



# Isotopic offset between unconfined water and water adsorbed to organic matter in equilibrium

Guo Chen, Karl Auerswald, Hans Schnyder

Lehrstuhl für Grünlandlehre, Technische Universität München, Alte Akademie 12, Freising-Weihenstephan 85354, Germany.

Correspondence to: Karl Auerswald (auerswald@wzw.tum.de)

**Abstract.** Hydrophilic surfaces influence the structure of water close to them and may thus affect the isotope composition of water. Such an effect should be relevant and detectable for materials with large surface areas and low water contents. The relationship between the volumetric solid:water ratio and the enrichment of heavy isotopes in adsorbed water compared with unconfined water was investigated for the materials silage, hay, organic soil (litter), filter paper, cotton, casein and flour. Each of these materials was equilibrated via the gas phase with unconfined water of known isotopic composition to quantify the isotopic difference between adsorbed water and unconfined water. Across all materials, enrichment of the adsorbed water was significant and negative (on average  $-0.91\text{ ‰}$  for  $^{18}\text{O}$  and  $-20.6\text{ ‰}$  for  $^2\text{H}$  at an average solid:water ratio of 0.9). The observed enrichment was not caused by solutes, volatiles or old water because the enrichment did not disappear for washed or oven dried silage, the enrichment was also found in filter paper and cotton, and the enrichment was independent of the isotopic composition of the unconfined water. Enrichment became linearly more negative with increasing volumetric solid:water ratio and even exceeded  $-4\text{ ‰}$  for  $^{18}\text{O}$  and  $-44\text{ ‰}$  for  $^2\text{H}$ . This enrichment behavior could be modeled by assuming two water layers: a thin layer that is in direct contact and influenced by the surface of the solid and a second layer of varying thickness depending on the total moisture content that is in equilibrium with the surrounding vapor. When we applied the model to soil water under grassland, the soil water extracted from 7 cm and 20 cm depth was significantly closer to local meteoric water than without correction for the surface effect. This study has major implications for the interpretation of the isotopic composition of water extracted from organic matter, especially when the volumetric solid:water ratio is larger than 0.5 or for processes occurring at the solid-water interface.

**Key-words:** discrimination; protein; cellulose; surface effect; O-18; H-2

## 1 Introduction

The  $^{18}\text{O}$  and  $^2\text{H}$  isotope composition of water reflects climate and many processes within the water cycle (Bowen, 2010; Gat, 1996). Changes in the isotope composition of water can either result from the mixing of water with differing isotopic composition or from the change in isotopic composition by fractionation, especially between vapor and liquid. Such fractionation can be affected by ion hydration. In aqueous solutions, ions change the activities of the isotopologues of water ( $\text{H}_2\text{O}$ ,  $\text{HDO}$ , and  $\text{H}_2^{18}\text{O}$ ) due to their hydration. This, in turn, causes the isotopic fractionation between aqueous solutions and water vapor to differ from the fractionation between pure water and vapor (Kakiuchi, 2007; Stewart and Friedman, 1975). Similar to salt, the surface of hydrophilic materials also interacts with water molecules creating a two-dimensional ice like water layer near the surface and a three dimensional liquid layer far from the surface (Asay and Kim, 2005; Miranda et al., 1998). Additionally, adsorption, may cause an energetic difference between water molecules at the surface of solids and the bulk water molecules. These structural and energetic differences may cause a difference in isotopic composition between



these two layers of water. If existent, such a surface effect should be strongest in materials with large specific surface area and with low water content. There are some indirect hints from studies of plant water uptake from soil, which show that mobile water differs isotopically from immobile water (Brooks et al., 2010; Evaristo et al., 2015; Tang and Feng, 2001) but to the best of our knowledge, such a surface effect has only been directly studied for clay (Oerter et al., 2014) and silica surfaces (Richard et al., 2007). It is not known how large the effect is for organic matter, which are associated with practically all mineral surfaces in the critical zone or form major constituents of other surfaces in the biosphere (Chorover et al., 2007; Nordt et al., 2012; Vazquez-Ortega et al., 2014).

A surface effect may be detected by establishing an equilibrium between water adsorbed to a material and air vapor created by unconfined water with known isotope composition in a closed chamber. If there is no surface effect, then the  $^{18}\text{O}$  and  $^2\text{H}$  isotope composition of the adsorbed water and unconfined water should be identical after equilibration. This is because the isotope composition of water under steady conditions is determined by the isotope composition of the water vapor, air humidity, equilibrium fractionation and kinetic fractionation (Helliker and Griffiths, 2007; Welhan and Fritz, 1977). All of these parameters are identical for adsorbed water and unconfined water when they both share for a sufficiently long enough time the same atmosphere, as is the case in a closed chamber.

We examined the hypothesis that the surfaces of organic materials influence the isotopic composition of adsorbed water and we choose materials of broad relevance. Silage is an important feedstuff delivering water to the animal and thus influencing the body water composition (Kohn, 1996) and animal products like milk. Hay in particular for example, has a low water content. Organic horizons at the soil surface (we call them litter thereafter) provide the interface where most vapor and water flows have to pass (Haverd and Cuntz, 2010). More pure materials like filter paper, cotton, casein powder and flour were included to identify whether the chemical identity causes or influences the effect. Finally we had to exclude that the effect resulted from artifacts like old water or volatiles and solutes interfering with the isotope measurements (Martín-Gómez et al., 2015; Schmidt et al., 2012; Schultz et al., 2011; West et al., 2011). Silage, which likely is a source of volatiles and solutes in rather large amounts (e.g., lactic acid, acetic acid, propionic acid, ethanol, and propanol; Porter and Murray, 2001), was also pretreated by washing and heating to remove potentially interfering substances. Water of contrasting isotope composition was used to identify any old water. Finally, we derived a simple prediction model for the effect and demonstrated its versatility in an application case with environmental samples.

## 2 Materials and Methods

We performed three equilibration experiments. Each equilibration experiment involved the exposure of samples to water vapor which originated from unconfined water, followed by cryogenic water extraction from samples and isotope composition measurement. We use  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  to describe the isotope composition of oxygen ( $^{18}\text{O}$ ) and hydrogen ( $^2\text{H}$ ) in water (with  $\delta^{18}\text{O}$  or  $\delta^2\text{H} = R_{\text{sample}}/R_{\text{standard}} - 1$ , where  $R_{\text{sample}}$  and  $R_{\text{standard}}$  denote the ratio of the abundances of heavy and light isotopes in samples following the international SMOW standard).

### 2.1 Preparation of samples

The materials comprised fresh silage, oven dried silage, washed silage, hay, fibric litter (slightly decomposed organic material; for definition see Schoeneberger et al., 2012), hemic litter (decomposed organic material of low fiber content), filter paper, cotton, casein and wheat flour. The silage and hay were obtained from a farm near Freising and were cut in pieces (4 cm to 8 cm). The silage was stored in a  $-18\text{ }^{\circ}\text{C}$  deep freezer while the hay was kept in a dark and dry place. The hemic and fibric horizons were gathered from a conifer forest near Freising (Germany) from a Haplic Podzol (according to IUSS Working Group WRB, 2014) area and stored in air tight bags in a refrigerator until use. In order to create a relative big range of water content, half of the litter samples were oven dried ( $16\text{ h}$  for  $100\text{ }^{\circ}\text{C}$ ) before the equilibration experiment. Filter



paper (Rotilabo®-round filters, type 11A, Germany), made of 100 % cellulose, and bleached medical cotton (Hartmann, Germany) were prewetted by spraying because the initially dry filter paper and cotton hardly adsorbed any humidity from air. Both materials were then slightly oven dried for different times (ranging from 0 to 60 min) at 50°C before the equilibration experiment. The casein powder contained 90 % natural casein and a small amount of carbohydrate. The wheat flour contained 70.9 % carbohydrates, most of which was starch. Casein and flour were oven dried for 6 h at 50 °C before use.

## 2.2 Unconfined water

Five isotopically distinct, unconfined waters were used. We term them very heavy, heavy, tap, light and very light waters according to their relative ranking of  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$ . These waters were produced from deionized water ( $\delta^{18}\text{O} = -10\text{‰}$ ,  $\delta^2\text{H} = -70\text{‰}$ ) by means of a rotary evaporator. Very heavy, heavy, light and very light waters had  $\delta^{18}\text{O}$  values of 15, 2, -15 and -22 ‰, and  $\delta^2\text{H}$  values of 125, 21, -113 and -160 ‰ respectively with slight deviations between individual experiments.

## 2.3 Set-up of the equilibration procedure

The different materials were placed in closed chambers (glass exsiccator vessels with a volume of approximate 20 L with drying agent removed) to equilibrate with unconfined water (Fig. 1). In a preliminary experiment, the effectiveness of the chambers' air seal was verified by flushing the containers with  $\text{N}_2$ , followed by monitoring the concentration of  $\text{CO}_2$  and water vapour inside the vessels. The concentrations after closing the chamber remained constant, which indicated that leaks were negligible. In another preliminary experiment we assessed the development of humidity in the chamber. The humidity reached 100 % within 20 min (half-life 1.8 min) after we put 200 mL of water at bottom of the chamber (Fig. 1), closed it and started the recycling pump (Laboport, Germany). All equilibration experiments lasted for 100 h. Sun et al. (2014) have shown that even for moist samples equilibration is relatively fast (half-life 20 h).

In each experiment, 200 mL of (unconfined) water was placed in a glass bowl (15 cm in diameter) on the bottom of the chamber and dishes containing the material samples under focus (about 3 g fresh matter per dish) were placed on a perforated sill in the chamber. We flushed the chamber with nitrogen gas to remove the air vapor and the oxygen to prevent the decay of the samples. After that we immediately closed the chamber and started the recycling pump to ensure homogeneity within the airspace of the chamber. After 100 h of equilibration, samples were quickly removed from the chamber, placed in 12 mL glass vials sealed with a rubber stopper and wrapped with parafilm. The samples were then stored in a -18 °C freezer until water extraction by cryogenic vacuum distillation, as described by Sun et al. (2014). In addition, the weight of samples was recorded before and after extraction. Unconfined water (1 mL) was also sampled at the end of equilibration, underwent cryogenic vacuum distillation and was stored in a refrigerator.

The extracted water was analyzed with a Cavity Ring Down (CRD) Spectrometer using a L2120 – i Analyzer (Picarro, USA). Measurements were repeated until values became stable around a mean. Mean analytical uncertainties quantified as SD of different replicate measurements for each sample were  $\pm 0.06\text{‰}$  for  $\delta^{18}\text{O}$ , and  $\pm 0.27\text{‰}$  for  $\delta^2\text{H}$ . Post-processing correction was made by running the ChemCorrect™ v1.2.0 (Picarro Inc.) to exclude the influence of volatiles according to Martín-Gómez et al. (2015).

## 2.4 Experiment A: Influence of materials

This experiment focused on the enrichment between water in different materials and unconfined water after equilibration. Dishes containing oven dried silage, hay, oven dried and fresh hemic litter, oven dried and fresh fibric litter, filter paper, bleached medical cotton, casein powder, or flour were all placed in different chambers for equilibration with unconfined water to avoid interference of volatiles in different materials. Eight samples for each material that differed in solid:water ratio were put in one chamber. Some materials (i.e., litter, filter paper, silage) were replicated in different experiments. The



118 maximum number of samples for one material (silage) was 72. Flour and casein were powders and prone to form dust during  
119 vacuum water extraction. To prohibit this, the opening of vials containing flour and casein powder was covered by parafilm  
120 with tiny holes.

## 121 2.5 Experiment B: Influence of isotopic composition in unconfined water

122 This experiment aimed to find evidence that the enrichment was independent of the isotopic composition of the unconfined  
123 water. This independence will also prove that the enrichment cannot be caused by old water within the materials due to  
124 insufficient equilibration. Eight samples of oven dried silage in each case were placed into chambers to equilibrate with five  
125 different unconfined waters.

## 126 2.6 Experiment C: Pretreatment of silage

127 This experiment investigated the influence of volatiles on the isotope measurement and it assessed the effect of silage solutes  
128 on isotopic fractionation between silage water and vapor.  
129 Fresh silage was divided into three groups (8 samples each): The first group did not undergo any pretreatment. For the  
130 second group, about 20 g of silage was immersed in 7 L of deionized water for about 2 min, stirred during immersion, then  
131 taken out using a colander and flushed with distilled water. After that we squeezed the silage by hand until no water drained  
132 off. This washing process was repeated three times. Finally, we reduced the water content of the washed silage by drying at  
133 80 °C for 40 min. For the third group, silage was oven dried for 16 h at 100 °C to remove water and organic volatiles. These  
134 three groups (we call them fresh silage, washed silage and oven dried silage, respectively, thereafter) were placed in  
135 individual chambers and equilibrated with tap water for 100 h.

## 136 2.7 Statistics

137 For statistical evaluation we report two-sided 95% limits of confidence (abbreviated CL) to separate between treatments and  
138 OLS regression to describe relations between two variables. Measured values were fitted to expected relations by  
139 minimizing the root mean squared error (RMSE). Statistical requirements (normal distribution) were met in all cases.

## 140 2.8 Modelling

141 Conceptually, we assumed water to be part of one of two pools, which are arranged in a shell-like structure around the solid:  
142 an inner shell (or layer) which is in immediate contact or close to the surface of the solid and an outer layer that differs in  
143 thickness depending on the moisture content or solid:water ratio of the sample. Assuming that the outer layer has the same  
144 isotopic composition as the unconfined water once equilibrium was attained and that the inner layer has an isotopic  
145 composition that is influenced by the solid, the isotope composition of total adsorbed water ( $\delta_T$ ) was defined as:

$$146 \quad \delta_T = f_O \times \delta_U + (1 - f_O) \times \delta_S, \quad (1)$$

147 where  $f_O$  is the fraction of water in the outer layer isotopically identical to the unconfined water,  $\delta_U$  and  $\delta_S$  are the isotope  
148 compositions of unconfined water and water influenced by the surface.

149 We defined enrichment ( $\epsilon_S$ ) between  $\delta_S$  and  $\delta_U$  as

$$150 \quad \epsilon_S = (\delta_S - \delta_U) / (1000 + \delta_U) \times 1000 \quad (2)$$

151 Combining eq. (1) and (2) leads to:

$$152 \quad \delta_T = \frac{1000 + \epsilon_S f_O}{1000} \delta_U + \epsilon_S f_O \quad (3)$$

153 From this it follows that the apparent enrichment ( $\epsilon_a$ ) between the total water in the material and unconfined water is given  
154 as:

$$155 \quad \epsilon_a = (\delta_T - \delta_U) / (1000 + \delta_U) \times 1000 = (1 - f_O) \times \epsilon_S = f_I \times \epsilon_S \quad (4)$$



The fraction constituted by the inner layer  $f_i$  in eq. (4) can be replaced by the ratio between  $R_i$ , the volumetric ratio of water:solid associated with the layer that is influenced by the surface, and  $R_T$ , the volumetric water:solid ratio of total adsorbed water:

$$\varepsilon_a = \varepsilon_s \times R_i/R_T \quad (5).$$

Assuming that the size of the inner layer  $R_i$  as well as  $\varepsilon_s$  are constant for a certain material,  $\varepsilon_a$  should be related linearly to the inverse of  $R_T$ , which is the volumetric solid:water ratio for the total adsorbed water.

## 2.9 Application case

Soil at 7 cm and 20 cm depths and rain water were sampled at the grassland in Grünschaige Experimental Station, Germany (48°23'N, 11°50'E, pasture #8 in Schnyder et al. (2006); 8.3 % organic matter, 30 % clay, 22 % sand) following the MIBA protocol (Moisture Isotopes in Biosphere and Atmosphere) at monthly intervals during the vegetation period (April to November) from 2006 to 2012 and at weekly intervals during the cold season (October to February) in 2015/2016. Soil sampling was always carried out on dry days at midday (between 11 a.m. and 16 p.m.). Two replicates of soil samples were collected on each sampling date. The data was used to verify if there was a consistent offset between soil water and rain water.

Further, the winter data, effects of soil evaporation from the vegetation covered soil, can be excluded. We verify if the offset can be corrected by accounting for the volumetric solid:water ratio of the soil according to our model. To this end, the sand content of the soil was not considered in the calculation of the solid:water ratio given that the contribution of sand to water storage is marginal (Walczak et al., 2002) and sand grains usually are coated by clay, sesquioxides, organic matter and biofilms and do not directly interact with water (Bisdorf et al., 1993; Bolster et al., 2001).

## 3 Results

### 3.1 Experiment A: Influence of materials

The apparent enrichment (*sensu* eq. 4) of  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  was negative and significant for all materials, except for  $^{18}\text{O}$  with filter paper and cotton and for  $^2\text{H}$  in a few samples of cotton. The volumetric solid:water ratios differed between materials but also between different samples within the materials providing a wide range.  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  apparent enrichments decreased significantly with volumetric solid:water ratio over the range of materials. The decrease was also significant for the different samples within each material (Fig. 2).

### 3.2 Experiment B: Influence of isotopic composition in unconfined water

The isotope composition of adsorbed water correlated closely with the unconfined water due to the wide range compared to the measurement errors ( $R^2 = 0.9990$  and  $0.9989$  for O and H, respectively; Table 1). However, the regressions showed that the intercept differed significantly from zero and the slope from one, which indicated that the isotope composition of adsorbed water was significantly different from that of unconfined water.

Equation (3) predicted a linear relation between  $\delta_T$  and  $\delta_U$  similar to the linear regressions shown in Table 1. Different to a regression, however, the slope and the intercept of eq. (3) are not independent but depend on  $\varepsilon_s \times f_0$ . To account for this dependency, the slope and the intercept of the linear equations were estimated by adjusting  $\varepsilon_s \times f_0$  in eq. (3) to minimize RMSE, while fitting the measured  $\delta_T$  and  $\delta_U$  values. The optimal fits lead to:

$$\begin{aligned} ^{18}\text{O}: \quad \delta_T &= \frac{1000-1.23}{1000} \times \delta_U - 1.23 \\ ^2\text{H}: \quad \delta_T &= \frac{1000-22.6}{1000} \times \delta_U - 22.6 \end{aligned} \quad (6)$$



192 The  $R^2$  between the predictions resulting from the two-layer model and the measurement were similar to that of the linear  
 193 regression ( $R^2 = 0.9990$  for  $^{18}\text{O}$  and  $0.9989$  for  $^2\text{H}$ ), although the model has one degree of freedom less than the regression.  
 194 The resulting optimal  $\epsilon_s \times f_o$  values were  $-1.23 \text{ ‰}$  for  $^{18}\text{O}$  and  $-22.6 \text{ ‰}$  for  $^2\text{H}$  meaning that the effect was 18 times stronger  
 195 for  $^2\text{H}$  than for  $^{18}\text{O}$ .

196  
 197 Equation (5) predicted that the apparent enrichment changes linearly with the solid:water ratio. This relation was highly  
 198 significant also in the case when waters with very differently isotopic composition were used ( $R^2$ :  $0.7589$  and  $0.8599$  for O  
 199 and H, respectively; Fig. 3). These relations were identical for very heavy, heavy, tap, light and very light water.

### 200 3.3 Experiment C: Pretreatment of silage

201 There was no significant difference between mean gravimetric water contents (based on dry matter) of washed silage ( $153 \text{ ‰}$   
 202  $\pm 33 \text{ ‰}$ ) and fresh silage ( $128 \text{ ‰} \pm 10 \text{ ‰}$ ) after 100 h equilibration. The water content of oven dried silage did not reach again  
 203 the same water content as fresh silage but was significantly lower ( $81 \text{ ‰} \pm 13 \text{ ‰}$ ). The apparent enrichment of washed silage,  
 204 oven dried silage and fresh silage all decreased with the solid:water ratio (Fig. 4), as already noted in the experiment with  
 205 different materials (Fig. 2) or in investigations with unconfined waters of different isotopic composition (Fig. 3). Washing  
 206 and oven drying should have removed most solutes and volatiles respectively and thus have created a large variation in the  
 207 amount of solutes and volatiles among the treatments. Still, the relationship between enrichment of three types of silage and  
 208 solid:water ratio followed the same line and the areas overlapped each other for the three types of silage (Fig. 4). This  
 209 implied that neither the volatiles, which possibly could have adulterated the measurements, nor the solutes, which possibly  
 210 could have influenced water activity in the silage, were the reason of enrichment. The different treatments, however,  
 211 separated along the common line due to their differences in water content, which again corroborated the prediction that the  
 212 apparent enrichment should linearly change with solid:water ratio.

### 213 3.4 Combining experiments A, B and C

214 When combining all experiments with different materials, different pretreatments and different unconfined waters, apparent  
 215 enrichments covered a wide range of about  $5 \text{ ‰}$  for  $^{18}\text{O}$  and  $46 \text{ ‰}$  for  $^2\text{H}$  (Fig. 5). Even within the same materials, the range  
 216 was up to  $2.5 \text{ ‰}$  for  $^{18}\text{O}$  and  $25 \text{ ‰}$  for  $^2\text{H}$ . Apparent enrichments within materials linearly decreased with the volumetric  
 217 solid:water ratio.

218 In order to exclude that the enrichment was caused by incomplete extraction, we compared the observed enrichments with  
 219 predictions based on Rayleigh fractionation resulting from incomplete extraction (Fig. 5). The enrichments predicted for  
 220 Rayleigh fractionation fell far apart the observed enrichments. The average deviation between the expected and the observed  
 221  $^2\text{H}$  enrichment was about  $15 \text{ ‰}$ . Furthermore, the slope of the relation between the enrichment of  $^2\text{H}$  and  $^{18}\text{O}$  was  
 222 significantly steeper for the observed enrichment than the slope predicted for a Rayleigh process. Additionally, an  
 223 unrealistically small fraction of water would have to be extracted (far below  $0.8$ ) to cause the same enrichment of  $^2\text{H}$  as  
 224 observed for most of the samples.

### 225 3.5 Application

226 For the growing season, soil water at 20 cm depth and 7 cm depth showed a distinct deviation from the local meteoric water  
 227 line (mean deviation for  $^2\text{H}$ :  $-8.1 \text{ ‰}$ ) with a slope almost identical to that of the meteoric water line (regression lines in Fig.  
 228 6, top panel). An identical mismatch was detected for the winter season (markers in Fig. 6, top panel) for which confounding  
 229 effects of evaporation are minimal (on average potential evaporation  $0.65 \text{ mm/d}$ ; actual evaporation  $0.54 \text{ mm/d}$ ; precipitation  
 230  $1.86 \text{ mm/d}$ ) and because the soil was completely covered by grass.



The deviation between the winter season data and the local meteoric water line correlated significantly ( $p < 0.001$ ) with the solid:water ratio for 7 cm depth but not for 20 cm depth. For both depths, the data moved closer to the local meteoric water line when the influence of confined water was removed by applying the general regression with solid:water ratio from Fig. 2 (Fig. 6, bottom panel). The mean deviation for  $^2\text{H}$  changed from  $-8.1\text{‰}$  to  $1.0\text{‰}$  due to this correction.

## 4 Discussion

The extraction of water from solid-water mixtures can be biased by incomplete extraction (Araguás-Araguás et al., 1995) or by the exchange of hydrogen or oxygen from the soil material with water molecules (Meißner et al., 2014). Here we add another confounding effect, which is the inhomogeneous isotopic composition of water above a solid surface. In the following we will discuss (1) whether the observed effect can be due to measuring errors or other reasons than the proposed surface effect, (2) what could be possible reasons of the surface effect, (3) which fields of application will this surface effect likely be important, and (4) which further work related to the surface effect may follow.

### 4.1 Excluding other mechanisms than the proposed surface effect

The study provided clear evidence that the water adsorbed by organic surfaces differed from what would be expected from the isotopic composition of the source water and it showed that this deviation became larger with decreasing water content. Alternative mechanisms leading to an isotopic offset other than the proposed surface effect could be (A) volatiles adulterating the measurements; (B) solutes influencing the isotopic composition of adsorbed water; (C) insufficient equilibration time; (D) incomplete extraction of water; (E) metabolically produced water from microorganisms adhering to the materials; (F) exchange of hydrogen and oxygen between the organic matter and the adsorbed water.

(A) The surface effect was largest for flour and casein that do not produce volatiles. Also the filter paper and cotton, which contain no volatiles, had the decreasing trend between apparent enrichment and solid:water ratio (Fig. 2). Even for silage the influence of volatiles was not evident because washed or oven-dried silage, which should have lost all their volatiles, behaved identical to fresh silage. Also the error in water content caused by not accounting for volatile losses was negligible. Using the correction function by Porter and Murray (2001) to calculate the true water content from the loss of weight, moves the respective data points of silage in Fig. 5 only invisibly (about  $0.03\text{ L/L}$  towards right side).

(B) Solutes in water can influence the isotopic fractionation between water and vapor because the energy stage of water molecules bound in the primary hydration sphere of cations and anions differs from that of the remaining bulk water molecules (Kakiuchi, 2007). This effect has been shown for many salts (e.g.,  $\text{KCl}$ ,  $\text{NaCl}$ ,  $\text{Na}_2\text{SO}_4$  and  $\text{ZnSO}_4$ ). The strength of this effect varies between different ions and may be small (Kakiuchi, 2007; Sofer and Gat, 1975; Stewart and Friedman, 1975).  $\text{NaCl}$  even does not have a measurable effect on  $^{18}\text{O}$  (O'Neil and Truesdel, 1991). Most of the solutes in our materials were organics for which the effect is unknown. However, this effect must have been small as the washed silage did not show a different pattern in enrichment compared to fresh silage (Fig. 4). Also the filter paper of analytical grade and bleached cotton that both should not carry any solutes did not show a different pattern.

(C) Insufficient time for equilibration may especially be relevant for silage and litter, which had the highest initial water content. For silage we could show that the apparent enrichment was independent of the isotopic composition in the unconfined water (Experiment B) despite the wide range of differently labelled unconfined waters (range for  $^{18}\text{O}$ :  $32\text{‰}$ ; range for  $^2\text{H}$ :  $285\text{‰}$ ). However, any old water would have led to a separation in the apparent enrichment. In contrast, our results were in accordance with the general rule that isotopic enrichment is independent of the isotope composition of the source, which is also underlying eq. (4) and (5). Furthermore, all our experiments used deionized water prepared from tap water, except for the experiment with labelled waters for which we can exclude the existence of old water. Our deionized water was similar in isotopic composition to silage water and soil water. The mean  $\delta^{18}\text{O}$  of our water was  $-10\text{‰}$  while the





mean for 52 fresh silage samples analyzed by Sun et al. (2014) was -11 ‰ (SD 3 ‰). A small fraction of old water thus cannot cause the large observed effects.

(D) An incomplete extraction should cause a large error at low moisture content, similar to the general relation between solid:water ratio and enrichment that we have observed (Fig. 6). However, the predicted enrichment by incomplete extraction based on a Rayleigh fractionation fell far apart from the observed enrichment (Fig. 5). In addition, no significant weight difference before and after oven drying of the samples was observed after vacuum extraction. Incomplete extraction is thus an unlikely explanation.

(E) Kreuzer-Martin et al. (2005) found that 10 % of the total water extracted from *Escherichia coli* cells during the log-phase of growth was generated by metabolism from atmospheric oxygen. Thus, intracellular water was distinguishable from extracellular water in  $\delta^{18}\text{O}$ . We flushed the chambers with nitrogen gas before equilibration to reduce availability of atmospheric oxygen and minimize microbial growth. For materials like silage dried at 100 °C or filter paper, any significant microbial growth is unlikely. Furthermore, isotopic adulteration caused by microorganisms should have caused  $^{18}\text{O}$  and  $^2\text{H}$  deviations in the opposite direction for the very heavy and the very light labeled experiments akin to the experiments by Kreuzer-Martin et al. (2005). In contrast to this  $^{18}\text{O}$  and  $^2\text{H}$  were always depleted in our experiments regardless of the isotope composition of unconfined water.

(F) Hydrogen bound to oxygen and nitrogen in many organic materials like bitumen, cellulose, chitin, collagen, keratin or wood may exchange isotopically with ambient water hydrogen (Bowen et al., 2005; Schimmelmann, 1991). At room temperature, this isotopic exchange occurs rapidly in water and an exchange with vapor is even several orders of magnitude faster (Bowen et al., 2005; Schimmelmann et al., 1993). Thus the exchange and the subsequent equilibration with the unconfined water will happen within 100 h. Furthermore, an exchange of hydrogen would not explain the observed offset in  $^{18}\text{O}$ .

## 4.2 Possible reason for the surface effect

The enrichments became more negative with increasing solid:water ratio and they followed the predictions of eq. 5. This implied that similar enrichments existed in different materials and that the simple two-layer model sufficiently described the experimental values. Abundant evidence exists that the properties of water change close to a surface (Anderson and Low, 1957; Goldsmith and Muir, 1960; Miranda et al., 1998). A hydrogen-bonded ice like network of water grows up as the relative humidity increases. Above 60% relative humidity, the liquid water configuration grows on top of the ice like layer (Asay and Kim, 2005). This transition from a two-dimensional ice-like water to a three-dimensional water-like layer has been already been shown in several cases (Kendall and Martin, 2005). As we used 100% relative humidity in our chamber, both layers should have been present.

The anomalies of water close to a surface appear not to be particularly affected by the detailed chemical nature of the solid substrates with which the water is in contact. This is referred to as the “paradoxical effect” and is tentatively interpreted in terms of an energy delocalization phenomenon (Drost-Hansen, 1978). This agrees with our observation that the difference between materials was small compared to the large variation of the effect caused by a varying solid-water ratio. The small differences between materials that appear in Fig. 2 may hence only be an effect due to differences between the different materials in their specific surface area per volume of solid but not due to their chemical nature.

In accordance with our study, Richard et al. (2007) found that water adsorbed in porous silica tubes was depleted in  $^2\text{H}$  compared to unconfined water and depletion increased with decreasing water quantity as a result of the interplay of molecular vibrational frequencies and intermolecular H-bonding. This mostly depends on the difference in zero-point energy between the  $^{16/18}\text{O}-^{1/2}\text{H}$  bonds, which is compressed at the transition between the bulk liquid and the confined liquid influenced by the surface (Richard et al., 2007). Our data show, that the effect is much larger for  $^2\text{H}$  than for  $^{18}\text{O}$  and it





312 practically disappears for  $^{18}\text{O}$  when the solid:water ratio decreases below 0.5 (Fig. 6). This may explain why the effect has  
313 been previously described for  $^2\text{H}$  but not for  $^{18}\text{O}$ .

#### 314 4.3 Fields of application

315 The isotopic composition of water in porous samples is usually determined by extracting all water in order to avoid any shift  
316 caused by Rayleigh fractionation. Hence, the inner layer close to the surface and the outer layer will be mixed. For many  
317 processes, especially in the transport of liquid water (e.g., groundwater recharge, stream flow discharge, water uptake by  
318 plants) only the outer, mobile layer will be relevant. The extraction of total water will then give a biased estimate of the  
319 mobile water. In accordance with our hypothesis, Brooks et al. (2010) even suggested two different soil water worlds to  
320 explain their data (mobile water and tightly bound water), which were not identical in terms of isotope composition. Tang  
321 and Feng (2001) also found isotopic differences between mobile and immobile water in soil and explained this by  
322 incomplete replacement of soil water by rainwater. Our laboratory experiments aimed to exclude such an effect. In our  
323 application case we also found a consistent offset between rain water and soil water that cannot result from incomplete  
324 replacement of old rain water in soil with new rain water because soil water had an offset from the meteoric water line. Such  
325 an offset has been shown for many locations around the world (Brooks et al., 2010; Evaristo et al., 2015), which challenges  
326 the assumption in land surface models that plants and streams derive their water from a single, well mixed subsurface water  
327 reservoir.

328 In other cases, which focus on the liquid-solid interface, only the water of the inner layer, which is influenced by the surface  
329 effect, will be relevant. For example, in studies of cell wall formation or degradation, the total water should be a biased  
330 estimate of the isotopic composition near the cell wall. Due to the change in apparent enrichment with water content, the  
331 total cell water will change just by a variation in vacuole volume even if the isotopic composition near the cell wall and in  
332 the vacuole remain unchanged.

#### 333 4.4 Further work

334 Solid:water ratio is clearly not the best parameter to describe the two-layer model. The relation should be influenced by  
335 specific surface area and by wettability. Hence, the water volume per wetted surface area would likely be a better parameter.  
336 For instance, when we wet the filter paper inhomogeneously, we got random results because the average solid:water ratio  
337 neither reflected the situation of the wet spots nor that of the dry spots. Also the increasing scatter for solid:water ratios  $>1.5$   
338 (Fig. 6) likely resulted from an inhomogeneous water distribution in these rather dry samples that may have left some parts  
339 of the sample completely dry and thus underestimated the water content of other parts. Still, our model was easy to apply  
340 and it worked sufficiently for the wide variety of materials examined. More materials varying in hygroscopic/hydrophobic  
341 behavior and in surface area should be included to better understand the rule behind the variation of enrichment and to  
342 expand the model.

#### 343 5 Conclusions

344 There was an abundance of evidence to suggest that the surface effect influenced the enrichment between water adsorbed by  
345 organic matter and unconfined water. Many hypothetical reasons for an erroneous enrichment could be excluded. The  
346 variation of apparent enrichment with water content was well described by a simple, easy to apply two-layer model. This  
347 enrichment should not be neglected when the surface area is huge and the water content is low. The surface effect will  
348 become especially relevant for processes happening at the liquid-surface interface like the growth or degradation of the  
349 organic materials.



### 350 Author contribution

351 G.C. and K.A. designed the experiments and analyzed the data. G.C. carried out the experiments and wrote a first draft. All  
352 authors developed and approved the manuscript.

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### 356 References

- 357 Anderson, D. M. and Low, P. F.: Density of water adsorbed on Wyoming Bentonite, *Nature*, 180, 1194, doi:  
358 10.1038/1801194a0, 1957.
- 359 Araguás-Araguás, L., Rozanski, K., Gonfiantini, R., and Louvat, D.: Isotope effects accompanying vacuum extraction of  
360 soil-water for stable-isotope analyses, *J. Hydrol.*, 168, 159-171, doi: 10.1016/0022-1694(94)02636-p, 1995.
- 361 Asay, D. B. and Kim, S. H.: Evolution of the adsorbed water layer structure on silicon oxide at room temperature, *J. Phys.*  
362 *Chem.*, 109, 16760-16763, doi: 10.1021/jp053042o, 2005.
- 363 Bisdom, E. B. A., Dekker, L. W., and Schouite, J. F. T.: Water repellency of sieve fractions from sandy soils and  
364 relationships with organic material and soil structure, *Geoderma*, 56, 105-118, doi: 10.1016/0016-7061(93)90103-r,  
365 1993.
- 366 Bolster, C. H., Mills, A. L., Hornberger, G. M., and Herman, J. S.: Effect of surface coatings, grain size, and ionic strength  
367 on the maximum attainable coverage of bacteria on sand surfaces, *J. Contam. Hydrol.*, 50, 287-305, doi:  
368 10.1016/s0169-7722(01)00106-1, 2001.
- 369 Bowen, G. J.: Isoscapes: spatial pattern in isotopic biogeochemistry, *Annu. Rev. Earth Planet. Sci.*, 38, 161-187, doi:  
370 10.1146/annurev-earth-040809-152429, 2010.
- 371 Bowen, G. J., Chesson, L., Nielson, K., Cerling, T. E., and Ehleringer, J. R.: Treatment methods for the determination of  $\delta^2\text{H}$   
372 and  $\delta^{18}\text{O}$  of hair keratin by continuous-flow isotope-ratio mass spectrometry, *Rapid Commun. Mass Spectrom.*, 19,  
373 2371-2378, doi: 10.1002/rcm.2069, 2005.
- 374 [Brooks, J. R., Barnard, H. R., Coulombe, R., and McDonnell, J. J.: Ecohydrologic separation of water between trees and](#)  
375 [streams in a Mediterranean climate, \*Nature Geoscience\*, 3, 100-104, doi: 10.1038/ngeo722, 2010.](#)
- 376 Chorover, J., Kretschmar, R., Garcia-Pichel, F., and Sparks, D. L.: Soil biogeochemical processes within the Critical Zone,  
377 *Elements*, 3, 321-326, doi: 10.2113/gselements.3.5.321, 2007.
- 378 Drost-Hansen, W.: Water at biological interfaces - structural and functional aspects, *Phys. Chem. Liq.*, 7, 243-248, doi:  
379 10.1080/00319107808084734, 1978.
- 380 [Evaristo, J., Jasechko, S., and McDonnell, J. J.: Global separation of plant transpiration from groundwater and streamflow,](#)  
381 [Nature, 525, 91-94, doi: 10.1038/nature14983, 2015.](#)
- 382 Gat, J. R.: Oxygen and hydrogen isotopes in the hydrologic cycle, *Annu. Rev. Earth Planet. Sci.*, 24, 225-262, doi:  
383 10.1146/annurev.earth.24.1.225, 1996.
- 384 Goldsmith, B. J. and Muir, J.: Surface ion effects in the dielectric properties of adsorbed water films, *Trans. Faraday Soc.*,  
385 56, 1656-1661, doi: 10.1039/TF9605601656 1960.
- 386 Haverd, V. and Cuntz, M.: Soil-Litter-Iso: A one-dimensional model for coupled transport of heat, water and stable isotopes  
387 in soil with a litter layer and root extraction, *J. Hydrol.*, 388, 438-455, doi: 10.1016/j.jhydrol.2010.05.029, 2010.



- 388 Helliker, B. R. and Griffiths, H.: Toward a plant-based proxy for the isotope ratio of atmospheric water vapor, *Global*  
389 *Change Biol.*, 13, 723-733, doi: 10.1111/j.1365-2486.2007.01325.x, 2007.
- 390 Kakiuchi, M.: Hydrogen isotope fractionation in aqueous alkaline earth chloride solutions, *J. Phys. Sci.*, 62, 721-728, doi:  
391 10.1515/zna-2007-1208, 2007.
- 392 Kendall, T. A. and Martin, S. T.: Mobile ions on carbonate surfaces, *Geochim. Cosmochim. Acta*, 69, 3257-3263, doi:  
393 10.1016/j.gca.2005.02.010, 2005.
- 394 Kohn, M. J.: Predicting animal  ~~$\delta^{18}\text{O}$~~  $\delta^{18}\text{O}$ : Accounting for diet and physiological adaptation, *Geochim. Cosmochim.*  
395 *Acta*, 60, 4811-4829, doi: 10.1016/s0016-7037(96)00240-2, 1996.
- 396 Kreuzer-Martin, H. W., Ehleringer, J. R., and Hegg, E. L.: Oxygen isotopes indicate most intracellular water in log-phase  
397 *Escherichia coli* is derived from metabolism, *Proc. Natl. Acad. Sci. U.S.A.*, 102, 17337-17341, doi:  
398 10.1073/pnas.0506531102, 2005.
- 399 Martín-Gómez, P., Barbeta, A., ~~and~~ Voltas, J., ~~Peñuelas, J., Dennis, K., Palacio, S., Dawson, T. E., and Ferrio, J. P.~~: Isotope-  
400 ratio infrared spectroscopy: a reliable tool for the investigation of plant-water sources?, *New Phytol.*, 207, 914-927,  
401 doi: 10.1111/nph.13376, 2015.
- 402 Meißner, M., Koehler, M., Schwendenmann, L., Hoelscher, D., and Dyckmans, J.: Soil water uptake by trees using water  
403 stable isotopes ( ~~$\delta^2\text{H}$~~  $\delta^2\text{H}$  and  ~~$\delta^{18}\text{O}$~~  $\delta^{18}\text{O}$ ), - a method test regarding soil moisture, texture and carbonate,  
404 *Plant Soil*, 376, 327-335, doi: 10.1007/s11104-013-1970-z, 2014.
- 405 Miranda, P. B., Xu, L., Shen, Y. R., and Salmeron, M.: Icelike water monolayer adsorbed on mica at room temperature,  
406 *Phys. Rev. Lett.*, 81, 5876-5879, doi: 10.1103/PhysRevLett.81.5876, 1998.
- 407 Nordt, L. C., Hallmark, C. T., Driese, S. G., Dworkin, S. I., and Atchley, S. C.: Biogeochemical characterization of a  
408 lithified paleosol: Implications for the interpretation of ancient Critical Zones, *Geochim. Cosmochim. Acta*, 87,  
409 267-282, doi: 10.1016/j.gca.2012.03.019, 2012.
- 410 O'Neil, J. R. and Truesdell, A. H.: Oxygen isotope fractionation studies of solute-water interactions. In: *Stable Isotope*  
411 *Geochemistry*, edited by Taylor, H. P., Geochemical Society Press, 17-25, 1991.
- 412 Oerter, E., Finstad, K., Schaefer, J., Goldsmith, G. R., Dawson, T., and Amundson, R.: Oxygen isotope fractionation effects  
413 in soil water via interaction with cations (Mg, Ca, K, Na) adsorbed to phyllosilicate clay minerals, *J. Hydrol.*, 515,  
414 1-9, doi: 10.1016/j.jhydrol.2014.04.029, 2014.
- 415 Porter, M. G. and Murray, R. S.: The volatility of components of grass silage on oven drying and the inter-relationship  
416 between dry-matter content estimated by different analytical methods, *Grass Forage Sci.*, 56, 405-411, doi:  
417 10.1046/j.1365-2494.2001.00292.x, 2001.
- 418 Richard, T., Mercury, L., Massault, M., and Michelot, J.: Experimental study of D/H isotopic fractionation factor of water  
419 adsorbed on porous silica tubes, *Geochim. Cosmochim. Acta*, 71, 1159-1169, doi: 10.1016/j.gca.2006.11.028, 2007.
- 420 Schimmelmann, A.: Determination of the concentration and stable isotopic composition of nonexchangeable hydrogen in  
421 organic matter, *Anal. Chem.*, 63, 2456-2459, doi: 10.1021/ac00021a013, 1991.
- 422 Schimmelmann, A., Miller, R. F., and Leavitt, S. W.: Hydrogen isotopic exchange and stable isotope ratios in cellulose,  
423 wood, chitin, and amino compounds. In: *Climate Change in Continental Isotopic Records*, edited by Swart, P. K.,  
424 Lohmann, K. C., McKenzie, J., and Savin, S., American Geophysical Union Press, Washington DC, 367-374, 1993.
- 425 Schmidt, M., Maseyk, K., Lett, C., Biron, P., Richard, P., Bariac, T., and Seibt, U.: Reducing and correcting for  
426 contamination of ecosystem water stable isotopes measured by isotope ratio infrared spectroscopy, *Rapid Commun.*  
427 *Mass Spectrom.*, 26, 141-153, doi: 10.1002/rcm.5317, 2012.
- 428 Schnyder, H., Schwertl, M., Auerswald, K., and Schaefele, R.: Hair of grazing cattle provides an integrated measure of the  
429 effects of site conditions and interannual weather variability on  ~~$\delta^{13}\text{C}$~~  $\delta^{13}\text{C}$  of temperate humid grassland,  
430 *Global Change Biol.*, 12, 1315-1329, doi: 10.1111/j.1365-2486.2006.01169.x, 2006.



- 431 Schoeneberger, P. J., Wysocki, D. A., and Benham, E. C.: Field book for describing and sampling soils, National Soil  
432 Survey Center Press, Lincoln UK, 300, 2012.
- 433 Schultz, N. M., Griffis, T. J., Lee, X., and Baker, J. M.: Identification and correction of spectral contamination in  $^2\text{H}/^1\text{H}$  and  
434  $^{18}\text{O}/^{16}\text{O}$  measured in leaf, stem, and soil water, Rapid Commun. Mass Spectrom., 25, 3360-3368, doi:  
435 10.1002/rcm.5236, 2011.
- 436 Sofer, Z. and Gat, J. R.: Isotope composition of evaporating brines - effect of isotopic activity ratio in saline solutions, Earth.  
437 Planet. Sci. Lett., 26, 179-186, doi: 10.1016/0012-821x(75)90085-0, 1975.
- 438 Stewart, M. K. and Friedman, I.: Deuterium fractionation between aqueous salt-solutions and water-vapor, J. Geophys. Res.,  
439 80, 3812-3818, doi: 10.1029/JC080i027p03812, 1975.
- 440 Sun, L., Auerswald, K., Schaeufele, R., and Schnyder, H.: Oxygen and hydrogen isotope composition of silage water, J.  
441 Agric. Food Chem., 62, 4493-4501, doi: 10.1021/jf405703g, 2014.
- 442 [Tang, K. L. and Feng, X. H.: The effect of soil hydrology on the oxygen and hydrogen isotopic compositions of plants'  
443 source water. Earth. Planet. Sci. Lett., 185, 355-367, doi: 10.1016/s0012-821x\(00\)00385-x, 2001.](#)
- 444 Vazquez-Ortega, A., Hernandez-Ruiz, S., Amistadi, M. K., Rasmussen, C., and Chorover, J.: Fractionation of dissolved  
445 organic matter by (oxy) hydroxide-coated sands: competitive sorbate displacement during reactive transport,  
446 Vadose Zone J., 13, 1539-1663, doi: 10.2136/vzj2013.10.0179, 2014.
- 447 Walczak, R., Rovdan, E., and [Witkowska-Walczak](#) Witkowska, B.: Water retention characteristics of peat and sand mixtures,  
448 International Agrophysics, 16, 161-165, 2002.
- 449 Welhan, J. A. and Fritz, P.: Evaporation pan isotopic behavior as an index of isotopic evaporation conditions, Geochim.  
450 Cosmochim. Acta, 41, 682-686, doi: 10.1016/0016-7037(77)90306-4, 1977.
- 451 West, A. G., Goldsmith, G. R., Matimati, I., and Dawson, T. E.: Spectral analysis software improves confidence in plant and  
452 soil water stable isotope analyses performed by isotope ratio infrared spectroscopy (IRIS), Rapid Commun. Mass  
453 Spectrom., 25, 2268-2274, doi: 10.1002/rcm.5126, 2011.
- 454 WRB: World reference base for soil resources 2014, FAO Press, Rome, 106-191, 2014.



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475 **Table 1: Regressions between water adsorbed by silage ( $\delta_T$ ) and unconfined water ( $\delta_U$ ) for five types of water (very heavy, heavy,**  
 476 **tap, light and very light water) based on equation  $\delta_T = \text{slope} \times \delta_U + \text{intercept}$ ;  $n = 40$ ; values in parenthesis denote the 95%**  
 477 **confidence level.**

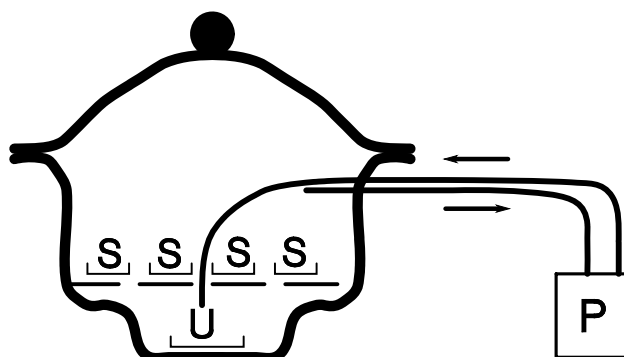
	$\delta^{18}\text{O}$	$\delta^2\text{H}$
Intercept	-1.30 ( $\pm 0.14$ )	-22.9 ( $\pm 1.1$ )
Slope	0.987 ( $\pm 0.010$ )	0.968 ( $\pm 0.011$ )
$R^2$	0.9990	0.9989

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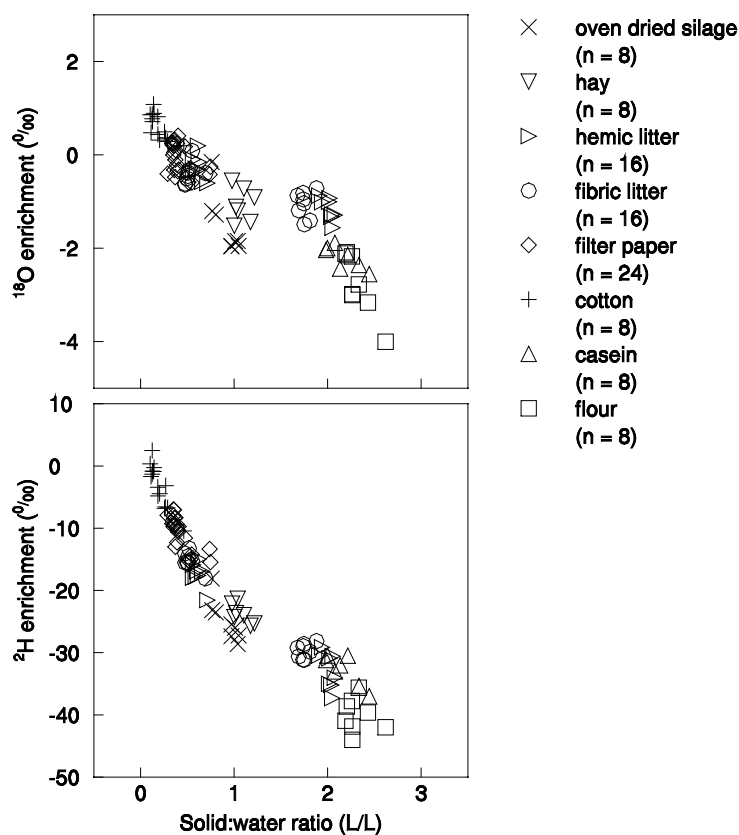
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Fig 1: Experimental set-up with an exsiccator vessel as the equilibration chamber. P: recycling pump ensuring air mixing and air movement within the chamber; U: unconfined water filled in the bottom part of the chamber; S: samples placed on top of the perforated middle plate. The arrows indicate the direction of air flow. Vaseline was used as sealant between the lid and the vessel.



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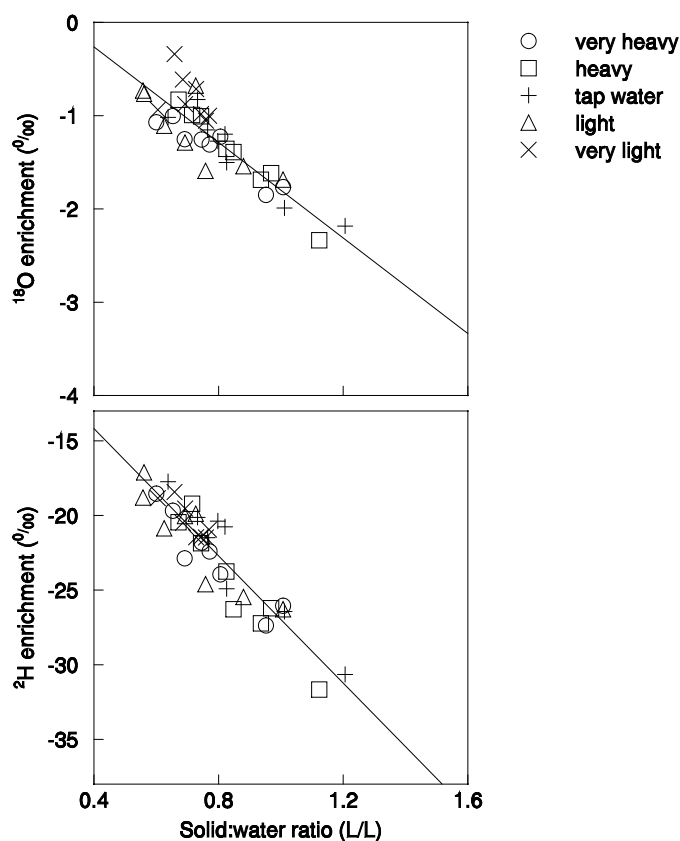
489 Fig. 2: Relationship between volumetric solid:water ratio and apparent enrichment of  $^{18}\text{O}$  and  $^2\text{H}$  between unconfined water and  
 490 water adsorbed by different materials. Taken together, the regressions are  $y = -0.906 x$  ( $R^2 = 0.6789$ ;  $N = 96$ ) for the enrichment of  
 491  $^{18}\text{O}$  and  $y = -17.75 x$  ( $R^2 = 0.8355$ ) for the enrichment of  $^2\text{H}$ .  
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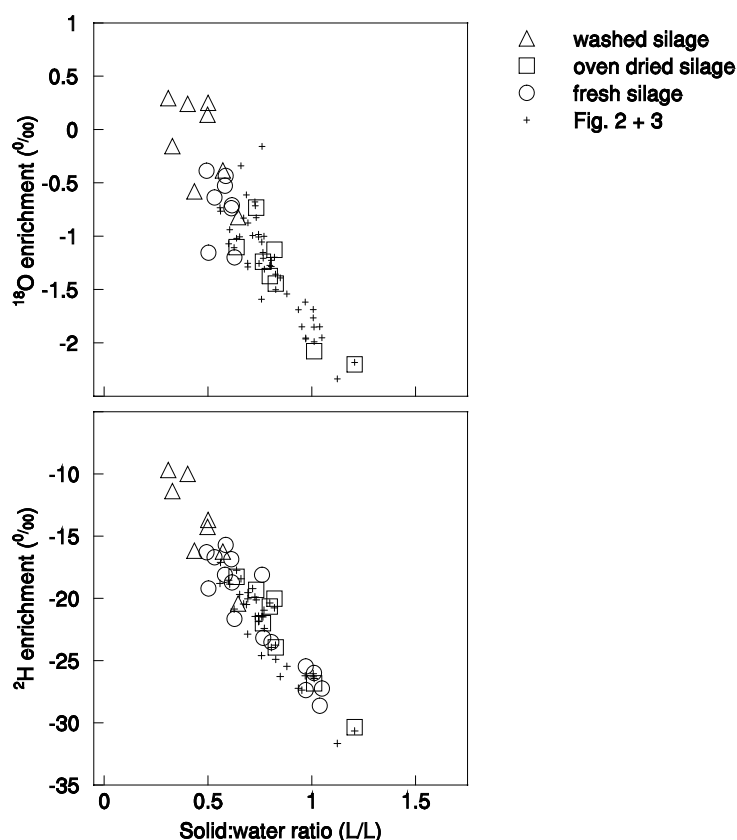
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496 Fig. 3: Relationship between volumetric solid:water ratio and apparent  $^{18}\text{O}$  and  $^2\text{H}$  enrichment of water absorbed by silage  
 497 compared to unconfined waters with different isotopic composition. The lines show the best fit (see eq. 6).  
 498

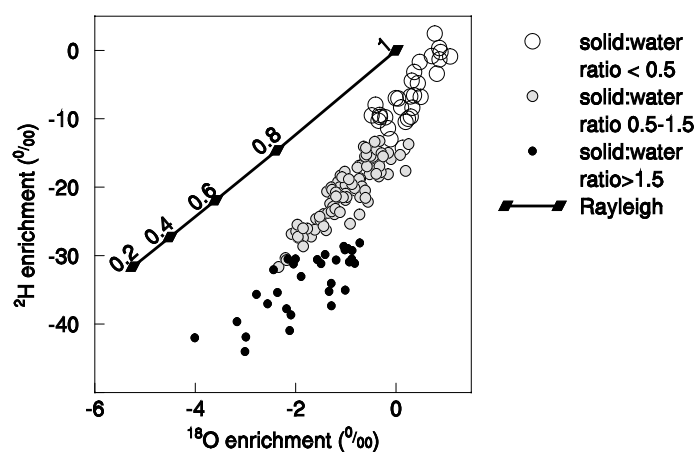


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 500 Fig. 4: Relationship between volumetric solid:water ratio and the apparent enrichment of  $^{18}\text{O}$  and  $^2\text{H}$  between unconfined water  
 501 and water adsorbed by silage with different pretreatments ( $N = 8$  each). The data of Fig. 2 and Fig. 3 (both oven dried silage,  $N =$   
 502 32) are provided for comparison.  
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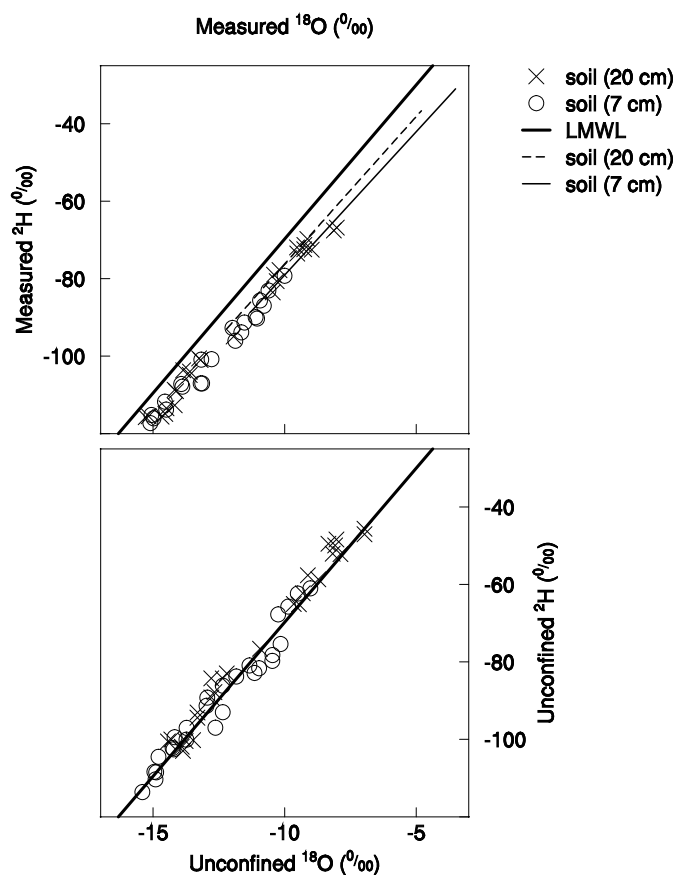
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507 Fig. 5: Apparent enrichment ( $^2\text{H}$  versus  $^{18}\text{O}$ ) of extracted water as observed in all experiments (markers indicate three groups of  
 508 solid:water ratios) and enrichment as expected from Rayleigh fractionation (line; numbers denote the fraction of extracted water).  
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Fig. 6: Isotope composition of measured total soil water (top) and estimated unconfined water (bottom) in soil at 7 cm depth (circles,  $N = 26$ ) and 20 cm depth (crosses,  $N = 26$ ) during the winter season. The bold solid line denotes the local meteoric water line ( $N = 79$ ;  $y = (8.0 \pm 0.2) x + (10 \pm 2)$ ,  $R^2 = 0.99$ ). The thin solid line and the dashed line indicate the range and the linear regressions for soil water at 7 cm ( $N = 84$ ;  $y = (7.7 \pm 0.5) x - (0 \pm 4)$ ,  $R^2 = 0.92$ ) and 20 cm depth ( $N = 85$ ;  $y = (7.4 \pm 0.5) x - (5 \pm 3)$ ,  $R^2 = 0.92$ ) during the growing season.