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# Relationship between the shell geochemistry of the modern aquatic gastropod *Radix* and water chemistry of lakes of the Tibetan Plateau

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Abstract Fossil shells of the aquatic mollusk *Radix* are common in the exposed sediments of lake terraces on the Tibetan Plateau. However, the living environment of Radix, and the geochemical characteristics of its shells, is unclear. Here, we report the results of an investigation of the occurrence of modern Radix in lakes of the southeastern and central Tibetan Plateau, as well as measurements of various geochemical characteristics of the shells. The results indicate that the nutritional status of the lake waters is the main limiting factor for the survival of *Radix* in these lakes. The Sr/Ca ratio of the Radix shells is significantly positively correlated with both the Sr/Ca ratio and the conductivity of the lake water. Initially,  $Kd_{Sr}$ decreases rapidly with low values of Sr/Cawater; however, in the case of Sr/Cawater values above 0.0076,  $Kd_{Sr}$  exhibits only a small range of variation. The  $\delta^{13}C_{\text{shell}}$  values are controlled by the  $\delta^{13}C$  of lake water dissolved inorganic carbon (DIC). In addition,

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F. Chen · H.-P. Hu University of Chinese Academy of Sciences, Beijing 100049, China the contribution of DIC of organic origin to the *Radix* shells increases when the lake water is deficient in DIC of inorganic origin. The  $\delta^{18}$ O values of the *Radix* shells provide useful information about the isotopic composition of the ambient waters.

**Keywords** Tibetan Plateau  $\cdot$  Gastropod *Radix*  $\cdot$  Hydrochemistry  $\cdot$  Shell geochemistry  $\cdot$  Oxygen and carbon isotopes  $\cdot$  Sr/Ca ratio

## Introduction

The Tibetan Plateau is one of the major regions of alpine lakes in the world, and although the statistics are incomplete, there are at least 800 lakes with an area  $>1 \text{ km}^2$  (Ma et al., 2011; Fig. 1A). The aquatic gastropod *Radix* (Montfort, 1810) is common in many of these lakes (von Oheimb et al., 2011); however, its ecological requirements in the lakes of the Tibetan Plateau are not fully understood (Liu, 1963; Zhao et al., 2005; von Oheimb et al., 2011; Taft et al., 2012, 2013). Thus, the limnological characteristics of the sites in which it occurs, as well as the modern *Radix* biology, warrant further investigation.

*Radix* is one of the few continental gastropod taxa which inhabit extreme boreal and arctic environments (White et al., 2008). Among the aquatic gastropods, besides *Radix*, only *Gyraulus* (Pulmonata, Planorbidae) can be found on the Tibetan Plateau (Taft et al., 2012).



Fig. 1 A Location of the sampled lake sites on the Tibetan Plateau. Sites where living specimens of *Radix* sp. occur at the present day are indicated by triangles, and sites where *Radix* sp. is absent are indicated by crosses. The Arabic numerals correspond to the Site no. in Table 1; **B** fossil shells of *Radix* 

sp. in exposed lake sediments of terraces of Lake Yamdrok Yumtso, Tibetan Plateau; C shell of *Radix* sp. from Lake Chen Co; the hole indicates the location of the sample obtained using a dental drill

The pulmonate gastropod *Radix* prefers calm, shallow water, such as lakes, wetlands, and slow-flowing sections of streams (Økland, 1990). Gaten (1986) measured the shell growth of *Radix* during all seasons and observed growth rates of 2.41–2.86 mm per month in summer and 0.35 mm per month in winter. Thus, although shell growth during winter is much slower, it

does not cease completely. The life span of *Radix* is approximately one year (Young, 1975; Glöer, 2002). During the winter months, *Radix* tend to move from shallow areas to deeper water (Gaten, 1986) or remain active under the ice cover (Glöer, 2002). Protozoa, weeds, cyanobacteria, chlorophyta, and diatoms constitute the primary diet of *Radix* (Knecht & Walter, 1977; Gittenberger et al., 1998). The pH tolerance range of *Radix* is from 5.8 to 9.9, while values from 7.0 to 9.6 are optimal (Økland, 1990).

Many lake basins on the Tibetan Plateau are characterized by multiple paleo-shorelines and/or lake terraces (e.g., Li, 2000; Zhao et al., 2003; Zheng et al., 2006; Lee et al., 2009; Long et al., 2012; Pan et al., 2012; Stauch et al., 2014), which potentially provide information on the history of past lake level fluctuations. However, there tends to be a lack of suitable archives for studying the paleohydrology and paleoclimatology of these lakes. Our field investigation indicates that fossil shells of Radix are common in the exposed sediments of lake terraces on the Tibetan Plateau (Fig. 1B). The geochemical information recorded in a single Radix shell may reflect annual hydrological and hydrochemical variations which are controlled by distinct climatic parameters. Thus, the study of fossil Radix shells in the lake terraces is potentially useful for reconstructing the paleohydrology and paleoclimatology of these sites.

In the past few decades, an increasing number of studies have focused on the use of  $\delta^{13}$ C and  $\delta^{18}$ O records from non-marine mollusk shells to infer changes in climatic and environmental conditions (e.g., Linz & Müller, 1981; Abell & Williams, 1989; Zanchetta et al., 1999; Curry & Fallick, 2002; Jones et al., 2002; Baroni et al., 2006; Tütken et al., 2006; Mii et al., 2012). In addition, a large number of studies have been performed on the trace element geochemistry of gastropod shells (e.g., Faure et al., 1967; Eisma et al., 1976; Dodd & Crisp, 1982; Dettman et al., 1999; Anadón et al., 2006; Carroll & Romanek, 2008). However, geochemical studies of the aragonite shells of Radix, including Sr/Ca ratios and the partition coefficient (Kd<sub>Sr</sub>), as well as their  $\delta^{13}$ C and  $\delta^{18}$ O isotope values, are comparatively rare.

Taft et al. (2012, 2013) were the first to demonstrate that *Radix* shells were a useful archive for reconstructing hydrological and climatic conditions on the Tibetan Plateau. They investigated the sclerochronological  $\delta^{13}$ C and  $\delta^{18}$ O patterns of the *Radix* shells on a sub-seasonal scale in several modern lakes across the Tibetan Plateau and discussed the possible mechanisms affecting carbon and oxygen stable isotopes in the shells. Nevertheless, the geochemical characteristics of *Radix* shells, and their relationship with the hydrochemistry of lakes on the Tibetan Plateau, are largely unknown. In addition, the average elevation of the Tibetan Plateau is above 4 km, and it covers a region of almost 3 million km<sup>2</sup> (Royden et al., 2008; Yao et al., 2013); thus, the lake environments on the Tibetan Plateau are diverse and complex. Therefore, further studies are needed to document and improve our understanding of the environmental implications of the Sr/Ca,  $\delta^{13}$ C, and  $\delta^{18}$ O characteristics of *Radix* shells in order to provide a sound basis for their use for environmental reconstruction.

The present study is an investigation of modern *Radix* and its ambient aquatic environment in lakes on the Tibetan Plateau. The objectives were (1) to characterize the modern environments of *Radix*; (2) to measure the Sr/Ca,  $Kd_{\rm Sr}$ ,  $\delta^{13}$ C, and  $\delta^{18}$ O values of the *Radix* shells and their habitat water; and thereby (3) to assess the potential of geochemical analyses of fossil *Radix* shells as paleo-hydrochemical and paleo-environmental proxies on the Tibetan Plateau.

## Materials and methods

## Field sampling

Twenty-two lakes on the southeastern and central Tibetan Plateau were investigated during two field trips from May to October 2013 (Fig. 1A). We collected water samples from all of the lakes and obtained specimens of living Radix (Fig. 1C) from 10 lakes. The specimens were collected from shallow water (<1.5 m) close to the shoreline. In addition, we collected 5 lake water samples for  $\delta^{13}$ C measurements of dissolved inorganic carbon (DIC) in the Yamdrok Yumtso lake basin, during July 2015. All water samples were filtered through a 0.45-µm membrane *in situ*. Samples for  $\delta^{13}$ C analysis of DIC were treated with trace amounts of HgCl<sub>2</sub> in situ to minimize the influence of microorganisms on the  $\delta^{13}$ C values (Wachniew & Różański, 1997). The locations of the lakes are shown in Fig. 1A, and more detailed information about the sampling sites and the measured geochemical parameters are listed in Table 1.

## Analytical methods

## Hydrochemical analysis

In situ electrical conductivity (EC), pH, and water temperature (T) were measured using a YSI ProPlus

**Table 1** Sampling localities and hydrochemical parameters [pH value, electric conductivity (EC), water temperature (T), non-purgeable organic carbon (NPOC), total nitrogen (TN) and

dissolved inorganic carbon (DIC), Mg/Ca (molar ratio), Sr/Ca (molar ratio),  $\delta^{18}O,~\delta D$ , and  $\delta^{13}C_{\rm DIC}]$  of the sampled lake waters

| Site no | Sample no. | Lake name      | рН                       | EC<br>(µs/cm)   | <i>Т</i><br>(°С)         | NPOC<br>(ppm) | TN<br>(ppm)                | DIC<br>(ppm)     | Sr/Ca<br>(molar ratio) | Mg/Ca<br>(molar ratio) |
|---------|------------|----------------|--------------------------|-----------------|--------------------------|---------------|----------------------------|------------------|------------------------|------------------------|
| 1       | NCS-1      | Nam Co         | 9.70                     | 1,704           | 12.5                     | 4.69          | 0.39                       | 151.9            | 0.0135                 | 11.50                  |
| 2       | NCS-2      | Nam Co         | 9.46                     | 1,367           | 13.7                     | 3.95          | 0.50                       | 121.6            | 0.0095                 | 6.07                   |
| 3       | NCS-3      | Nam Co         | 9.38                     | 1,668           | 12.6                     | 3.63          | 0.30                       | 157.4            | 0.0102                 | 7.53                   |
| 4       | NCS-4      | Nam Co         | 9.36                     | 1,731           | 13.5                     | 3.33          | 0.36                       | 160.0            | 0.0156                 | 12.00                  |
| 5       | BMNC130824 | Baima Namco    | 8.96                     | 1,045           | 15.2                     | 11.80         | 1.26                       | 33.3             | 0.0084                 | 1.81                   |
| 6       | KM130703   | Gongmo Tso     | 7.15                     | 421             | 13.4                     | $NM^{a}$      | $\mathrm{NM}^{\mathrm{a}}$ | NM <sup>a</sup>  | 0.0030                 | 0.21                   |
| 7       | KMC130917  | Gongmo Tso     | 9.09                     | 430             | 17.5                     | 6.94          | 0.69                       | 12.9             | 0.0032                 | 0.26                   |
| 8       | CHW-130629 | Chen Co        | 8.45                     | 1,040           | 17.9                     | 2.12          | 0.29                       | 26.2             | 0.0089                 | 1.04                   |
| 9       | BJ-130630  | Pagyü Tso      | 8.52                     | NM <sup>a</sup> | 17.3                     | 8.97          | 0.87                       | 36.3             | 0.0097                 | 1.99                   |
| 10      | YH-W-3     | Yamdrok Yumtso | 8.82                     | 371             | 14.1                     | 4.50          | 0.57                       | 109.0            | 0.0023                 | 12.70                  |
| 11      | YH-130630  | Yamdrok Yumtso | 8.95                     | 385             | 19.0                     | 4.66          | 0.52                       | 115.0            | 0.0019                 | 15.30                  |
| 12      | NZC-1      | Nongzhen Tso   | 9.76                     | 181             | 16.4                     | 8.56          | 0.78                       | 8.8              | 0.0047                 | 0.45                   |
| 13      | MCBN130824 | Mucuobingni    | 9.57                     | 251             | 12.6                     | 1.50          | 0.24                       | 121.8            | 0.0015                 | 18.40                  |
| 14      | QGC130826  | Qiagui Co      | 9.89                     | 425             | 14.5                     | 3.46          | 0.43                       | 34.5             | 0.0076                 | 2.39                   |
| 15      | SLC130826  | Selin Co       | 9.58                     | 703             | 15.7                     | 2.73          | 0.26                       | 36.7             | 0.0072                 | 1.91                   |
| 16      | LC130824   | Lang Co        | 9.57                     | 2,509           | 12.6                     | 12.10         | 0.94                       | 268.8            | 0.0006                 | 20.80                  |
| 17      | XRC130824  | Xuru Co        | 9.19                     | 3,890           | 10.8                     | 2.31          | 0.26                       | 133.7            | 0.0083                 | 2.01                   |
| 18      | ceh130826  | Cuoe lake      | 9.18                     | 387             | 14.5                     | 5.63          | 0.58                       | 44.1             | 0.0024                 | 2.63                   |
| 19      | CJN130830  | Cuo Jia Na     | 9.81                     | 1,045           | 14.2                     | 19.90         | 1.63                       | 98.2             | 0.0033                 | 7.17                   |
| 20      | arc130823  | Angren Co      | 9.80                     | 7,980           | 16.0                     | 48.50         | 3.29                       | 1344.0           | 0.0059                 | 1.98                   |
| 21      | DZC130826  | Daze Co        | 10.1                     | 21,765          | 13.4                     | 12.30         | 1.32                       | 1359.0           | 0.0019                 | 53.60                  |
| 22      | 13#        | 13#            | 9.87                     | 269             | 8.4                      | 9.75          | 0.97                       | 31.3             | 0.0055                 | 2.91                   |
| 23      | PM-W-2     | Puma Yumco     | 8.99                     | 452             | 15.5                     | 2.11          | 0.35                       | 44.3             | 0.0048                 | 1.92                   |
| 24      | DQC130825  | Dangqiong Co   | 9.53                     | 144,842         | 16.9                     | 40.60         | 3.20                       | 2107.0           | $ND^{b}$               | $ND^{b}$               |
| 25      | DL1-130825 | Tangre Yumco   | 9.43                     | 8,978           | 15.7                     | 2.55          | 0.43                       | 347.8            | 0.0015                 | 45.40                  |
| 26      | SLC2       | Selin Co       | 9.41                     | 12,630          | 13.1                     | 5.56          | 0.48                       | 268.6            | 0.0025                 | 24.40                  |
| 27      | RWL130910  | Ranwu lake     | 8.68                     | NM <sup>a</sup> | 11.5                     | 0.28          | 0.15                       | 8.2              | 0.0014                 | 0.28                   |
| Site no | Sample no. | Lake name      | δ <sup>18</sup> O<br>(‰) | δD<br>(‰)       | $\delta^{13}C_{DIC}$ (‰) | Latitu<br>(N) | de                         | Longitude<br>(E) | Altitude<br>(m)        | Note                   |
| 1       | NCS-1      | Nam Co         | -6.5                     | -70.5           | NM <sup>a</sup>          | 30°47′        | 16.57″                     | 90°16′50.39″     | 4,728                  | Found Radix            |
| 2       | NCS-2      | Nam Co         | -9.8                     | -91.0           | NM <sup>a</sup>          | 30°53′5       | 5.53″                      | 90°14′58.71″     | 4,728                  | Found Radix            |
| 3       | NCS-3      | Nam Co         | -8.0                     | -77.4           | NM <sup>a</sup>          | 30°55′3       | 35″                        | 90°57′03″        | 4,728                  | Found Radix            |
| 4       | NCS-4      | Nam Co         | -6.9                     | -70.5           | NM <sup>a</sup>          | 30°53′3       | 36″                        | 90°52′08″        | 4,728                  | Found Radix            |
| 5       | BMNC130824 | Baima Namco    | -6.7                     | -73.2           | NM <sup>a</sup>          | 30°46′3       | 31″                        | 90°57′40″        | 4,728                  | Found Radix            |
| 6       | KM130703   | Gongmo Tso     | -13.6                    | -117.4          | -3.8                     | 28°59′4       | 42″                        | 90°23′40″        | 4,438                  | Found Radix            |
| 7       | KMC130917  | Gongmo Tso     | -12.4                    | -108.9          | NM <sup>a</sup>          | 28°59′4       | 42″                        | 90°23'40"        | 4,438                  | Found Radix            |
| 8       | CHW-130629 | Chen Co        | -4.2                     | -62.8           | 0.2                      | 28°57′5       | 57″                        | 90°29′56″        | 4,434                  | Found Radix            |
| 9       | BJ-130630  | Pagyü Tso      | -6.3                     | -70.2           | 1.9                      | 28°44′4       | 44.90″                     | 90°50′40.44″     | 4,511                  | Found Radix            |
| 10      | YH-W-3     | Yamdrok Yumtso | -6.5                     | -74.7           | -0.2                     | 28°46′4       | 48″                        | 90°31′56″        | 4,436                  | Found Radix            |
| 11      | YH-130630  | Yamdrok Yumtso | -5.7                     | -71.0           | NM <sup>a</sup>          | 28°50′0       | 05″                        | 90°02′04″        | 4,444                  | Found Radix            |
| 12      | NZC-1      | Nongzhen Tso   | -6.1                     | -75.7           | -14.7                    | 28°53′2       | 29″                        | 90°24′21″        | 4,564                  | Found Radix            |
| 13      | MCBN130824 | Mucuobingni    | -9.9                     | -97.2           | NM <sup>a</sup>          | 30°33′5       | 59.56″                     | 86°17′7.03″      | 4,680                  | Found Radix            |
| 14      | QGC130826  | Qiagui Co      | -6.8                     | -77.5           | NM <sup>a</sup>          | 31°51′4       | 42.37″                     | 88°17′45.65″     | 4,552                  | Found Radix            |
| 15      | SLC130826  | Selin Co       | -7.3                     | -78.9           | NM <sup>a</sup>          | 31°47′5       | 56.94″                     | 88°26′11.62″     | 4,545                  | Found Radix            |
| 16      | LC130824   | Lang Co        | -6.4                     | -84.5           | NM <sup>a</sup>          | 29°12′3       | 51.36″                     | 87°24′17.47″     | 4,294                  | Not found Radix        |

Table 1 continued

| Site no | Sample no. | Lake name    | δ <sup>18</sup> Ο<br>(‰) | δD<br>(‰) | $\delta^{13}C_{DIC}$ (‰) | Latitude<br>(N) | Longitude<br>(E) | Altitude<br>(m) | Note            |
|---------|------------|--------------|--------------------------|-----------|--------------------------|-----------------|------------------|-----------------|-----------------|
| 17      | XRC130824  | Xuru Co      | -6.8                     | -80.6     | NM <sup>a</sup>          | 30°13′37.06″    | 86°27′9.05″      | 4,721           | Not found Radix |
| 18      | ceh130826  | Cuoe lake    | -7.5                     | -77.9     | NM <sup>a</sup>          | 31°41′30.98″    | 88°41′32.55″     | 4,550           | Not found Radix |
| 19      | CJN130830  | Cuo Jia Na   | -4.7                     | -66.3     | NM <sup>a</sup>          | 30°53'42"       | 90°52′11″        | 4,728           | Not found Radix |
| 20      | arc130823  | Angren Co    | -6.3                     | -83.8     | $NM^{a}$                 | 29°18′52.36″    | 87°12′44.59″     | 4,308           | Not found Radix |
| 21      | DZC130826  | Daze Co      | -6.8                     | -72.5     | NM <sup>a</sup>          | 31°49′34.5″     | 87°32′35.29″     | 4,469           | Not found Radix |
| 22      | 13#        | 13#          | -7.1                     | -78.7     | $NM^{a}$                 | 31°28′16.71″    | 89°54′1.74″      | 4,740           | Not found Radix |
| 23      | PM-W-2     | Puma Yumco   | -6.7                     | -73.3     | $NM^{a}$                 | 28°35′50.94″    | 90°29'38.14"     | 5,030           | Not found Radix |
| 24      | DQC130825  | Dangqiong Co | -5.3                     | -69.7     | $NM^{a}$                 | 31°32′15.44″    | 86°43′50.24″     | 4,477           | Not found Radix |
| 25      | DL1-130825 | Tangre Yumco | -6.8                     | -78.8     | $NM^{a}$                 | 31°20′13.72″    | 86°45′5.74″      | 4,542           | Not found Radix |
| 26      | SLC2       | Selin Co     | -4.0                     | -53.9     | $NM^{a}$                 | 31°42′25.81″    | 88°37′51.75″     | 4,569           | Not found Radix |
| 27      | RWL130910  | Ranwu lake   | -15.9                    | -113.6    | $NM^{a}$                 | 29°28′54.98″    | 96°38′46.19″     | 3,913           | Not found Radix |

The isotope data are reported using the  $\delta$ % notation relative to V-SMOW and the  $\delta^{13}C_{DIC}$  relative to V-PDB. The Site no. is corresponding to the Arabic numerals in Fig. 1A

<sup>a</sup> NM not measured

<sup>b</sup> ND none detected

Multi-probe Water Quality Sonde made by YSI., USA. The accuracies of the EC, pH, and T measurements are  $\pm 0.001$ ,  $\pm 0.10$ , and  $\pm 0.2$ , respectively. The concentrations of dissolved cations (K<sup>+</sup>, Na<sup>+</sup>, Mg<sup>2+</sup> and Ca<sup>2+</sup>) were measured by Inductively Coupled Plasma Optical Emission Spectrometry (Prodigy, Teledyne Leeman Labs), with analytical relative standard deviation (RSD) <0.5%. The concentrations of dissolved anions (F<sup>-</sup>, Cl<sup>-</sup>, NO<sub>3</sub><sup>-</sup> and SO<sub>4</sub><sup>2-</sup>) were determined by ion chromatography (Metrohm Compact IC 761), with RSD <1%. The concentrations of  $HCO_3^-$  and  $CO_3^{2-}$  were measured by automatic titration (Metrohm), with relative error <1.8% and RSD <1.2%. The concentrations of non-purgeable organic carbon (NPOC), total nitrogen (TN), and DIC in the lake water were measured using a Shimadzu SSM 5000A/TOC-V CPH meter, with RSD <3%. The concentration of Sr was measured by Inductively Coupled Plasma Optical Mass Spectrometry (X-7 series; Thermo Elemental); the detection limit was 0.004 ppb for Sr, and RSD <1%.  $\delta^{18}$ O and  $\delta$ D were analyzed using a Wave Scan-Cavity Ring Down Spectrometer (L1102i), and the results are reported using the  $\delta$ % notation relative to V-SMOW; RSD for  $\delta^{18}$ O and  $\delta$ D are <0.1‰ and <0.6‰, respectively. The  $\delta^{13}C_{DIC}$  was determined using the MAT253 + Gas-Bench at the State Key Laboratory of Hydrology-Water Resources and Hydraulic Engineering, Hohai

University. The results are reported using the  $\delta$ % notation relative to V-PDB, with RSD <0.2%.

Analysis of shell geochemistry

Because *Radix* has not so far been identified to species level on the Tibetan Plateau (Taft et al., 2012), we use *Radix* sp. to represent all of the species belonging to the genus. The following procedure was used to prepare the shells for geochemical analysis. Firstly, all obvious inorganic and organic material was removed from the shells using a fine brush; secondly, the shells were treated in  $H_2O_2$  for 24 h to remove any dispersed organic material; and thirdly, the shells were rinsed with ultrapure water in an ultrasonic bath and dried for 6 h at 60°C.

Since juvenile shell portions often may not be in geochemical equilibrium with ambient conditions (e.g., Schöne, 2008), the adult shells were selected for analysis. Two shells of similar shape and length were selected from each sampling site. In order to obtain a mass of  $200 \pm 50 \,\mu\text{g}$  of shell sample for  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  analysis, a portion of ca. 0.1–0.5 mm in diameter was removed from the aperture of each shell using a dental drill (Fig. 1C). Taft et al. (2012) suggested that each shell section represents a clearly defined growth period. We therefore assumed that the shell fragment from the aperture ('shell-aperture')

would record the isotopic characteristics of the water during the last ca. sub-weekly period prior to the date on which the shells were collected. The remaining shells were homogenized with a mortar and pestle and analyzed as powdered bulk samples ('shell-AVG').

The  $\delta^{13}$ C and  $\delta^{18}$ O values of the *Radix* sp. shell samples were measured using a GasBench II linked to a MAT-253 ThermoFisher Scientific Isotope Ratio Mass Spectrometer at the Key Laboratory of Cenozoic Geology and Environment, Institute of Geology and Geophysics, Chinese Academy of Sciences. Data are reported using the  $\delta$ ‰ notation relative to V-PDB. The instrumental precisions of the isotopic measurements determined from simultaneous replicate analysis of IVA standards are <0.16‰ for  $\delta^{18}$ O and <0.07‰ for  $\delta^{13}$ C.

Powdered shell samples were dissolved in 5 ml of dilute ultrapure  $HNO_3$  acid (3%) for major and trace element analyses. The solutions were analyzed for Ca, Mg, and Sr using Inductively Coupled Plasma Optic Mass Spectrometry (X-7 series; Thermo Elemental). The detection limits were 0.003 ppm for Ca, 0.05 ppm for Mg, and 0.004 ppb for Sr.

## Results

Physical and chemical characteristics of the lake water

The hydrochemical parameters of lakes where *Radix* sp. was found are shown in Table 1. pH and EC range from 7.15 to 9.89 and from 181 to 1731  $\mu$ S/cm, respectively. *T* at the time of collection ranged from 12.5 to 19.0°C. The NPOC values ranged from 1.50 to 11.80 ppm, TN values from 0.24 and 1.26 ppm, and DIC values from 8.8 to 160.0 ppm. The Sr/Ca molar ratio (Sr/Ca) ranged from 0.0015 to 0.0156 and the Mg/Ca molar ratio (Mg/Ca) from 0.21 to 18.40. The  $\delta^{18}$ O values ranged from -13.6 to -4.2% and the  $\delta$ D values from -117.4 to -62.8%. The  $\delta^{13}C_{\text{DIC}}$  values ranged from -14.7 to 1.9%.

The hydrochemical parameters of lakes where *Radix* sp. were not found are also shown in Table 1. In the case of these sites, pH and EC range from 8.68 to 10.1 and from 269 to 144,842  $\mu$ S/cm, respectively. Unfortunately, EC of sample RWL130910 (Ranwu lake) was not measured. At the time of collection, *T* ranged from 8.4 to 16.9°C. The NPOC values range

from 0.28 to 48.50 ppm and TN from 0.15 to 3.29 ppm. The DIC values range from 8.2 to 2107.0 ppm. Sr/Ca ranges from 0.0014 to 0.0083 and Mg/Ca from 0.28 to 53.60. The  $\delta^{18}$ O values range from -15.9 to -4.0‰ and  $\delta$ D from -113.6 to -53.9‰.

Cation and anion concentrations (ppm) for all the sampled lake waters are listed in Table 2. Piper diagrams (Fig. 2) are used to illustrate the range of water properties of the sampled lakes. In general, the concentrations of most cations and anions exhibit significant variability.  $Mg^{2+}$  and/or Na<sup>+</sup> are the dominant cations in all of the sampled lakes, except for Nongzhen Tso, Gongmo Tso, and Ranwu lake, which were Ca-dominated. In contrast, the predominant anion depends on the specific sampling locality. It is noteworthy that the lakes containing living *Radix* sp. have low Cl<sup>-</sup> lake water concentrations (Fig. 2A).

#### Sr concentration in the *Radix* sp. shells

The analytical data for the *Radix* sp. shells, and for the corresponding lake waters in which they grew, are listed in Tables 1 and 3. The Sr/Ca<sub>shell</sub> plots against Sr/ Ca<sub>water</sub>, EC, water T, and pH are illustrated in Fig. 3. A significant linear positive correlation is observed between Sr/Cashell and Sr/Cawater and EC (Sr/Cashell versus Sr/Ca<sub>water</sub>;  $R^2 = 0.78$ , n = 15, P < 0.001; Fig. 3A; Sr/Ca<sub>shell</sub> versus EC;  $R^2 = 0.71$ , n = 14, P < 0.001; Fig. 3B), while no significant linear correlation is observed between Sr/Ca<sub>shell</sub> and water T and pH (Sr/Ca<sub>shell</sub> versus water T;  $R^2 = 0.10$ , n = 15, P = 0.249; Fig. 3C; Sr/Ca<sub>shell</sub> versus water pH;  $R^2 = 0.08$ , n = 15, P = 0.318; Fig. 3D). The Sr concentrations in the Radix sp. shells range from 849.5 to 2383.0 ppm. The Sr/Ca values of the shells range from 0.0010 to 0.0028. The partitioning of Sr between the mollusk shell and the ambient water is usually expressed as a partition coefficient, defined as  $Kd_{Sr} = (Sr/Ca_{shell})/(Sr/Ca_{water})$ . The  $Kd_{Sr}$  values obtained in the present study range from 0.167 to 0.867 with mean  $Kd_{Sr} = 0.317$  (*n* = 15). Moreover, there is a variable sensitivity evident in the plot of  $Kd_{Sr}$ versus Sr/Ca<sub>water</sub> (Fig. 4).

# $\delta^{13}$ C and $\delta^{18}$ O values of the *Radix* sp. shells

The carbon and oxygen isotopic data for the shell samples are listed in Table 4.

Table 2 Cation and anion concentrations (ppm) for the sampled lake waters

| Sample no. | Na <sup>+</sup><br>(ppm) | K <sup>+</sup> | Mg <sup>2+</sup> | Ca <sup>2+</sup> | Sr <sup>2+</sup> | $F^{-}$ | $Cl^{-}$ | NO <sub>3</sub> <sup>-</sup> | $SO_{4}^{2-}$ | $CO_{3}^{2-}$ | $HCO_3^-$ |
|------------|--------------------------|----------------|------------------|------------------|------------------|---------|----------|------------------------------|---------------|---------------|-----------|
| NCS-1      | 351.7                    | 37.5           | 86.97            | 12.53            | 0.371            | 4.81    | 64.34    | 0.00                         | 185.4         | 203           | 542       |
| NCS-2      | 247.7                    | 26.9           | 68.55            | 18.61            | 0.385            | 4.03    | 44.79    | 0.00                         | 138.1         | 103           | 489       |
| NCS-3      | 322.2                    | 35.6           | 82.15            | 17.98            | 0.401            | 5.07    | 62.46    | 2.41                         | 181.8         | 124           | 651       |
| NCS-4      | 346.2                    | 38.3           | 85.08            | 11.73            | 0.400            | 6.55    | 85.08    | 10.5                         | 214.1         | 126           | 697       |
| BMNC130824 | 598.8                    | 76.4           | 212.40           | 193.90           | 3.567            | 5.96    | 123.05   | 0.00                         | 1973.7        | 27            | 123       |
| KM130703   | 9.2                      | 1.1            | 14.83            | 117.22           | 0.773            | 0.13    | 0.56     | 0.00                         | 316.3         | 0             | 62        |
| KMC130917  | 7.6                      | 0.1            | 11.87            | 74.19            | 0.523            | 0.14    | 0.12     | 0.00                         | 159.4         | 9             | 48.8      |
| CHW-130629 | 154.8                    | 16.2           | 65.58            | 104.06           | 2.029            | 0.00    | 6.67     | 0.00                         | 639.2         | 0             | 126       |
| BJ-130630  | 151.0                    | 10.1           | 149.46           | 123.66           | 2.634            | 0.00    | 16.56    | 0.00                         | 945.3         | 13            | 170       |
| YH-W-3     | 212.0                    | 21.7           | 165.15           | 21.41            | 0.110            | 0.53    | 71.03    | 4.21                         | 727.4         | 39            | 522       |
| YH-130630  | 264.4                    | 23.7           | 182.17           | 19.65            | 0.080            | 1.43    | 74.03    | 0.00                         | 733.1         | 85            | 482       |
| NZC-1      | 5.1                      | 0.2            | 5.32             | 19.70            | 0.203            | 0.77    | 2.05     | 0.00                         | 6.7           | 21            | 33        |
| MCBN130824 | 292.4                    | 41.4           | 111.60           | 10.02            | 0.034            | 2.28    | 36.69    | 0.00                         | 349.3         | 98            | 521       |
| QGC130826  | 56.6                     | 6.0            | 23.60            | 16.26            | 0.271            | 0.71    | 16.42    | 0.00                         | 40.5          | 38            | 139       |
| SLC130826  | 53.8                     | 7.5            | 19.39            | 16.73            | 0.264            | 0.74    | 18.71    | 0.38                         | 37.6          | 17            | 168       |
| LC130824   | 644.2                    | 11.8           | 71.82            | 5.68             | 0.008            | 2.43    | 149.3    | 0.00                         | 18.9          | 327           | 1,069     |
| XRC130824  | 898.7                    | 80.0           | 29.03            | 23.83            | 0.433            | 13.64   | 707.5    | 0.00                         | 379.1         | 261           | 410       |
| ceh130826  | 25.9                     | 4.2            | 32.03            | 20.10            | 0.106            | 0.61    | 7.68     | 0.41                         | 22.4          | 21            | 204       |
| CJN130830  | 188.2                    | 25.4           | 66.98            | 15.41            | 0.111            | 2.45    | 43.61    | 0.00                         | 67.9          | 147           | 348       |
| arc130823  | 2063.5                   | 7.9            | 9.05             | 7.54             | 0.098            | 9.23    | 265.13   | 0.00                         | 1058.4        | 756           | 1,270     |
| DZC130826  | 5891.2                   | 540.2          | 122.45           | 3.77             | 0.015            | 1.51    | 974.64   | 0.00                         | 4465.9        | 4231          | 2,416     |
| 13#        | 31.3                     | 6.2            | 23.13            | 13.09            | 0.157            | 0.71    | 2.92     | 0.06                         | 6.2           | 62            | 99        |
| PM-W-2     | 20.9                     | 5.1            | 38.30            | 32.88            | 0.346            | 0.76    | 2.10     | 0.00                         | 80.9          | 9             | 211       |
| DQC130825  | 42684.8                  | 8524.0         | 67.58            | 0.00             | 0.030            | 0.00    | 72839.60 | 0.00                         | 5704.7        | 10,971        | 793       |
| DL1-130825 | 2330.8                   | 248.0          | 276.54           | 10.05            | 0.032            | 6.97    | 1523.50  | 0.00                         | 2770.7        | 666           | 938       |
| SLC2       | 2879.8                   | 327.4          | 183.64           | 12.39            | 0.067            | 0.00    | 1807.55  | 0.00                         | 4048.3        | 431           | 828       |
| RWL130910  | 0.5                      | 0.5            | 2.62             | 15.61            | 0.048            | 0.09    | 0.11     | 0.00                         | 8.2           | 0             | 46        |

The  $\delta^{13}$ C and  $\delta^{18}$ O values of both shell-aperture and shell-AVG exhibit significant variability. The  $\delta^{13}$ C total values of shell-AVG range from -11.8 to -0.7%, while those of shell-aperture range from -11.6 to 0.0%. The relationships between the  $\delta^{13}$ C values of the shells and the  $\delta^{13}$ C<sub>DIC</sub>, DIC, and NPOC values of the lake water are illustrated in Fig. 5. A significant positive correlation is observed between both shell-AVG and shell-aperture  $\delta^{13}$ C value and the  $\delta^{13}$ C<sub>DIC</sub> of the lake water ( $\delta^{13}$ C<sub>shell-AVG</sub> versus  $\delta^{13}$ C<sub>DIC</sub>;  $R^2 = 0.73$ , n = 10, P = 0.0017;  $\delta^{13}$ C<sub>shell-aperture</sub> versus  $\delta^{13}$ C<sub>DIC</sub>;  $R^2 = 0.90$ , n = 10, P < 0.0001; Fig. 5A). There is a variable sensitivity between the shell  $\delta^{13}$ C values and lake water DIC (Fig. 5B). Moreover, the  $\delta^{13}$ C of shells tends to decrease with increasing NPOC in lake water  $(\delta^{13}C_{shell-AVG}$  versus NPOC;  $R^2 = 0.37$ , n = 28, P < 0.001;  $\delta^{13}C_{shell-aperture}$  versus NPOC;  $R^2 = 0.26$ , n = 28, P = 0.005; Fig. 5C). The  $\delta^{18}O$  total values of shell-AVG range from -12.1 to -0.9%, while those of shell-aperture range from -12.4 to 3.3%. The relationships between the  $\delta^{18}O$  values of shell-AVG and shell-aperture and the  $\delta^{18}O$  values of the ambient lake water are illustrated in Fig. 6. In both cases, the shell  $\delta^{18}O$  values are significantly positively correlated with the corresponding lake water values ( $\delta^{18}O_{shell-AVG}$  versus  $\delta^{18}O_{water}$ ;  $R^2 = 0.90$ , n = 30, P < 0.0001;  $\delta^{18}O_{shell-aperture}$  versus  $\delta^{18}O_{water}$ ;  $R^2 = 0.81$ , n = 30, P < 0.0001; Fig. 6).



Fig. 2 Piper diagrams illustrating the range of geochemical characteristics of the sampled lake waters: A sites where living specimens of *Radix* sp. occur at the present day and B sites where *Radix* is absent

| Sample no. | Water       |             |             |                        | Shell     | <i>Kd</i> <sub>Sr</sub> |             |                        |                               |
|------------|-------------|-------------|-------------|------------------------|-----------|-------------------------|-------------|------------------------|-------------------------------|
|            | Ca<br>(ppm) | Mg<br>(ppm) | Sr<br>(ppm) | Sr/Ca (molar<br>ratio) | Ca<br>(%) | Mg<br>(ppm)             | Sr<br>(ppm) | Sr/Ca (molar<br>ratio) |                               |
| NCS-1      | 12.53       | 86.97       | 0.371       | 0.0135                 | 39.67     | 109.90                  | 1959.0      | 0.0023                 | 0.167                         |
| NCS-2      | 18.61       | 68.55       | 0.385       | 0.0095                 | 37.52     | 130.60                  | 1444.0      | 0.0018                 | 0.186                         |
| NCS-3      | 17.98       | 82.15       | 0.401       | 0.0102                 | 39.21     | 151.00                  | 1901.0      | 0.0022                 | 0.217                         |
| NCS-4      | 11.73       | 85.08       | 0.400       | 0.0156                 | 38.90     | 156.00                  | 2383.0      | 0.0028                 | 0.179                         |
| BMNC130824 | 193.90      | 212.40      | 3.567       | 0.0084                 | 36.73     | 53.73                   | 2083.0      | 0.0026                 | 0.308                         |
| KM130703   | 117.22      | 14.83       | 0.773       | 0.0030                 | 38.67     | 31.78                   | 849.5       | 0.0010                 | 0.333                         |
| KMC130917  | 74.19       | 11.87       | 0.523       | 0.0032                 | 39.56     | 65.68                   | 910.2       | 0.0011                 | 0.327                         |
| CHW-130629 | 104.06      | 65.58       | 2.029       | 0.0089                 | 36.52     | 36.40                   | 1593.0      | 0.0020                 | 0.224                         |
| BJ-130630  | 123.66      | 149.46      | 2.634       | 0.0097                 | 36.70     | 42.14                   | 1360.0      | 0.0017                 | 0.174                         |
| YH-W-3     | 21.41       | 165.15      | 0.110       | 0.0023                 | 37.83     | 121.80                  | 903.2       | 0.0011                 | 0.466                         |
| YH-130630  | 19.65       | 182.17      | 0.080       | 0.0019                 | 37.05     | 164.60                  | 911.1       | 0.0011                 | 0.607                         |
| NZC-1      | 19.70       | 5.32        | 0.203       | 0.0047                 | 38.55     | 24.35                   | 1089.0      | 0.0013                 | 0.274                         |
| MCBN130824 | 10.02       | 111.60      | 0.034       | 0.0015                 | 36.77     | 78.07                   | 1072.0      | 0.0013                 | 0.867                         |
| QGC130826  | 16.26       | 23.60       | 0.271       | 0.0076                 | 35.80     | 75.22                   | 1343.0      | 0.0017                 | 0.225                         |
| SLC130826  | 16.73       | 19.39       | 0.264       | 0.0072                 | 38.19     | 82.34                   | 1233.0      | 0.0015                 | 0.205                         |
|            |             |             |             |                        |           |                         |             |                        | Mean<br>$Kd_{\rm Sr} = 0.317$ |

Table 3 Analytical data for the shells of Radix sp. shells and ambient lake waters

## Discussion

Living environments of Radix sp.

We noted two obvious characteristics of all of the sampling sites where *Radix* sp. was found: the

presence of living aquatic plants in the immediate vicinity, and co-occurrence of the aquatic snail *Gyraulus*.

In their survey of the occurrence of *Radix* sp. in Tibetan Plateau lakes, Taft et al. (2013) found that lakes containing living *Radix* sp. were characterized



Fig. 3 The relationship between Sr/Ca molar ratio data in *Radix* sp. shells and A with the Sr/Ca molar ratio in the host water; B with the host water EC; C with the host water T; and D with host water pH

by pH values ranging from 7.4 to 10.4 and EC values from 142 to 10,300  $\mu$ s/cm. The EC values of most of our sampling sites lie within these ranges, and therefore, it seems that EC has only a minor influence on the survival of *Radix* sp. On the other hand, White et al. (2008) noted that *Radix* sp. inhabit extremely cold lake environments with ice cover for more than half of the year. Taft et al. (2012) concluded that there is no critical water temperature which prevents shell accretion in *Radix* sp. In the present study, the ranges of pH and *T* values for lakes with and without *Radix* sp. were similar, and consequently, we conclude that these two variables have only a minor influence on the survival of *Radix* sp. on the Tibetan Plateau.

In the case of those lakes in which *Radix* sp. was not found, the following observations can be made: On the



Fig. 4 The relationship between  $Kd_{Sr}$  and lake water Sr/Ca molar ratio

Table 4 Oxygen and carbon isotope values of the Radix sp. shells

| Sample no. | AVG             |                 |                 | Aperture        |                 |                 |                 |                |  |
|------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|----------------|--|
|            | Shell-1         |                 | Shell-2         |                 | Shell-1         |                 | Shell-2         |                |  |
|            | $\delta^{18}$ O | $\delta^{13}$ C | $\delta^{18}$ O | $\delta^{13}$ C | $\delta^{18}$ O | $\delta^{13}$ C | $\delta^{18}$ O | $\delta^{13}C$ |  |
| NCS-1      | -4.9            | -2.3            | -4.9            | -2.1            | -4.1            | -1.8            | -4.3            | -1.1           |  |
| NCS-2      | -6.4            | -1.5            | -7.3            | -2.6            | -5.3            | -1.3            | -5.3            | -1.4           |  |
| NCS-3      | -5.0            | -3.1            | -5.1            | -3.3            | -4.8            | -2.3            | -4.8            | -3.6           |  |
| NCS-4      | -3.8            | -3.7            | -4.1            | -2.2            | -4.2            | -3.4            | -4.4            | -1.4           |  |
| BMNC130824 | -4.1            | -6.0            | -4.2            | -6.2            | -4.5            | -5.5            | -4.4            | -6.0           |  |
| KM-130703  | -10.9           | -4.5            | -11.9           | -5.2            | -11.4           | -4.5            | -12.4           | -4.4           |  |
| KMC130917  | -12.1           | -11.8           | -11.3           | -10.1           | -11.3           | -11.6           | -10.5           | -9.2           |  |
| CHW-130629 | -1.8            | -2.1            | -0.9            | -0.7            | -1.8            | -4.2            | -1.9            | -0.8           |  |
| BJ-130630  | -4.1            | -3.2            | -4.1            | -3.7            | -0.3            | -1.0            | -3.2            | -2.1           |  |
| YH-W-3     | -3.8            | -2.6            | -4.6            | -2.0            | -2.5            | -3.2            | -2.2            | -3.5           |  |
| YH-130630  | -3.5            | -2.5            | -3.1            | -2.6            | -2.6            | -2.1            | 3.3             | -1.5           |  |
| NZC-1      | -5.9            | -7.2            | -5.9            | -6.6            | -3.0            | -8.5            | -3.0            | -9.9           |  |
| MCBN130824 | -6.5            | -2.0            | -6.9            | -3.4            | -6.9            | -2.4            | -7.6            | -3.8           |  |
| QGC130826  | -5.0            | -1.4            | -4.4            | -1.0            | -4.8            | 0.0             | -5.0            | -1.1           |  |

The shell data are reported using the  $\delta$ % notation relative to V-PDB

basis of both the measured hydrochemical data and field observations, these lakes were often characterized by high Cl<sup>-</sup> concentrations (Fig. 2B) and/or by the absence of living aquatic plants in the lake margins. Although the geochemical characteristics of water samples ceh130826, 13# and PM-W-2 (Table 2) are favorable for the growth of Radix sp., in the case of ceh130826 (Cuoe lake), the steeply sloping lake bottom topography may have been disadvantageous, and in lake 13#, the presence of a large number of small white shrimps may indicate that in this case competition with other organisms for food was an important factor. In the case of Puma Yumco (sample PM-W-2), the very high altitude (5030 m) may have been a limiting factor; however, in sample RWL130910 (Ranwu lake), the NPOC, TN, and DIC values were the lowest of all of the sampled sites and thus may have restricted the shell growth of *Radix* sp.

SLC130826

-4.4

-3.0

-4.6

-2.5

Based on the foregoing evidence, we suggest that the living environment of *Radix* sp. is probably controlled by specific lake ecosystem attributes such as temperature, pH, altitude, nutrition (NPOC, TN and DIC values), conductivity, lake bottom topography, and competition for food. Moreover, Radix sp. tend to live in oligohaline to mesohaline water bodies, and thus, the nutrient content of the lake waters may be the main limiting factor for their survival, rather than the high pH values observed in the Tibetan Plateau lakes.

-4.1

-1.2

-4.4

-2.2

Sr/Ca and  $Kd_{Sr}$  values of the *Radix* sp. shells

Several previous studies have analyzed the Sr/Ca ratio in mollusk shells and have noted significant relationships with temperature (e.g., Dodd, 1965; Surge & Walker, 2006) or salinity (e.g., Wolf et al., 1967; Eisma et al., 1976). On the other hand, many lines of evidence indicate that the Sr/Ca ratio in mollusk shells is controlled by biological factors (e.g., Gillikin et al., 2005; Poulain et al., 2015), such as growth rate (e.g., Takesue & van Geen, 2004) or metabolism (e.g., Purton et al., 1999).

In the present study, the plot of the Sr/Ca ratio of the shells of *Radix* sp. (Sr/Ca<sub>shell</sub>) versus the corresponding value of the lake water (Sr/Cawater) reveals a significant positive correlation (Fig. 3A), implying that Sr/Cawater is a major factor controlling Sr/Cashell. Sr/Ca<sub>shell</sub> is also well correlated with EC (Fig. 3B). While our results indicate that the aragonite shells formed in a range of values of water T (12.5–19°C), the relationship between  $Sr/Ca_{shell}$  and water T is unclear (Fig. 3C). Similarly, there is no significant correlation between Sr/Ca<sub>shell</sub> and water pH (Fig. 3D).



Fig. 5 The relationship between the shell  $\delta^{13}$ C values of *Radix* sp. and lake water  $\delta^{13}C_{DIC}$ , and lake water DIC (ppm), and lake water NPOC (ppm). A  $\delta^{13}C_{shell}$  versus  $\delta^{13}C_{DIC}$ ; B  $\delta^{13}C_{shell}$  versus DIC; C  $\delta^{13}C_{shell}$  versus NPOC. The shell  $\delta^{13}C$  and water  $\delta^{13}C_{DIC}$  values are reported using the  $\delta$ ‰ notation relative to V-PDB

Previous studies have reported a range of 0.22 to 0.31 for the partition coefficient of Sr  $(Kd_{Sr})$  between the shells of diverse non-marine aragonitic



Fig. 6 Relationship between the  $\delta^{18}$ O values of the shells of *Radix* sp. and the  $\delta^{18}$ O values of the lake water. The  $\delta^{18}$ O values of the shells data are reported using the  $\delta^{\infty}$  notation relative to V-PDB and those of the water to V-SMOW

gastropods and bivalves and the surrounding water (Odum, 1951; Faure et al., 1967; Buchardt & Fritz, 1978; Rosenthal & Katz, 1989; Bailey & Lear, 2006; Anadón et al., 2010). In the present study, the mean  $Kd_{Sr}$  for *Radix sp.* shells is 0.317 (n = 15). It is noteworthy that the MCBN130824 sampling site (Mucuobingni) has the very high  $Kd_{Sr}$  value of 0.867 (Table 3). This anomaly probably results from the fact that lake water contains a low Sr concentration (0.034 ppm; Table 3).

Various factors may affect  $Kd_{Sr}$ , such as temperature, salinity, and growth rate (Rosenthal & Katz 1989). Only a few workers have discussed the factors affecting  $Kd_{Sr}$  variations in the aragonite shells of freshwater mollusks; however, no clear relationship between  $Kd_{Sr}$  and water T has been reported for aragonitic shells (Buchardt & Fritz, 1978; Bailey & Lear, 2006; Carroll & Romanek, 2008; Anadón et al., 2010). For Radix sp. shells, our results support previous findings. Nevertheless, Bailey & Lear (2006) and Carroll & Romanek (2008) suggested that in freshwater bivalves, apart from the strong biological control of the trace element incorporation,  $Kd_{Sr}$ decreased for values of high Sr/Ca<sub>water</sub>. In the present study, it is noteworthy that the plot of  $Kd_{Sr}$  versus Sr/ Cawater reveals a variable sensitivity. For values of Sr/  $Ca_{water}$  less than ~0.007,  $Kd_{Sr}$  decreases rapidly with increasing Sr/Cawater; however, in the case of Sr/ Ca<sub>water</sub> values above ~ 0.007 (Table 3),  $Kd_{Sr}$  exhibits only a small range of variation (Fig. 4).

# $\delta^{13}$ C and $\delta^{18}$ O values of the *Radix* sp. shells

In view of the fact that the analyzed shell fragments ('shell-aperture' in Table 4) were formed prior to sampling, the isotopic characteristics of the lake water during the few weeks prior to the sampling date are clearly reflected by the shell-aperture isotopic data (Taft et al., 2012). A similar pattern is also found in other aragonitic shells (Anadón et al., 2010). In addition, each analyzed powdered bulk sample ('shell-AVG' in Table 4) represents the isotopic value of a *Radix* shell averaged over the life span of the individual.

McConnaughey et al. (1997) inferred that DIC in water can be directly used for aquatic mollusk shell accretion, and that 90% of the carbonate in the shell is derived from this source. The DIC ingested by aquatic mollusks is likely to have high  $\delta^{13}$ C values, between -3% and +3% (Leng & Marshall, 2004). In addition, mollusks also ingest organic matter in the form of algae and plant debris which have low  $\delta^{13}$ C values, between -16% and -35% (Talbot & Johannessen, 1992). The modern Radix sp. shells analyzed in the present study have  $\delta^{13}C$  values ranging from -11.8 to 0.0‰. Overall, our data are consistent with those for aquatic mollusks utilizing carbon derived from both the water's inorganic pool and dietary organic carbon (Fritz & Poplawski, 1974) but are indicative of DIC being the main source. This is further demonstrated by the significant positive correlation between both shell-AVG and shell-aperture  $\delta^{13}$ C values and the  $\delta^{13}$ C<sub>DIC</sub> of the lake water (Fig. 5A). This indicates that the  $\delta^{13}$ C values of *Radix* sp. shells mainly reflect the  $\delta^{13}C_{DIC}$  of the lake water.

 $\delta^{13}C_{DIC}$  in a lake is controlled by photosynthesis, mineralogical substrate, decomposition of organic matter, the isotopic composition of inflowing waters, and exchange with atmospheric CO<sub>2</sub> (Yan et al., 2009; Li et al., 2012). Our results indicate that Nongzhen Tso and Gongmo Tso have significantly low  $\delta^{13}C$  values in shells and lake water. In addition, both lakes, especially Nongzhen Tso are characterized by an abundance of aquatic vegetation, shallow water depth, and small lake area. The low  $\delta^{13}C$  values in lake water DIC probably reflect an abundance of aquatic vegetation which through aerobic decay or respiration adds isotopically light CO<sub>2</sub> to the lake water (Fritz et al., 1975).

It is noteworthy that overall the plot of shell  $\delta^{13}$ C against water DIC reveals a variable sensitivity. Shell  $\delta^{13}$ C changes rapidly during low water DIC at the early stages of the reaction; however, in the case of water DIC values above 33 ppm, the corresponding shell  $\delta^{13}$ C values exhibit only a small range of variation (Fig. 5B). In addition, the  $\delta^{13}$ C of both shell-AVG and shell-aperture tends to decrease with increasing NPOC in the lake water (Fig. 5C). Therefore, a more reasonable explanation is that the  $\delta^{13}$ C of the Radix sp. shells is probably limited by the lake water DIC which is derived from the decomposition of organic matter when the water total DIC content is low (<33 ppm; Fritz & Poplawski, 1974; Fritz et al., 1975; Yan et al., 2009). We suggest, however, that more work is needed to understand the mechanisms by which carbon is incorporated into the Radix sp. shells.

In general, the  $\delta^{18}$ O values of freshwater mollusk shells depend primarily on the temperature and oxygen isotopic composition of the water in which they grew (e.g., Fritz & Poplawski, 1974; Leng et al., 1999). Leng & Marshall (2004) have systematically reviewed the controls on the oxygen isotope composition of lacustrine skeletal deposits and illustrate how oxygen isotope studies contribute to an understanding of changes in temperature, precipitation patterns, and evaporation. Over the Tibetan Plateau, the  $\delta^{18}$ O value of precipitation reflects information about the interaction between the westerlies and Indian monsoon, combined with local recycling which is characterized by evaporation, convection, and droplet re-evaporation (Yao et al., 2013). The lake water  $\delta^{18}$ O is controlled by distinct climatic parameters such as precipitation, evaporation, temperature, humidity, and particular lake system parameters such as size, depth, water residence time, and water composition of inflows (Taft et al., 2012). Monsoon-influenced exoreic lakes display a characteristic monsoonal isotopic pattern which is marked by a negative correlation between shell  $\delta^{18}$ O and the amount of rainfall, resembling the 'amount effect' in precipitation (Lee & Fung, 2007; Taft et al., 2012). Taft et al. (2013) indicated that the  $\delta^{18}$ O values of *Radix* sp. shells from lakes on the eastern and central Tibetan Plateau reflect monsoon signals.

In the present study, the fractionation factor between  $\delta^{18}O_{shell-AVG}$  and  $\delta^{18}O_{water}$  ( $\alpha_{shell-AVG-water}$ ) ranges from 1.031 to 1.034 with a mean of 1.033, while the fractionation factor between  $\delta^{18}O_{shell-aperture}$ 

and  $\delta^{18}O_{water}$  ( $\alpha_{shell-aperture-water}$ ) ranges from 1.032 to 1.040 with a mean of 1.034. These values are close to the observations on natural samples of Patterson et al. (1993) and to the calculated results for the aragonite–water system by Zheng (1999).

The temperature dependence of the oxygen isotopic fractionation between aragonite (natural biogenic and synthesized aragonite) and water has also been reported (Grossman & Ku, 1986; Patterson et al., 1993; Zhou & Zheng, 2000). With regard to the Radix sp. on the Tibetan Plateau, however, our data suggest that there is no significant relationship between either the  $\alpha_{shell-AVG-water}$  or the  $\alpha_{shell-aperture-water}$  with the measured ambient water  $T (10^3 \ln \alpha_{\text{shell-AVG-water}})$  $= 10.329 \times 10^{3}/T - 3.1357, R^{2} = 0.088, n = 30,$  $P = 0.111; \ 10^3 \ln \alpha_{\text{shell-aperture-water}} = -16.246 \times 10^3 / 10^$  $T + 89.918, R^2 = 0.072, n = 30, P = 0.152$ ). In the present study, lake water conditions during Radix sp. growth were not monitored continuously, and despite the fact that we measured the lake water temperature, these temperature values are unlikely to represent seasonal and/or inter-seasonal average Radix sp. growth conditions. Moreover, we speculate that the effect of temperature on fractionation may be relatively weak for contemporary Radix sp. (Tyler et al., 2008). We suggest that the temperature dependence of the oxygen isotopic fractionation in the aragonitewater system for Radix sp. needs to be further studied.

When  $\delta^{18}O_{shell}$  is plotted against  $\delta^{18}O_{water}$  (Fig. 6), the slopes of the regression line are  $\approx 1$ . This implies that the changes in  $\delta^{18}O_{shell}$  may predominantly reflect changes in lake water  $\delta^{18}O_{water}$ , for example, due to evaporation or the changing  $\delta^{18}O$  of precipitation (Tyler et al., 2008). Thus, our data suggest that the  $\delta^{18}O$ values of *Radix* sp. shells can be used to reflect climatic and hydrologic variability at the large regional scale of the Tibetan Plateau. In addition, the good correlation between shell-aperture  $\delta^{18}O$  and the  $\delta^{18}O$  of the lake water (Fig. 6) to some degree confirms that the *Radix* sp. shells can be used as high-resolution archive for analysis of sclerochronological  $\delta^{13}C$  and  $\delta^{18}O$  patterns in lakes across the Tibetan Plateau.

## Conclusions

Our results lead to the following conclusions:

1. *Radix* sp. tend to live in oligohaline to mesohaline water bodies. The nutritional status of the lake

water is possibly the main factor limiting the survival of *Radix* sp, rather than the high pH values typical of the lakes of the Tibetan Plateau.

- 2. The observed strong correlation between the Sr/ Ca ratio of the *Radix* sp. shells and that of the ambient waters, as well as with lake water conductivity, makes the Sr/Ca ratio a valuable tool for paleohydrological studies. However, pH and water *T* have only a minor influence on the Sr/ Ca ratio of the *Radix* sp. shells. The mean  $Kd_{\rm Sr}$ value obtained in the present study (0.317, n = 15) is close to the mean values obtained for diverse non-marine aragonitic gastropods and bivalves. In addition, the  $Kd_{\rm Sr}$  decreases rapidly when Sr/Ca<sub>water</sub> is low; however, in the case of Sr/ Ca<sub>water</sub> values above 0.0076,  $Kd_{\rm Sr}$  exhibits only a small range of variation.
- 3. The shell  $\delta^{13}$ C of *Radix* sp. depends on the  $\delta^{13}$ C of lake water DIC. The  $\delta^{13}$ C values of lake water DIC are primarily controlled by that of the aqueous carbonate species. Our results suggest that the contribution of DIC originated from the degradation of organic matter to the *Radix* sp. shells increases when the water is deficient in DIC of inorganic origin.
- 4. The oxygen isotope ratios of the shells of *Radix* sp. provide information on the isotopic composition of the water in which the shells were formed, which in turn relates to the climatic conditions prevailing during the life span of individuals of *Radix* sp.
- 5. Overall, the fossil shells of the gastropod *Radix* sp. in the terraces of the lakes on the Tibetan Plateau are a potentially valuable archive for reconstructing the paleohydrology and paleoclimatology of the region.

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