



Soil CO₂ and CH₄ emissions and their carbon isotopic signatures linked to saturated and drained states of the Three Gorges Reservoir of China[☆]

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ABSTRACT

Human activities such as dams disturb the structure and function of wetlands, triggering large soil CO₂ and CH₄ emissions. However, controls over field CO₂ and CH₄ emissions and their carbon isotopic signatures in reservoir wetlands are not yet fully understood. We investigated in situ CO₂ and CH₄ emissions, the δ¹³C values of CO₂ and CH₄, and associated environments in the saturated and drained states under four elevations (i.e., the water column, <147 m, permanent inundation area without plants; the low, 145–160 m, frequently flooded area with revegetation; the high, 160–175 m, rarely flooded area with revegetation; and the upland area as the control, >175 m, nonflooded area with original plants) in the Three Gorges Reservoir area. The CO₂ emissions were significantly higher in high elevation, and they also significantly differed between the saturated and drained states. In contrast, the CH₄ emissions on average (41.97 μg CH₄ m⁻² h⁻¹) were higher at high elevations than at low elevations (22.73 μg CH₄ m⁻² h⁻¹) during the whole observation period. CH₄ emissions decreased by 90% at low elevations and increased by 153% at high elevations from the saturated to drained states. The δ¹³C of CH₄ was more enriched at high elevations than in the low and upland areas, with a more depleted level under the saturated state than under the drained state. We found that soil CO₂ and CH₄ emissions were closely related to soil substrate quality (e.g., C: N ratio) and enzyme activities, whereas the δ¹³C values of CO₂ and CH₄ were primarily associated with root respiration and methanogenic bacteria, respectively. Specifically, the effects of the saturated and drained states on soil CO₂ and CH₄ emissions were stronger than the effect of reservoir elevation, thereby providing an important basis for assessing carbon neutrality in response to anthropogenic activities.

1. Introduction

Increased CO₂ and CH₄ emissions are the main causes of global warming (IPCC, 2013), with approximately 44% and 60% of CO₂ and CH₄ emissions being emitted into the atmosphere by anthropogenic activities (WMO, 2017). Recently, human activities such as dams/reservoirs have been identified as an unignorable fraction of anthropogenic CO₂ and CH₄ sources (Deemer et al., 2016; Hao et al., 2019; Zhang et al., 2021). The global reservoir CO₂ and CH₄ emissions from water surface were estimated to be 36.8 CO₂-C Tg and 13.4 CH₄-C Tg per year, respectively (Deemer et al., 2016). In particular, dam-triggered periodic flooding, which is distinct from natural systems, could enhance gas emissions (Barros et al., 2011; Tan et al., 2019). However, the contribution of CO₂ and CH₄ emissions in reservoir wetlands to the

atmosphere is still an ongoing scientific debate (Marani and Alvala, 2007; Jacinthe, 2015; Zhang et al., 2021).

CO₂ emissions from reservoir wetlands are directly influenced by continued organic matter inputs from inflowing rivers and the regrowth of plants along riparian conditions during the drained state (Deemer et al., 2016; Li et al., 2018). It has been suggested that the respiration of flooded organic matter could lead to greater anaerobic CO₂ emissions from the water surface (McNicol and Silver, 2014). Some studies have reported that CO₂ emissions at the air-water interface tend to decline more rapidly in cold-water environments than in warm waters (Abril et al., 2005; Barros et al., 2011; Teodoru et al., 2011). In the riparian zone, changes in vegetation usually affect the amount and quality of soil organic matter, leading to considerable changes in substrate-stimulated soil microbial activity, which can in turn influence soil respiration

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(Morrissey et al., 2014; Moon et al., 2016). In addition, water-level fluctuation strongly affects CO₂ emissions in riparian zones by changing the soil moisture and temperature conditions (McNicol and Silver, 2014; Poblador et al., 2017; Luan et al., 2018).

In contrast, soil CH₄ emissions in reservoir wetlands are difficult to accurately estimate and predict, as complex controls over CH₄ processes generate large spatial and temporal variations (Barros et al., 2011; Deemer et al., 2016; Zhang et al., 2021). In riparian soils of reservoir wetlands, water-level fluctuation with wetting and drying cycles greatly influences CH₄ emissions by changing the aerobic/anaerobic conditions in the soil profile (Hao et al., 2019; Müller et al., 2019) and affects CH₄-metabolizing microorganism (i.e., methanotrophic and methanogens) activity (Li et al., 2020). Microorganisms involved in CH₄ metabolism rely on simple substrates released from respiring or fermenting microbes, which are often regulated by enzyme activity and the degradation of complex organic matter (Morrissey et al., 2014; Wang et al., 2020a). Thus, dam-triggered hydrological fluctuations are an overwhelming force driving soil CH₄ emission processes in the riparian zone, but how soil CH₄ emissions respond to fluctuations in wetting and drying and the associated drivers remain unclear.

Stable carbon isotopes are used extensively to quantify ecological processes related to carbon cycling (Bowling et al., 2008; Brownlow et al., 2017). The δ¹³C of the CO₂ value after microbial respiration could deviate, possibly because microbes utilized different substrates. If the δ¹³C value of the substrates changes with different litter inputs (Lorenz et al., 2020), so could the δ¹³C of CO₂. Meanwhile, the δ¹³C of CH₄ was associated with CH₄ production processes, with CH₄ produced by acetate cleavage typically having δ¹³C values between -65 and -50‰, whereas CH₄ from CO₂ production had δ¹³C values between -110 and -60‰ (Vaughn et al., 2016). Water fluctuation can alter soil microclimatic conditions and substrates and potentially affect soil-respired CO₂ and CH₄ isotope signatures.

The Three Gorges Reservoirs were designed for multiple functions, with a large spectrum of water-level fluctuation from elevations of 147 m–175 m during damming operation (Bao et al., 2015). Water-level fluctuation could strongly impact soil water conditions and plant biomass (Ye et al., 2013). Our previous studies also found that water-level fluctuation predominantly altered plant functional species (Zhang et al., 2020a), soil C dynamics and microbial community and enzyme activities (Yang et al., 2019; Zhang et al., 2020b). However, there are few studies on the patterns of soil CO₂ and CH₄ emissions and their carbon isotopic signatures under water-level fluctuations. Thus, in the present study, we selected four elevations (i.e., >175 m, 160–175 m, 145–160 m and the water column) under the riparian zone of the Three Gorges Reservoir, in order to quantify the pattern of CO₂ and CH₄ emissions and their carbon isotopic signatures, as well as associated controls under the saturated and drained states. We hypothesized that 1) changes in soil conditions (e.g., soil substrate quality, soil moisture and temperature) would alter CO₂ emission and the δ¹³C value of CO₂ because of shifts in dominant plant species under flooding (Zhang et al., 2020a); 2) the pattern of CH₄ emission and its isotopic signature would be more sensitive to the flooding, reflecting the increased soil anaerobic environment (Zhao et al., 2020); and 3) the different flooding status (i.e., the saturated and drained states) would result in alterations in enzyme expression and microbial attributes, which would greatly affect CO₂ and CH₄ emissions (Shi et al., 2021).

2. Materials and methods

2.1. Site description

The research site was located at Zhongxian in Chongqing (30°42'N, 108°18'E), which is located in the riparian zone of the Three Gorges Reservoir area. The climate in this region is a southeast subtropical monsoon climate, with an annual mean temperature of approximately 16.5–19 °C and monthly average temperatures of 28–32 °C during

summer. The mean precipitation is 1100 mm, 80% of which falls in the hot-wet season (April to October). The soil is classified as purple soil in the Chinese soil classification, equivalent to Regosols in the FAO taxonomy and Entisol in the USDA taxonomy (Ye et al., 2017). The vegetation is dominated by *Cynodon dactylon*, *Setaria viridis*, *Echinochloa crusgalli*, *Digitaria sanguinalis* and *Xanthium sibiricum*.

2.2. Field experimental design and sample collection

This study was conducted in June–August 2017. Soil samples were taken when the reservoir's water level fell to 145 m, and the whole riparian zone was exposed to air after inundation. In this experiment, four transects (the water column, the low, the high and the upland area) were selected, and each transect was approximately 60 m × 15 m. The distance between two adjacent blocks was approximately 15 m. We collected samples from three plots along the transects, representing the following elevation range and flooding regimes: the water column, <147 m, permanent inundation area without plants, where is flooded the whole year on average; the low elevation, 145–160 m, frequently flooded area with revegetation; where is flooded 6–8 months per year on average, the high elevation 160–175 m, rarely flooded area with revegetation; where is flooded 3–4 months per year on average, and the upland area as the control, >175 m, nonflooded area with original plants, where has not been flooded since the establishment of the Three Gorges Reservoir (Fig. 1b). Three 1 m × 1 m subplots were selected randomly per elevation. Then, three cores (10 cm depth, 5 cm diameter) were sampled at random and homogenized within each subplot. A total of twelve soil samples were harvested to represent each elevation. A small subsample (approximately 50 g) was removed from each soil sample, immediately sieved through a 2-mm mesh, and stored at -80 °C for phospholipid fatty acid (PLFA) extraction. Another subsample was removed and stored at 4 °C for soil enzyme activities. The remaining soil samples were air-dried, stored in airtight plastic bags, and then returned to the laboratory for chemical and biochemical analyses.

2.3. Gas emission measurements and soil analyses

The exchange rates of CO₂ and CH₄ were measured at the soil/water-atmosphere interface via the floating and static chamber method in eleven 1-week campaigns from June 2017 to August 2017, respectively. The soil-atmosphere system consisted of two parts: a cylindrical bottom pedestal (diameter = 0.3 m, height = 0.2 m) inserted 0.1 m into the soil, installed one week before the first measurement (vegetation within the rings was clipped); and a removable opaque static chamber (diameter = 0.3 m, height = 0.3 m) equipped with a gasket to ensure airtightness during sampling that was removed afterwards. In addition, a silicone tube (0.5 cm inner diameter and 0.12 m long) and a battery-operated fan were installed through the top of the chamber to mix the air when the sample was collected. A floating chamber (diameter = 0.3 m, height = 0.3 m) was designed to collect gas samples at water surfaces. Plastic foam collars were fixed onto the bottom outside of the floating chamber to maintain the waterline at 10 cm from the chamber. Gas sampling was conducted between 9:00 and 11:00 a.m. The two chambers were maintained without leaking by a bulldog clip during the measuring period. Generally, when sampling began, 140-ml air samples were collected every 10 min (0, 10, 20 and 30 min) using 100-ml plastic syringes. The concentration and δ¹³C value of CO₂ and CH₄ were analyzed with a gas isotope analyzer (912-0003, LGR, America).

The CO₂ and CH₄ emissions were calculated using linear model regression analysis as follows:

$$F = dc/dt \times M/V_0 \times 273/(273 + T) \times V/A \quad (1)$$

where F is the CH₄ emission (μg·m⁻²·h⁻¹) or CO₂ emission (mg·m⁻²·h⁻¹), respectively. dc/dt is the rate of gas concentration change inside the chamber; M is the molecular weight of the analyzed gas; V₀ is the

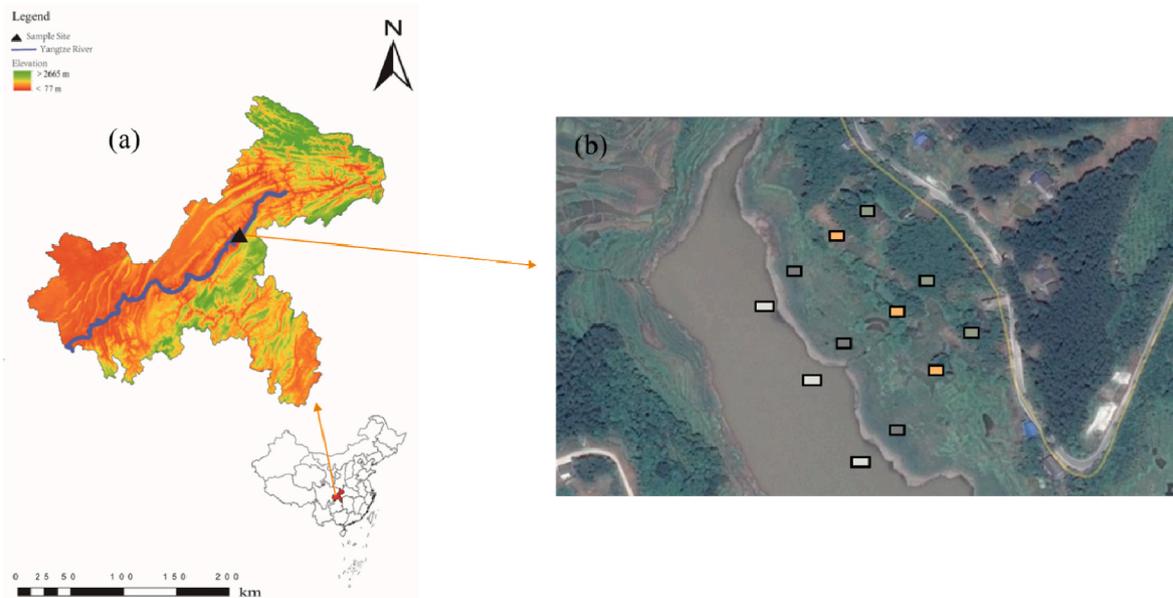


Fig. 1. Location of the study area in Chongqing Zhongxian; (a), satellite imagery of sampling sites and detailed view of the static flux chamber placement along the elevation gradient (b) in the riparian zone of the Three Gorges Reservoir region.

ideal gas constant at standard temperature and pressure (1 mol^{-1}); T is the air temperature inside the chamber at the sampling time; V is the chamber volume (m^3); and A is the chamber area (m^2). Almost all the coefficients of determination (r^2) of linear regression were greater than 0.9 ($P < 0.05$). Occasionally, data from individual chambers were excluded if the regression coefficients (r^2) were below 0.9 ($P > 0.05$).

2.4. Auxiliary measurements

The variations in daily precipitation and daily mean air temperature were obtained from a nearby weather station during the observation period and are presented in Fig. 1S. During gas sampling by the static chamber, the air temperature was measured with a mercurial thermometer. Simultaneously, soil temperature and moisture were measured outside each chamber with a portable instrument that measured soil temperature and moisture (SINTH8, SinoMeasure, China). Soil pH was measured in a water suspension at a ratio of 1:2.5. Zhang et al. (2020) described the details of the methods regarding soil organic C (SOC) contents, soil recalcitrant C (RC), and soil enzyme activities. Briefly, the SOC concentration was determined by a C/N analyzer. RC was obtained by two-step acid hydrolysis (Rovira and Ramón Vallejo, 2007). The enzyme activities of α -glucosidase (AG, EC 3.2.1.20); β -glucosidase (BG, EC 3.2.1.21); 1,4- β -cellobiosidase (CB, EC 3.2.1.91); 1,4- β -xylosidase (XS, EC 3.2.1.37); acid phosphatase (AP, EC 3.1.3.2); leucine aminopeptidase (LAP, EC 3.4.11.1); and β -1,4-N-acetylglucosaminidase (NAG, EC 3.1.6.1) were measured with fluorometric MUB-linked substrates (DeForest, 2009; German et al., 2011).

PLFAs were extracted using the method described by Bossio and Scow (1998) with taxonomic groups ascribed to individual PLFAs using the Sherlock PLFA Method and Tools Package. Briefly, lipids were extracted from 8 g of freeze-dried soils in a single-phase mixture of chloroform: methanol: phosphate buffer (1:2:0.8 by volume). Next, phospholipids were split into neutral, glyco- and phospholipids by silica acid column chromatography (SI column, Bond Elut, 500 mg, 3 ml; Varian; USA). After mild-alkali methanolysis, the extraction was transferred to a separatory funnel to separate overnight. Then, the extraction was analyzed with a gas chromatograph equipped with a flame ionization detector (Agilent 6890, Agilent Technologies, Palo Alto, CA, USA) and coupled to a mass spectrometer (Agilent MS 5975 C, USA).

Nonadecanoic acid methyl ester (19:0) was used as an internal standard for converting the peak area into nanomoles of each resultant fatty acid. The specific PLFA marker 18:1 ω 7c was used to quantify the relative abundance of methanotrophic bacteria (Smith et al., 2015).

2.5. Statistical analysis

All data analyses were performed using IBM SPSS Statistics 21.0 (Armonk, NY, USA). One-way analysis of variance (ANOVA) with Duncan's test was used to determine the statistical significance of elevation on parameters including plant traits (plant C, plant N, and plant C: N ratios), soil C pools (SOC, C:N ratio, LC, RC and RIC), soil properties (pH, SM and ST), enzyme activities (AG, BG, CB XS, AP, NAG and LAP), the specific PLFA marker 18:1 ω 7c, gas emissions (CH_4 and CO_2) and the isotope values ($\delta^{13}\text{C}$ -plant, $\delta^{13}\text{C}$ -soil, $\delta^{13}\text{C}$ - CO_2 and $\delta^{13}\text{C}$ - CH_4). The statistical significance of the redundancy analysis (RDA) was performed to test the relationship between the greenhouse gas emissions and environmental variables. The partial Mantel test was used to explore the relationship between the $\delta^{13}\text{C}$ of CO_2 and CH_4 and environmental variable indices. The RDA and Mantel test analysis were completed by the vegan package in R3.3.3.

3. Results

3.1. Spatial variation in plant, soil, microbe and climate parameters

The plant C: N ratios, the RC content and soil enzyme activities were significantly higher at low elevations compared with the upland area. However, the soil C: N ratios, LC content and the methanotrophic bacteria biomass showed the opposite trend (Tables 1S and 2S). During the whole study period, precipitation was significantly higher in the earlier saturated state than in the later drained state ($P < 0.05$, Fig. 1S), and air temperature differed between the two states ($P < 0.05$, Fig. 1S). The effect of rainfall increased soil temperature by $>10\%$ and decreased soil moisture by $>30\%$ under the saturated state compared to the drained state (Table 1). Soil moisture was significantly lower under the drained state, with the highest values at low elevations and the lowest level at high elevations (Fig. 2S; Table 1).

Table 1

Mean CO₂ emissions, net CH₄ emissions, δ¹³C values of CO₂, and CH₄, soil temperature and soil moisture during saturated and drained state in different elevations in the riparian zone of Three Gorges Reservoir region.

Elevations	Soil water State	CO ₂ emissions (mg·m ⁻² ·h ⁻¹)	CH ₄ emissions (ug·m ⁻² ·h ⁻¹)	δ ¹³ C-CO ₂	δ ¹³ C-CH ₄	ST (°C)	SM (%)
Low	Saturated	391.65 ± 43.74 A	44.65 ± 18.05 A	-18.95 ± 1.43 A	-40.14 ± 12.13 B	26.44 ± 0.24 B	39.38 ± 0.82 A
	Drained	292.63 ± 19.59C	0.58 ± 4.25 A	-15.7 ± 1.21 B	-37.24 ± 6.79 A	29.13 ± 0.35C	29.73 ± 1.08 A
High	Saturated	432.49 ± 32.21 A	21.91 ± 8.68 A	-18.88 ± 1.81 A	-43.59 ± 6.63 B	27.43 ± 0.23 B	35.94 ± 0.70 B
	Drained	461.03 ± 24.48 A	14.64 ± 8.50 A	-20.08 ± 1.65 A	-57.66 ± 4.57 B	30.69 ± 0.17 B	25.05 ± 1.52 B
Upland	Saturated	397.30 ± 27.33 A	-17.21 ± 5.3 B	-22.72 ± 2.33 A	-34.82 ± 2.76 A	29.71 ± 0.54 A	35.05 ± 0.67 B
	Drained	387.08 ± 24.76 B	5.79 ± 3.58 A	-23.33 ± 1.97 A	-71.86 ± 1.83C	32.17 ± 0.29 A	26.6 ± 1.68AB

Note: ST: soil temperature, SM: soil moisture, Values are Means ± SE. Different capital letters in the same column for each variable indicate statistically significant differences among different elevations in the same state at $P < 0.05$ based on the one-way ANOVA.

3.2. Temporal variation in CO₂ emissions and the δ¹³C value of soil-respired CO₂

Soil CO₂ emissions varied from 185.24 ± 6.41 to 561.40 ± 45.10 mg CO₂ m⁻² h⁻¹ during the whole period (Fig. 2a). Meanwhile, the average CO₂ emissions was significantly higher in high elevations than upland area ($P > 0.05$, Fig. 2b). The water column area below an elevation of 147 m acted as the weak CO₂ pool (22.15 ± 2.11), with no obvious temporal changes in CO₂ emissions being observed during the whole period (Fig. 2). The δ¹³C value of soil-respired CO₂ exhibited significant variation among elevations (Fig. 4a). In general, δ¹³C was most depleted in the upland area and enriched in the water column area ($P < 0.05$, Fig. 4a).

The CO₂ emissions and δ¹³C value of soil-respired CO₂ between the saturated and drained states varied with elevation (Fig. 5). The CO₂ emissions did not significantly change between the saturated and drained states in the high and upland areas (429 vs. 402 mg CO₂ m⁻² h⁻¹), but CO₂ emissions were lower under the drained state than under the saturated state at the low elevation (Table 1, Fig. 5a). The δ¹³C value of soil-respired CO₂ showed a similar pattern of CO₂ emissions, with a lower δ¹³C value of soil-respired CO₂ at low elevations during the drained state (Table 1, Fig. 6a).

3.3. Temporal variation in CH₄ emissions and the δ¹³C value of CH₄

Soil CH₄ emissions ranged from weekly 162.54 ± 32.94 and 290.50 ± 88.57 μg CH₄ m⁻² h⁻¹ and ended at approximately weekly 15.77 ± 4.31 and 50.26 ± 18.12 μg CH₄ m⁻² h⁻¹ at low and high elevations, respectively (Fig. 3a). However, CH₄ emissions changes dramatically in the middle of the whole period at low elevation due to the summer

flooding (Fig. 3a). Based on the annual average values, soil CH₄ emissions were all above zero in the riparian zone. With all elevations considered, the water column area exhibited a significantly higher mean CH₄ emission compared with other elevations (Fig. 3b). The δ¹³C of CH₄ with a signature of -38.11 ± 5.16‰ was more enriched in the high elevation than in the low and upland areas, with the δ¹³C of CH₄ between -53.21 ± 3.96‰ and -51.17 ± 4.57‰ (Fig. 4b).

During the saturated state, the net CH₄ emission rate was 44.64 ± 18.04 μg CH₄ m⁻² h⁻¹ at low elevations and 21.90 ± 8.67 μg CH₄ m⁻² h⁻¹ at high elevations, accounting for approximately 97% and 59.94% of the total net CH₄ emissions, respectively (Fig. 5b). In contrast, during the drained state, the net CH₄ emissions significantly ($P < 0.05$) decreased by 90% at low elevations and increased by 153% at high elevations compared with the upland area. The δ¹³C of CH₄ for all elevations was significantly more depleted under the saturated state than under the drained state except at low elevations. Regardless of the elevation, flooding had a significant effect on CH₄ emissions and δ¹³C values (Figs. 5b and 6b).

3.4. Controls on CO₂ and CH₄ emissions

The RDA showed that soil greenhouse gas emissions were significantly dependent on environmental factors (i.e., soil enzyme activities, C:N ratio, pH), accounting for 75.90% of the variation ($p = 0.002$, Fig. 7). For CO₂ emissions, the RDA revealed that the contribution of soil enzyme activities to CO₂ emissions was greater than that of other environmental factors (Fig. 7). CO₂ emissions were negatively correlated with C:N ($P < 0.05$). CH₄ emissions were negatively correlated with enzyme activities (e.g., BG, LAP and NAG) and maintained a positive correlation with methanotrophic bacteria ($P < 0.05$, Fig. 7). The

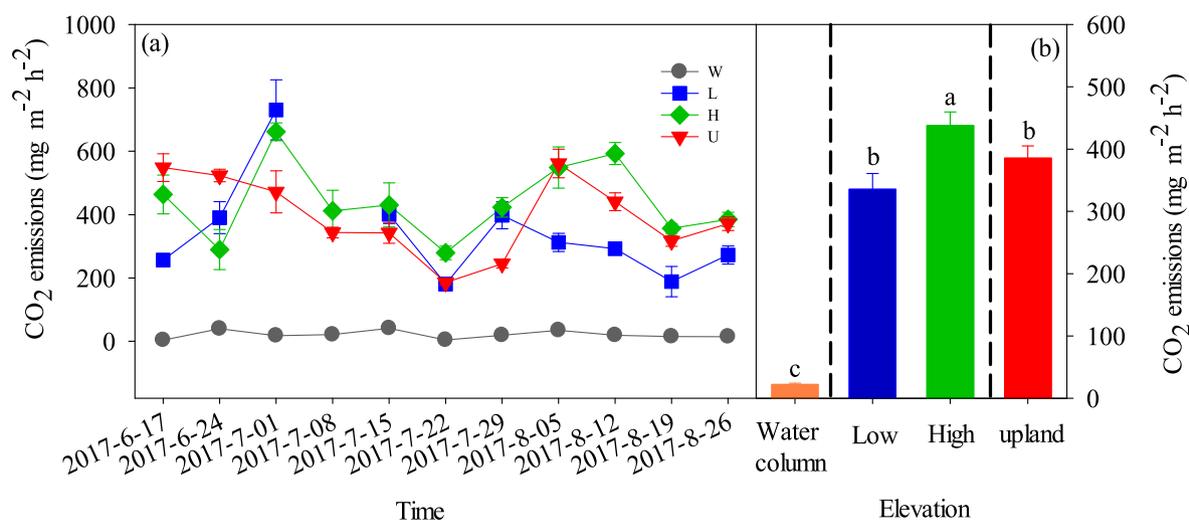


Fig. 2. Soil CO₂ emissions of the weekly average (a) and the whole no-flooding period (b) under different elevations (i.e., water column, low, high and upland) in the riparian zone. Error bars represent the standard errors of the mean values, and different lowercase letters over the bars indicate statistically significant differences at $P < 0.05$ among different elevations. Abbreviations: W, water column; L, low; H, high; A, upland.

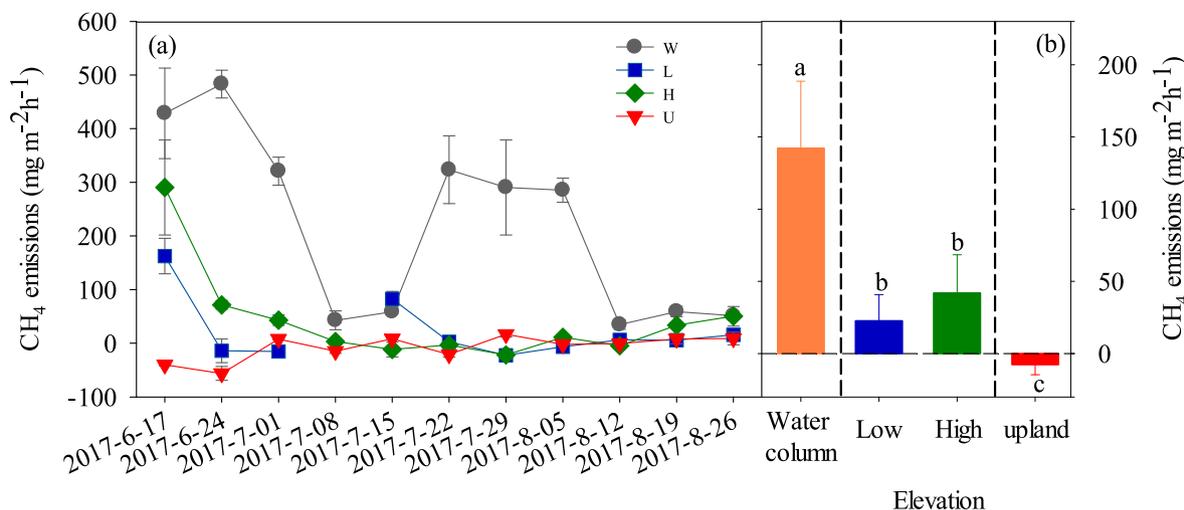


Fig. 3. CH₄ emissions of the weekly average (a) and the whole no-flooding period (b) under different elevations (i.e., water column, low, high and upland) in the riparian zone. Error bars represent the standard errors of the mean values, and different lowercase letters over the bars indicate statistically significant differences at $P < 0.05$ among different elevations. See Fig. 2 for the abbreviations.

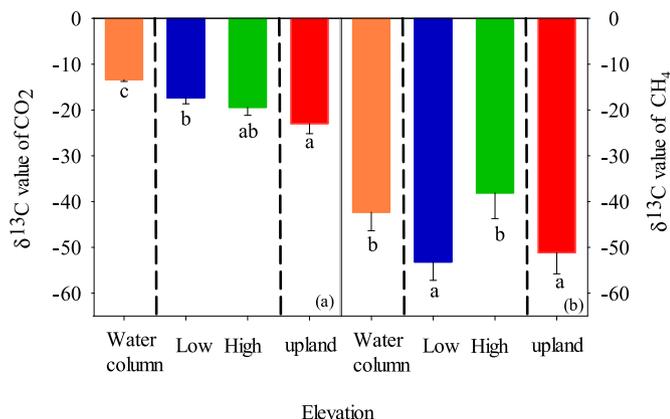


Fig. 4. The $\delta^{13}\text{C}$ of soil-respired CO₂ (a) and $\delta^{13}\text{C}$ of CH₄ (b) during the whole no-flooding period under different elevations (i.e., water column, low, high and upland) in the riparian zone. Error bars represent the standard errors of the mean values, and different lowercase letters over the bars indicate statistically significant differences at $P < 0.05$ among different elevations.

$\delta^{13}\text{C}$ of CO₂ showed a significant correlation with soil enzyme activities ($P < 0.05$, $r > 0.60$, Fig. 8), and the $\delta^{13}\text{C}$ of CH₄ was only correlated with Mb ($P < 0.05$, $r > 0.60$, Fig. 8) based on partial Mantel tests.

4. Discussion

Variations in soil CO₂ and CH₄ emissions and their carbon isotopic signatures estimated from the riparian zone were strongly affected by periodic flooding, which could determine the strength of the source/sink of CO₂ and CH₄ in the Three Gorges Reservoir. We found that soil CO₂ and CH₄ emissions were closely related to microbial attributes (soil enzyme activities and methanotrophic bacteria) and edaphic factors such as soil substrate quality (C:N ratio) and soil properties (i.e., soil moisture and temperature; Fig. 7). Thus, our results highlighted that dam-triggered periodic flooding resulted in alterations in enzyme expression and microbial strategies for using C (Keane et al., 2021) and methanotrophic processing (Prananto et al., 2020) and potentially changed CO₂ and CH₄ emissions and their carbon isotopic signatures.

Consistent with the first hypothesis, the relatively higher CO₂ emissions at riparian zone could be due to the lower substrates quality and higher soil enzyme activities (Tables 1S and 2S). Our previous study

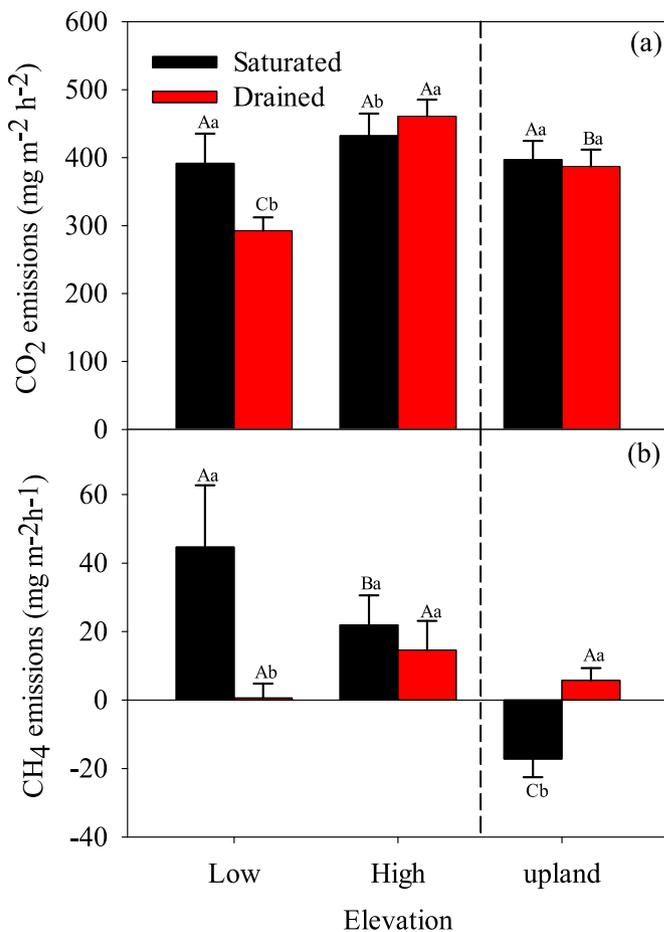


Fig. 5. Mean CO₂ emissions (a) and CH₄ emissions (b) in the soil water saturated and drained states at different elevations (i.e., low, high and upland). Error bars represent the standard errors of the mean values, and different capital letters over the bars indicate statistically significant differences at $P < 0.05$ among different elevations. Different lowercase letters over the bars indicate statistically significant differences at $P < 0.05$ between saturated and drained samples.

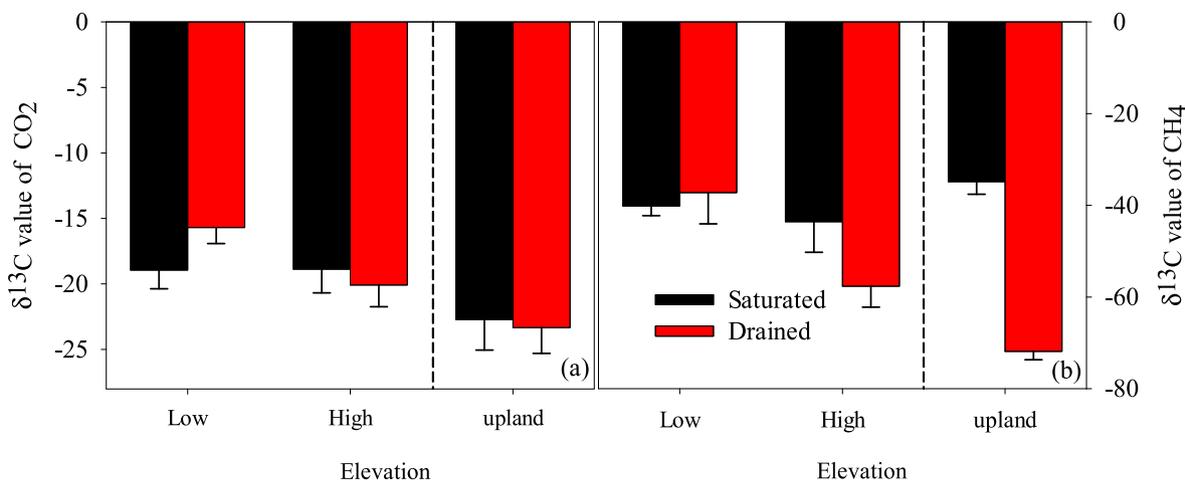


Fig. 6. Mean $\delta^{13}\text{C}$ of soil-respired CO_2 (a) and $\delta^{13}\text{C}$ of CH_4 (b) at soil water saturated and drained states under different elevations (i.e., low, high and upland). Error bars represent the standard errors of the mean values, and different capital letters over the bars indicate statistically significant differences at $P < 0.05$ among different elevations. Different lowercase letters over the bars indicate statistically significant differences at $P < 0.05$ between saturated and drained samples.

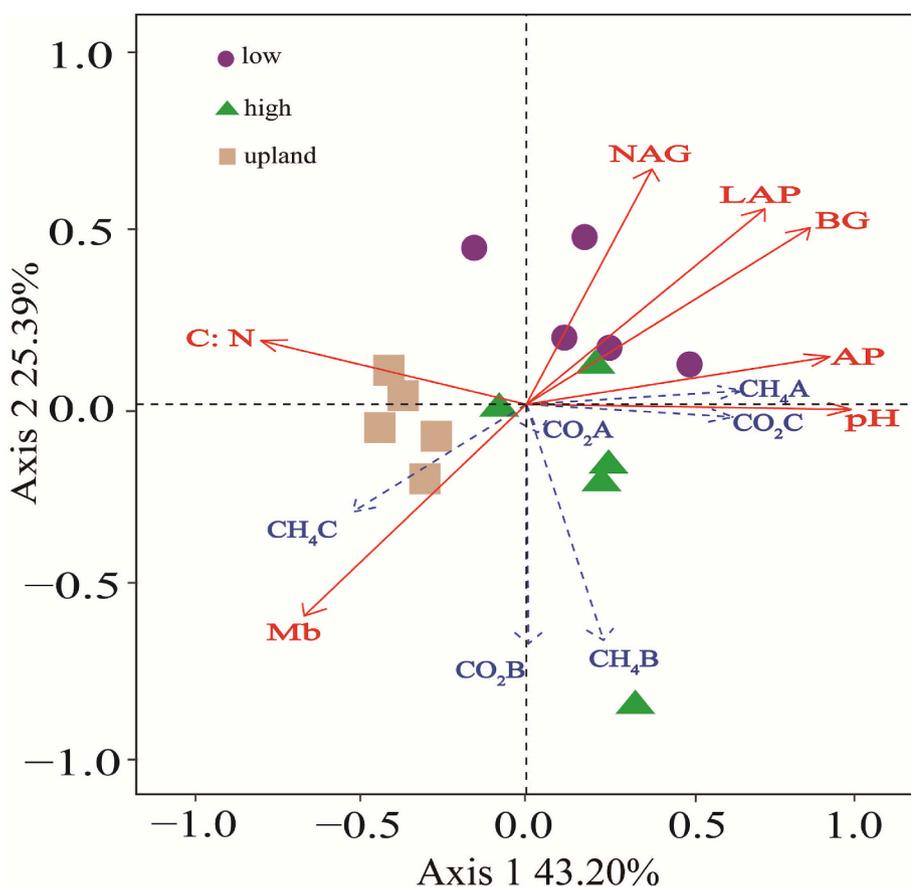


Fig. 7. Redundant analysis (RDA) of a two-dimensional sequencing diagram between greenhouse gases (CO_2 and CH_4) and soil physicochemical properties. The red solid lines represent different soil physicochemical indexes, and the purple dashed lines represent different states of greenhouse gas. Abbreviations: BG, β -glucosidase; LAP, leucine aminopeptidase; NAG, N-acetyl- β -glucosaminidase; AP, acid phosphatase; Mb, methanotrophic bacteria; CO_2A , soil CO_2 emission in the soil water saturated state; CO_2B , soil CO_2 emission in the soil water drained state; CO_2C , soil CO_2 emission during the whole drained state; CH_4A , CH_4 emission in the soil water saturated state; CH_4B , CH_4 emission in the soil water drained state; CH_4C , CH_4 emission during the whole drained state. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

in the same area showed that dam-triggered flooding strongly altered the functional plant groups (C3 and C4 plants), soil quality and micro-environments (Zhang et al., 2020a), which could potentially affect soil CO_2 emissions. Fresh litter of the C4 plant (*Cynodon dactylon*) at low elevations with a higher C:N ratio could aggravate the limitation of nutrients for microbial decomposition. Interestingly, we found that soil CO_2 emissions significantly differed between the saturated and drained states with different patterns at different elevations. In the early saturated state, the weekly CO_2 emissions were significantly higher at low elevations than at other elevations, possibly due to increased soil water

content (Figs. 2a and 1S), which could be beneficial to soil respiration decomposition by roots and microbes (Poblador et al., 2017). However, during the drained state, soil CO_2 emissions declined at low elevations, probably due to the decrease in root respiration after regular summer flooding in our study area (Fig. 5a).

The $\delta^{13}\text{C}$ values of soil-respired CO_2 were enriched at low elevations in the drained state, partly due to the change in the components of soil respiration (Fig. 6a). Root respiration is an especially important component of soil CO_2 emissions and can contribute 10–90% of the total CO_2 emissions across different ecosystems (Subke et al., 2006). The

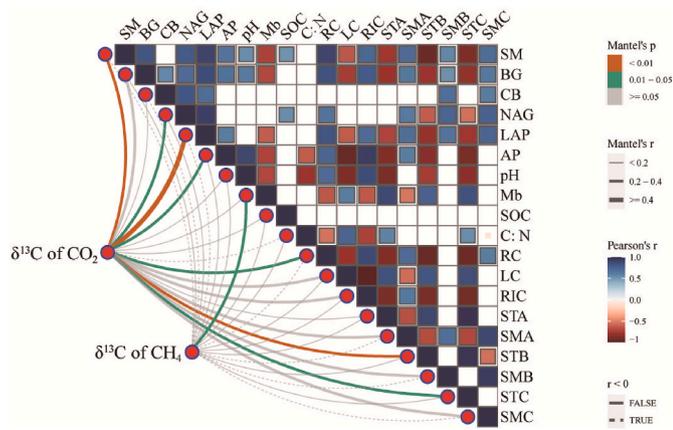


Fig. 8. Pairwise comparisons between $\delta^{13}\text{C}$ of CO_2 and CH_4 with soil physicochemical properties by partial Mantel tests. The color gradients denote Spearman's correlation coefficients; the line edge width corresponds to Mantel's r statistic for the corresponding distance correlations, and the edge color denotes the statistical significance based on 9999 permutations; the dotted line indicates that r is negative. Abbreviations: CB, cellobiohydrolase; SOC, soil organic carbon; LC, soil labile carbon; RC, soil recalcitrant carbon; RIC, RC/SOC; STA, mean soil temperature during summer flooding; STB, mean soil temperature after summer flooding; STC, mean soil temperature during the drained state; SMA, mean soil volumetric water content during summer flooding; SMB, mean soil volumetric water content after summer flooding; STC, mean soil volumetric water content during the drained state; SWC, soil water content. See Fig. 7 for the abbreviations. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

inundated plants at this elevation directly decreased root respiration, which could decrease soil CO_2 emissions. We also found that the $\delta^{13}\text{C}$ of CO_2 also showed a close relationship with soil enzyme activities (Fig. 8). Previous studies have found that the decomposition of SOC via soil enzymes can lead to isotopic fractionation and enrichment in soil ^{13}C (Peri et al., 2012). Soil-respired CO_2 from microbial respiration carries a $\delta^{13}\text{C}$ isotopic signature that could be very similar to the utilized substrate (Lorenz et al., 2020). Thus, the $\delta^{13}\text{C}$ signatures of soil-respired CO_2 further confirmed a consistent interpretation that the shift in substrate quality and enzyme activities across periodic flooding was the main contributor to CO_2 emissions. It is worth noting that the mean CO_2 emissions in our study area were generally higher than those in other riparian regions (Table 3S, Jacinthe et al., 2015; Pinto et al., 2020; Wang et al., 2020b). This substantially high CO_2 emission could be attributed to high microbial respiration rates associated with soil substrate quality, and this point was supported by the negative correlation of soil CO_2 emissions with the soil C: N ratio (Table 3S; Miao et al., 2013; Miller et al., 2019; Wang et al., 2020b).

The average CH_4 emissions were positive in the riparian zone of the reservoir wetland in our study area (Fig. 3). This result agreed well with the evidence that the periodic flooding soils were CH_4 sources compared with the upland area (Table 3S; Marani and Alvola, 2007; Jacinthe, 2015; Hao et al., 2019). It has been suggested that BG activity is well correlated with CH_4 production in wet-dry cycle soils (Morrissey et al., 2014; Wang et al., 2020a), and we did observe strong relationships between CH_4 production and soil enzyme activity (Fig. 7). One of the most important mechanisms is that these enzymes produce monomers and oligomers in anaerobic environments, with the end products of fermentation used as substrates for methanogenesis microorganisms (Neubauer et al., 2013), which in turn potentially contribute to higher CH_4 emission (Morrissey et al., 2014; Medvedeff et al., 2015).

Conversely, low CH_4 emissions with high soil enzyme activities and soil C content were observed at low elevations (Fig. 3). This phenomenon is probably attributed to complex relationships between nutrient and soil enzyme activities. On the one hand, enzyme activities could

accelerate soil organic matter decomposition rates, leading to substrate accumulation (Burns et al., 2013). On the other hand, a greater nutrient content could increase enzyme activities, forming a wider range of substrates for decomposers, not only for methanogenesis (Cenini et al., 2016) but also methanotrophs. Thus, our results could be attributed to the second explanation. In the low riparian zone, residual plants can provide greater sources of C or nutrients for soil microbes (Zhang et al., 2020b). However, the microenvironment or substrate type terminated the supply to methanogenic microbes and thus restrained the net CH_4 emission.

During the saturated state, the average CH_4 emissions decreased with increasing elevation because the lower elevation suffered a longer inundated period, which strengthened the soil anaerobic conditions and thus increased CH_4 emissions. Compared to the upper high elevation zone, the low elevation zone with a significantly higher CH_4 emission seemed to be the most important CH_4 source during the earlier water-level recession period. Interestingly, we found that soil CH_4 emissions greatly decreased from the saturated ($44.64 \mu\text{g CH}_4 \text{ m}^{-2} \text{ h}^{-1}$) to drained state ($0.5794 \mu\text{g CH}_4 \text{ m}^{-2} \text{ h}^{-1}$) at low elevations (Fig. 5b). With the water level dropping, the soil microenvironments of the riparian zone became similar across the elevation gradients (Yuan et al., 2012). During the drained state, CH_4 emissions were probably jointly affected by soil nutrient content and microbial activities under a constant water moisture state. Based on our isotope data, the $\delta^{13}\text{C}$ of CH_4 values in all riparian zones ranged from -65 to -50‰ and were characterized by acetate cleavage (Table 1, Fig. 4) (Whiticar et al., 1986; Vaughn et al., 2016). Acetate cleavage dominates methanogenesis when organic matter is abundant and surface emission is high. When the soil is submerged, an important reason for methane production is the anaerobic environment, but when soil moisture is relatively stable and low, soil nutrient limitation and acetate cleavage are critical limiting factors affecting methane production.

5. Conclusion

To conclude, variations in soil CO_2 and CH_4 emissions and their carbon isotopic signatures estimated from the riparian zone were strongly affected by periodic flooding, which could determine the strength of the source/sink of CO_2 and CH_4 in the Three Gorges Reservoir. Compared to upland area, the tolerable soil environments, higher enzyme activities as well as lower substrate quality at riparian zone maintained higher CO_2 emissions. The $\delta^{13}\text{C}$ signatures of soil-respired CO_2 further confirmed that the shift in substrate quality and enzyme activities was the main contributor to CO_2 emissions. The accumulative CH_4 emissions increased from low to high elevations in the riparian zone with CH_4 uptake in the upland area. Based on the $\delta^{13}\text{C}$ value of CH_4 , we tentatively concluded that the higher CH_4 emission during the saturated state was characterized by an acetate cleavage process in stronger anaerobic environments. Thus, our results highlighted that dam-triggered periodic flooding resulted in alterations in soil quality (i.e., C:N ratio), enzyme expression and microbial strategies for using C, and methanotrophic processing and potentially changed CO_2 and CH_4 emissions and their carbon isotopic signatures.

Author statement

Zhang Dandan: Conceptualization, Data curation, Writing – original draft, Formal analysis. Li Jinsheng: Formal analysis, Visualization. Wu Junjun: Visualization, Cheng Xiaoli: Supervision, Resources, Reviewing and Editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.envpol.2021.118599>.

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