Accumulation of Terrestrial Dissolved Organic Matter Potentially Enhances Dissolved Methane Levels in Eutrophic Lake Taihu, China

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ABSTRACT: Inland waters play an important role for the storage of chromophoric dissolved organic matter (CDOM) and outgassing of methane (CH4). However, to date, linkages between the optical dynamics of CDOM and dissolved CH4 levels remain largely unknown. We used multi-year (2012–2014) seasonal data series collected from Lake Taihu and 51 connecting channels to investigate how CDOM optical dynamics may impact dissolved CH4 levels in the lake. High dissolved CH4 in the northwestern inflowing river mouths coincided with high underwater UV–vis light availability, dissolved organic carbon (DOC), chemical oxygen demand (COD), DOM aromaticity, terrestrial humic-rich fluorescence, and in situ measured terrestrial CDOM, depleted dissolved oxygen (DO), stable isotopic δ2H, and δ18O compared with other lake regions. Our results further revealed positive relationships between dissolved CH4 and CDOM absorption at 350 nm, i.e. al(350), COD, DOC, terrestrial humic-rich fluorophores, and DOM aromaticity, and negative relationships between dissolved CH4 and DO, δ2H, and δ18O. The central lake samples showed a major contribution of terrestrial-sourced molecular formulas to the ultrahigh resolution mass spectrometry data, suggesting the presence of allochthonous DOM sources even here. We conclude that an elevated terrestrial CDOM input likely enhances dissolved CH4 levels in Lake Taihu.

INTRODUCTION

Inland waters (rivers, lakes, ponds, etc.) play an important role for the storage and outgassing of greenhouse gases.1,2 Lake ecosystems, especially the most common shallow lake types, are hot spots of organic carbon transformation and methane (CH4) emission.3 Globally, lakes cover 3.7% of the Earth’s ice-free land surface area and contribute approximately 20% of the CH4 outgassing from all natural ecosystems.4 Eutrophication and climate warming have together fueled the outgassing of CH4 from lake ecosystems.3,5 The net outgassing of CH4 from lakes to the atmosphere is the difference between methanogenesis and oxidation of CH4.6,7 Anaerobic fermentation of acetate by certain archaea (acetoclastic methanogenesis) in carbon-rich environments serving as the terminal step in the decomposition of dissolved organic matter (DOM) has traditionally been considered as the primary pathway of biological CH4 production.6,7 However, recent studies have highlighted that a large fraction of CH4 oversaturation in aquatic environments can be produced in oxygenated surface waters.8,9 The paradoxical supersaturation of CH4 in oxygenated surface waters has been suggested to be a result of microbial transformation of DOM.8,9 Recent studies have also suggested that photochemical degradation may be more
important than microbial degradation in controlling the decomposition of newly exposed terrestrial DOM, further stimulating the outgassing of greenhouse gases in Arctic waters. Irradiation experiments revealed significant photochemical CH$_4$ production in the anoxic water column, especially in the inflowing river mouths in this case.

DOM plays a vital role in biogeochemical processes in inland waters and the composition of lake DOM is highly variable, depending on its source, biological reactivity, and time of processing. Terrestrial humic-rich substances usually contribute primarily to the DOM pool in lakes in the monsoon region, while autochthonous inputs from algae and macrophytes can also be important substrates for microbial degradation and are important sources of carbon in lake ecosystems. The mixing of terrestrial and autochthonous DOM can be utilized and transformed by microbial and photochemical degradation and are important sources of carbon in lake ecosystems.

The role of DOM in controlling CH$_4$ emissions from an augmented terrestrial DOM input has, however, lowered bacterial and archaea oxidation of CH$_4$ (aerobic and anaerobic, respectively) to CO$_2$, a major pathway of biological CH$_4$ oxidation, consequently reducing the levels of dissolved CH$_4$. To date, however, the role of the composition of DOM for the levels of dissolved CH$_4$ in lake ecosystems remains to be elucidated.

Lake Taihu is well-suited for the undertaking of studies to reveal the linkage between CDOM optical dynamics and dissolved CH$_4$ levels as it has high inputs of both allochthonous and autochthonous DOM. The Lake Taihu watershed is located in the Yangtze River Delta, the most urbanized area in China with a high population density and extensive economic development. Excessive input of nutrients has led to the occurrence and persistence of nuisance blooms of cyanobacteria and high heterogeneity of DOM quality and dissolved CH$_4$ concentrations in the lake. The direction of the water flow of the channels connecting Lake Taihu with the fluvial plain is primarily controlled by meteorological and anthropogenic disturbances (e.g., floodgates). However, to date, the relative importance of allochthonous and autochthonous CDOM, regulated by the inflow and backflow discharge ($Q_{\text{net}}$, net inflow discharge), for the dissolved CH$_4$ levels has not been assessed.

We studied how the optical compositional dynamics of CDOM mediated by net inflow discharge quantitatively drives the levels of dissolved CH$_4$. A total of 700 CDOM samples and 552 headspace CH$_4$ samples were collected in Lake Taihu and the connecting channels in the watershed from February 2012 to November 2014. We expected to find large variations in dissolved CH$_4$ levels among lake regions and hydrological seasons due to different net inflow of terrestrial CDOM.

**MATERIALS AND METHODS**

**Study Sites.** Lake Taihu has an area of 2338.1 km$^2$ and a water retention time of $\sim$309 days, and the lake watershed covers 36 500 km$^2$. A total of 172 channels are connected with the lake, and the elevation of the water surface of the connecting channels is mostly lower than 5 m. The water exchange between the lake and the connecting channels is complex and similar to that of lagoons. This leads to large
variations in the concentration and composition of DOM in different areas of the lake. Information about hydraulic subwatersheds and hydrological data collection, multiyear variations of daily δ²H and δ¹⁸O (Figure S2), and monthly Qnet (inflow – backflow runoff) in each hydraulic subwatershed (Figure S3; Table S1) can be found in the Supporting Information.

Light Availability, Secchi Disk Depth (SDD), Dissolved Oxygen (DO), and CDOM Optical Sampling Collection. Water depth (WD) was measured in situ using a scaled straight stick, while Secchi disk depth (SDD) was determined in situ using a 30 cm diameter black and white Secchi disk. The dissolved oxygen (DO) concentration was determined in situ at 0.5 m depth using a YSI 6600 V2 multisensor sonde. WD, SDD, and DO data were only available from the seasonal campaign conducted in Lake Taihu from February 2012 to November 2014 (Table S2).

CDOM absorption and fluorescence measurements were conducted to trace the sources and optical properties of CDOM and to investigate the impacts of CDOM optical compositional dynamics on the levels of dissolved CH₄. Detailed information about the seasonal CDOM sampling collection in the lake and the connecting channels from 2012 to 2014 can be found in the Supporting Information (Table S2).

Previous studies have revealed that relatively high concentrations of dissolved organic carbon (DOC) are exported to Lake Taihu from the northwestern subwatershed via Zhushan Bay. An extensive sampling campaign (n = 61) was therefore carried out in Zhushan Bay, located in northwestern Lake Taihu, in April 2014 with the aim to trace the sources of CDOM and assess light availability (Figure 1). Detailed information about the measurement of CDOM fluorescence using an in situ CDOM fluorescence sensor, light availability indices including the diffuse attenuation coefficient, Kd, of light in the UV region and the photosynthetically active radiation (PAR, 400–700 nm), as well as measurements of total suspended matter (TSM) and inorganic suspended matter (ISM), can be found in the Supporting Information (Table S2).

Stable Isotopic δ²H and δ¹⁸O Sampling Collection and Measurements. Stable isotopic composition of surface water provides information on hydrological and meteorological processes as evaporation fuels the accumulation of the heavier isotopic water molecules (H²¹⁸O and H²¹⁹O) in the liquid water, and these can therefore be used to trace the sources of riverine CDOM and evaporation in Lake Taihu. We assumed that the source of riverine CDOM in Lake Taihu is consistent with the depletion of δ²H and δ¹⁸O. Detailed information about the stable isotopic δ²H and δ¹⁸O sampling collection in the lake and the connecting channels can be found in the Supporting Information (Table S2).

Chlorophyll a (Chl-a), Chemical Oxygen Demand (COD), DO, and CDOM Optical Measurements. Previous studies have indicated that the degradation of algal mats can impact the levels of dissolved CH₄. In this study, Chl-a was used as a surrogate for algal biomass, and the linkage between Chl-a and dissolved CH₄ concentrations in Lake Taihu was investigated. DOC, COD, and optical measurements of CDOM were used to trace the sources and optical composition of CDOM (for information about Chl-a, DOC, COD, and CDOM optical measurements, see the Supporting Information, Table S2).

CDOM absorption indices included the absorption coefficient at 350 nm, a(350), the ratio of a(250)/a(365), the spectral slope (S₈₋₉), and the slope ratio (S₉), which can be used to trace the amount of CDOM and the relative molecular size and aromaticity of CDOM. Detailed information about the measurement and calculation of CDOM absorption indices is given in the Supporting Information (Table S2).

CDOM fluorescence (excitation–emission matrixes, EEMs) measurements and the calibration processes including corrections of Raman and Rayleigh peaks, inner-filter effects, and normalization of Raman units (RU) can be found in the Supporting Information. PARAFAC is a three-way multivariable data analysis technique that statistically decomposes the complex mixture of CDOM fluorophores into trilinear components with a single emission maximum and corresponding to a group of highly covarying analytes. Using PARAFAC modeling, a six-component model was well validated (Figure 2; Figures S4 and S5) (see details about the modeling and validation in the Supporting Information, Table S2).

Dissolved CH₄ Sampling Collection and Measurements. In total, dissolved CH₄ concentrations were determined in 552 headspace samples (29 lake sites × 4 seasons × 3 years and 51 river sites × 4 seasons) (Figure 1; Table S2). Direct measurement of dissolved CH₄ was performed by headspace equilibration following the method detailed in Xiao et al. (see the Supporting Information, Table S2). We determined only the levels of dissolved CH₄ but not...
CH₄ emission as wind speed and flow velocity data for the river samples were not available. Although the relationship between the levels of dissolved CH₄ and CH₄ emissions is not 1:1, in this study we used dissolved CH₄ concentrations as a proxy for the potential CH₄ emission flux in different areas of Lake Taihu since the levels of dissolved CH₄ largely control the emission from the lake²¹ (Figure S1).

Fourier Transform Ion Cyclotron Resonance Mass Spectrometry (FT-ICR MS) Analyses. Details on the pretreatment of the FT-ICR MS samples can be found in the Supporting Information. Briefly, in November 2016, FT-ICR MS samples collected from central Lake Taihu and headwater streams of the lake basin as well as algal-derived samples to determine degradation were solid-phase-extracted with PPL Bond Elut (Agilent) resins before analysis by negative-ion electrospray FT-ICR MS³³,³⁴ to trace the sources of DOM in the lake. Different sources of CDOM exhibit distinct molecular signatures,³⁵ and FT-ICR MS can be a useful tool in tracing the sources of CDOM in Lake Taihu. The assigned molecular formulas were categorized according to Ohno et al.³⁶ Briefly, the chemical classes classified by the van Krevelen diagrams include the following: (i) lipids (O/C = 0−0.3, H/C = 1.5−2.0), (ii) proteins and amino sugars (O/C = 0.3−0.67, H/C = 1.5−2.2), (iii) lignins (O/C = 0.1−0.67, H/C = 0.7−1.5), (iv) carbohydrates (O/C = 0.67−1.2, H/C = 1.5−2.2), (v) unsaturated hydrocarbons (O/C = 0−0.1, H/C = 0.7−1.5), (vi) condensed aromatics (O/C = 0.2−0.7), and (vii) tannins (O/C = 0.67−1.2, H/C = 0.5−1.5).³⁵

Statistical Analyses. Statistical analyses, including mean values, standard deviations (SD), and t-tests, were performed applying R-studio 0.97.551 (R i386 2.15.2) software. Linear regressions and van Krevelen diagrams were made using MATLAB R2016b. Results with p < 0.05 for t-tests and linear regressions were reported as significant. Means were shown ±SD.

■ RESULTS

PARAFAC Modeling Results. Fluorescent DOM (FDOM) is composed of humic substances and amino acids, both free and bound in proteins, and alterations in PARAFAC-derived FDOM components are sometimes used as a surrogate for changes in the composition of the wider DOM pool.³² The six-component PARAFAC model explains >99.9% of the variables of the whole EEMs data set. The spectral characteristics of the six components (C1−C6, Figure 2; Figure S4) were compared with PARAFAC results for samples collected from other aquatic ecosystems using an online spectral library called Openflouor.³⁷ C1 had two Ex maxima at ≤230 and 285 nm and an Em maximum (340 nm) as have amino acid
Figure 4. Spatial variations of dissolved organic carbon (a), Fmax of terrestrial humic-like C2 (b), terrestrial CDOM concentrations measured using an in situ fluorescence sensor (c), and diffuse attenuation coefficient of PAR (Kd(PAR) (d)) in Zhushan Bay, northwestern Lake Taihu, in April 2014. Examples of van Krevelen diagrams based on the distribution of headwater DOM (e), algal-derived DOM (f), and central Lake Taihu DOM (g) samples. Dashed boxes in panels e–g tentatively assigned as protein and lignin molecules are marked. Relationship between Kd(PAR) and terrestrial CDOM concentrations measured using fluorescence sensor (h). The colored dots in the lower three panels represent the percent relative abundance of the FT-ICR MS signals. Relationship between Kd(PAR) and in situ measured CDOM concentrations (h).

associated tryptophan-like components.38,39 C2 displayed two Ex maxima at 240 and 350 nm and an Em maximum (468 nm) and is categorized as a typical terrestrial humic-like substance.38,40,42 C3 exhibited Ex/Em maxima at ≤230/420 nm, similar to those of agricultural-soil-derived humic-like or fulvic-like materials.43 C4 peaked at 380 nm and is categorized as a microbial humic-like substance.38,44,45 C5 (Ex/Em ≤230 (275)/316 nm) and C6 (Ex/Em ≤230 (270)/≤300 nm) had spectral characteristics similar to those of redshifted tyrosine and typical tyrosine substances, respectively.38,46 The multiyear (2012–2014) mean Fmax ranged from 0.24 ± 0.09 RU (microbial humic-like C4) to 5.04 ± 2.04 RU (tryptophan-like C1) for the PARAFAC-derived C1–C6 in Lake Taihu with the relative contribution percentages for the six components ranging from 2.2 ± 0.8% (C4) to 46.9 ± 19.0% (C1) (Table S3).

Spatial Variability of Light Availability, Stable Isotopic δ2H and δ18O, CDOM-Related Indices, COD, DO, Dissolved CH4, and Chl-a. SDD in the lake ranged from 0.0 to 1.7 m with a mean of 0.4 ± 0.3 m, high values being observed in the northern and northwestern bays for the campaigns conducted seasonally from February 2012 to November 2014 (Figure S6). The ratio of SDD to WD ranged from 0.06 to 0.61 with a mean of 0.19 ± 0.12 and, similarly, high values of SDD to WD were recorded in the northern and northwestern bays of the lake (Figure S6).

As evaporation enhances the accumulation of heavier water isotopic molecules of δ2H and δ18O in the liquid water, more depleted δ2H and δ18O imply a strong riverine signal, while enriched δ2H and δ18O indicate evaporation induced by prolonged water retention time in the lake.27 For all samples collected from 2012 to 2014, δ2H and δ18O ranged from −63.4 to −16.5‰ (−33.9 ± 8.2‰) and from −8.9 to 0.4‰ (−4.6 ± 1.3‰), respectively. Multiyear mean δ2H and δ18O increased notably from the northwestern bays to the southeastern bays in all seasons (Figure 3; Figure S7). δ2H and δ18O in the samples collected from the inflowing rivers, i.e., in the northwestern subwatersheds (e.g., Huxi and Wuchengxiyu) were generally lower than in the corresponding bays and coastal lake regions.

In all samples collected from 2012 to 2014, DOC and a(350) ranged from 1.0 to 17.2 mg L−1 (4.4 ± 1.3 mg L−1) and from 0.4 to 16.0 m−1 (4.0 ± 1.6 m−1), respectively. Multi-year (February 2012 to November 2014) mean concentrations of DOC and a(350) and the Fmax of C1–C4 and C6 decreased markedly from the northwestern part of the lake, especially Zhushan Bay, to the southeastern lake regions (Figure 3; Figure S7). In comparison, a(250)/a(365), S295−235, and SR increased notably from the northwestern part of the lake to the southeastern bays (Figure S7). COD concentrations in the samples collected from the lake from 2012 to 2014 ranged from 2.43 to 15.56 mg L−1 with a mean of 4.27 ± 1.28 mg L−1, and COD decreased gradually from the northwestern regions to the southeastern bays in all seasons (Figure S6). DO concentrations in the samples collected from the lake ranged from 2.47 to 12.28 mg L−1 with a mean of 8.86 ± 1.69 mg L−1, and depleted DO was observed in the northwestern inflowing river mouths (Figure S6).

Zhushan Bay located in northeastern Lake Taihu has relatively high concentrations of terrestrial, agricultural, and microbial humic-like fluorophores as well as depleted DO (Figure 3; Figure S6), and the sources of CDOM in the bay were investigated based on more comprehensive samplings.25 The exclusively terrestrial humic-like C2 decreased from the northwestern Zhusuan Bay and the northeastern coastal line to the southeastern bays in all seasons from 2012 to 2014 (Figures 3 and 4), coinciding with the higher inflow runoff from the tributaries in the Huxi and Wuchengxiyu subwatersheds than in the remaining subwatersheds. During the Zhushan Bay sampling campaign in April 2014, DOC ranged from 3.7 to 7.1 mg L−1 with a mean of 5.3 ± 0.8 mg L−1.
In comparison, $F_{\text{max}}$ of terrestrial humic-like C2 and in situ measured fluorescence intensity (370/460 nm) ranged from 0.1 to 2.7 RU (0.8 ± 0.6 RU) and from 21.5 to 200.0 μg L$^{-1}$ (90.8 ± 54.2 μg L$^{-1}$), respectively (Figure 4). DOC concentrations, $F_{\text{max}}$ of terrestrial humic-like C2, and measured in situ fluorescence intensity (370/460 nm) all decreased notably from the northwestern inner Zhushan Bay toward the south entrance of the bay (Figure 4). The sampling in Zhushan Bay therefore indicated that high concentrations of terrestrial humic-like and anthropogenic tryptophan-like fluorophores were discharged into the lake via the bay, potentially enhancing the levels of dissolved CH$_4$, $K_d$ of light in the UV region and the PAR regions was fairly low, corresponding to a relatively low TSM and high SDD at the inflowing river mouth compared with the open water region (Figure 4; Figures S6 and S8). Correspondingly, a significant negative relationship was found between the $K_d$ of light in the UV region and the PAR regions and SDD ($r^2 = 0.89$, $p < 0.001$) and with in situ measured fluorescence intensity (370/460 nm) ($r^2 = 0.32$, $p < 0.001$) (Figure 4; Figure S8).

Dissolved CH$_4$ samples collected from the macrophyte-dominated lake regions (Figure 1) were excluded from our analyses since macrophytes influence the dynamics of the greenhouse gas exchange. Submerged macrophytes affect biological and chemical processes in shallow lakes, including the biomass of organisms across trophic levels, and can therefore affect the dynamics of greenhouse gas. Submerged macrophytes fuel the transport of dissolved and ebbullitive CH$_4$ from the sediment to the water column and can thus increase dissolved CH$_4$ concentrations in the water. This is supported by a recent study showing that organic sediments in macrophyte-dominated lake areas strongly enhanced the levels of dissolved CH$_4$. In this study, the boundaries of macrophyte-dominated lake regions were delineated following the results detailed in Dong et al. and Liu et al. (Figure 1). The field investigation conducted by Dong et al. showed that East Lake Taihu Bay, Xukou Bay, and Guangfu Bay in the southeastern part of the lake were macrophyte-dominated, which was well validated by the remote sensing results obtained by Liu et al. Dissolved CH$_4$ in all the remaining lake samples collected from 2012 to 2014 ranged from 4.2 to 618.3 nmol L$^{-1}$, with a mean of 64.9 ± 92.3 nmol L$^{-1}$ (Figure 3). In the nonmacrophyte-dominated lake regions, dissolved CH$_4$ decreased notably from the northwestern bays or the western coastal regions to the southeastern lake regions in all hydrological seasons from 2012 to 2014 (Figure 3). Dissolved CH$_4$ in the river samples ranged from 7.9 to 1597 nmol L$^{-1}$, with a mean of 307.8 ± 347.5 nmol L$^{-1}$ (Figure 3), and was significantly higher than in the lake samples from the Huxi, Wuchengxiyu, Zhexi, and Hangjiahu subwatersheds in all seasons ($t$-test, $p < 0.05$; Figure 3; Table S4).

As the temporal resolution of the sampling campaign was seasonal, we found highly depleted δ$^2$H and δ$^{18}$O, and enhanced input of terrestrial C2, and thereby high concentrations of dissolved CH$_4$ in the northwestern lake regions in November compared with August due to drought on hot August days in the Lake Taihu watershed (Figure 3). An increase in the temporal resolution of the sampling campaign can probably capture the rainy season enhancement of dissolved CH$_4$ while in the present results the seasonal enrichment signal was surpassed by the spatial signal.

Significantly higher Chl-a concentrations were observed in August (37.8 ± 27.6 μg L$^{-1}$) and November (21.0 ± 18.1 μg L$^{-1}$) than in February (9.4 ± 3.8 μg L$^{-1}$; $t$-test, $p < 0.005$) and May (12.4 ± 7.1 μg L$^{-1}$; $t$-test, $p < 0.05$). Chl-a decreased from the northern regions of the lake to the southeastern bays in all seasons except in February from 2012 to 2014 (Figure 3), coinciding with higher external loading from the northwestern subwatersheds, for instance Huxi and Wuchengxiyu. Chl-a in the river samples ranged from 0.5 to 770 μg L$^{-1}$, with a mean of 42.9 ± 74.3 μg L$^{-1}$, and showed no significant difference from the mean Chl-a in the lake samples.

Relationships between Dissolved CH$_4$ and Stable Isotopic, CDOM-Related Indices, Chl-a, and Hydrological Indices. Significant positive relationships were found between mean dissolved CH$_4$ and mean $a(350)$ ($r^2 = 0.11$, $p < 0.001$), DOC ($r^2 = 0.36$, $p < 0.001$), $F_{\text{max}}$ of PARAFAC-derived C1–C3 ($r^2 = 0.07$–0.50, $p < 0.01$), COD ($r^2 = 0.43$, $p < 0.001$), and monthly net inflow runoff ($Q_{\text{net}}$) ($p < 0.001$) (Table SS) for all the samples collected in the lake watershed. It should be noted that the coefficients of determination between mean dissolved CH$_4$ and mean terrestrial PARAFAC-derived C2 and the products of C2 and $Q_{\text{net}}$, i.e., $Q_{\text{net}} \times C2$, can be as strong as 0.50 and 0.27, respectively (Table SS).

In comparison, significant negative relationships were found between dissolved CH$_4$ and $a(250)/a(365)$ ($r^2 = 0.14$, $p < 0.001$), $S_{275–295}$ ($r^2 = 0.31$, $p < 0.001$), $S_R$ ($r^2 = 0.28$, $p < 0.001$), stable isotopic δ$^2$H ($r^2 = 0.25$, $p < 0.001$) and δ$^{18}$O ($r^2 = 0.10$, $p < 0.001$), and DO ($r^2 = 0.11$, $p < 0.001$) for all the samples collected (Table SS; Figure S9). No significant relationship emerged between dissolved CH$_4$ and Chl-a or with C4–C6 if all data were used during linear fitting, but exclusion of river samples resulted in significant positive relationships between dissolved CH$_4$ and Chl-a ($r^2 = 0.18$, $p < 0.001$; Figure S9), C4 ($r^2 = 0.10$, $p < 0.001$), and C6 ($r^2 = 0.08$, $p < 0.01$). A close relationship was found between dissolved CH$_4$ and Chl-a if we include only August samples ($r^2 = 0.47$, $p < 0.001$; Figure S10).

Significant positive relationships appeared between mean DOC and mean $a(350)$ ($r^2 = 0.55$, $p < 0.001$), PARAFAC-derived C2–C4 ($r^2 = 0.10$–0.62, $p < 0.001$), COD ($r^2 = 0.69$, $p < 0.001$), net inflow runoff ($Q_{\text{net}}$) ($r^2 = 0.43$, $p < 0.001$), and the product of C2 and $Q_{\text{net}}$, i.e., $Q_{\text{net}} \times C2$ ($r^2 = 0.56$, $p < 0.001$) in all the samples collected in the lake and the connecting channels (Table SS). Additionally, the coefficient of determination of linear fitting between mean terrestrial C2 and mean DOC can be as high as 0.62 (Table SS). No significant relationship was found between DOC and Chl-a, C1, CS, and C6 (Table SS). In comparison, significant negative relationships were observed between mean DOC and mean $a(250)/a(365)$ ($r^2 = 0.30$, $p < 0.001$), $S_{275–295}$ ($r^2 = 0.28$, $p < 0.001$), $S_R$ ($r^2 = 0.26$, $p < 0.001$), stable isotopic δ$^2$H ($r^2 = 0.30$, $p < 0.001$) and δ$^{18}$O ($r^2 = 0.20$, $p < 0.05$), and DO ($r^2 = 0.15$, $p < 0.001$) in all the samples collected (Table S5). When excluding river samples, we further discovered significant positive relationships between terrestrial humic-like C2 and COD ($r^2 = 0.42$, $p < 0.001$) and between C2 and Chl-a ($r^2 = 0.18$, $p < 0.001$) and a negative relationship between C2 and DO ($r^2 = 0.15$, $p < 0.001$) (Figure S9).

FT-ICR MS Results. Lignin molecules exhibited 3331, 1802, and 3245 assigned formulas, corresponding to 77, 50, and 68% of the total assigned formulas for the samples collected from the headwater of the Lake Taihu watershed, algal-derived samples, and the central lake, respectively, in November 2016 (Figure 4; Table S6). In comparison, protein-like assigned formulas displayed 214, 897, and 691 peaks,
corresponding to 5, 25, and 16%, for the headwater, algal-derived, and central lake samples, respectively (Figure 4; Table S6). It is apparent that terrestrial humic-rich lignin molecular formulas have a higher relative abundance than other compound classes assigned to the headwater and the central lake samples (Figure 4; Table S6). In comparison, biological protein-rich molecular formulas have a greater relative abundance than formulas assigned to other compounds for the algal-derived samples (Figure 4; Table S6).

**DISCUSSION**

Our results indicated that the terrestrial CDOM input likely overrode algal blooms in enhancing dissolved CH$_4$ levels in large, eutrophic Lake Taihu. First, we found dissolved CH$_4$ to be highest in the northwestern regions, especially the inflowing river mouths, coinciding with high levels of DOC, terrestrial humic-rich C$_2$–C$_4$, in situ measured terrestrial humic-like fluorophores, and Chl-$a$ (May, August, and November) compared with the southeastern region of the lake (Figures 3 and 4; Figure S7). The significantly higher mean concentrations of dissolved CH$_4$ in the river samples than in lake samples in the western and northern subwatersheds (Huxi, Wuchengxiyu, Zhexi, and Hangjiahu) in all seasons (Figure 3; Table S4) likely provide further evidence of this. Although upwelling groundwater may enhance the advection of CH$_4$ to the inflowing channels, it contributes only a little to the water budget in the lake watershed. The discrepancy between the spatial distribution of high values of riverine and lacustrine dissolved CH$_4$, especially in the southwestern river mouths (Figure 3), suggested that the dynamics of dissolved CH$_4$ in the lake itself might not simply reflect the river flow. Moreover, the highly depleted $\delta^{2}H$ and $\delta^{18}O$ and the low $a(250)/a(365)$, $S_{275-295}$, and $S_R$ in the northwestern river mouths (Figure 3; Figure S7) supported the occurrence of a higher terrestrial contribution here. This is substantiated by $a(250)/a(365)$, $S_{275-295}$, and $S_R$, which were all negatively related to DOM aromaticity. Inflow water discharge usually has depleted $\delta^{2}H$ and $\delta^{18}O$, while enhanced evaporation resulting from a prolonged water retention time in the lake will enrich these isotopes. Second, we found significantly positive relationships between dissolved CH$_4$ versus $a(350)$, DOC, COD, $F_{\text{max}}$ of C$_1$–C$_3$, DOM aromaticity, and net inflow runoff ($Q_{\text{net}}$), as well as $Q_{\text{net}}\times C_2$ for all the samples collected (Figure 5; Table S5). $F_{\text{max}}$ of C$_2$ merely denotes the fluorescence intensity of terrestrial humic-like fluorophores, while $Q_{\text{net}}\times C_2$ serves as a surrogate of the loading of terrestrial humic-rich substances. Furthermore, the dissolved CH$_4$ was negatively related to DO and $\delta^{2}H$ and $\delta^{18}O$ (Figure 5; Table S5).

Microbial and photochemical degradation of the terrestrial humic-like C$_2$ and agricultural humic-like C$_3$ in the northwestern inflowing river mouths and shorelines of the lake likely enhanced the DO depletion observed in these lake regions (Figures 3 and 4; Figure S6), while the relatively high SDD and the low TSM and $K_d$ of underwater light in the UV and PAR ($K_d$(PAR)) regions in the DO-depleted river mouths probably increased the photochemical degradation of CDOM (Figure 4; Figures S6, S8, and S9) and, therefore, potentially also increased the dissolved CH$_4$ concentrations. This is directly supported by the significant positive relationship between terrestrial CDOM and $K_d$(PAR) and between $K_d$ of underwater light in the UV and PAR ($K_d$(PAR)) regions (Figure 4; Figure S8). Recent studies demonstrated notably higher SDD and $K_d$(PAR) in the northwestern than in the remaining lake regions apart from those dominated by macrophytes. Higher UV and PAR light availability in the DO-depleted water column (Figure 4; Figure S8) can lead to mineralization of CDOM to dissolved CH$_4$ bioavailable...
organic substances, and refractory CDOM in the northwestern inflowing river mouths of the lake. This agrees with the higher SDD and low K_c(PAR) and TSM reported from this area. Recent studies have further revealed elevated terrestrial CDOM and nutrient accumulation and enhanced DO depletion in the northwestern part of the lake, which may result in enhanced dissolved CH_4 levels.

Although there were positive relationships between Chl-a and dissolved CH_4 when excluding river samples or only examining August samples (Figures S9 and S10), these were less compared with the relationship between terrestrial C2 and dissolved CH_4 (Figure 5), and the spatial distribution patterns of Chl-a and dissolved CH_4 differed in all seasons (Figure 3). This was particularly evident in August and November when there were algal blooms in the northwestern shapes characterize the dissolved CH_4, and it appeared that humic-rich allowed us, in part, to elucidate the potential carbon sources of the algal degradation DOM samples, further suggesting that the emission of CH_4 under anaerobic and aerobic conditions.

DOM in Lake Taihu is primarily of allochthonous origin (Figure 4).

The correlation between terrestrial humic-like substances and dissolved CH_4 in Lake Taihu and the connecting channels does not necessarily prove causation as no information was available on DOM composition of pore water in the sediment where a large fraction of the CH_4 is likely produced. Although the relationship between Chl-a and dissolved CH_4 was weak compared with the relationship between terrestrial C2 and dissolved CH_4 during 2012–2014 (Figure 5), the massive algal mats recorded in the northwestern bay during the past three decades may enrich the sediment with particulate carbon. The subsequent degradation of particulate organic carbon in the sediment may provide a labile DOM source for methanogens. DOM optical measurements coupled with FT-ICR MS and δ^{13}C-DOM can be used to quantify the relative importance of autochthonous and allochthonous DOM for CH_4 production at ecosystem scale. Acetoclastic methanogenic archaea have been found at the molecular level in diverse oxygenated aquatic environments where they release CH_4 aerobically. Follow-up experimental work is, therefore, needed to quantify the contribution of DOM degradation to the emission of CH_4 under anaerobic and aerobic conditions.

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.est.8b02163.

Additional information on methods; data including the gas transfer coefficient (k) determined by wind speed and temperature in different lake regions is remarkably uniform, and the emission from the lake is largely controlled by the levels of dissolved CH_4 across the lake (Figure S10). Our results revealed that six individual fractions differing in spectral shapes characterize the fluorescent properties of CDOM in Lake Taihu and the S1 major channels connecting to it (Figure 2). Dividing CDOM into the six fluorescence components allowed us, in part, to elucidate the potential carbon sources of dissolved CH_4, and it appeared that humic-rich fluorophores derived from terrestrial environments correlated with the levels of dissolved CH_4. Despite the fact that the protein-like fluorescence components C1, C5, and C6 contributed primarily to the fluorescence characteristics of CDOM, dissolved CH_4 was weakly or not correlated with these three components, and DOC was not correlated with them at all (Table S5). In comparison, the humic-like components, C2–C4, likely deriving from the terrestrial environment (e.g., soil organic matter), explained a large proportion of the variability of dissolved CH_4 and predominantly the variability of DOC (Figure 5; Table S5). Headwater and central lake water samples share higher relative abundances of terrestrial lignin molecular formulas identified via FT-ICR MS compared with the algal degradation DOM samples, further suggesting that DOM optical measurements coupled with FT-ICR MS and δ^{13}C-DOM can be used to quantify the relative importance of autochthonous and allochthonous DOM for CH_4 production at ecosystem scale. Acetoclastic methanogenic archaea have been found at the molecular level in diverse oxygenated aquatic environments where they release CH_4 aerobically. Follow-up experimental work is, therefore, needed to quantify the contribution of DOM degradation to the emission of CH_4 under anaerobic and aerobic conditions.

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Notes

The authors declare no competing financial interest.

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