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Variations in $\delta^{13}C_{DIC}$ and Influencing Factors in a Shallow Macrophytic Lake on the Qinghai–Tibetan Plateau: Implications for the Regional Carbon Cycle and Sustainable Development

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Abstract: Lake carbon cycle in lake ecosystems is critical for regional carbon management. The application of carbon isotope techniques to terrestrial and aquatic ecosystems can accurately elucidate carbon flow and carbon cycling. Lake ecosystems on the Qinghai–Tibetan Plateau are fragile and sensitive to climate and environment changes, and the carbon cycle impact on the carbon isotopic composition ($\delta^{13}C$) of dissolved inorganic carbon (DIC) in these systems has not been well studied, limiting the ability to devise effective management strategies. This study explored the relationship among the $\delta^{13}C$ position of the DIC ($\delta^{13}C_{DIC}$) in Genggahai Lake, the lake environment, and the climate of the watershed based on the observed physicochemical parameters of water in areas with different types of submerged macrophyte communities, combined with concomitant temperature and precipitation changes. Overall, the Genggahai Basin $\delta^{13}C_{DIC}$ exhibited a large value range; the average $\delta^{13}C_{DIC}$ for inflowing spring water was the most negative, followed by the Shazhuyu River, and then lake water. Variations in the photosynthetic intensity of different aquatic plants yielded significantly changing $\delta^{13}C_{DIC\text{-}L}$ values in areas with varied aquatic plant communities. Hydrochemical observations revealed that $\delta^{13}C_{DIC\text{-}I}$ and aquatic plant photosynthesis primarily affected the differences in the $\delta^{13}C_{DIC\text{-}L}$ values of Genggahai Lake, thereby identifying them as the key carbon cycle components in the lake. This improves the understanding of the carbon cycle mechanism of the Qinghai–Tibetan Plateau Lake ecosystem, which is beneficial to improving sustainable lake development strategies.

Keywords: dissolved inorganic carbon; aquatic plant photosynthesis; lake carbon cycle; Genggahai Lake; Qinghai–Tibetan Plateau

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1. Introduction

Accounting for approximately 3.7% of the global land area, lakes are an important component of inland water systems that regulate the carbon cycle by storing, transporting, and transforming carbon [1–4]. Information on the lake carbon cycle can contribute to a comprehensive understanding of the terrestrial carbon cycle [5,6], which is anticipated to play an important role in China's 2060 carbon neutral commitment [7]. Understanding the lake carbon cycle is of great significance to research regional and global carbon budget, which is a foundational work to establish the regional carbon management and sustainable development policy.

Dissolved inorganic carbon (DIC) occurs in water as CO_2, CO_3^{2-}, HCO_3^-, and H_2CO_3. Carbon isotopes ($\delta^{13}C$) record the cycling of carbon during each of these links, which involves equilibrium and kinetic fractionation [8,9]. Thus, $\delta^{13}C$ analyses of DIC ($\delta^{13}C_{DIC}$) provide a powerful tool for tracing the carbon cycle in lakes and elucidating carbon fluxes [10,11].

Lake ecosystems on the Qinghai–Tibetan Plateau are fragile and sensitive to the changes in the climate and environment [12]. Lakes on the Qinghai–Tibetan Plateau account for approximately 57% of the total lake area in China [13]. Understanding the carbon cycling process of lakes on the Qinghai–Tibetan Plateau is of profound significance for regional ecological environment protection and sustainable development. However, most studies on the $\delta^{13}C_{DIC}$ in lakes on the Tibetan Plateau have focused on the climatic and environmental implications of the $\delta^{13}C$ of lake sediments and have not considered the regional carbon cycle. Furthermore, variations in the $\delta^{13}C_{DIC}$ of various lake types may have different responses to carbon cycle processes. For example, Lei et al. [10] analysed the characteristics of the $\delta^{13}C_{DIC}$ of 24 lakes (mainly closed lakes) across the Qiangtang Plateau, revealing that the high $\delta^{13}C_{DIC}$ values of closed lakes could be primarily attributed to significant catchment-scale contributions from carbonate weathering and the evasion of dissolved CO_2 induced by enhanced lake water evaporation. Therefore, performing a comprehensive study on the changes in the $\delta^{13}C_{DIC}$ values of lakes on the Qinghai–Tibetan Plateau is necessary.

Genggahai (GGH) Lake is a shallow macropytic lake on the Qinghai–Tibetan Plateau. Due to less influence by human activity and aquatic plants grow flourishingly, variations in $\delta^{13}C_{DIC}$ of the GGH Lake is related to carbon cycle in the basin, which is affected by regional climate and environment change and biological activity. This study explored the relationships among the $\delta^{13}C_{DIC}$ values in the GGH Lake, the lake environment, and the watershed climate based on the observed water physicochemical parameters in areas with different types of submerged macrophyte communities, as well as the changes in the temperature and precipitation during the same period. Our objective was to identify the influencing factors of $\delta^{13}C_{DIC}$ in the GGH Lake and reveal its implications for the carbon cycle in an closed lake system.

2. Study Area

Gonghe Basin (35.45°–36.93° N, 98.77°–101.37° E) is located in the north-eastern Tibetan Plateau (Figure 1a) at a mean elevation of approximately 3000 m. This region is characterised by an alpine arid and semi-arid continental climate [14] (pp. 1–166). From the meteorological data from Gonghe Station from 2000 to 2020, the mean annual temperature was ~5.14 °C and the mean annual precipitation was ~325 mm; the annual potential evaporation precipitation was ~1183 mm (http://data.cma.cn/, 1 November 2021). The basin has an elongated shape, which is surrounded by the Xiqing, Heka, Ela, Wahong, Waligong, and South Qinghai mountains (Figure 1a) [14] (pp. 1–166).

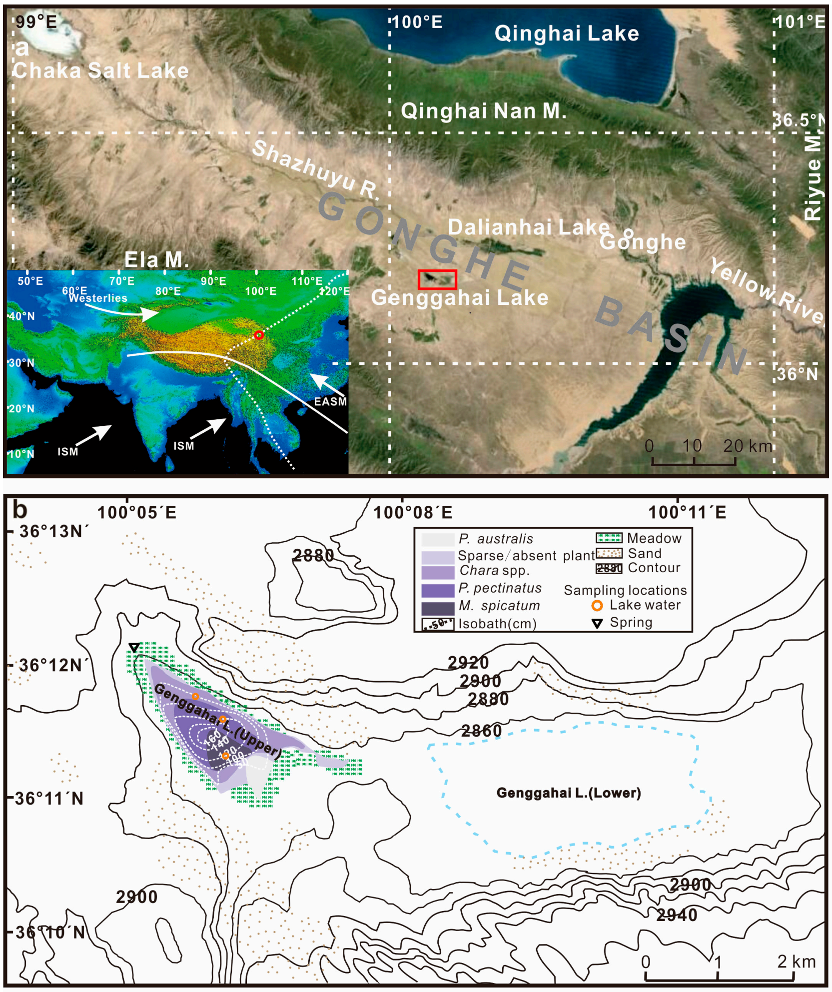

Figure 1. Setting and location. (**a**) Location of the Gonghe Basin (Google Earth^{TM}). The red rectangle and red circle indicate the study site. Climate circulation systems influencing the study area are also shown. Dashed and solid lines indicate the modern extent of the East Asian summer monsoon (EASM) and Indian Ocean summer monsoon (ISM), respectively [15] (pp. 221–264). (**b**) GGH Lake, its surrounding physical environments, and the spatial distribution of modern aquatic vegetation [16].

The GGH Lake (36.18° N, 100.1° E) is located in the central Gonghe Basin (Figure 1a). The present-day area of the lake is approximately 2.0 km^2, with a surface elevation of 2860 m, maximum depth of 1.8 m, water salinity of 1 g L^{-1}, and pH of 9.2 ± 0.5 [16]. The lithology of the GGH Basin is dominated by limestone, sandstone, marble, slate, and schist [14] (pp. 1–166). The lake has no natural discharge outlets or direct surface inflows and is fed mainly by groundwater. Within the lake basin, springs emerge as artesian water. Small spring-water streams emanate from sediment outcrops in the northwest part of the catchment and feed the GGH Lake and sustain the grassland [17] (Figure 1b). The lake is inhabited by numerous submerged macrophytes, such as *Chara* spp., *Myriophyllum spicatum* L., and *Potamogeton pectinatus* L. (Figure 1b), as well as gastropod molluscs [16]. The entire lake is surrounded by grassland, particularly the desert grassland ecosystem dominant in the GGH Basin. This region is characterised by strong wind-blown sand movement. Human activity remains limited, with only a small number of Tibetan herdsmen, who graze their livestock on the grasslands surrounding the lake.

3. Materials and Methods

Between 2012 and 2015, from May to September of each year, lake water samples were collected at a depth of 20 cm in three different areas to characterise the *Chara* spp., *M. spicatum*, and *P. pectinatus* communities. Meanwhile, watershed groundwater samples and surface water samples from the Shazhuyu River were also collected multiple times. Due to the freezing of lake water, water samples were not collected from October to the following April. The collected samples were stored in 500 mL polyethylene plastic bottles; after adding 1 mL of $HgCl_2$ solution, each bottle was sealed with a sealing membrane. A total of 53 lake water samples, 19 groundwater samples, and 17 river water samples were collected. The physical and chemical parameters (i.e., the temperature, dissolved oxygen (DO), and pH) (Table 1) were measured in situ using a portable water quality analyser (AquaRead-1000, Aquaread, Broadstairs, UK).

Table 1. The detailed list of water samples and measured parameters.

Water Sample Types	Measured Parameters
Lake water Groundwater River water	Water temperature pH DO carbon isotopes oxygen isotopes

The plant samples, including *Chara* spp., *M. spicatum*, and *P. pectinatus*, were sampled monthly adjacent to the lake water sampling points. Additionally, *Gyraulus sibiricus* samples adhered to *Chara* spp. were collected. The fresh plant samples and shells were stored in polyethylene plastic bags and numbered before being transported to the lab for further pre-treatment and analysis.

The water samples were subjected to the following laboratory pre-treatment procedure before DIC isotope determination. First, each sample was filtered through a 0.45 μm glass-fibre filter, and 4 mL of a saturated $BaCl_2$ solution was added to induce $BaCO_3$ precipitation [18]. After the samples settled, the clear liquid was siphoned away; the remaining solids were placed in a drying oven to dry at a constant low temperature of 30 °C. Finally, the dried samples were ground to a uniform size, and appropriate amounts were weighed for the DIC isotope determination. The prepared DIC samples were tested using a MAT-253 isotope ratio mass spectrometer (Thermo-Finnigan, Bremen, Germany), together with a micro-carbonate sampling device (Kiel IV). Tests were performed at the Key Laboratory of Western China's Environmental Systems, MOE, Lanzhou University. Test results were reported relative to the PDB standard. Analytical precision values for $\delta^{13}C$ and $\delta^{18}O$ were ±0.03‰ and ±0.05‰, respectively.

To analyse the $\delta^{13}C$ of the plant bulk organic matter ($\delta^{13}C_{org}$), samples were pre-treated based on the methods reported by Song et al. [19]. The $\delta^{13}C_{org}$ was analysed using an on-line Conflo III-DeltaPlus isotope ratio mass spectrometry, combined with a Flash EA1112 elemental analyser at Lanzhou University (Lanzhou, China). The results were reported in ‰ relative to PDB. The standard samples were glycine, puge, and wheat, whose standard deviations were 0.04, 0.07, and 0.05, respectively. All values are reported and discussed in the following section. Temperature data from the Gonghe Station were obtained from the China Meteorological Data Service Centre: Daily data from surface meteorological stations in China.

The Grapher 8 and Coreldraw X8 software were used for drawings. Pearson correlation analysis was applied by SPSS 17.0 statistical software. Pearson correlation analysis is a statistical method used to evaluate the strength and direction of a linear relationship between two variables. Based on covariance, it calculates a correlation coefficient between −1 and 1. The results of Pearson correlation analysis include two values: correlation coefficient and *p*-value. The correlation coefficient shows the direction and strength of the linear relationship between two variables. A value close to 1 indicates a high positive correlation between the two variables and a value close to −1 indicates a high negative correlation, and a value close to 0 indicates that the two variables are independent of each other and have no correlation. The *p*-value represents an indicator of whether two variables are significantly correlated. If the *p*-value is less than 0.05, the two variables are considered to be significantly correlated.

4. Results

4.1. Physical and Chemical Parameters of Lake Water

The overall temperature and pH trends of the lake water were relatively consistent in all three plant communities with the previously published physical and chemical parameters of water from the GGH Lake measured from 2012 to 2013 [20] (Figure 2a,b). Specifically, the overall temperature remained relatively constant throughout the entire monitoring period, but its annual fluctuations were more notable, with higher temperatures in June, July, and August, and relatively low temperatures in May and September (Figure 2a). From 2012 to 2015, the lake water pH exhibited an overall gradually increasing trend from May to September, with only a few exceptions (e.g., June and July 2015; Figure 2b). Beginning in May, both the number of aquatic plants and water temperature of the lake increased, which led to the increased consumption of CO_2 and HCO_3^- via plant photosynthesis. Moreover, the variation in DO was not notable, with all three plant communities showing inconsistencies, which may have been related to the high sensitivity of DO to temperature changes (Figure 2c).

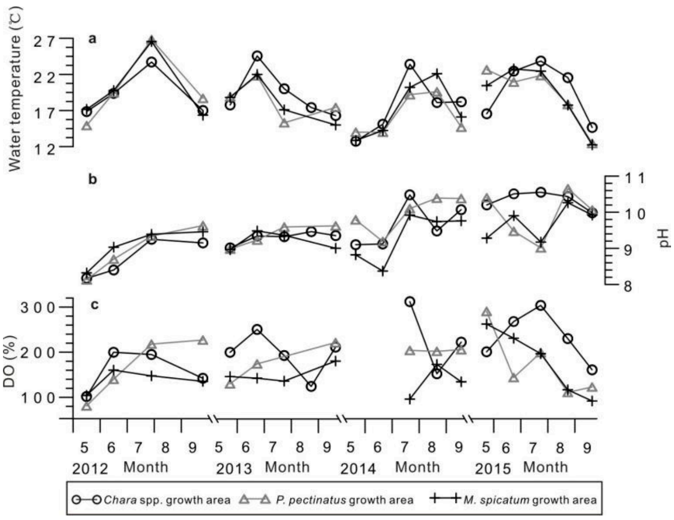

Figure 2. Seasonal variations in the physical and chemical parameters of the lake water in different plant growth areas of the GGH Lake from May to September (2012–2015). (**a**) Water temperature; (**b**) pH value; and (**c**) DO concentrations.

A comparison of the three plant communities showed that, except for 2012, the pH values in the *Chara* spp. community were slightly higher than those in the *P. pectinatus* and *M. spicatum* communities throughout the testing period (Figure 2a,b). Differences in the DO values of lake water in the three communities were more prominent; the *Chara* spp. community had relatively higher DO values in June and July (Figure 2c). Compared to vascular plants (*P. pectinatus* and *M. spicatum* communities), *Chara* spp. community has higher photosynthesis rates and lower respiration rates [21].

4.2. Spatial Variations in DIC Isotopic Composition in the GGH Basin Waterbodies

From 2012 to 2015, the $\delta^{13}C_{DIC}$ values of water from the GGH Lake ($\delta^{13}C_{DIC\text{-}L}$) ranged from −17.3 to 1.6‰, with a mean value of −6.91‰. The $\delta^{13}C_{DIC}$ of water from the Shazhuyu River ($\delta^{13}C_{DIC\text{-}R}$) ranged from −15.9 to −5.6‰, with a mean value of −10.8‰. The $\delta^{13}C_{DIC}$ for the groundwater spring, a lake water source, ranged from −17.3 to −1.1‰, with a mean value of −11.1‰. Overall, the $\delta^{13}C_{DIC\text{-}L}$ values were the most positive, followed by the $\delta^{13}C_{DIC\text{-}R}$ values, whereas the $\delta^{13}C_{DIC}$ values of the inflowing spring water ($\delta^{13}C_{DIC\text{-}I}$) were the most negative (Figure 3).

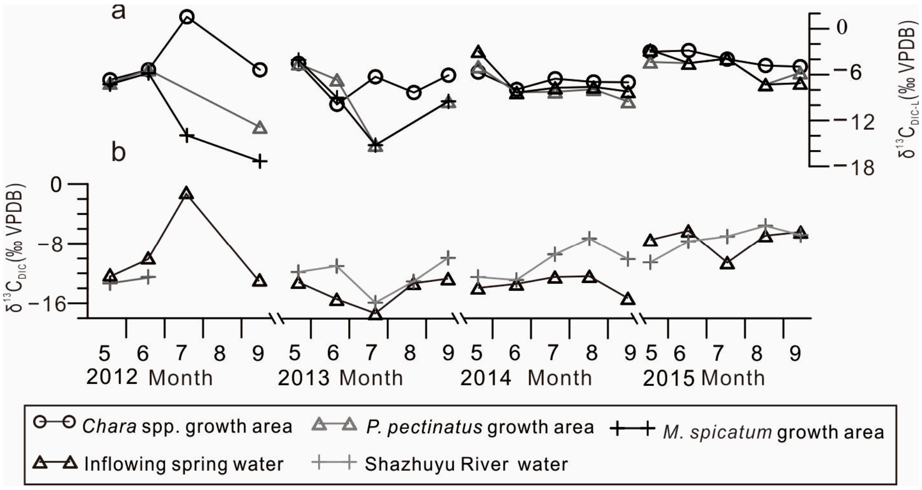

Figure 3. Variations in the carbon isotopes of DIC precipitated from the (**a**) surface water in the GGH Lake and (**b**) waterbodies within its catchment from May to September (2012–2015).

The $\delta^{13}C_{DIC-L}$ values in the *Chara* spp. community were more positive than those in the *M. spicatum* and *P. pectinatus* communities. From 2012 to 2015, the $\delta^{13}C_{DIC-L}$ values in the *Chara* spp. community ranged from −9.9 to 1.6‰, with a mean value of −5.4‰. In the *P. pectinatus* community, the $\delta^{13}C_{DIC-L}$ values ranged from −15.2 to −3.9‰, with a mean value of −7.4‰. In the *M. spicatum* community, the $\delta^{13}C_{DIC-L}$ values ranged from −17.3 to −2.9‰, with a mean of −7.9‰. Except for one month (September 2012), the *P. pectinatus* and *M. spicatum* communities had similar $\delta^{13}C_{DIC-L}$ values and both communities had more negative values than the *Chara* spp. community (Figure 3a).

4.3. Temporal Variations in DIC Isotopic Compositions in the GGH Basin Waterbodies

In the *Chara* spp. community, the $\delta^{13}C_{DIC-L}$ values were relatively positive in July from 2012 to 2014, while, in 2015, there was a gradual decrease. In contrast, the $\delta^{13}C_{DIC-L}$ values of the *P. pectinatus* and *M. spicatum* communities did not show a seasonal bias of more positive values (Figure 3a). Figure 4a shows the interannual variations in the $\delta^{13}C_{DIC-L}$ values. From 2012 to 2015, the mean $\delta^{13}C_{DIC-L}$ values of the *Chara* spp. community were all more positive (−4.0‰), whereas in 2013 and 2014, these values were more negative (−6.8‰). From 2012 to 2015, the $\delta^{13}C_{DIC-L}$ values in the *P. pectinatus* and *M. spicatum* communities showed a gradual positive trend. In comparison, the $\delta^{13}C_{DIC-I}$ values from 2012 to 2015 showed an overall increase; however, in 2013 and 2014, these values became increasingly negative (Figure 4b). During the same period, the $\delta^{13}C_{DIC-R}$ values gradually increased (Figure 4b).

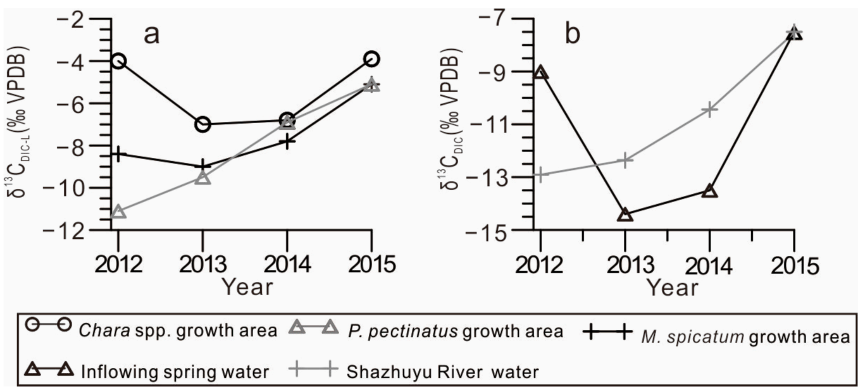

Figure 4. Annual variations in the carbon isotopes of DIC precipitated from the surface water in the GGH Lake (**a**) and waterbodies within its catchment (**b**) from 2012 to 2015.

5. Discussion

5.1. Factors Affecting $\delta^{13}C_{DIC}$ in the GGH Basin Waterbodies

Several major processes affect the stable carbon isotope compositions of lake water DIC, such as in-lake processes (including lake metabolism, organic matter decomposition, calcite precipitation, and exchange with atmospheric CO_2) and the climatic and geographical environment of the catchment (including carbonate rock weathering, dissolution, and soil respiration) [22]. The climatic and geographical environment in the catchment can alter the carbon isotope composition of the lake water DIC by influencing the aqueous CO_2 and alkalinity of inflowing water. Generally, the five following factors affect the isotope composition of the DIC.

(1) Lake inflow carbon isotope composition. The isotopic composition of the lake inflow directly affects the isotopic composition of the lake. This is especially significant in exorheic lakes or lakes with a short water retention time. In the *Chara* spp. community and *P. pectinatus* growth area, the $\delta^{13}C_{DIC\text{-}L}$ and $\delta^{13}C_{DIC\text{-}I}$ values were positively correlated, whereas in the *M. spicatum* community growth area, these values showed no correlation (Figure 5). This is because the *M. spicatum* community was located far from the small spring-water streams, where weak exchange with spring water took place (Figure 1b). This reveals that the $\delta^{13}C_{DIC\text{-}I}$ values were key factors influencing the $\delta^{13}C_{DIC\text{-}L}$ values.

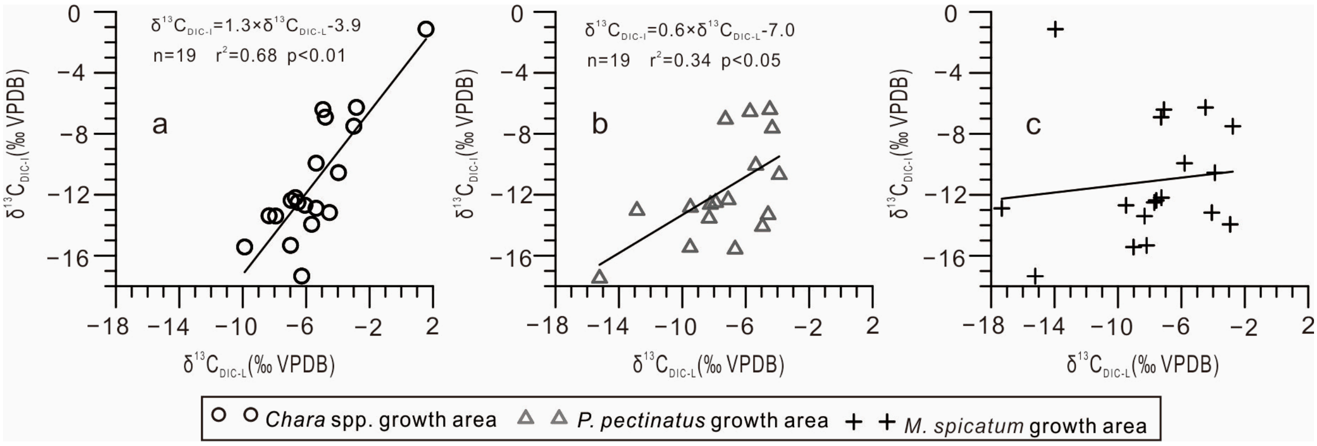

Figure 5. Correlation between the carbon isotopes of inflowing water DIC and lake water DIC in (**a**) *Chara* spp., (**b**) *Potamogeton pectinatus*, and (**c**) *Myriophyllum spicatum* growth areas.

(2) Exchange with atmospheric CO_2. The DIC pool in the lake tends to be in isotopic equilibrium with the atmosphere via CO_2 exchange. During this process, ^{12}C-rich CO_2 is preferentially released from the lake surface into the atmosphere, resulting in a DIC pool that is enriched in ^{13}C. This exchange process is slow; therefore, its effect on the $\delta^{13}C_{DIC\text{-}L}$ values is more notable in endorheic lakes with a long retention time. However, it is not easily observable in lakes with short retention times or rapid circulation [23]. When the exchange between lake water and atmospheric CO_2 reaches an equilibrium, the $\delta^{13}C_{DIC\text{-}L}$ values range from 1 to 3‰ [24,25].

The exchange between the DIC of lake water and atmospheric CO_2 is continuous in exorheic lakes. At equilibrium, isotope fractionation occurs between atmospheric CO_2 and dissolved carbonate species, i.e., CO_2 (aq), HCO_3^-, and CO_3^{2-} [8], as follows:

$$\varepsilon_{aq\ g} = -(0.0049 \pm 0.003) \times T\ (°C) - (1.31 \pm 0.06), \quad (1)$$

$$\varepsilon_{HCO3\ g} = -(0.141 \pm 0.003) \times T\ (°C) + (10.78 \pm 0.05), \text{ and} \quad (2)$$

$$\varepsilon_{CO3\ g} = -(0.052 \pm 0.03) \times T\ (°C) + (7.22 \pm 0.46) \quad (3)$$

Temperatures recorded using a water level data logger showed that from May to September of 2013–2015, the mean water surface temperature of the GGH Lake was 17.0 °C. Based on Equations (1)–(3), at the mean water surface temperature in the GGH Lake, the carbon isotope fractionation factors between H_2CO_3, HCO_3^-, and CO_3^{2-} and atmospheric CO_2 are −1.5, 8.38, and 5.89‰, respectively. The isotopic composition of global atmospheric CO_2 is approximately −8.1‰ [26]. Therefore, at equilibrium, the isotopic values of H_2CO_3, HCO_3^-, and CO_3^{2-} in the lake water are −9.6, 0.28, and −2.21‰, respectively. Different forms of DIC have varying $\delta^{13}C$ values at isotopic equilibrium; the magnitude of the $\delta^{13}C_{DIC}$ value depends on the proportions of the different DIC forms in lake water, which is related to its pH value [27] (p. 583). When the pH value is 5.5, 80% of the DIC in a water body has an aqueous CO_2 form (aq). When the pH is 8.5, CO_2 (aq) accounts for <1% of the DIC, which predominantly takes HCO_3^- and CO_3^{2-} forms. When the pH reaches 10, HCO_3^- accounts for <50% of the DIC; however, CO_3^{2-} dominates the DIC [27] (p. 583). The pH value of the GGH Lake varied from 8.1 to 10.6, indicating that HCO_3^- and CO_3^{2-} were the dominant forms of DIC. Notably, the actual $\delta^{13}C_{DIC}$ values of the GGH Lake were generally more negative than the atmosphere-equilibrated $\delta^{13}C_{DIC}$.

(3) Organic matter decomposition in lake sediments. Sedimentary organic matter in lakes includes native aquatic plants and terrestrial organic debris transported into the lake from the surrounding watershed. Once degraded, this organic matter increases the ^{12}C-enriched DIC composition of lake water [9]. Organic matter decomposition in Qingmuke Lake (a freshwater lake located on the Qiangtang Plateau) resulted in a DIC isotope value equal to, or even lower than, that of river water [10]. In contrast, the $\delta^{13}C_{DIC\text{-}L}$ values in the GGH Lake were significantly more positive than that of the Shazhuyu River, indicating that organic matter decomposition may have had a relatively small effect on the DIC lake composition. Additionally, methane produced by organic matter decomposition resulted in a more negative $\delta^{13}C_{DIC}$ value. Organic matter decomposition can cause a decrease in the $\delta^{13}C_{DIC}$ values to −50‰ [28]. This value is significantly lower than the mean $\delta^{13}C_{DIC\text{-}L}$ value of the GGH Lake, which indicates that the CO_2 or methane produced via decomposition did not have a significant effect on seasonal or interannual changes in the $\delta^{13}C_{DIC\text{-}L}$ values.

During the observation period, the mean carbon isotopic composition of organic matter ($\delta^{13}C_{org}$) in the *Chara* spp. community was −16.0‰, whereas the mean values of $\delta^{13}C_{org}$ in the *P. pectinatus* and *M. spicatum* communities were −12.7 and −11.4‰, respectively. If we neglect the carbon isotope fractionation effect owing to organic matter decomposition, the $\delta^{13}C$ of the CO_2 released via organic matter decomposition is then equal to $\delta^{13}C_{org}$. According to Equation (2), if HCO_3^- is the dominant form of DIC in the lake, then the equilibrium isotopic value of HCO_3^- in the lake water is 0.28‰. In this case, the mean $\delta^{13}C_{DIC\text{-}L}$ value of the *Chara* spp., *P. pectinatus*, and *M. spicatum* communities would be −15.72, −12.42, and −11.12‰, respectively. However, the observed mean $\delta^{13}C_{DIC\text{-}L}$ values for these three communities were −5.4, −7.4, and −7.9‰, confirming that organic matter decomposition has a limited effect on the $\delta^{13}C_{DIC\text{-}L}$ of the GGH Lake.

(4) Lake photosynthetic activity. In highly productive lakes, photosynthesis is a key factor that affects the $\delta^{13}C_{DIC}$ lake water values [29] (pp. 197–207), [30] (pp. 99–118). During photosynthesis, plants preferentially uptake ^{12}C, which yields more negative $\delta^{13}C$ values for plants and the $\delta^{13}C_{DIC}$ of the water body becomes more positive [31]. Charaphytes are an important submerged aquatic macrophyte. Compared with vascular plants, charaphytes have a higher photosynthetic rate and lower respiration rate. The preferential uptake of $^{12}CO_2$ for photosynthetic purposes could have led to the ^{13}C-enrichment of DIC in the lake water [32]. During intense photosynthesis, dissolved CO_2 in lake water is limited [33]. When this occurs, charaphytes use HCO_3 for photosynthetic activity. Compared with vascular plants, charaphytes can use HCO_3^- for photosynthetic activity more effectively [21]. According to Equations (1)–(3), the $\delta^{13}C$ values of HCO_3^- were more positive than those of H_2CO_3 and CO_3^{2-} in the

lake water. In contrast, the photosynthetic activity of charaphytes results in carbonate precipitation in the surrounding waters, forming thick $CaCO_3$ encrustations [33]. This also leads to ^{13}C-enriched water in the charaphyte growth area.

We found that the seasonal bias in the $\delta^{13}C_{DIC-L}$ values of the *Chara* spp. community were more positive in July (2012–2014) (Figure 4a). Additionally, at the beginning of *Chara* spp. growth (in May), the $\delta^{13}C_{DIC-L}$ value of the *Chara* spp. community was equal to the value at the end of *Chara* spp. growth (in September), especially in 2012. In contrast, the $\delta^{13}C_{DIC-L}$ values in the *P. pectinatus* and *M. spicatum* communities showed no seasonality (Figure 4a). This phenomenon may have occurred because *Chara* spp. has a higher photosynthetic rate and a lower respiration rate than that of the other communities [21]. Pentecost et al. [34] also observed seasonal variations in the $\delta^{13}C_{DIC-L}$ values of the *Chara* spp. community in the UK. Pełechaty et al. [33] suggested that an increase in $\delta^{13}C_{DIC}$ results from the intense photosynthetic activity of *Chara rudis* during the early summer. Moreover, we found that the differences in the $\delta^{13}C_{DIC-L}$ values between *Chara* spp. and vascular plants were smaller at the beginning and end of the growing season in the GGH Lake but larger during the mid-growth season (July) (Figure 4a). There may have been a limited impact from photosynthesis on the $\delta^{13}C_{DIC-L}$ values in areas with submerged vascular plants. This trend was not evident during certain months, e.g., in July 2015. Due to data limitations, we cannot provide an explanation for this phenomenon. Nevertheless, we can reasonably conclude that the variations in the $\delta^{13}C_{DIC-L}$ values of the lake water were related to the intensity of photosynthetic activity in different aquatic plants.

(5) Water retention time. In arid regions, with extended lake water residence times, strong evaporation leads to the preferential loss of the light $^{12}CO_2$ and $^{16}O_2$ isotopes, yielding more positive $\delta^{13}C_{DIC-L}$ and oxygen ($\delta^{18}O$) isotopic lake water compositions; furthermore, a significant positive correlation was observed between $\delta^{13}C_{DIC-L}$ and $\delta^{18}O_L$ [35]. Monitoring results revealed that the $\delta^{18}O_L$ values of the GGH Lake significantly deviated from the global meteoric water line but were consistent with the local evaporation line, indicating that evaporation affected the $\delta^{18}O_L$ lake water composition [19,36]. However, this study found that the $\delta^{13}C_{DIC-L}$ and $\delta^{18}O_L$ values of the GGH Lake were not correlated (Figure 6), indicating that evaporation may have had only a minimal effect on the $\delta^{13}C_{DIC-L}$ value of the lake.

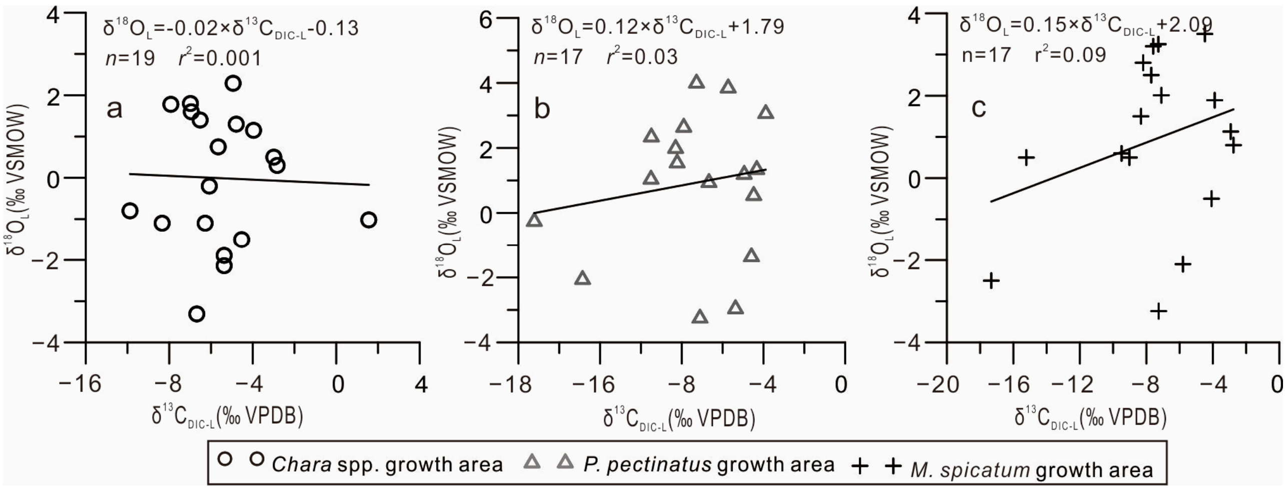

Figure 6. Correlation between the carbon isotopes of the lake water DIC and oxygen isotopes of the lake water in the (**a**) *Chara* spp., (**b**) *Potamogeton pectinatus*, and (**c**) *Myriophyllum spicatum* growth areas.

The preceding analysis demonstrates that the lake surface to atmospheric exchange of CO_2 and evaporation had a relatively minimal effect on $\delta^{13}C_{DIC-L}$; $\delta^{13}C_{DIC-I}$ primarily influenced the changes in $\delta^{13}C_{DIC-L}$. The high photosynthetic efficiency of *Chara* spp. indicates that its corresponding $\delta^{13}C_{DIC-L}$ values showed a seasonal trend of more positive values, which were more positive than the $\delta^{13}C_{DIC\text{-}L}$ values of areas with vascular plants. This shows that the photosynthetic activity of vascular plants has a negligible effect on the $\delta^{13}C_{DIC\text{-}L}$ of lake water.

5.2. Isotopic Composition of DIC in the GGH Basin Groundwater and Shazhuyu River

The $\delta^{13}C_{DIC\text{-}I}$ and $\delta^{13}C_{DIC\text{-}R}$ values were significantly more negative than $\delta^{13}C_{DIC\text{-}L}$ during the same period, suggesting that isotopic fractionation due to atmospheric exchange or the photosynthesis of aquatic plants occurred after groundwater inflow into the lake [25]. Compared with the $\delta^{13}C_{DIC\text{-}I}$ value, $\delta^{13}C_{DIC\text{-}R}$ was significantly more positive, likely because the Shazhuyu River forms via surface runoff, as well as the more frequent exchange of atmospheric CO_2 with surface water than groundwater.

A positive correlation was observed between $\delta^{13}C_{DIC\text{-}I}$ and $\delta^{13}C_{DIC\text{-}R}$ ($n = 17$, $r^2 = 0.46$, and $p < 0.01$). Additionally, given that groundwater is the main water source for the GGH Lake, $\delta^{13}C_{DIC\text{-}I}$ was the main factor affecting the $\delta^{13}C_{DIC\text{-}L}$ values of the lake water (see Section 5.1). Therefore, we further analysed the influencing factors of $\delta^{13}C_{DIC\text{-}I}$.

Three main species of DIC in water bodies are CO_2, CO_3^{2-}, and HCO_3^-. In this study, the titration method was used to determine the DIC composition of groundwater in the GGH Basin. HCO_3^- was the dominant form of DIC. Additional research has shown that the primary source of HCO_3^- in the groundwater of the Genggahai Basin and the Shazhuyu River is the chemical weathering of rocks, particularly carbonate rocks [37]. Based on the reaction equation for chemical weathering, CO_2 is an essential component of this process. Dissolved CO_2 in groundwater originates from the atmospheric flux, watershed soil respiration, and organic matter decomposition. Atmospheric CO_2 normally has $\delta^{13}C$ values of approximately −8‰ [25], which is relatively stable. The $\delta^{13}C$ values of soil CO_2 from areas with C_3 and C_4 plants range from −32 to −20‰ [38] and −17 to −19‰, respectively. The $\delta^{13}C$ value of HCO_3^- from carbonate dissolution during subsurface weathering is approximately 0‰ [39]. The isotopic equilibrium between CO_2 and HCO_3^- from different sources can be attained through the following exchange reaction:

$$^{13}CO_2(g) + H_2{}^{12}CO_3(aq) = {}^{12}CO_2(g) + H_2{}^{13}CO_3(aq). \quad (4)$$

The fractionation factor of CO_2 and HCO_3^- in the CO_2–HCO_3^- system in soil is approximately 10‰ [25]. After soil CO_2 is dissolved in water, its $\delta^{13}C$ value becomes more negative than that of HCO_3^-. However, carbonate rock dissolution increases the $\delta^{13}C$ value of HCO_3^-. The carbon source that affects the isotopic composition of DIC in groundwater encompasses two components: (1) ^{13}C from the weathering and dissolution of carbonate rocks, which possesses a more positive isotope ratio; and (2) ^{12}C from CO_2 generated through soil respiration, which has a more negative isotope ratio. Therefore, the relative contribution of these two carbon sources to the groundwater DIC determines the composition of $\delta^{13}C_{DIC\text{-}I}$ in the GGH Basin.

5.3. Implications of Lake Water DIC Isotopic Composition on the Carbon Cycle

Lake ecosystems are active components of the global carbon cycle as they continually fix and release carbon through various biological processes, including photosynthesis, food web activity, and bacterial degradation [1,40]. The DIC and its isotopes are an important tool to elucidate the carbon cycle of lake ecosystems [10,11,18,30,41] (pp. 99–118). Striegl et al. [42] found that during the ice melting period, the average $\delta^{13}C_{DIC\text{-}L}$ value in 132 freshwater lakes in temperate and cold regions was −14‰. Similarly, Bade et al. [22] reported that the average $\delta^{13}C_{DIC\text{-}L}$ value in 108 freshwater lakes in different regions was −15‰. In freshwater lakes, the photosynthesis of aquatic plants and organic matter decomposition in sediments are active components of the carbon cycle [10]. In freshwater lakes on the Qiangtang Plateau, variations in $\delta^{13}C_{DIC\text{-}L}$ also showed that organic matter decomposition significantly contributed to the carbon cycle of the lake ecosystems [10]. However, Lei et al. [10] found that the $\delta^{13}C_{DIC\text{-}L}$ values of endorheic lakes on the Qiangtang Plateau were relatively high, approaching the $\delta^{13}C_{DIC\text{-}L}$ values of equilibrated CO_2 in water to atmospheric exchange. Therefore, water surface to atmospheric CO_2 exchange drives the carbon cycle in endorheic lakes on the Qiangtang Plateau [10].

The $\delta^{13}C_{DIC\text{-}L}$ values of the GGH Lake significantly exceeded the $\delta^{13}C_{DIC}$ values of freshwater lakes (>8‰) but were more negative than those of lakes on the Qiangtang Plateau (>−5.71‰) [10]. This indicates that the carbon cycle of the GGH Lake significantly differs from those of freshwater lakes and lakes on the Qiangtang Plateau. Variations in the $\delta^{13}C_{DIC\text{-}L}$ values of the GGH Lake indicate that organic matter decomposition and water–atmospheric CO_2 exchange are not likely the main components of its carbon cycle (Section 5.1). Carbon input from inflowing groundwater and the photosynthesis of aquatic plants may be the main components of its carbon cycle.

Recent vegetation surveys have shown that the main terrestrial plants in the GGH watershed are *Artemisia desertorum*, *Oxytropis aciphylla*, *Achnatherum splendens*, *Orinus kokonorica*, and *Agropyron cristatum*. This is identical to that in the Qinghai Lake watershed [43]. The $\delta^{13}C_{org}$ values of ^{3}C plants in the Qinghai Lake watershed varied from −27.7 to −24.5‰ and those of soil in the Qinghai Lake watershed varied from −26.9 to −24.8‰ [43]. When the DIC in groundwater only originates from soil respiration and organic matter decomposition, if we neglect the effect of carbon isotope fractionation due to organic matter decomposition, the $\delta^{13}C_{DIC}$ of groundwater ranges from −27.7 to −24.5‰. Considering the isotope fractionation between CO_2 (aq) and HCO_3^- (approximately −10‰) [8], the $\delta^{13}C_{DIC}$ of groundwater ranges from −17.7 to −14.5‰. As previously mentioned, most $\delta^{13}C_{DIC}$ values of groundwater in the GGH Lake watershed were more positive than −14.5‰. This indicates that soil respiration and organic matter decomposition may not be the only carbon source for the groundwater DIC in the watershed.

Significantly higher $\delta^{13}C_{DIC}$ (approximately −3 to +3‰) values in groundwater can occur in karstic regions where a proportion of the carbon atoms derive from the dissolution of catchment limestones [44]. For example, the $\delta^{13}C_{DIC}$ of groundwater from the Donggi Cona catchment on the north-eastern Qinghai–Tibetan Plateau varies from 0.9 to 2.0‰ [39]. Paleolake sediments that formed in the early and middle Pleistocene are widely distributed throughout the Gonghe Basin [14] (pp. 1–166). Here, HCO_3^- originates from the weathering of paleolake sediments and subsequent mineral dissolution, which is enriched in ^{13}C. Groundwater flows transport the HCO_3^-, thus yielding more positive $\delta^{13}C_{DIC}$ values. We suggest that HCO_3^- originates from paleolake sediment weathering, which affects the groundwater DIC pool in the GGH watershed and yields relatively positive groundwater $\delta^{13}C_{DIC}$ values. Thus, variations in the $\delta^{13}C_{DIC}$ values of groundwater may reflect the relative contributions of two carbon sources to the groundwater DIC pool. One carbon source is CO_2 that derives from soil respiration and organic matter decomposition while the other is HCO_3^- from paleolake carbonate sediments. Furthermore, we elucidated that variations in $\delta^{13}C_{DIC\text{-}I}$ may be affected by carbonate weathering and soil respiration related to vegetation succession. Therefore, variations in the $\delta^{13}C_{DIC\text{-}L}$ values of the GGH Lake may reflect the lake productivity and carbon cycle of the watershed. In the future, we should consider the carbon flux generated from aquatic plants photosynthesis, carbon sink capacity caused by rock weathering process in the watershed and carbon flux produced by soil respiration, when estimate the regional carbon budget.

6. Conclusions

(1) For the overall DIC isotopic composition in the GGH Basin, we found that $\delta^{13}C_{DIC\text{-}I}$ was the most negative, followed by $\delta^{13}C_{DIC\text{-}R}$; $\delta^{13}C_{DIC\text{-}L}$ was the most positive. This was caused by isotope fractionation resulting from the photosynthesis of aquatic plants after spring water inflow into the lake.

(2) Owing to variations in the photosynthetic activity intensity of different aquatic plants, there were also significant variations in the $\delta^{13}C_{DIC\text{-}L}$ values in areas with different aquatic plants. This likely occurred because *Chara* spp. plants have a higher photosynthetic rate and are more capable of using CO_2 for photosynthetic activity, converting them into plant organisms.

(3) Variations in the $\delta^{13}C_{DIC-L}$ were primarily affected by the $\delta^{13}C_{DIC-I}$ and aquatic plant photosynthesis. The change in $\delta^{13}C_{DIC-I}$ to a more positive value resulted from carbon isotope equilibration between ^{13}C from carbonate weathering in the watershed and $^{12}CO_2$ from soil respiration.
(4) The changes in the $\delta^{13}C_{DIC-L}$ composition of the GGH Lake indicated that the DIC from lake inflow and the photosynthesis of aquatic plants were the key components in the carbon cycle of the lake. This provides more supportive evidence to estimate the regional carbon budget and sustainable development.

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