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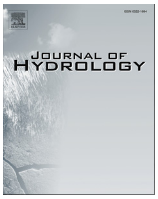

A deuterium-based labeling technique for the investigation of rooting depths, water uptake dynamics and unsaturated zone water transport in semiarid environments

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SUMMARY

Non- or minimum-invasive methods for the quantification of rooting depths of plants are rare, in particular in (semi-)arid regions; yet, this information is crucial for the parameterization of SVAT (Soil–Vegetation–Atmosphere Transfer) models and understanding of processes within the hydrological cycle. We present a technique utilizing the stable isotope deuterium (^2H) applied as artificial tracer to investigate the vertical extent of the root zone, characterize water uptake dynamics of trees and shrubs at different depths and monitor transport of water through the unsaturated zone of dry environments.

One liter of 35% deuterated water (^2H_2O) was punctually applied at several depths (0.5 m, 1 m, 2 m, 2.5 m and 4 m) at six different plots at a natural forested site in the Cuvelai–Etosha Basin (CEB), Namibia/Angola. Subsequently, uptake of the tracer was monitored by collecting plant samples (xylem and transpired water) up to seven days after tracer injection. Soil profiles at the plots were taken after the campaign and again after six months in order to evaluate the transport and distribution of ^2H within the unsaturated zone.

Of 162 plant samples taken, 31 samples showed clear signals of artificially introduced ^2H, of which all originate from the plots labeled up to 2 m depth. No artificially injected ^2H was found in plants when tracer application occurred deeper than 2 m. Results further indicate a sharing of water resources between the investigated shrubs and trees in the upper 1 m whilst tree roots seem to have better access to deeper layers of the unsaturated zone. The soil profiles taken after six months reveal elevated ^2H-concentrations from depths as great as 4 m up to 1 m below surface indicating upward transport of water vapor. Purely diffuse transport towards the soil surface yielded an estimated 0.4 mm over the dry season.

Results are of particular significance for a more precise parameterization of SVAT models and the formulation of water balances in semiarid areas. The developed methodology is beneficial for site-specific investigations in complex and data scarce environments, where the root zone plays a major role for the overall water balance. For arid and semiarid environments experiencing low recharge rates, water transported in its vapor phase is found to play an important role for the overall soil water balance. The use of ^2H_2O is cost-effective and provides the opportunity to investigate multiple effects along the soil–vegetation interface that have been difficult to deal with previously.

1. Introduction

In recent history, two scientific articles raised the recognition of the role of plants within the hydrological cycle to a new level. Jasechko et al. (2013) stated that "transpiration is the largest water flux from Earth's continents, representing 80–90 percent of terrestrial evapotranspiration" (Jasechko et al., 2013). One year later, McDonnell (2014) published his idea of what he called "*two water worlds*" hypothesis which is based on the findings that in certain watersheds streams and trees return different pools of water to the hydrosphere (McDonnell, 2014). Even though heavily debated (e.g. Brooks, 2015; Evaristo et al., 2015; Good et al., 2015) and partially rectified (e.g. Coenders-Gerrits et al., 2014; Schlaepfer et al., 2014), both studies emphasize the urgent need for an

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increased number of studies along the soil–plant–atmosphere interface. Despite of advances in applied techniques, measuring root activity is recognized as one of the greatest challenges in plant ecology (Casper et al., 2003; Kulmatiski et al., 2010; Schenk, 2008) and ecohydrology in general. In dry environments, where plant adaptations to the local conditions such as the development of deep tap roots or hydraulic redistribution are common (Canadell et al., 1996; Dawson, 1993; Richards and Caldwell, 1987), it is crucial to obtain information on the lateral and vertical extent of the root zone in order to parameterize SVAT (Soil Vegetation Atmosphere Transfer) and climate models (Schulze et al., 1996). Furthermore, the knowledge of rooting depth and distribution is critical for the calculation of deep percolation and for the identification of thresholds of precipitation duration and intensity that support groundwater recharge (Seyfried et al., 2005). From an ecohydrological perspective, the dominance of certain functional groups (e.g. trees, shrubs, herbaceous plants) or species (Canadell et al., 1996) and effects such as bush encroachment (Hipondoka et al., 2003) or hydraulic redistribution (e.g. Dawson, 1996, 1993; Prieto et al., 2012; Schulze et al., 1998) are of particular interest. Information on rooting depth and distribution is also highly beneficial for any investigation along the soil–vegetation–atmosphere interface because water is the limiting factor in arid and semiarid environments (Canadell et al., 1996; Oliveira et al., 2005; Schenk and Jackson, 2005, 2002a; Schulze et al., 1996; Seyfried et al., 2005).

An overview of studies on maximum rooting depths of plants is presented by Canadell et al. (1996). The authors pointed out that plants from environments experiencing a long dry season on average have deepest roots (15 ± 5.4 m). The deepest recorded rooting depths were found in the Kalahari (*Boscia albitrunca* – 68 m, *Acacia erioloba* – 60 m; Jennings, 1974). Deepest roots can be expected where thick sandy soils are present (Canadell et al., 1996). For such soils, a dependence of maximum rooting depth on rainfall amount and distribution has been found previously (Burke, 2006; Schenk and Jackson, 2002b). On a global scale, results from existing studies have been used in order to map the global distribution of deep roots in relation to climate and soil characteristics (Schenk and Jackson, 2005), to derive parameters for modeling (Zeng, 2001) or to investigate the lateral zone of influence of plants (Casper et al., 2003). Schenk and Jackson (2002b) evaluated existing data and derived relationships for below- and aboveground allometries in water-limited ecosystems.

1.1. Different approaches for investigating rooting depths

Traditionally, approaches for investigating root structure and functioning included manual digging, excavation techniques and even the use of dynamite throughout the 20th century (Canadell et al., 1996). Schulze et al. (1996), for example, excavated trenches of 5–10 m length and 3 m depth to investigate rooting depths in Patagonia. Jennings (1974) made a rather coincidental finding during borehole drilling in Botswana: They found roots of *B. albitrunca* and *A. erioloba* at 68 m and 60 m depth, respectively. In contrast, Ringrose et al. (2000) found no evidence of roots appearing deeper than 6 m in a similar environment. Since the beginning of the new century, innovative techniques have developed with different approaches.

Ground-penetrating radar (GPR), for instance, has been utilized in several studies (Bassuk et al., 2011; Butnor et al., 2003; Hruska et al., 1999; Raz-Yaseef et al., 2013; Stokes et al., 2002). The technique was shown to be a reliable, non-invasive method for the location of roots and the determination of bulk root densities. However, bulk root density has been found to be not a good indicator for root activity (Kulmatiski et al., 2010). Jackson et al. (1999) used caves and DNA to derive ecosystem rooting depths and were able to assign roots to plant species up to 65 m depth. Regardless of these recent developments, traditional methods are still common (Hipondoka and Versfeld, 2006; Rings et al., 2013) and challenges remain.

1.2. Approaches based on stable isotopes of water

Techniques utilizing stable isotopes of water (deuterium, ^{2}H and oxygen-18, ^{18}O) as environmental or artificial tracer (subsequently, we refer to as ‘labeling’) have been found to be particularly suitable for plant-related studies and are used extensively (Casper et al., 2003). In early studies, Allison and Barnes (1984) evaluated the effect of climate and vegetation on water stable isotope compositions (Allison and Barnes, 1984). Other applications of environmental isotopes include the identification of water use strategies of certain species (i.e. Brunel et al., 1995; Chimner and Cooper, 2004; Edwin et al., 2014; Li et al., 2007), changes in water-use strategies (Wu et al., 2013), quantifications of preferential flow (Stumpp and Maloszewski, 2010) and investigations on hydraulic lift (Ceperley et al., 2014). Dawson et al. (2007) used measurements of stable isotopes in woody plants to identify nighttime transpiration.

Artificial applications of the stable isotope deuterium (^{2}H) are only now becoming increasingly popular in hydrology and soil sciences. Labeling with deuterated water (^{2}H$_2$O) is of advantage because it is not radioactive and has no toxicological concerns during both labeling and measurement (Becker and Coplen, 2001), compared to the previously used tritium (^{3}H). Being part of the water molecule, ^{2}H is considered a conservative tracer and therefore ideal for studies in the unsaturated zone (Koeniger et al., 2010). It can be measured in very low concentrations (Becker and Coplen, 2001); therefore only small amounts of tracer are necessary and experiments become economically feasible. It has been shown previously that the use of artificial ^{2}H enables a quantification of transpiration (i.e. Calder et al., 1986, 1992; Calder, 1992; Lambs and Saenger, 2011; Marc and Robinson, 2004). Meinzer et al. (2006) used ^{2}H$_2$O to study the dynamics of water transport in conifers and found maximum sap flow velocities of 2.4–5.3 m d^{-1}. Studies directly related to root distribution are, however, rare and limited to shallow depths (e.g. Bishop and Dambrine, 1995; Hawkins et al., 2009; Peñuelas and Filella, 2003; Plamboeck et al., 1999; Sternberg et al., 2005). Only one attempt was made to use artificial ^{2}H$_2$O to investigate the spatiotemporal distribution of plant water use of trees and grasses in a tropical savanna (Kulmatiski et al., 2010) to a maximum depth of 120 cm. It is this latter investigation by Kulmatiski et al. (2010) that prompted the idea for the present study. The authors injected ^{2}H$_2$O at four different depths (5, 20, 50 and 120 cm) and analyzed plant samples taken from up to five meters distance from the labeled plots in order to test the two-layer hypothesis (Walter, 1971). This theory suggests that woody plant species develop deep roots to escape competition with grasses at shallow depths. The findings by Kulmatiski et al. (2010) do not support this theory and are of high relevance for future research. Based on their studies, one could also investigate water uptake by plants from deeper regions of the unsaturated zone (which is essentially the purpose of this study).

In numerous studies ^{2}H$_2$O was applied as artificial tracer to investigate water movement in the unsaturated zone. Applications include studies of preferential flow (Hangen et al., 2005; Schumann and Herrmann, 2001; van der Heijden et al., 2013), transport velocities (Blume et al., 1967; Koeniger et al., 2010; Mali et al. (2007); Saxena, 1984; Zimmermann et al., 1966) and capillary rise (Grünberger et al., 2011). Koeniger et al. (2010) conclude ^{2}H$_2$O to be a suitable water tracer extending possibilities for field studies in the field of biogeosciences. Allison et al. (1994) state that in dry areas with sandy soils and high porosity water vapor transport might diffuse a tracer into soil and groundwater systems (Allison