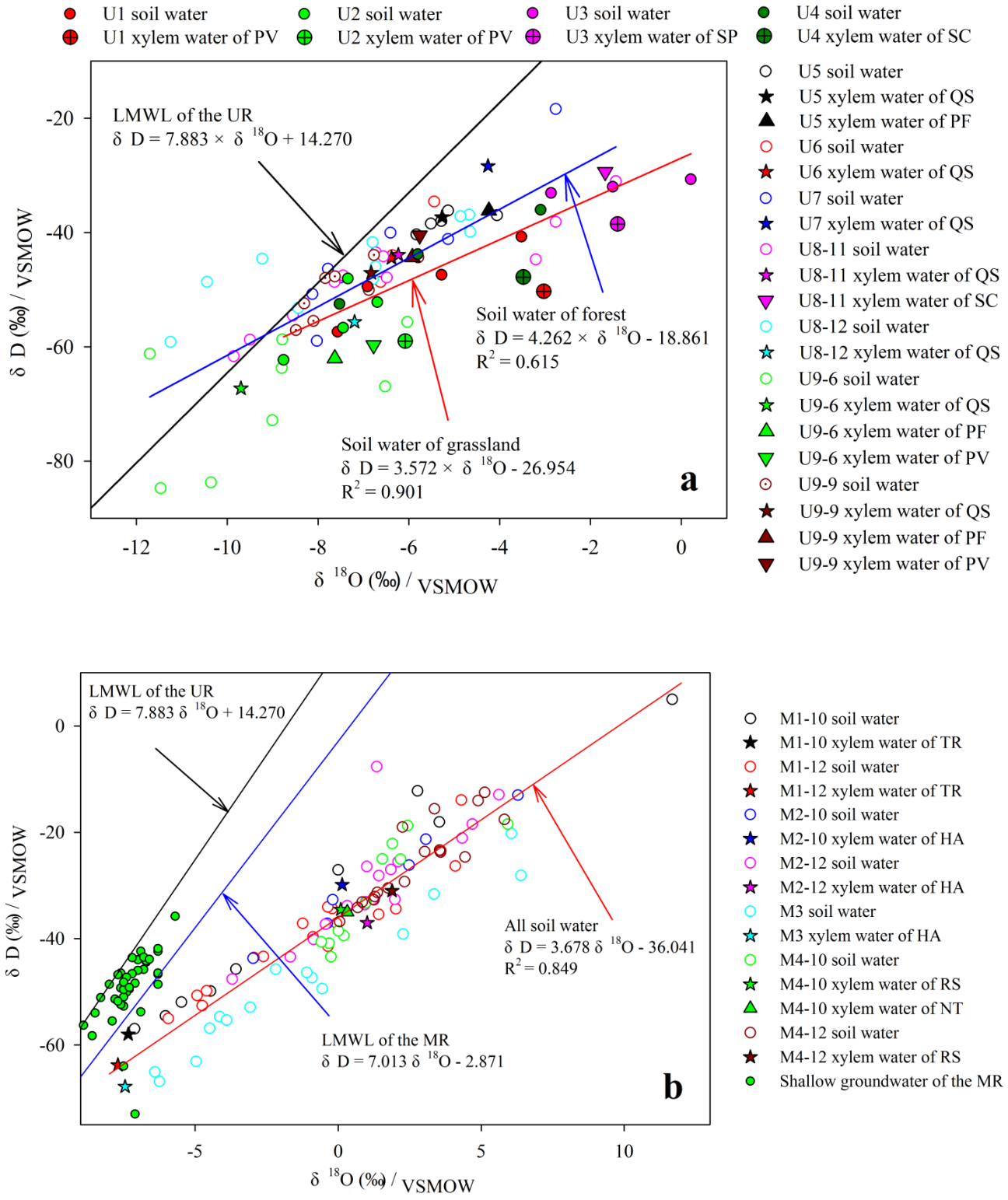
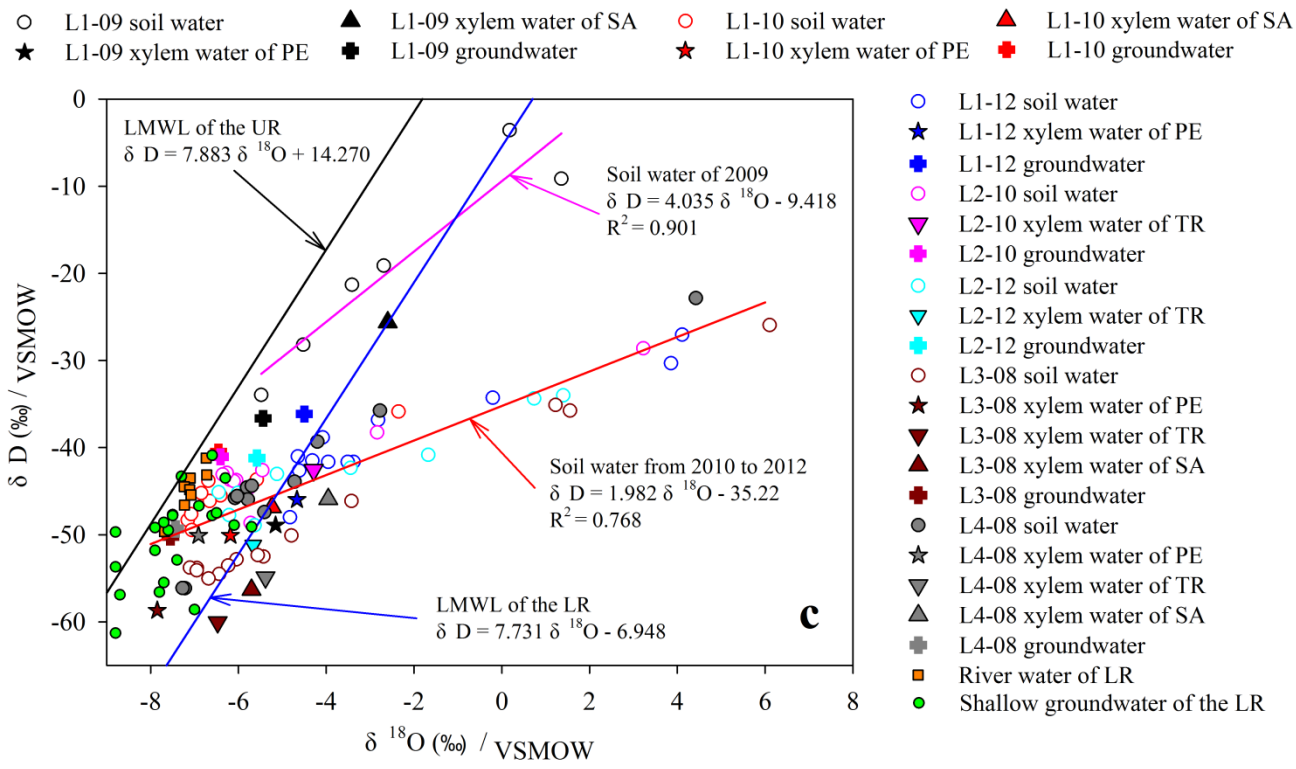

h
i
L5-10
L5-12
L6-10
L6-12
The planted shrubland, LR
The Gobi, LR
Depth (cm)
Soil water content (%)
0
50
100
150
200
250
300
10
20
30

Fig. 3


U1 soil water
U1 xylem water of PV
U2 soil water
U2 xylem water of PV
U3 soil water
U3 xylem water of SP
U4 soil water
U4 xylem water of SC
U5 soil water
U5 xylem water of QS
U5 xylem water of PF
U6 soil water
U6 xylem water of QS
U7 soil water
U7 xylem water of QS
U8-11 soil water
U8-11 xylem water of QS
U8-11 xylem water of SC
U8-12 soil water
U8-12 xylem water of QS
U9-6 soil water
U9-6 xylem water of QS
U9-6 xylem water of PF
U9-6 xylem water of PV
U9-9 soil water
U9-9 xylem water of QS
U9-9 xylem water of PF
U9-9 xylem water of PV
LMWL of the UR
δ D = 7.883 × δ 18O + 14.270
Soil water of forest
δ D = 4.262 × δ 18O - 18.861
R2 = 0.615
Soil water of grassland
δ D = 3.572 × δ 18O - 26.954
R2 = 0.901
a
δ D (‰) / VSMOW
δ 18O (‰) / VSMOW
-20
-40
-60
-80
-12
-10
-8
-6
-4
-2
0
LMWL of the UR
δ D = 7.883 δ 18O + 14.270
All soil water
δ D = 3.678 δ 18O - 36.041
R2 = 0.849
LMWL of the MR
δ D = 7.013 δ 18O - 2.871
b
M1-10 soil water
M1-10 xylem water of TR
M1-12 soil water
M1-12 xylem water of TR
M2-10 soil water
M2-10 xylem water of HA
M2-12 soil water
M2-12 xylem water of HA
M3 soil water
M3 xylem water of HA
M4-10 soil water
M4-10 xylem water of RS
M4-10 xylem water of NT
M4-12 soil water
M4-12 xylem water of RS
Shallow groundwater of the MR
0
-20
-40
-60
-5
0
5
10


L1-09 soil water
L1-09 xylem water of PE
L1-09 xylem water of SA
L1-09 groundwater
L1-10 soil water
L1-10 xylem water of PE
L1-10 xylem water of SA
L1-10 groundwater
LMWL of the UR
δ D = 7.883 δ 18O + 14.270
Soil water of 2009
δ D = 4.035 δ 18O - 9.418
R2 = 0.901
Soil water from 2010 to 2012
δ D = 1.982 δ 18O - 35.22
R2 = 0.768
LMWL of the LR
δ D = 7.731 δ 18O - 6.948
c
δ D (‰) / VSMOW
δ 18O (‰) / VSMOW
L1-12 soil water
L1-12 xylem water of PE
L1-12 groundwater
L2-10 soil water
L2-10 xylem water of TR
L2-10 groundwater
L2-12 soil water
L2-12 xylem water of TR
L2-12 groundwater
L3-08 soil water
L3-08 xylem water of PE
L3-08 xylem water of TR
L3-08 xylem water of SA
L3-08 groundwater
L4-08 soil water
L4-08 xylem water of PE
L4-08 xylem water of TR
L4-08 xylem water of SA
L4-08 groundwater
River water of LR
Shallow groundwater of the LR

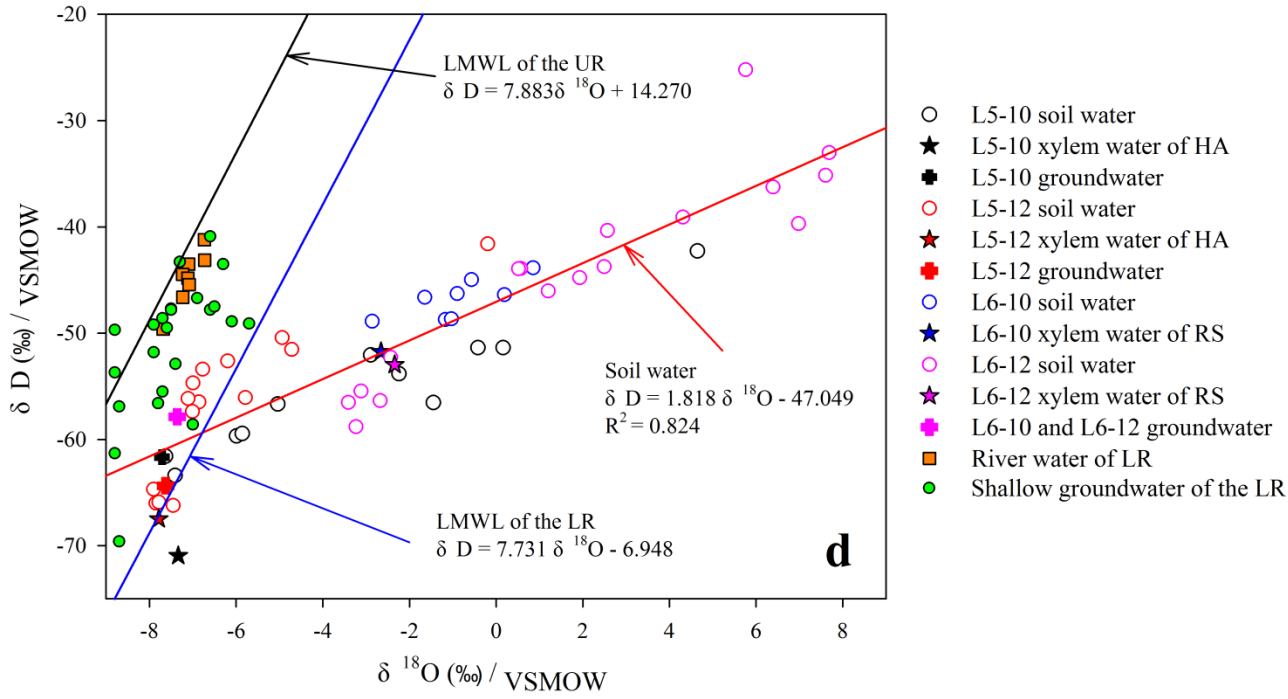
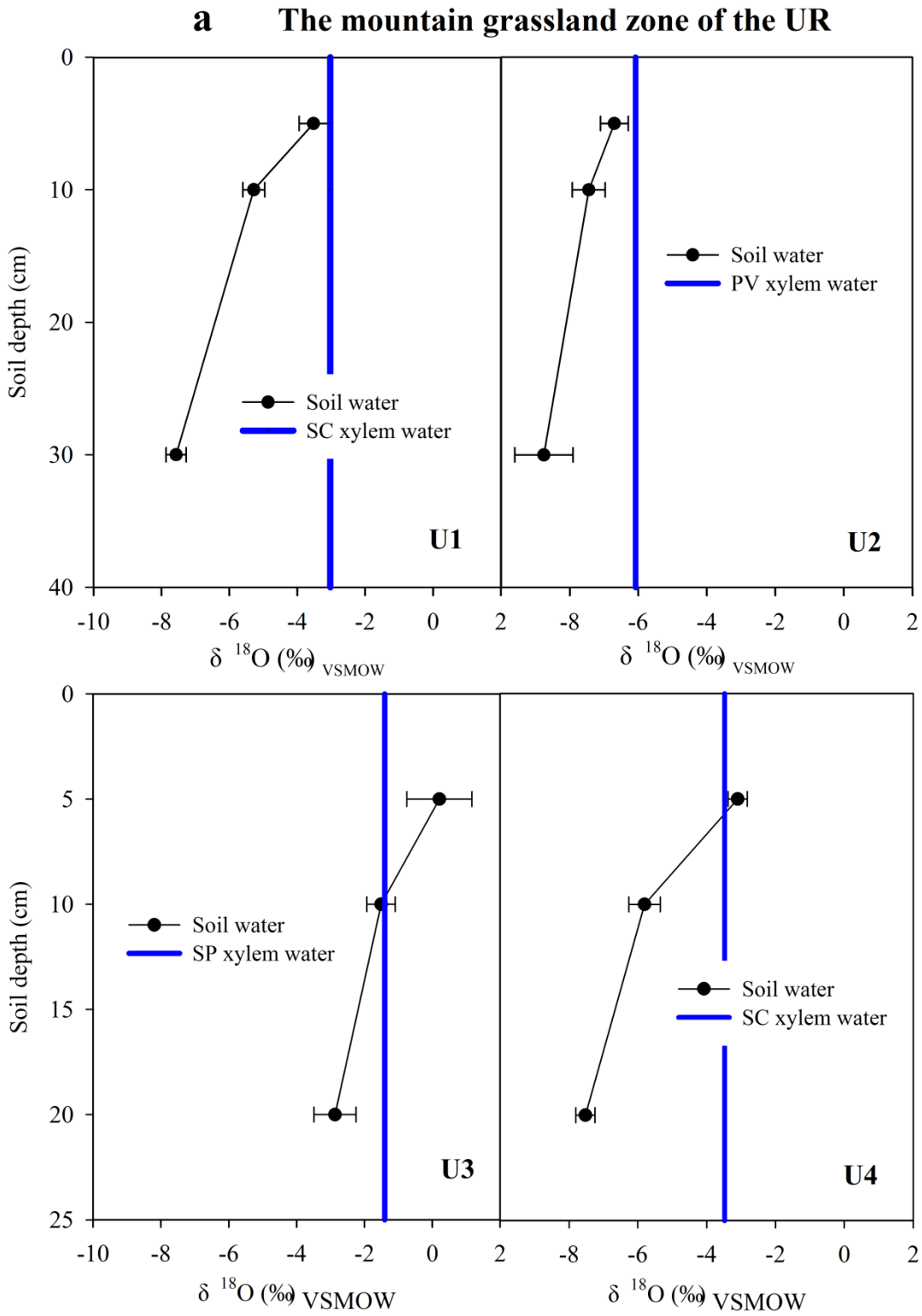
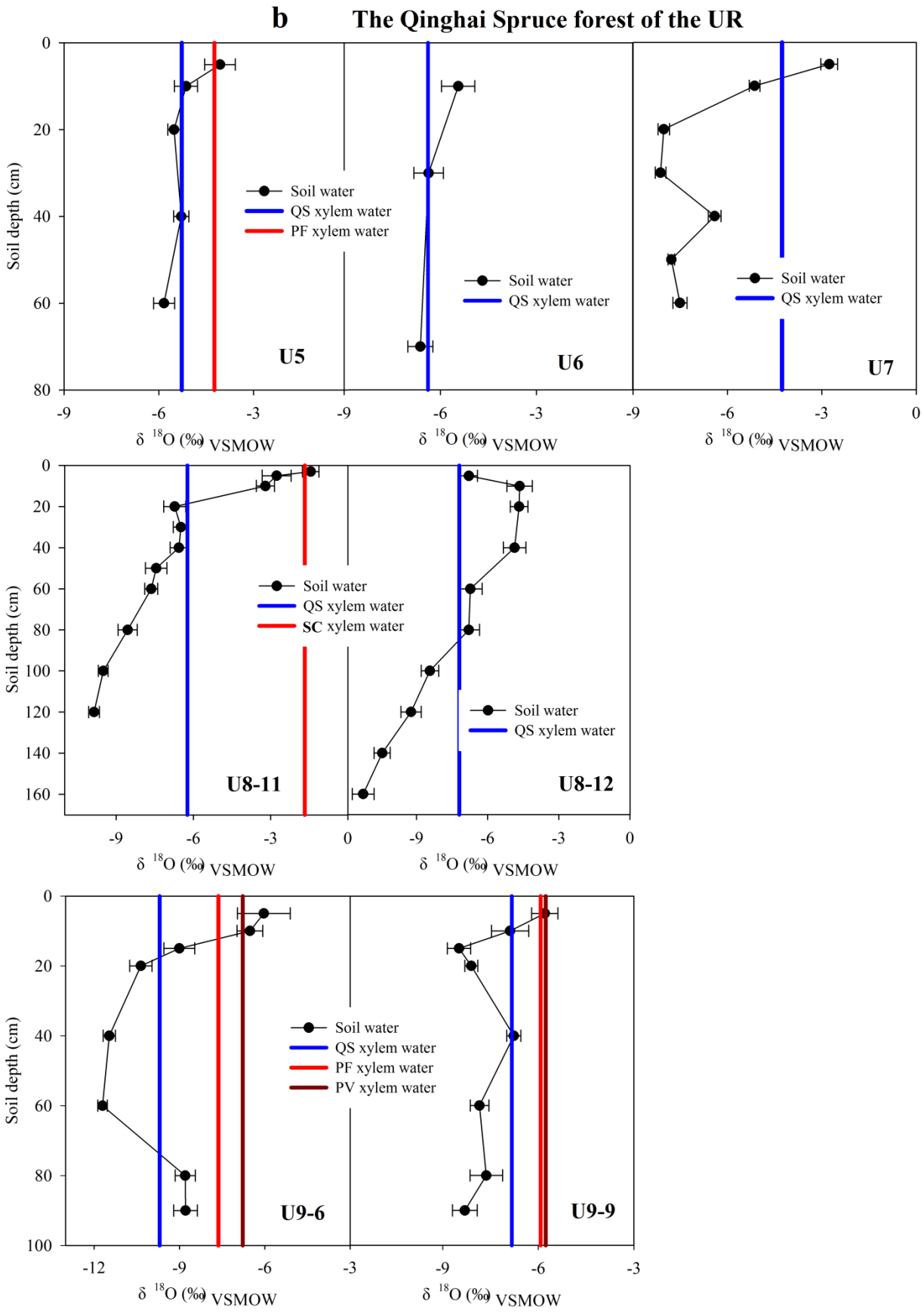
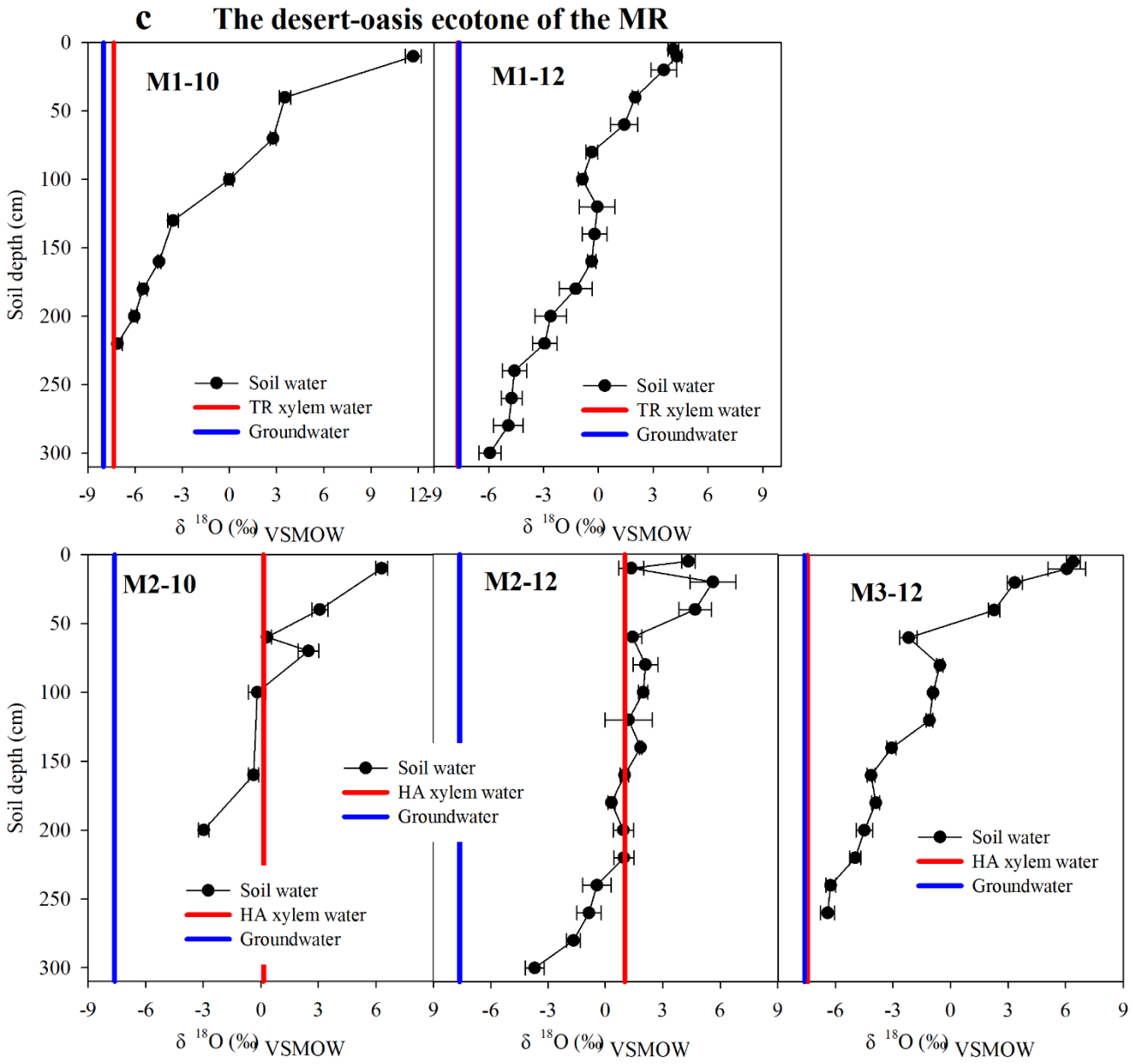
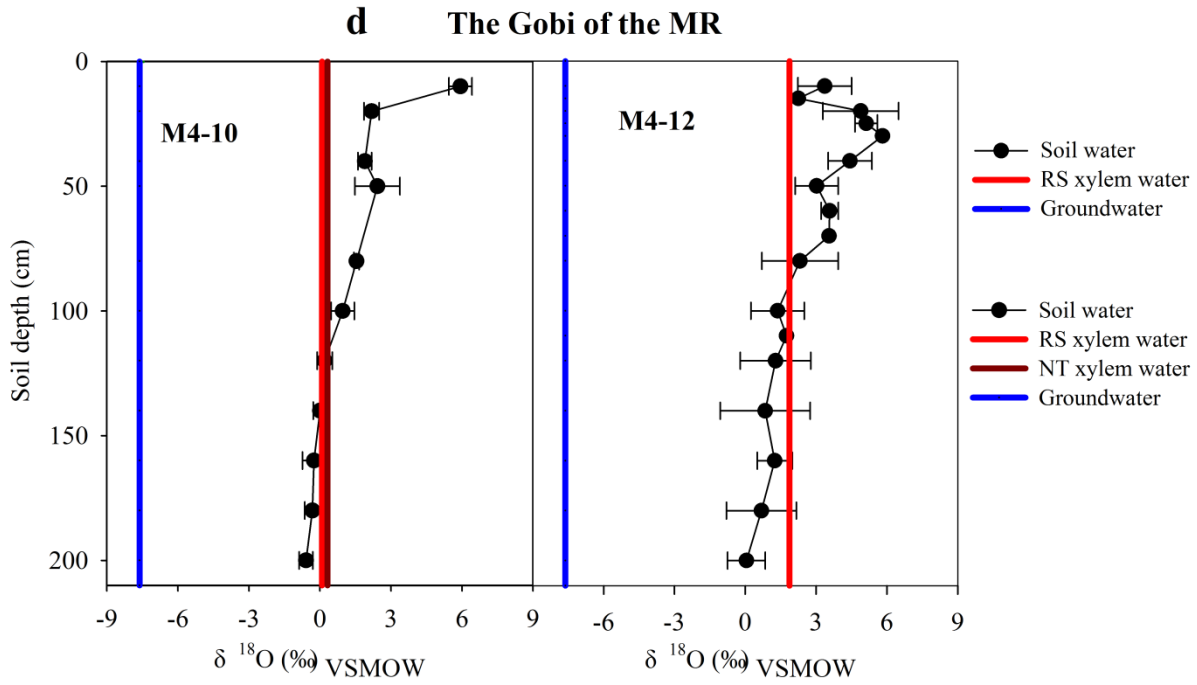

LMWL of the UR
δ D = 7.883δ 18O + 14.270
Soil water
δ D = 1.818 δ 18O - 47.049
R2 = 0.824
LMWL of the LR
δ D = 7.731 δ 18O - 6.948
d
δ D (‰) / VSMOW
δ 18O (‰) / VSMOW
L5-10 soil water
L5-10 xylem water of HA
L5-10 groundwater
L5-12 soil water
L5-12 xylem water of HA
L5-12 groundwater
L6-10 soil water
L6-10 xylem water of RS
L6-12 soil water
L6-12 xylem water of RS
L6-10 and L6-12 groundwater
River water of LR
Shallow groundwater of the LR

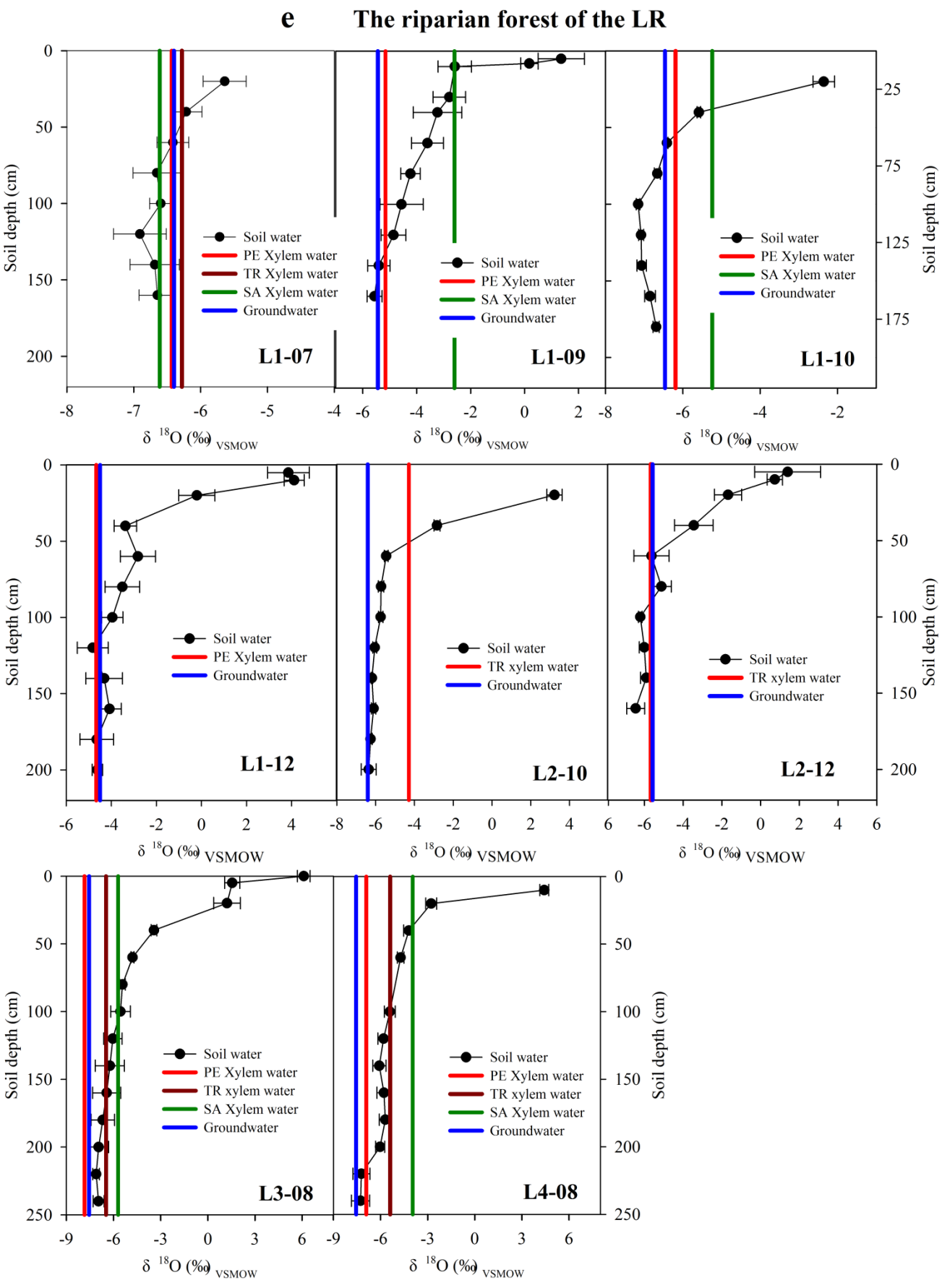
Fig. 4

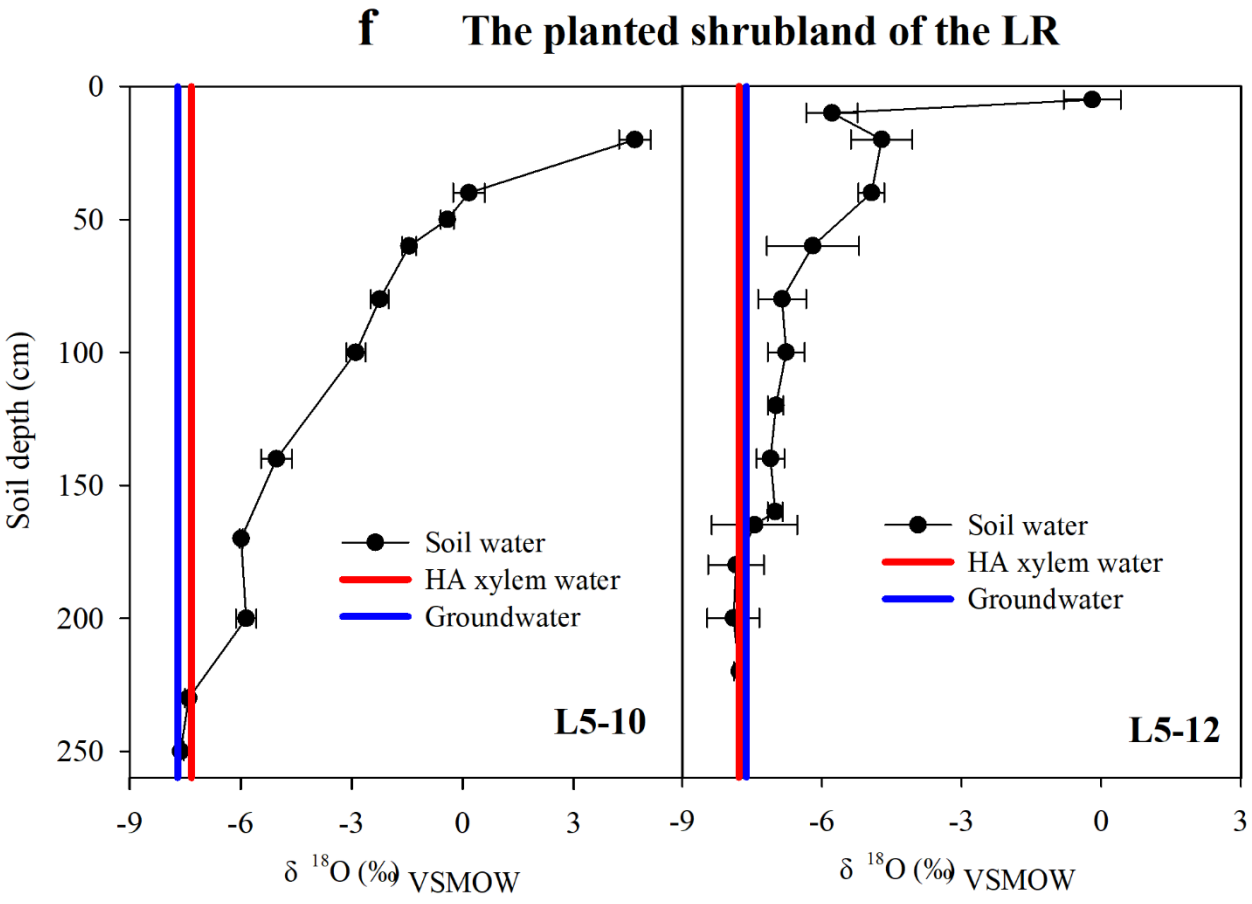

a The mountain grassland zone of the UR
Soil depth (cm)
Soil water
SC xylem water
PV xylem water
SP xylem water
U1
U2
U3
U4
δ 18O (‰) VSMOW


b
The Qinghai Spruce forest of the UR
Soil depth (cm)
Soil water
QS xylem water
PF xylem water
U5
U6
U7
δ 18O (‰) VSMOW
SC xylem water
U8-11
U8-12
PV xylem water
U9-6
U9-9


c The desert-oasis ecotone of the MR
M1-10
M1-12
M2-10
M2-12
M3-12
Soil depth (cm)
Soil water
TR xylem water
HA xylem water
Groundwater
δ 18O (‰) VSMOW


d
The Gobi of the MR
M4-10
M4-12
Soil depth (cm)
0
50
100
150
200
-9
-6
-3
0
3
6
9
δ 18O (‰) VSMOW
Soil water
RS xylem water
Groundwater
Soil water
RS xylem water
NT xylem water
Groundwater


e The riparian forest of the LR
Soil depth (cm)
δ 18O (‰) VSMOW
Soil water
PE Xylem water
TR Xylem water
SA Xylem water
Groundwater
L1-07
L1-09
L1-10
L1-12
L2-10
L2-12
L3-08
L4-08


f The planted shrubland of the LR
Soil depth (cm)
0
50
100
150
200
250
Soil water
HA xylem water
Groundwater
L5-10
L5-12
-9
-6
-3
0
3
δ 18O (‰) VSMOW

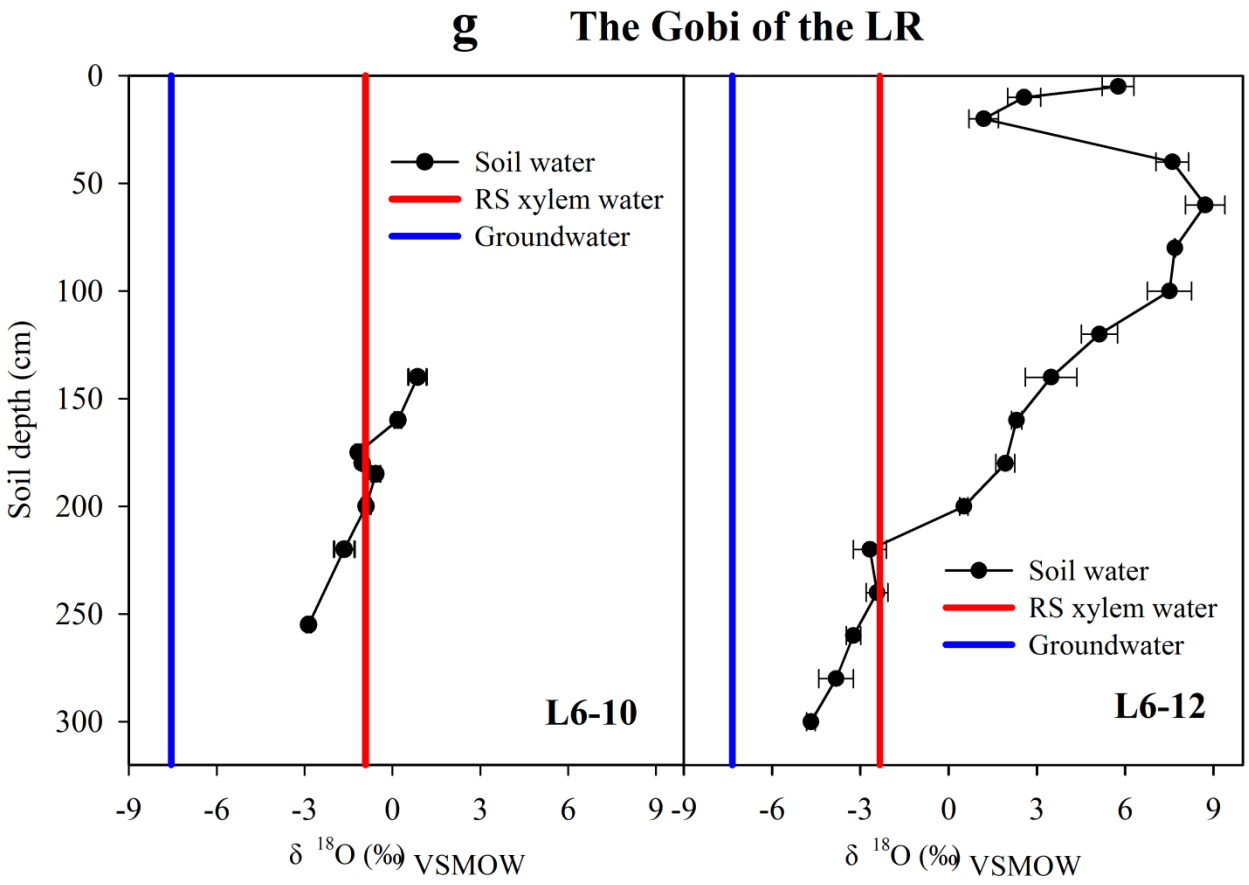

g
The Gobi of the LR
Soil depth (cm)
0
50
100
150
200
250
300
Soil water
RS xylem water
Groundwater
L6-10
L6-12
-9
-6
-3
0
3
6
9
δ 18O (‰) VSMOW

Fig. 5

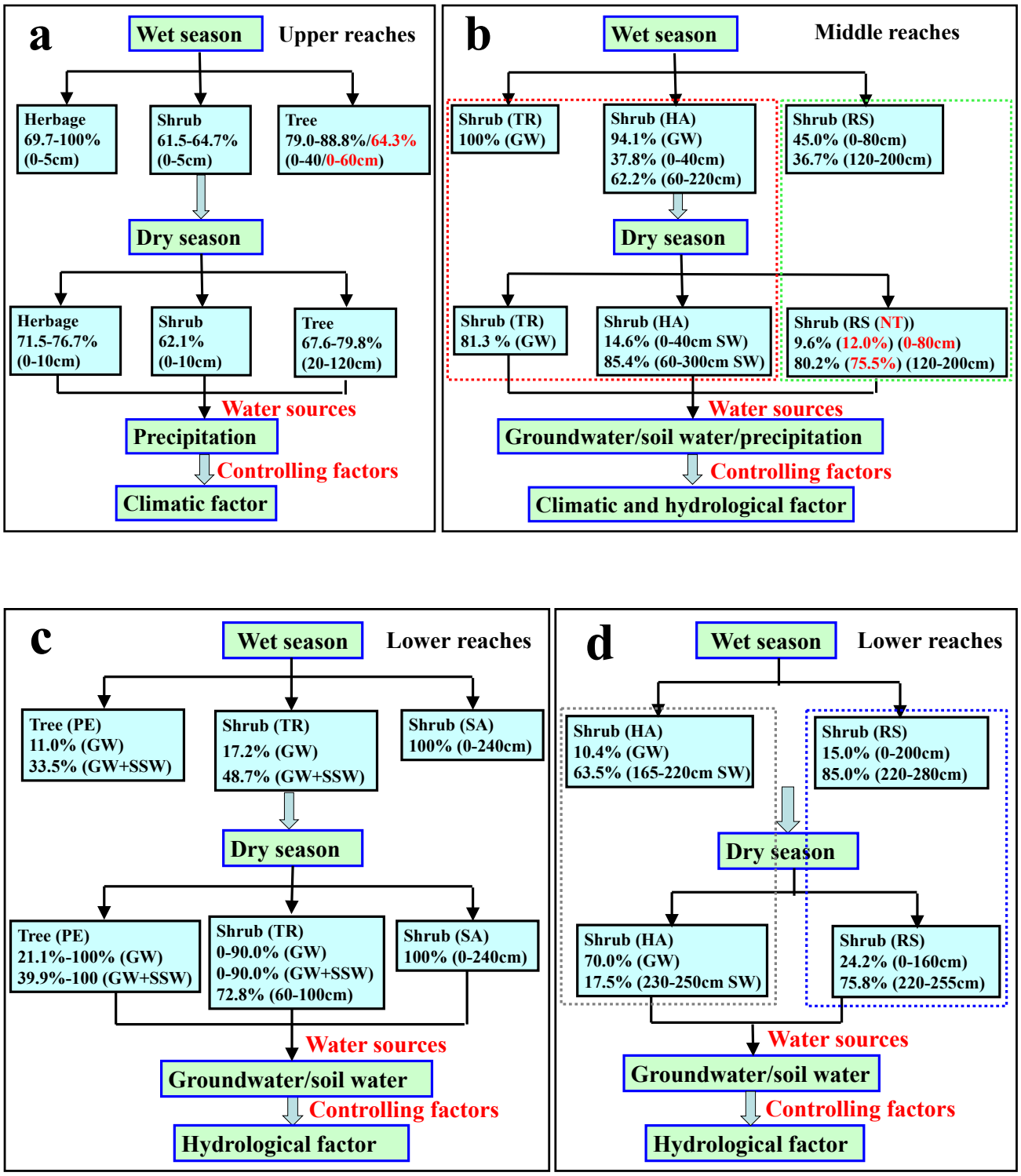

a
Wet season
Upper reaches
Herbage
69.7-100%
(0-5cm)
Shrub
61.5-64.7%
(0-5cm)
Tree
79.0-88.8%/64.3%
(0-40/0-60cm)
Dry season
Herbage
71.5-76.7%
(0-10cm)
Shrub
62.1%
(0-10cm)
Tree
67.6-79.8%
(20-120cm)
Water sources
Precipitation
Controlling factors
Climatic factor
b
Wet season
Middle reaches
Shrub (TR)
100% (GW)
Shrub (HA)
94.1% (GW)
37.8% (0-40cm)
62.2% (60-220cm)
Shrub (RS)
45.0% (0-80cm)
36.7% (120-200cm)
Dry season
Shrub (TR)
81.3 % (GW)
Shrub (HA)
14.6% (0-40cm SW)
85.4% (60-300cm SW)
Shrub (RS (NT))
9.6% (12.0%) (0-80cm)
80.2% (75.5%) (120-200cm)
Water sources
Groundwater/soil water/precipitation
Controlling factors
Climatic and hydrological factor
c
Wet season
Lower reaches
Tree (PE)
11.0% (GW)
33.5% (GW+SSW)
Shrub (TR)
17.2% (GW)
48.7% (GW+SSW)
Shrub (SA)
100% (0-240cm)
Dry season
Tree (PE)
21.1%-100% (GW)
39.9%-100 (GW+SSW)
Shrub (TR)
0-90.0% (GW)
0-90.0% (GW+SSW)
72.8% (60-100cm)
Shrub (SA)
100% (0-240cm)
Water sources
Groundwater/soil water
Controlling factors
Hydrological factor
d
Wet season
Lower reaches
Shrub (HA)
10.4% (GW)
63.5% (165-220cm SW)
Shrub (RS)
15.0% (0-200cm)
85.0% (220-280cm)
Dry season
Shrub (HA)
70.0% (GW)
17.5% (230-250cm SW)
Shrub (RS)
24.2% (0-160cm)
75.8% (220-255cm)
Water sources
Groundwater/soil water
Controlling factors
Hydrological factor

Table S1 Comparison of contribution rates (%±SD) of different water sources to plant species between deuterium fractionation (DF) and deuterium non-fractionation (NDF) using the Bayesian isotope mixing model (MixSIAR). The acronyms of plants are the same as the Table 1.

	$\delta^{18}O$ and $\delta^{2}H$ of soil water (‰)							$\delta^{18}O/\delta^{2}H$ of
U7-DNF	5cm SW	10cm SW	20cm SW	30cm SW	40cm SW	50cm SW	60cm SW	xylem water (‰)
$\delta^{18}O$	-2.76	-5.14	-8.02	-8.13	-6.41	-7.78	-7.51	$\delta^{18}O_{QS}$ = -4.26
$\delta^{2}H$	-18.39	-41.16	-58.97	-50.75	-40.03	-46.36	-47.89	$\delta^{2}H_{QS}$ = -28.37
Mix-$\delta^{18}O$	63.5±0.06	8.4±0.08	5.3±0.04	5.1±0.04	6.5±0.05	5.4±0.04	5.7±0.04	
Mix-^{2}H	64.7±0.03	6.9±0.05	4.8±0.05	5.3±0.04	6.6±0.05	6.0±0.04	5.6±0.04	
Mix-$\delta^{18}O/\delta^{2}H$	64.8±0.03	6.7±0.05	4.8±0.03	5.3±0.04	6.7±0.05	6.0±0.04	5.7±0.04	
U9-9- DNF	3~5cm SW	5~10cm SW	10~20cm SW	20~40cm SW	40-60cm SW	80~90cm SW		$\delta^{18}O_{QS}$ = -6.83
$\delta^{18}O$	-5.8	-6.88	-8.29	-6.77	-7.74	-8.31		$\delta^{2}H_{QS}$ = -47.06
$\delta^{2}H$	-44.36	-50.06	-56.28	-43.94	-47.83	-52.41		
Mix-$\delta^{18}O$	27.3±0.15	19.9±0.16	9.9±0.07	21.9±0.19	11.7±0.09	9.4±0.07		
Mix-^{2}H	26.4±0.18	11.8±0.09	7.5±0.05	27.4±0.17	17.3±0.15	9.5±0.06		
Mix-$\delta^{18}O/\delta^{2}H$	32.0±0.15	13.3±0.09	7.6±0.05	24.7±0.16	13.4±0.10	9.1±0.06		
Mix-$\delta^{18}O$	64.7±0.18	12.5±0.17	4.9±0.04	7.5±0.06	5.4±0.04	5.0±0.04		$\delta^{18}O_{PF}$ = -5.92
Mix-^{2}H	**33.4±0.31**	4.8±0.04	3.2±0.02	**48.9±0.31**	5.8±0.05	4.0±0.03		$\delta^{2}H_{PF}$=-44.32
Mix-$\delta^{18}O/\delta^{2}H$	76.8±0.09	4.3±0.03	2.8±0.02	8.2±0.08	4.5±0.04	3.3±0.02		
M4-12-DNF	10-15cm SW	20-40cm SW	50-70cm SW	80cm SW	100-120cm SW	120-160cm SW	180-200cm SW	$\delta^{18}O_{RS}$ = 1.88
$\delta^{18}O$	2.81	5.07	3.38	2.32	1.47	1.05	0.37	$\delta^{2}H_{RS}$ = -31.02
$\delta^{2}H$	-17.31	-17.2	-23.58	-29.29	-31.3	-32.92	-35.48	
Mix-$\delta^{18}O$	12.2±0.10	7.8±0.05	10.6±0.08	14.4±0.13	18.2±0.15	18.3±0.15	18.4±0.13	
Mix-^{2}H	5.0±0.03	5.1±0.04	6.7±0.05	11.5±0.10	17.0±0.17	23.8±0.21	30.9±0.19	
Mix-$\delta^{18}O/\delta^{2}H$	4.6±0.03	5.5±0.04	7.0±0.05	12.9±0.11	18.5±0.18	24.4±0.21	27.2±0.18	
L6-12- DNF	5-20cm SW	40-100cm SW	120cm SW	140-160cm SW	200cm SW	220-240cm SW	260-280cm SW	δ^{18}_{RS} = -2.34
$\delta^{18}O$	2.97	7.17	4.31	1.87	0.52	-2.55	-3.25	$\delta^{2}H_{RS}$ = -52.97

δ^2H	-36.5	-36.03	-39.08	-44.86	-43.94	-54.32	-56.93	
Mix-$\delta^{18}O$	2.9±0.02	2.2±0.01	2.7±0.02	3.3±0.03	3.8±0.03	18.4±0.26	66.6±0.25	
Mix-^2H	4.7±0.03	4.7±0.03	5.1±0.04	6.1±0.05	6.1±0.05	29.3±0.26	**44.0±0.26**	
Mix-$\delta^{18}O/\delta^2H$	3.2±0.02	2.3±0.02	2.7±0.02	3.5±0.03	4.0±0.03	20.2±0.26	64.2±0.25	
M1-10-DF	10cm SW	40-70cm SW	100cm SW	130-160cm SW	180-200cm SW	220cm SW	GW	$\delta^{18}O_{TR}$ =-7.34
$\delta^{18}O$	11.68	3.16	0.00	-4.02	-5.76	-7.12	-7.99	δ^2H_{TR} = -58.00
δ^2H	4.99	-15.14	-27.09	-47.81	-53.25	-56.96	-50.17	
Mix-$\delta^{18}O$	1.1±0.01	1.6±0.01	1.8±0.01	2.6±0.02	3.3±0.03	8.3±0.17	**81.3±0.16**	
Mix-^2H	0.6±0.00	0.8±0.01	0.9±0.01	1.5±0.01	1.9±0.02	92.6±0.03	1.7±0.02	
Mix-$\delta^{18}O/\delta^2H$	0.5±0.00	0.6±0.00	0.7±0.01	1.2±.0.01	1.6±0.01	93.8±0.03	1.7±0.02	
L2-12-DF	5-10cm SW	20cm SW	40cm SW	60-80cm SW	100-140cm SW	160cm SW	GW	$\delta^{18}O_{TR}$ = -5.66
$\delta^{18}O$	1.07	-1.68	-3.45	-5.38	-6.04	-6.45	-5.58	δ^2H_{TR} = -51.19
δ^2H	-34.19	-40.84	-42.33	-45.98	-46.11	-45.13	-41.2	
Mix-$\delta^{18}O$	3.4±0.22	4.5±0.03	6.8±0.07	13.5±0.15	23.0±0.22	31.5±0.22	**17.2±0.18**	
Mix-^2H	7.4±0.11	13.1±0.18	6.6±0.05	14.9±0.19	8.8±0.09	42.6±0.24	6.6±0.06	
Mix-$\delta^{18}O/\delta^2H$	4.5±0.04	6.7±0.09	6.1±0.05	14.2±0.18	10.4±0.12	51.1±0.20	7.0±0.07	
L5-10-DF	20cm SW	40-60cm SW	80-100cm SW	140-160cm SW	170-200cm SW	230-250cm SW	GW	$\delta^{18}O_{HA}$ = -7.34
$\delta^{18}O$	4.65	-0.57	-2.57	-5.04	-5.92	-7.52	-7.71	δ^2H_{HA} = -70.94
δ^2H	-42.28	-53.09	-52.94	-56.68	-59.56	-62.49	-61.66	
Mix-$\delta^{18}O$	1.5±0.01	1.9±0.01	2.3±0.02	3.1±0.03	3.8±0.03	**17.5±0.28**	**70.0±0.26**	
Mix-^2H	0.1±0.00	0.1±0.00	0.1±0.00	0.2±0.00	0.2±0.00	**99.1±0.00**	**0.2±0.00**	
Mix-$\delta^{18}O/\delta^2H$	0.1±0.00	0.1±0.00	0.1±0.00	0.1±0.00	0.2±0.00	**49.5±0.50**	**49.9±0.43**	
L3-08-DF	0-5cm SW	5-20cm SW	40-60cm SW	80-120cm SW	140-180cm SW	200-240cm SW	GW	$\delta^{18}O_{PE}$= -7.85
$\delta^{18}O$	6.11	1.39	-4.11	-5.68	-6.45	-7	-7.55	δ^2H_{PE} = -58.69
δ^2H	-25.94	-35.43	-48.12	-52.57	-54.35	-53.91	-50.2	
Mix-$\delta^{18}O$	1.3±0.01	1.6±0.01	2.5±0.02	3.2±0.03	**57.1±0.04**	**5.8±0.12**	28.6±0.38	
Mix-^2H	0.7±0.01	0.9±0.01	**68.0±0.40**	1.6±0.02	1.8±0.02	1.8±0.02	25.3±0.41	

Mix-$\delta^{18}O/\delta^{2}H$	0.3±0.00	0.3±0.00	0.5±0.00	0.7±0.01	0.8±0.01	0.8±0.01	**96.6±0.02**	
Mix-$\delta^{18}O$	2.5±0.00	3.1±0.00	5.4±0.00	9.1±0.10	***18.2±0.20***	**24.0±0.20**	37.7±0.30	$\delta^{18}O_{TR}$ = -6.47
Mix-^{2}H	0.3±0.00	0.3±0.00	0.5±0.00	0.6±0.01	***0.7±0.01***	0.6±0.01	**97.0±0.01**	$\delta^{2}H_{TR}$ = -60.03
Mix-$\delta^{18}O/\delta^{2}H$	0.3±0.00	0.4±0.00	0.6±0.01	0.7±0.01	***48.7±0.50***	0.7±0.01	48.6±0.50	
Mix-$\delta^{18}O$	3.6±0.02	4.7±0.03	8.7±0.07	18.3±0.19	**28.4±0.24**	**36.4±0.25**		$\delta^{18}O_{SA}$ = -5.70
Mix-^{2}H	0.8±0.01	1.0±0.01	1.6±0.01	2.2±0.02	92.1±0.04	2.4±0.02		$\delta^{2}H_{SA}$= -56.31
Mix-$\delta^{18}O/\delta^{2}H$	0.8±0.01	1.0±0.01	1.6±0.01	2.2±0.02	91.9±0.04	2.5±0.02		

Note: Data of red bold indicate both $\delta^{18}O$ and $\delta^{2}H$ values of plant xylem water were not within the range of values of all potential water sources.

Table S2 Contribution rates (%±SD) of different water sources to plant species with deuterium fractionation (DF) based on $\delta^{18}O$ alone, $\delta^{2}H$ alone and both $\delta^{18}O$ and $\delta^{2}H$ by the Iso-Source model (Iso-$\delta^{18}O$, Iso-$\delta^{2}H$ and Iso-$\delta^{18}O/\delta^{2}H$) and the Bayesian isotope mixing model (Mix-$\delta^{18}O$, Mix -$\delta^{2}H$ and Mix -$\delta^{18}O/\delta^{2}H$). The acronyms of plants are the same as the Table 1.

	$\delta^{18}O$ and $\delta^{2}H$ of soil water (‰)							$\delta^{18}O/\delta^{2}H$ of
M1-10-DF	10cm SW	40-70cm SW	100cm SW	130-160cm SW	180-200cm SW	220cm SW	GW	xylem water (‰)
$\delta^{18}O$	11.68	3.16	0	-4.02	-5.76	-7.12	-7.99	$\delta^{18}O_{TR}$ = -7.34
$\delta^{2}H$	4.99	-15.14	-27.09	-47.81	-53.25	-56.96	-50.17	$\delta^{2}H_{TR}$ = -58.00
Iso-$\delta^{18}O$	0.3±0.01	0.8±0.01	1.3±0.02	3.1±0.03	5.9±0.05	15.9±0.14	72.6±0.12	
Mix-$\delta^{18}O$	**1.1±0.01**	**1.6±0.01**	**1.8±0.01**	**2.6±0.02**	**3.3±0.03**	**8.3±0.17**	**81.3±0.16**	
Iso-$\delta^{2}H$	/	/	/	/	/	/	/	
Mix-$\delta^{2}H$	**0.6±0.00**	**0.8±0.01**	**0.9±0.01**	**1.5±0.01**	**1.9±0.02**	**92.6±0.03**	**1.7±0.02**	
Iso-$\delta^{18}O/\delta^{2}H$	/	/	/	/	/	/	/	
Mix-$\delta^{18}O/\delta^{2}H$	**0.5±0.00**	**0.6±0.00**	**0.7±0.01**	**1.2±.0.01**	**1.6±0.01**	**93.8±0.03**	**1.7±0.02**	
L2-12-DF	5-10cm SW	20cm SW	40cm SW	60-80cm SW	100-140cm SW	160cm SW	GW	$\delta^{18}O_{TR}$ = -5.66
$\delta^{18}O$	1.07	-1.68	-3.45	-5.38	-6.04	-6.45	-5.58	$\delta^{2}H_{TR}$ = -51.19
$\delta^{2}H$	-34.19	-40.84	-42.33	-45.98	-46.11	-45.13	-41.2	
Iso-$\delta^{18}O$	1.6±0.02	2.8±0.03	4.7±0.04	13.5±0.12	25.8±0.20	35.3±0.18	16.3±0.14	
Mix-$\delta^{18}O$	**3.4±0.22**	**4.5±0.03**	**6.8±0.07**	**13.5±0.15**	**23.0±0.22**	**31.5±0.22**	**17.2±0.18**	
Iso-$\delta^{2}H$	/	/	/	/	/	/	/	
Mix-$\delta^{2}H$	**7.4±0.11**	**13.1±0.18**	**6.6±0.05**	**14.9±0.19**	**8.8±0.09**	**42.6±0.24**	**6.6±0.06**	
Iso-$\delta^{18}O/\delta^{2}H$	/	/	/	/	/	/	/	
Mix-$\delta^{18}O/\delta^{2}H$	**4.5±0.04**	**6.7±0.09**	**6.1±0.05**	**14.2±0.18**	**10.4±0.12**	**51.1±0.20**	**7.0±0.07**	
L5-10-DF	20cm SW	40-60cm SW	80-100cm SW	140-160cm SW	170-200cm SW	230-250cm SW	GW	$\delta^{18}O_{HA}$ = -7.34
$\delta^{18}O$	4.65	-0.57	-2.57	-5.04	-5.92	-7.52	-7.71	$\delta^{2}H_{HA}$ = -70.94
$\delta^{2}H$	-42.28	-53.09	-52.94	-56.68	-59.56	-62.49	-61.66	
Iso-$\delta^{18}O$	0.3±0.01	0.8±0.01	1.3±0.01	2.8±0.03	4.5±0.04	33.8±0.03	56.4±0.03	
Mix-$\delta^{18}O$	**1.5±0.01**	**1.9±0.01**	**2.3±0.02**	**3.1±0.03**	**3.8±0.03**	**17.5±0.28**	**70.0±0.26**	

Iso-δ^2H	/	/	/	/	/	/	/	
Mix-δ^2H	**0.1±0.00**	**0.1±0.00**	**0.1±0.00**	**0.2±0.00**	**0.2±0.00**	**99.1±0.00**	**0.2±0.00**	
Iso-$\delta^{18}O/\delta^2H$	/	/	/	/	/	/	/	
Mix-$\delta^{18}O/\delta^2H$	**0.1±0.00**	**0.1±0.00**	**0.1±0.00**	**0.1±0.00**	**0.2±0.00**	**49.5±0.50**	**49.9±0.43**	
L3-08-DF	0-5cm SW	5-20cm SW	40-60cm SW	80-120cm SW	140-180cm SW	200-240cm SW	GW	$\delta^{18}O_{PE}$= -7.85
$\delta^{18}O$	6.11	1.39	-4.11	-5.68	-6.45	-7.00	-7.55	δ^2H_{PE} = -58.69
δ^2H	-25.94	-35.43	-48.12	-52.57	-54.35	-53.91	-50.2	
Iso-$\delta^{18}O$	/	/	/	/	/	/	***100%****	
Mix-$\delta^{18}O$	**1.3±0.01**	**1.6±0.01**	**2.5±0.02**	**3.2±0.03**	**57.1±0.04**	**5.8±0.12**	**28.6±0.38**	
Iso-δ^2H	/	/	/	/	/	/	/	
Mix-δ^2H	**0.7±0.01**	**0.9±0.01**	**68.0±0.40**	**1.6±0.02**	**1.8±0.02**	**1.8±0.02**	**25.3±0.41**	
Iso-$\delta^{18}O/\delta^2H$	/	/	/	/	/	/	/	
Mix-$\delta^{18}O/\delta^2H$	**0.3±0.00**	**0.3±0.00**	**0.5±0.00**	**0.7±0.01**	**0.8±0.01**	**0.8±0.01**	**96.6±0.02**	
Iso-$\delta^{18}O$	1.2±0.01	2.0±0.02	5.8±0.05	10.9±0.09	17.8±0.15	26.4±0.20	36.0±0.19	$\delta^{18}O_{TR}$ = -6.47
Mix-$\delta^{18}O$	**2.5±0.00**	**3.1±0.00**	**5.4±0.00**	**9.1±0.10**	**18.2±0.20**	**24.0±0.20**	**37.7±0.30**	δ^2H_{TR} =- 60.03
Iso-δ^2H	/	/	/	/	/	/	/	
Mix-δ^2H	**0.3±0.00**	**0.3±0.00**	**0.5±0.00**	**0.6±0.01**	**0.7±0.01**	**0.6±0.01**	**97.0±0.01**	
Iso-$\delta^{18}O/\delta^2H$	/	/	/	/	/	/	/	
Mix-$\delta^{18}O/\delta^2H$	**0.3±0.00**	**0.4±0.00**	**0.6±0.01**	**0.7±0.01**	**48.7±0.50**	**0.7±0.01**	**48.6±0.50**	
Iso-$\delta^{18}O$	2.1±0.02	3.5±0.03	10.5±0.06	20.8±0.17	29.0±0.21	34.1±0.20		$\delta^{18}O_{SA}$ = -5.70
Mix-$\delta^{18}O$	**3.6±0.02**	**4.7±0.03**	**8.7±0.07**	**18.3±0.19**	**28.4±0.24**	**36.4±0.25**		δ^2H_{SA}= -56.31
Iso-δ^2H	/	/	/	/	/	/		
Mix-δ^2H	**0.8±0.01**	**1.0±0.01**	**1.6±0.01**	**2.2±0.02**	**92.1±0.04**	**2.4±0.02**		
Iso-$\delta^{18}O/\delta^2H$	/	/	/	/	/	/		
Mix-$\delta^{18}O/\delta^2H$	**0.8±0.01**	**1.0±0.01**	**1.6±0.01**	**2.2±0.02**	**91.9±0.04**	**2.5±0.02**		

Fig. S1 Precipitation of the Heihe River Basin during the study period of the upper reaches (a), middle reaches (b) and lower reaches (c).

Fig. S2 The local meteoric water lines of the Heihe River Basin.

Fig. S1

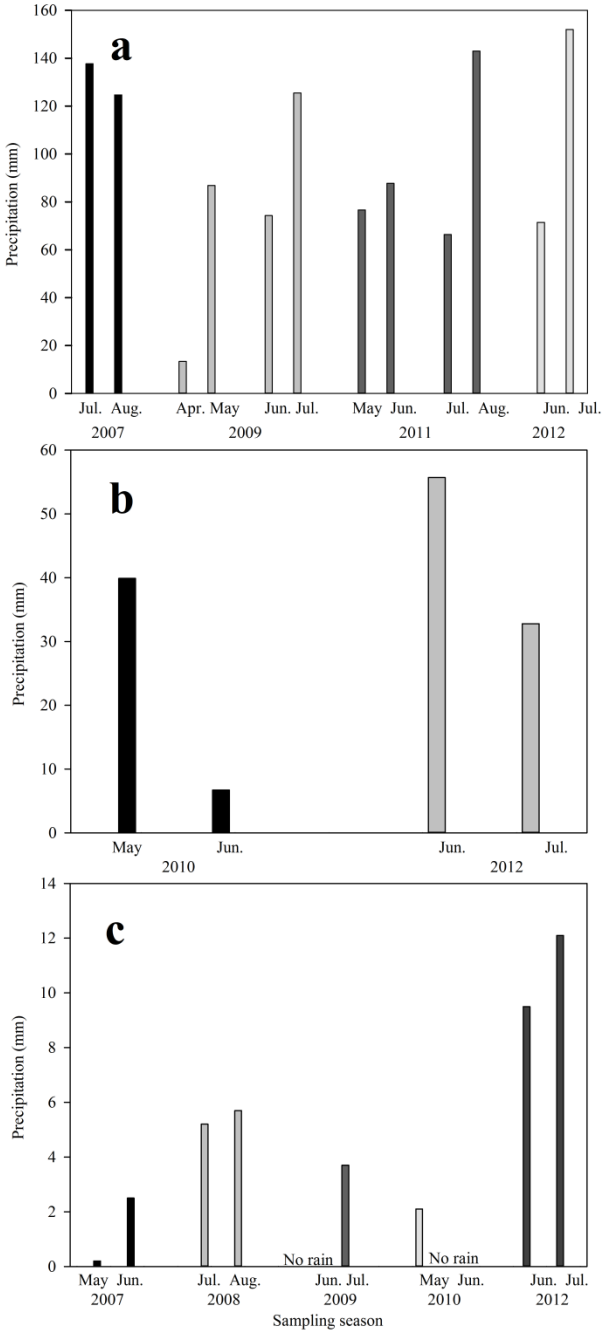
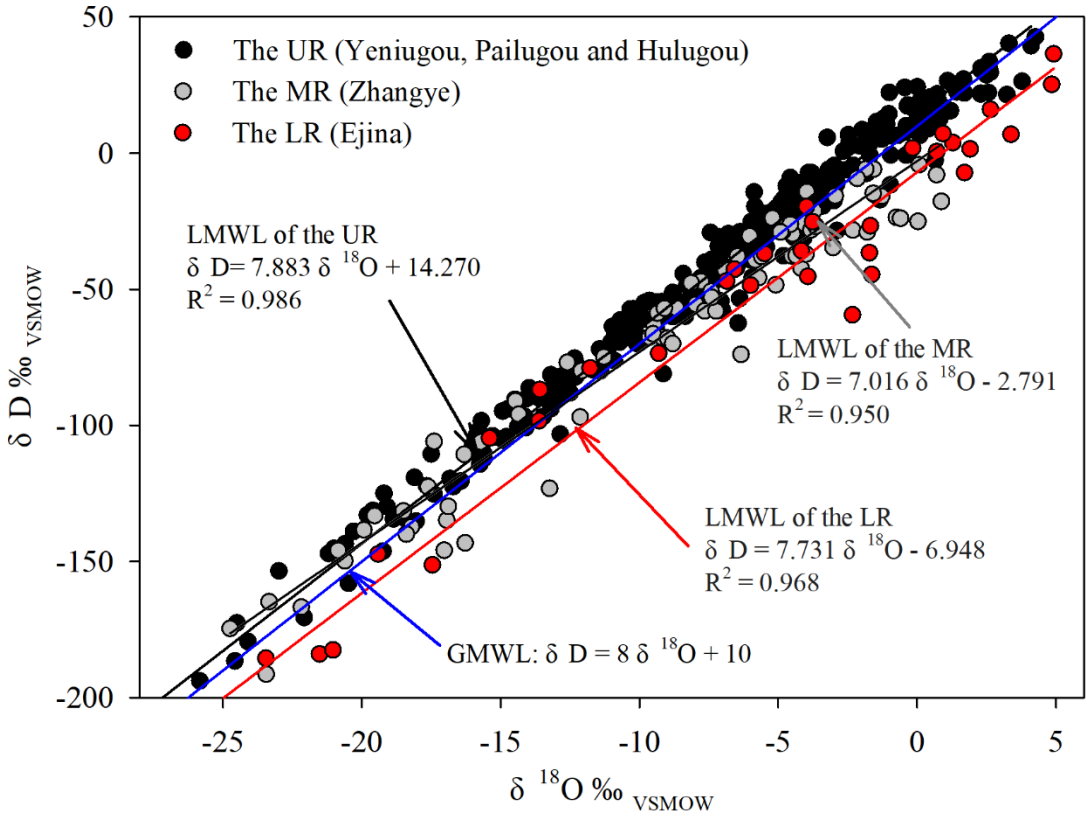


Fig. S2


The UR (Yeniugou, Pailugou and Hulugou)
The MR (Zhangye)
The LR (Ejina)
LMWL of the UR
δ D= 7.883 δ 18O + 14.270
R2 = 0.986
LMWL of the MR
δ D = 7.016 δ 18O - 2.791
R2 = 0.950
LMWL of the LR
δ D = 7.731 δ 18O - 6.948
R2 = 0.968
GMWL: δ D = 8 δ 18O + 10
δ D ‰ VSMOW
δ 18O ‰ VSMOW
50
0
-50
-100
-150
-200
-25
-20
-15
-10
-5
0
5