Introduction
Water shortages are a critical factor that constrains the distribution and abundance of plants in semi-arid areas of the world. The capacity of plants access different water resources is associated with their life forms and distribution (Dodd et al. 1998). Many perennial plants in semi-arid and seasonally dry areas have a dimorphic root system, which utilizes soil moisture in the upper soil layers during wet seasons and penetrates to deeper layers to take up water when the dry season arrives (Zapater et al. 2011, Kray et al. 2012). This seasonal shift of water resources is significant for species growing in semi-arid areas (Moreno and Bertiller 2012). Traditional studies to determine root distribution have used excavation methods, which are destructive, time-consuming and not suitable for long-term research (Meinzer et al. 2001). In addition, root distribution may not reflect the water uptake depths of plants (Moreira et al. 2000). Analysis of isotopic compositions of water from plant stems and soil materials has been applied in ecological studies as an alternative approach to study water uptake by roots (Zencich et al. 2002, Nie et al. 2011). There is limited isotopic fractionation by plant roots during water uptake (Dawson and Ehleringer 1991) and the isotopic compositions of xylem water are in accordance with the isotopic values of soil water where the plants absorb the water. By comparing hydrogen and oxygen isotopic compositions from xylem water with potential water sources, proportional contributions of water sources to the plants may be determined (Ehleringer and Dawson 1992). Snyder and
Williams (2000) found that Goodding’s willow (Salix gooddingii C.R. Ball) only utilized groundwater during the rainy season. Duan et al. (2008) showed that Chinese pine (Pinus tabulaeformis Carr.) derived 92.5% of its xylem water from rain water during the growing season. McCole and Stern (2007) found that Ashe’s juniper (Juniperus ashei J. Buchholz) used groundwater in the summer and soil water during the winter. However, these studies took advantage of a two- or three-compartment linear mixing approach, which is limited to calculating contributions of no more than three water sources (Burgess et al. 2000, Phillips and Gregg 2001). In fact, plants may have multiple sources of water, including different soil depths and rainwater. Phillips and Gregg (2003) proposed a multi-source mass balance approach, which resolved this restriction and could assess contributions of different water sources (Asbjornsen et al. 2007). Using the software IsoSource (Phillips and Gregg 2003), the fractional contribution of each potential source is determined and the results are presented as a distribution range of feasible solutions rather than unique solutions. This may provide a more realistic approach in plant water uptake studies and avoid misinterpretation of results (Benstead et al. 2006).

Chinese arborvitae (Platycladus orientalis [Linn.] Franco) is widely planted in semi-arid China (Chen et al. 2009). However, how it survives and even thrives in semi-arid or drought-prone areas is unknown and hence it is important to understand seasonal water use patterns and dynamics. In this study, stable isotope techniques were applied with the multi-source mass balance analysis approach to examine the depth of water uptake in the soil horizon and determine the proportional contributions of different water sources to total plant water uptake in different seasons. The goals of this study were: 1) to determine the proportional contribution of water from different soil depths to total plant water uptake; 2) to evaluate the utility of standard stable isotope analysis based on single-source direct inference and multi-source mass balance approaches; and 3) to enhance understanding of how seasonal change alters plant water use dynamics and the implications for hydrological functioning in semi-arid areas.

Materials and Methods

Site description

The study site was a 50-year-old plantation of Chinese arborvitae. Research was carried out July to November, 2011 at the Jufeng National Forest, Beijing (Fig. 1). This is an experimental forest established in the 1960s to increase forest cover in the western part of Beijing and planted with a variety of coniferous species, but predominantly P. orientalis. Stand density is 1176 trees/ha with a mean tree height of 10.7 m and a DBH of 20.9 cm. Soil type is sandy loam, with a texture consisting mainly of sand, silt and clay. The maximum depth is 120 cm above weathered bedrock. Soil pH ranges between 6.4 and 8.4; mean total nitrogen and mean total phosphorus are 4.87 g kg⁻¹ and 101.50 mg kg⁻¹, respectively. The climate is hot and rainy in summer (June to September), cold and dry in winter (November to February). Mean annual temperature is 11.6°C with an average of 193 frost-free days. Mean annual precipitation is approximately 630 mm, 70% to 80% falls between July and September.

Meteorological data

Mean daily and monthly precipitation was monitored by the Chinese Forest Ecosystem Research Network (CFERN) weather station located some 300 m from the sampling site (Fig. 1).

Collection of samples for isotopic analyses

Samples of each rain event were collected into a plastic tank to avoid evaporation and immediately put into 50 ml capped vials, sealed with Parafilm ‘M’ and frozen to -20°C until isotopic analysis. Branch samples of P. orientalis were collected from three plots at each of three sampling sites on July 16 and July 23 (wet season), October 16 and October 24 (transitional season), November 7 and November 19 (dry season) representing different periods of water use. The area of each plot was 10 m × 10 m. At each plot, three soil sampling points and three to five adjacent plants of P. orientalis were selected randomly for testing variations in isotopic compositions. Soil samples (two replicated samples/depth) were collected at 10-cm intervals to 120 cm using a soil auger. All plant and soil samples were placed immediately into 50 ml capped vials after collection, sealed with Parafilm ‘M’ and frozen to -20°C until isotopic analysis.

Isotopic analyses

Water was extracted from branches and soil samples using a cryogenic vacuum distillation approach (West et al. 2006). The δ²H and δ¹⁸O isotope ratios of the branch water, soil water and precipitation were measured with a liquid water isotope laser spectroscopy instrument (Model DLT-100, LGR Inc.) (Lis et al. 2008) at the Key Laboratory of Soil and Water Conservation, Beijing Forestry University. The δ²H and δ¹⁸O values are expressed as the ²H/H and ¹⁸O/¹⁶O relative to Vienna Standard Mean Ocean Water (VSMOW) in ‰, respectively. VSMOW is a water standard defining the isotopic composition of freshwater. This standard includes both the established values of stable isotopes found in water and calibration materials provided for standardization and inter-laboratory comparisons of instruments used to measure these values in experimental materials.
\[ \delta^{2}H \text{ or } \delta^{18}O = \left( \frac{R_{\text{sample}}}{R_{\text{standard}}} - 1 \right) \times 1000 \]

where \( R \) are the molar ratios of \(^2\text{H}/\text{H} \) or \(^{18}\text{O}/^{16}\text{O} \) of the sample and VSMOW. Measurement precision was better than \( \pm0.32 \% \) and \( \pm0.17 \% \) for \( \delta^{2}H \) and \( \delta^{18}O \), respectively (Berman et al. 2009).

**Analysis and statistics**

Based on similar \( \delta^{18}O \) values of different soil depths on each sampling day, soil profiles (0–120 cm) were subdivided into four or five depth intervals to facilitate root water uptake (Table 1). The isotopic composition for each depth was according to the average value of samples within each interval. The isotopic values of each interval and the stem water were analyzed by the IsoSource software (the multi-source mass balance approach) to evaluate the contribution of each soil depth to stem water. The multi-source mass balance approach analyzed the data more systematically and provided a more quantitative assessment of the estimated range of feasible contributions of potential water sources from different soil depths to total water uptake. The fractional increment was set at 1\%, and the uncertainty level was set at 0.2\%.

**Table 1. Subdivided soil depth intervals on different sampling dates**

<table>
<thead>
<tr>
<th>Soil depth intervals</th>
<th>Sampling dates</th>
</tr>
</thead>
<tbody>
<tr>
<td>0-20 cm</td>
<td>July 16</td>
</tr>
<tr>
<td>20-40 cm</td>
<td>July 23</td>
</tr>
<tr>
<td>40-80 cm</td>
<td>October 16</td>
</tr>
<tr>
<td>&gt;80 cm</td>
<td>October 24</td>
</tr>
<tr>
<td>0-20 cm</td>
<td>November 19</td>
</tr>
<tr>
<td>0-20 cm</td>
<td>November 7</td>
</tr>
<tr>
<td>20-30 cm</td>
<td></td>
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<tr>
<td>30-40 cm</td>
<td></td>
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<tr>
<td>40-80 cm</td>
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<tr>
<td>&gt;80 cm</td>
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</table>

**Results**

**Isotopic composition of rainwater**

Fig. 2 shows that \( \delta^{2}H \) and \( \delta^{18}O \) values ranged from -119.4 \% to -19.3 \% and -15.5 \% to -1.1 \%, respectively, with standard deviations of 23.5 \% and 3.4 \%, respectively. The larger standard deviation of \( \delta^{2}H \) and \( \delta^{18}O \) values indicated the significant seasonal variations in rainwater isotopic compositions. The Global Meteoric Water Line (GMWL) indicates the average relationship between hydrogen and oxygen isotope ratios in natural terrestrial waters, expressed as a worldwide average (Craig 1961). A meteoric water line may be calculated for a given area, and used as a baseline within that area. Kinetic fractionation will cause the isotope ratios to vary between localities within that area. This relationship is used within the field of isotope hydrology. Craig’s original assertion is that isotopic enrichments, relative to ocean water, display a linear correlation over the entire range for water that has not undergone excessive evaporation. The local meteoric water line for rainwater was \( \delta^{2}H = 6.82\delta^{18}O - 2.94 \) (\( R^2 = 0.90, n = 35 \)), for which the slope and the intercept were smaller than the GMWL \( \delta^{2}H = 8\delta^{18}O + 10 \), indicating isotopic evaporative enrichment in precipitation in this area.

**Isotopic compositions of soil and branch water**

Standard stable isotope analysis based on single-source direct inference assumed that plants were extracting water from a single dominant soil depth determined by the intersection of \( \delta^{18}O \) values of the stem water and the soil profile. Fig. 3 shows that in the wet season, soil isotopic values of \( \delta^{18}O \) near the surface did not show evaporative isotopic enrichment, and variations of \( \delta^{18}O \) values for different depths were not significant. However, before and during the dry seasons, isotopic values of \( \delta^{18}O \) in the upper horizon were less negative due to enrichment associated with greater evaporation and little precipitation. Isotopic values of the deeper soil water (below 40 cm) stayed relatively constant during all seasons, ranging from -8.9 \% to -6.9 \% with a standard deviation of 0.67 \%. Isotopic plant signatures corresponded to isotopic compositions in the soil; consequently, the variation in isotopic compositions of soil profiles in different seasons provided a direct way to study the depth plants were absorbing water.

Fig. 3 also shows that *P. orientalis* mainly obtained water from the upper 40 cm on July 16, October 16, and November 19. On other sampling dates, uptake was below 40 cm. However, specific depths of water uptake were less clear and not determined due to the more complicated isotopic gradient for soil water.

**Fig. 2.** Isotopic compositions of rainwater during different sampling periods.
Fig. 3. $\delta^{18}O$ values (±SD) of soil water and stem water on different sampling dates. ( ) corresponds to the $\delta^{18}O$ value of the stem.
Fig. 4. Frequency histograms at different sampling dates showing the range of water sources at different soil depths to total water uptake. P.F. is proportional frequency and S.P. source proportion.
Estimates of the contributions of water sources at different soil depths to total water uptake are provided by frequency histograms (Fig. 4). On each sampling day, the frequency histogram was clear for a certain subdivided soil depth, while others were broad and diffuse. *P. orientalis* received a significant portion (72% to 79%) of water from the 20–40 cm depth. As the wet season progressed, histogram patterns also became broad and diffuse. In the transitional and dry seasons, trees obtained moisture mainly from lower soil levels (40–80 cm) and the proportions of soil water to total plant water uptake were 41% to 84%. As the dry season increased, plants utilized deeper soil water. However, in the transitional and dry seasons with earlier rainfall, *P. orientalis* shifted to the surface soil (0–20 cm) to withdraw a relatively large proportion of water: 28% to 37% (mean 31.8%) and 53% to 61% (mean 57%) on October 16 and November 19, respectively (Fig. 4). Although there was rainfall on October 23 (Fig. 5), surface soil water contributed little to total plant water uptake (Fig. 4).

### Discussion

**Seasonal isotopic variations in soil water**

Variations in isotopic composition of upper soil water were due to combined effects of evaporation and earlier rainfall, while compositions in deeper soil layers were essentially determined by previous rainfalls (Song et al. 2009). Isotopic soil profiles varied significantly in the upper soil layers (0–40 cm) between seasons, while soil layers below 40 cm remained relatively constant (Fig. 3). The dissimilarities between upper and lower soil depths could be due to differences in soil moisture evaporation rates and infiltration rates at different depths in different seasons. Soil moisture evaporation rates decreased as soil depth increased (Barnes and Allison 1983, Gazis and Feng 2004), an important factor that led to isotopic fractionation of the soil profile. Infiltration rates at different soil depths could be partially due to the soil moisture deficit (Robinson 1999) before precipitation in different seasons. In the wet season, the isotopic values of δ18O at all soil depths did not vary much (Fig. 3b). This could be due to 1) relatively...
higher relative humidity (Fig. 5), which led to a decrease in vapor pressure deficit (Wilson et al. 2001, Farquhar et al. 2007) so that evaporation at the surface soil was not significant; 2) frequent precipitation recharged the soil water, which led to no significant variations (Zenchic et al. 2002); 3) the soil moisture content was above the capillary breaking point due to frequent precipitation and the movement of soil water to the evaporation surface was mainly as liquid, which slowed the evaporation rate (Benoit and Kirkham 1963); and/or 4) the possibility that water moved through the profile through large pores or “preferential flow” paths, which would have preserved the isotopic composition of the rain (Gazis and Feng 2004, Brooks et al. 2009). In contrast, the correlation between soil moisture evaporation rate and temperature was not significant (p > 0.1). Isotopic compositions of all soil depths were determined to be a mixture of previous rainfalls (Fig. 5 and Fig. 3, July 16 and July 23), so there were no remarkable variations in the δ18O values of the vertical gradients of the soil profile.

In the transitional (October 16 and 24) and dry (November 7 and 19) seasons, with long periods of drought and limited rain, the soil moisture deficit of the upper soil layers (0–40 cm) increased and the mean relative humidity decreased (Fig. 5). Thus, rainwater could stay near the soil surface for longer periods with longer exposure times for evaporation, resulting in a significant evaporative enrichment in the upper soil horizon (Fig. 3). However, the relatively less negative δ18O values of all depths on October 24 are due to the precipitation on October 23 after soil water recharged on October 13 (Fig. 5). Another finding was that deeper soil (below 40 cm) was barely affected by evaporation and the δ18O values were a mixture of previous rainfall, so the values of δ18O below 40 cm were more negative in the transitional and dry seasons than in the wet season (Fig. 3). As there was no rain between October 24 and November 16, δ18O values below 40 cm should be negative; however, the δ18O value at 40–60 cm November 7 was more negative than on October 24 (Fig. 3), which suggests the possibility of “hydraulic redistribution” (Brooks et al. 2002). Soil water uptake depth by roots on November 7 also supports this finding (Fig. 3e and Fig. 4e).

Seasonal isotopic variations in plant water

This study employed two different approaches in an effort to see how P. orientalis depends on upper or lower water sources, and as a result, interpretations are limited. In the wet season, both the direct inference approach and the multi-source mass balance approach suggest that water sources for P. orientalis was dominated by surface soil water (Fig. 3a and Fig. 4a). As the wet season progressed with greater precipitation, it was difficult to determine which depth of soil was the main water source. There was a possibility that a mixture of soil water from different depths was the principal water source. However, plants tend to utilize soil water that is more accessible (higher soil moisture content). There is also the possibility that the surface soil still dominated the water source up to 90% (Fig. 4b) due to frequent precipitation that fully recharged the surface layers (Fig. 5), suggesting that P. orientalis depends more on summer rains. P. orientalis is a conifer with a relatively shallow root system (Zhao et al. 2006). In the rainy season, coniferous species with developed lateral roots largely rely on rain water (Flanagan et al. 1992, Valentini et al. 1992). Li et al. (2007) found that Larix sibirica Lede. only used recent rainfall in July (the growing season) in semi-arid areas of Mongolia, which is similar to the results here. The high dependency on summer rains by P. orientalis suggests its high water use efficiency as well (Ehleringer et al. 1991, Wang et al. 2007).

In the dry season, the direct inference approach showed differential water uptake (Fig. 3c–f). IsoSource calculations showed the same patterns (Fig. 4c–f). This suggests that the main water source for P. orientalis was at lower soil depths after long periods without rain. This is similar to the findings of Romero-Saltos et al. (2005). Roots utilized water in the upper soil layers that was recharged by the earlier rainfall (Fig. 5). Furthermore, the contributions of upper soil layers to total water uptake were different on October 24 and November 19, although with similar previous precipitation (Fig. 4). These data suggest that previous summer rains that penetrated into the deeper soil layers contributed more to total plant water uptake in the transitional season than in the dry season (Fig. 4c). After a lengthy period of drought, deeper water from summer rains decreased and contributed less. White et al. (1985) found that Pinus strobus L. absorbed water from lower depths during a drought period but obtained water from the upper soil when rain occurred. This supports the findings in this study. However, rainfall on October 23 contributed little to plant water utilization (Fig. 4d), which suggests that precipitation reaches a minimum threshold to recharge the soil for roots in the upper layers. Soil moisture in drought-prone areas varies significantly even in adjacent areas and plants generally change their water acquisition spatial patterns in response (Schlesinger et al. 1990). The root zone can obtain more water where soil moisture availability is normal than where it is lacking to maintain the growth of plants (Simoneau and Habib 1994, Green and Clothier 1995). The findings in this study are consistent with other studies that show that a number of plants in arid and semi-arid areas obtain surface soil water during the wet season and shift to deeper soil layers during the dry season. They possess a “dimorphic root”, which is a root system with two distinct root forms adapted to perform different functions (Chimner and Cooper 2004, Hasselquist and Allen 2009). One of the most common manifestations is in plants with both lateral roots and a taproot that grows straight down to the water table. Dimorphic root systems are advantageous for large woody species where the acquisition of seasonally limited water is a premium.

The two approaches applied in this study have different characteristics. The direct inference approach provides visual assessment and indicates possible water sources but ignores the possibility that plants obtain water at different soil depths (Wang et al. 2010). The multi-source mass balance approach overcomes this. There are three advantages of this approach: 1) it provides a method that analyzes the data more systematically, which reduces misinterpretations from observers’ bias; 2) it permits sensitivity analysis by applying different standards; and 3) it provides a more quantitative assessment of the range of feasible contributions of water sources from different soil depths to total water uptake (Asbjornsen et al. 2007).

In addition, the data suggest that P. orientalis depends largely on summer rainfall and competes for water with...
annual shallow-rooted herbs and shrubs. Undergrowth vegetation in this forest ecosystem may grow deep roots to survive during the growing season. Water is a key factor that constrains the distribution and abundance of plants in semi-arid areas. From fieldwork carried out earlier in 2010 on the ecological biodiversity index of the *P. orientalis* forest ecosystem was low, suggesting the lack of available water for undergrowth vegetation and an inability to compete for water with *P. orientalis*. Furthermore, the high dependence on summer rainfall implies the low leaf water potential of *P. orientalis* (Ehleringer et al. 1991), suggesting the ability to survive from serious water stress.

**Conclusions**

This study examined seasonal water use patterns of a 50-year-old *P. orientalis* plantation. The results have important implications for plant species in semi-arid China. Both the direct inference approach and the multi-source mass balance approach demonstrated similar water use patterns despite the different characteristics between these two methods. There is generally a switch of water sources from shallow soil depths in the rainy season to lower depths in the dry season. This switch may be due to their dimorphic root systems and varied rainfall distribution between seasons. In addition, there is a minimum threshold of precipitation for *P. orientalis* to obtain water by roots in the upper soil in the dry season after long periods of drought. Furthermore, the ability to compete and high dependence on summer rainfall of *P. orientalis* should be one of the more widely planted species in water-limited areas of China.

We acknowledge that no replications were made. Further studies with more sampling periods and more plant species are needed to determine seasonal water use patterns and plant–soil–water cycling processes to better understand differential responses in water use patterns among species in drought-prone areas.

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**References**


