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ORIGINAL PAPER

Spectroscopic characterization and molecular weight distribution of dissolved organic matter in sediment porewaters from Lake Erhai, Southwest China

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Abstract Dissolved organic matter (DOM) in sediment porewaters from Lake Erhai, Southwest China was investigated using dissolved organic carbon (DOC) concentration, UV absorbance, fluorescence and molecular weight distribution. DOC exhibited a high concentration at the sediment–water interface with a rapid decrease to the oxic–anoxic interface at approximately 7 cm, and then increased with depth. Similar trends were also found for the UV absorption coefficients at 254 and 280 nm in the porewaters. DNA in the sediment was also measured, which confirmed the high abundance of aerobic bacteria in the upper layer of the sediment. Both humic-like (peaks A and C) and protein-like (peaks B and D) fluorescence were observed in the porewater DOM, and their fluorescence intensities exhibited a similar porewater profile as DOC concentration. A strong correlation was found between the peak fluorescence intensity ratio r(A, C) and r(D, B). Both the fluorescence index and UV absorption coefficient at 254 nm suggested a dramatic increase in aromaticity of porewater DOM across the oxic–anoxic interface. Porewater DOM exhibited a multimodal distribution of molecular weight with a relatively low polydispersity. The results of this study offer significant insight into the nature and properties of DOM in freshwater ecosystems.

Keywords Dissolved organic matter · Sediment · Porewater · DNA · Fluorescence · Molecular weight

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Introduction

Dissolved organic matter (DOM) is a ubiquitous constituent of sedimentary porewaters. It plays a significant role in many physical and biogeochemical processes in aquatic environments (O'Loughlin and Chin 2004). As a result, there has been a genuine interest over the past decades in studying the distribution and characterization of porewater DOM from coastal and marine systems (e.g., Orem and Gaudette 1984; Orem and Hatcher 1987; Alperin et al. 1994, 1999; Hedges and Keil 1995; Burdige and Gardner 1998; Burdige 2001, 2002; Sierra et al. 2001; Burdige et al. 2004), lakes (e.g., Orem et al. 1986;

Chin et al. 1994; Wolfe et al. 2002; O'Loughlin and Chin 2004), and wetlands (e.g., Chin et al. 1998). Since DOM is an extremely complex and heterogeneous mixture, it is essentially impossible to completely separate and identify its chemical composition. Instead, chemical characterization of natural DOM has been largely on its optical properties and molecular weight distribution.

A fraction of DOM is optically active and known as colored DOM (CDOM). Both UV–visible absorption properties and fluorescence properties of CDOM have been used for the characterization of the total DOM pool (e.g., Kalle 1966; Senesi 1990; Coble et al. 1990). Three-dimensional excitation emission matrix fluorescence spectroscopy (3DEEM) is a particularly powerful tool, which has been successfully used to probe chemical structures of DOM because of its ability to distinguish among different classes of DOM of different origins (Coble et al. 1990; Coble 1996; Wu and Tanoue 2001; Baker and Spencer 2004; Cammack et al. 2004). In seawater, several types of DOM fluorescence with excitation/emission wavelength peaks (Ex_{max}/Em_{max}) have been observed: humic-like fluorescence (denoted as peaks A, C and M) and protein-like fluorescence (peaks T and B) (e.g., Coble 1996; Yamashita and Tanoue 2003). Although a few studies were reported on the fluorescence properties of DOM in coastal and marine sediment porewaters (Chen and Bada 1989, 1994; Chen et al. 1993; Skoog et al. 1996; Coble 1996; Sierra et al. 2001; Burdige et al. 2004), there is a general lack of knowledge with respect to freshwater porewaters.

The measurement of molecular weight distribution provides another means of DOM characterization (Chin et al. 1994; O'Loughlin and Chin 2004). The majority of porewater DOM is believed to have molecular weight of >1 kDa, which consists largely of humic substances (Krom and Sholkovitz 1977; Orem et al. 1986; Burdige 2001) with minor amounts of soluble proteins, carbohydrates, and other non-humic macromolecules. Lower molecular-weight components (i.e., <1 kDa) can also be present in natural DOM, which mainly consist of fatty acids, sugars and amino acids (O'Loughlin and Chin 2004). However, so far only a few studies have examined the vertical profiles of the molecular weight distribution of DOM in freshwater sediment porewaters (Chin et al. 1998; O'Loughlin and Chin 2004).

Here we report the vertical profiles of dissolved organic carbon (DOC), UV absorbance, 3DEEM, and molecular weight distribution of DOM in freshwater sediment porewaters from Lake Erhai, Southwest China. The DNA properties in the sediment core were also investigated. To the best of our knowledge, there are very few studies focusing on the DNA distribution in lake sediments. The objective of this study was to characterize the nature and properties of the lake sediment porewater DOM, and to help to understand the role of sediment as a source of fluorescent organic matter to the porewaters and overlying water, and the biogeochemical cycling of DOM in lacustrine environments.

Materials and methods

All the chemicals used were of analytical grade or higher, unless otherwise specified. The laboratory water used was Milli-Q water (18.2 MΩ cm, Millipore).

Sampling sites

Lake Erhai (100°5′–17′ E, 25°35′–58′ N, altitude: 1,974 asl) is located in the north of Dali City in Yunnan Province, Southwest China, and is the largest fault lake in the western Yunnan Plateau (Fig. 1). The surface area of the lake is 249.8 km^2, with an average water depth of 10.5 m (max: 20.9 m) (Wan et al. 2003). The lake is located in the subtropical monsoon climate zone with an annual average temperature of 15°C, precipitation of 1,060 mm, and potential evaporation of 1,970 mm. The catchment area is mainly underlain by sedimentary and metamorphic rocks, specifically carbonate and siliciclastic rocks and gneisses. The surface water is characterized by a slightly alkaline pH (8.0–8.5) and a low salinity (Wan et al. 2003).

Six sediment cores were obtained simultaneously at a depth of 18 m at Station 2 (EH-2, Fig. 1) using a gravity corer (with a tube of 10 cm in diameter and 50 cm in length). The cores were

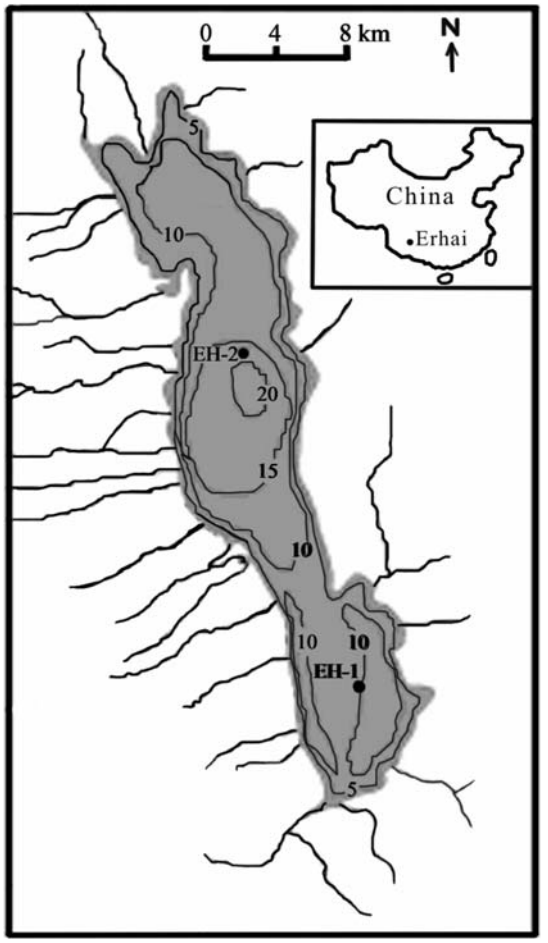

Fig. 1 Sampling sites of Lake Erhai

undisturbed during the sampling processes. They were sliced into 1 cm sections under N_2 atmosphere, and samples were then placed into and N_2-containing centrifuge bottles. Water samples were collected at depths of 1 and 18 m. All samples were transported to the laboratory within 30 h after sampling. In the laboratory, porewaters were extracted from the sediments using centrifugation (4,000 rpm for 20 min) at 4°C. The sediment samples were freeze-dried and ground with a mortar and pestle for DNA analysis. The surface water and porewater samples were filtered through pre-combusted GF/F glass fiber filters (5 h at 450°C, Whatman, U.K.) and were kept in well-sealed brown glass vials (16 ml, National Scientific Company) under N_2 atmosphere at 4°C in dark before analyses.

Porewater analyses

DOC

Porewater DOC concentrations were measured with a High TOC/N II Analyzer (Elementar, Germany). Potassium hydrogen phthalate was used as the standard.

UV absorbance

UV absorbance measurements were carried out on a Shimadzu UV-3000 double beam spectrophotometer at room temperature with 1 cm quartz cells. Milli-Q water was used as the reference. The measured absorbance at wavelength λ (A_λ) was converted to absorption coefficient (a_λ) with the equation $a_\lambda = 2.303A_\lambda/l$, where l is the cell path length, and specific UV absorption SUVA = a_λ/DOC.

Fluorescence

The fluorescence was determined on a spectrofluorometer (Hitachi, Model F-4500) at room temperature with a 150-W ozone-free xenon arc lamp and a 1 cm quartz cell. The 3DEEM fluorescence data were collected for 2 nm wavelength of emission scans at every 5 nm wavelength of excitation. The spectrofluorometer was operated under the conditions of 5 nm slit width for excitation and 10 nm slit width for emission, photomultiplier voltage of 700 V, scan speed of 1200 nm per min and auto response time. The scan range was 220–400 nm for excitation and 250–500 nm for emission. Triplicate measurements of each sample were carried out, and reported as their average. The spectra were blank subtracted. The software SigmaPlot (SPSS) was employed for the visualization of 3DEEM data, and the fluorescence intensity was expressed in arbitrary unit. The fluorescence index ($f_{450/500}$, the ratio of fluorescence intensity at the emission wavelength 450 nm to that at 500 nm at an excitation wavelength of 370 nm) (Battin 1998; McKnight et al. 2001) was determined.

Molecular weight

The molecular weight distribution was measured by high performance size exclusion chromatography (HPSEC) following the procedures of Chin et al. (1994) and Zhou et al. (2000). The HPSEC system was equipped with a HP 1100 controller with a pump in isocratic mode with a flow rate of 0.5 ml min^{-1}, and a UV detector set at a wavelength of 254 nm. A YMC-60 column (Waters, Milford, MA) packed with a silica diol modified material with 5 μm gel bead diameter and 60 Å pore size was used. The mobile phase was composed of 0.001 M Na_2HPO_4, 0.001 M NaH_2PO_4, and 0.03 M NaCl. Acetone was used to determine the permeation volume. The calibration of the molecular weight was based on polystyrene sulfonate (PSS) standards (13, 6.8, 4.3, 1.4 KDa, and 210 Da; American Polymer Standards, Mentor, OH). The number-averaged molecular weight (M_n) and weight-averaged molecular weight (M_w) for the porewater samples were calculated using the following equations (Chin et al. 1994):

$$M_n = \sum_{i=1}^{n} h_i / \sum_{i=1}^{n} (h_i / M_i) \quad (1)$$

$$M_w = \sum_{i=1}^{n} (h_i * m_i) / \sum_{i=1}^{n} h_i \quad (2)$$

where M_i is the molecular weight at eluted volume i, and h_i is the height of the sample HPSEC curve eluted at volume i.

DNA analysis

Two grams of dried sediment sample was added into 5 ml lysozyme solution (10 g l^{-1}) and was shaken five to eight times while being cultured at 37°C for 2 h. Five milliliters of DNA extractant (1 M Tris–HCl, 5.8 g l^{-1} NaCl, 7.5 g l^{-1} EDTA, and 20 g l^{-1} sodium dodecyl sulfate) was added to extract nucleic acids. The solution was centrifuged for 30 min at 4,000 rpm at 4°C to obtain the supernatant. The centrifugation step was repeated three times, and the mixed supernatant was then extracted with an equal volume of saturated hydroxybenzene and chloroform–isoamyl alcohol. The purified solution was centrifuged again to obtain the final supernatant, to which ethanol was added to precipitate DNA. After being air-dried, the DNA pellets were re-dissolved in 100 μl *N*-[tris(hydroxymethyl)methyl]-2-aminoethanesulfonic acid (TE) buffer. Ten microliters of the DNA solution was placed into agarose gel containing 0.5 mg g^{-1} ethidium bromide (EB). The agarose gel was analyzed by electrophoresis at 45 V in 0.5× TBE (Tris–boracic acid–EDTA) buffer, and imaged in UV light on GIS-2008 (Tanon, China) (Wei et al. 2005).

Results and discussion

Vertical distribution of DOC in sediment porewaters

The porewater DOC concentrations from Lake Erhai decreased rapidly from 40.0 mg l^{-1} near the sediment–water interface to 5.5 mg l^{-1} at 6–9 cm below the interface (Fig. 2A). Further down to the deeper layer of the sediment, the DOC concentration increased gradually, reaching 55 mg l^{-1} at 30 cm. This trend is similar to the porewater DOC profile from Peddock Island core (Boston Harbor, MA) (Chin and Gschwend 1991) and from the Gulf of Biscay (Sierra et al. 2001). But it is different from many other reports where porewater DOC concentrations were found to generally increase with depth (Chen et al. 1993; Alperin et al. 1994; Chin et al. 1998; Burdige and Gardner 1998; Burdige 2001; Simoneit and Sparrow 2002; O'Loughlin and Chin 2004).

The DOC concentrations in sediment porewaters are controlled by a variety of biogeochemical processes. The U-shaped DOC profile in Lake Erhai suggests that different processes occurred in the upper and deeper layers of the sediment. Previous studies on Fe and Mn speciation showed that the overlying water and upper layer of the sediment of Lake Erhai were oxic year-round and that the oxic–anoxic interface was located at approximately 7 cm below the sediment–water interface (Luo et al. 2000). Therefore, the sharp decrease in the DOC concentration in the upper layer of the sediment

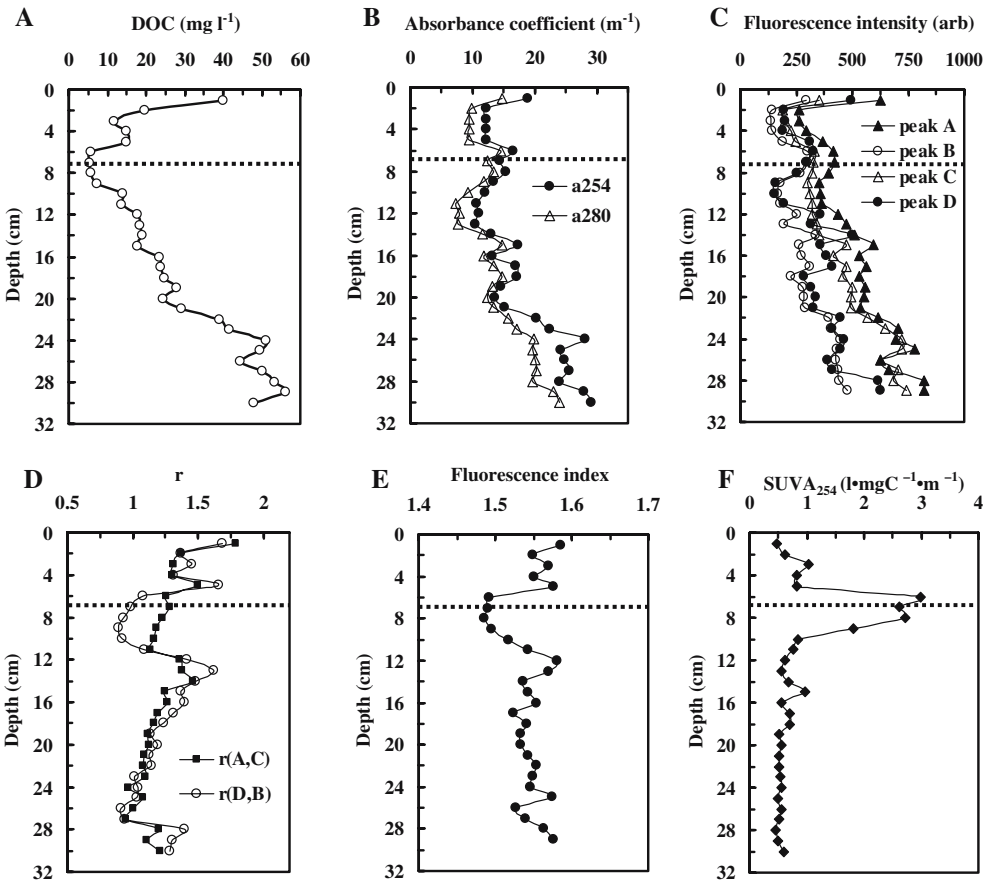

Fig. 2 Porewater profiles of DOM from Lake Erhai. (**A**) DOC concentrations; (**B**) UV absorption coefficients at 254 nm (a_{254}) and 280 nm (a_{280}); (**C**) fluorescence intensities; (**D**) fluorescence intensity ratios; (**E**) fluorescence index; (**F**) $SUVA_{254}$. The dashed line represents the oxic–anoxic interface as reported by Luo et al. (2000)

porewaters was likely due to the intensive aerobic decomposition of organic matter.

To probe the microbial activities in the sediments, the total amount of DNA was analyzed. DNA is considered as an indicator of bacterial quantities and the degradation extent of sedimentary organic matter (Wei et al. 2004, 2005). The agarose gel photo (Fig. 3) shows that DNA mainly occurred in the top 7 cm sediments, suggesting the abundance of aerobic bacteria within this oxic layer. Previous studies on the same sampling site showed that both particulate organic carbon (POC) and nitrogen concentrations also decreased rapidly, and POC decreased from 6% at the 1 cm depth to 2% at the 10 cm depth (Wei et al. 2004).

The downward increase in the DOC concentration below the oxic–anoxic interface indicates that DOC was produced and accumulated in the deeper anoxic environment. This can be from the accumulation of recalcitrant DOC (primarily humic substances) formed by abiotic polymerization of low-molecular-weight DOC, anaerobic degradation of particulate organic matter (Burdige 2002; O'Loughlin and Chin 2004), the reductive dissolution of Fe- and Mn-oxides in the anoxic environment and the subsequent release of the absorbed organic matter to porewaters (Sierra et al. 2001), and the complexation between DOC and dissolved Fe(II) (O'Loughlin and Chin 2004). Indeed, the concentrations of Fe- and Mn-oxides in sediments from Lake Erhai were found to decrease abruptly below 7 cm depth (Luo et al. 2000). Several studies have reported a strong correlation between DOC and dissolved Fe(II) in sediment porewaters in anoxic sediments (Chin et al. 1998; O'Loughlin and Chin 2004).

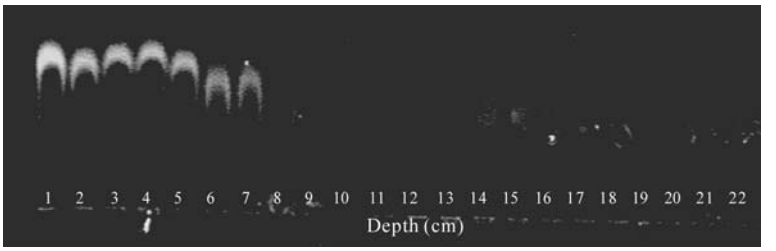

Fig. 3 Agarose gel photo of DNA in the sediments of Lake Erhai

UV absorption properties of DOM

Whereas UV light absorbance has been used to estimate DOC concentrations in a variety of surface waters with varying success (Stewart and Wetzel 1981; Yelverton and Hackney 1986), only a few studies have examined the relationship between UV absorbance and DOC concentrations in porewaters (e.g., O'Loughlin and Chin 2004).

Figure 2B shows the UV absorption coefficients at wavelengths 254 nm (a_{254}) and 280 nm (a_{280}) of porewaters from Lake Erhai. Two hundred and fifty-four and 280 nm wavelengths were commonly used in many DOM studies since the majority of DOM contains functional groups with structures similar to a large number of aromatic compounds (phenolic substances, anilines, benzoic acids, polyenes and polycyclic aromatic hydrocarbons with two or more rings) that absorb light in this UV region due to π–π^* electronic transitions (Traina et al. 1990; Chin et al. 1994; O'Loughlin and Chin 2004). The vertical depth profiles of a_{254} and a_{280} were similar to that of DOC concentration (Fig. 2A). Indeed, significant correlations were observed between the DOC concentrations and a_{254} ($R^2 = 0.77$, $p < 0.01$) and a_{280} ($R^2 = 0.69$, $p < 0.01$), suggesting that the proportion of UV-absorbing DOM in the porewaters relative to DOC was constant. O'Loughlin and Chin (2004) also reported a strong correlation between the DOC concentration and a_{280} in sediment porewaters from Green Bay, Lake Michigan. Therefore, similar in surface waters, a_{254} and a_{280} could be potentially used as a predictor for DOC concentration in sediment porewaters.

Fluorescence properties of porewater DOM

For the sediment porewater DOM from Lake Erhai, there were four distinguishable fluorescence peaks (peaks A, B, C and D) (Fig. 4), which were commonly observed for natural DOM samples (Coble 1996; Wu and Tanoue 2001; Leenheer and Croué 2003; Chen et al. 2003). Peaks A (Ex/Em = 240–255/426–454 nm) and C (Ex/Em= 310–360/408–456 nm) are considered to be humic-like fluorescence, while peaks B (Ex/Em =2 70–280/326–348 nm) and D (Ex/Em = 225–230/324–344 nm) are attributed to protein-like fluorescence (Mopper and Schultz 1993; Coble 1996; Mayer et al. 1999; Baker 2001; Wu and Tanoue 2001; Chen et al. 2003; Leenheer and Croué 2003; Wu et al. 2003; Yamashita and Tanoue 2003; Burdige et al. 2004). The fluorescence intensities of each peak in the porewaters were much higher than those in the overlying water (Fig. 4), and exhibited similar trends (Fig. 2C) as those of the DOC concentration (Fig. 2A) and the UV absorbance (Fig. 2B). Significant correlations between the DOC concentration and fluorescence intensities at peaks A, B, C and D were also observed (peak A: $R^2 = 0.78$, $p < 0.01$; peak B: $R^2 = 0.80$, $p < 0.01$; peak C:R^2=0.70, $p < 0.01$; peak D: $R^2 = 0.62$, $p < 0.01$), once again suggesting that the proportion of CDOM in the porewater samples in Lake Erhai relative to DOC was constant.

Protein-like fluorescence can be further divided into tryptophan-like (Ex/Em = 270–280/320–350 nm) and tyrosine-like fluorescence (Ex/Em = 270–280/300–320 nm) (Coble 1996; Mayer et al. 1999; Chen et al. 2003; Leenheer and Croué 2003; Wu et al. 2003; Yamashita and Tanoue 2003; Burdige et al. 2004) by their differences in emission wavelength. Tyrosine-like fluorescence was reported in porewaters from the continental shelf off the west coast of Mexico, and one sample at a depth of 0.5 cm was found to contain tryptophan-like fluorescence (Coble 1996). Mathews et al. (1996) observed a peak at Ex/Em = 280/

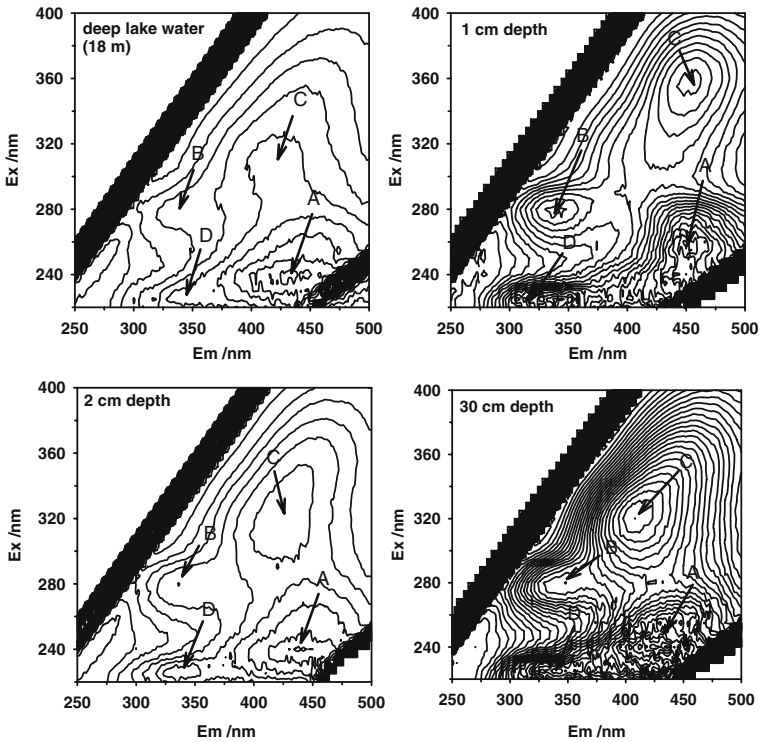

Fig. 4 3DEEM of DOM in the overlying water and sediment porewaters from Lake Erhai

320–350 nm in the EEM spectra of isolated organic matter from corals, which was attributed to tryptophan in proteins. Although the tryptophan-like and tyrosine-like fluorophores cannot be distinguished from peak B in porewaters from Lake Erhai, and still very few work to date has ever described protein-like fluorescence as actually derived from tyrosine, tryptophan or protein in DOM (Yamashita and Tanoue 2003), we still believe that both tryptophan- and tyrosine-like materials were present in the porewaters and the single peak B (the Ex/Em maxima ranged among 270–280/326–348 nm) represented their average, as our previous study (Wu and Tanoue 2002) showed the presence of both tryptophan (0.17–0.32 μmol g^{-1}) and tyrosine (0.91–5.14 μmol g^{-1}) in the sediments of Lake Erhai. The release of the sedimentary tryptophan-like and tyrosine-like organic material into the porewater during early diagenesis may co-contribute to the strong protein-like fluorescence of porewater DOM.

Ratios of fluorescence intensities of humic-like peaks A to C, r(A, C), in the sediment porewaters of Lake Erhai ranged from 1.79 to 0.94 (Fig. 2D). They are similar to the average values reported for marine sediment porewaters from the west coast of Mexico (0.77; Coble 1996) and the Chesapeake Bay (2.1; Burdige et al. 2004). The highest r(A,C) ratio of 1.79 was observed near the sediment–water interface (1 cm) and no systematic change in the depth profile (Fig. 2D). r(A, C) was reported to be strongly pH-dependent (Patel-Sorrentino et al. 2002), thus, r(A, C) variation with depth may indicate the change of pH in the sediment porewaters. Protein-like fluorescence peak D was sometimes neglected in previous studies, but can be reliably measured with the recent development of stable source lamp (Chen et al. 2003). The presence of both peak B and D allowed us to calculate the ratios of their fluorescence intensities, r(D, B) in Lake Erhai. r(D, B) ratios were very similar to r(A, C) and exhibited a similar vertical distribution trend (Fig. 2D). When plotting r(A, C) to r(D, B), a strong correlation between them was found ($R^2 = 0.85$, $p < 0.01$). There were also strong correlations between the protein-like fluorescence at peak B and the humic-like fluorescence at peak A and C ($R^2 = 0.84$, 0.85, respectively, $p < 0.01$, Fig. 5). These observations suggest that

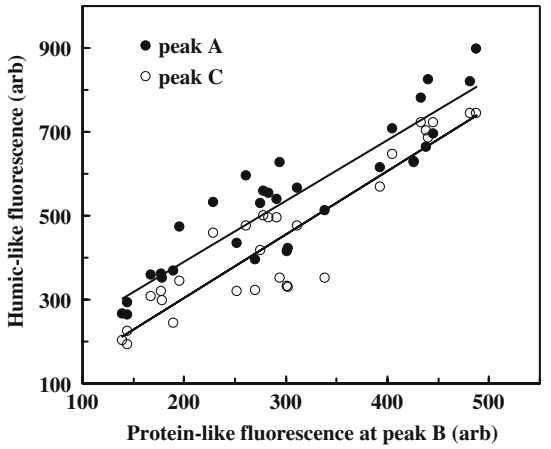

Fig. 5 Relationship between fluorescence intensity at peak B and peaks A and C for the porewater samples

an intrinsic relationship may exist between the origins of protein-like and humic-like fluorescence organic matter in sediment porewaters from Lake Erhai.

Aromaticity of porewater DOM

The aromaticity of porewater DOM from Lake Erhai was evaluated from the fluorescence index $f_{450/500}$ and the specific UV absorbance at 254 nm $SUVA_{254}$ (Fig. 6). McKnight et al. (2001) reported that $f_{450/500}$ related linearly with the aromaticity of organic matter, and thus can be used as a surrogate for general structural features

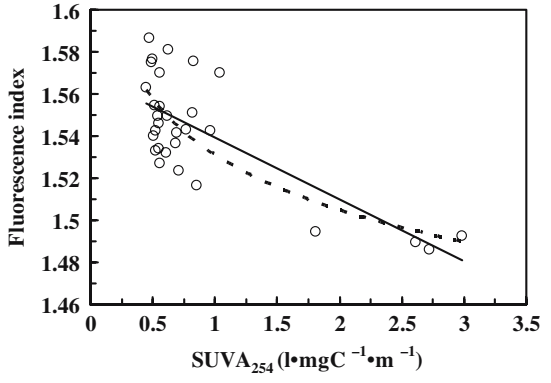

Fig. 6 Fluorescence index vs. $SUVA_{254}$ for the lake sediment porewater samples. The linear regression equation is $Y = -0.0294X + 1.5686$ ($R^2 = 0.56$, $p < 0.01$), and the power function regression is $Y = 1.531X^{-0.025}$ ($R^2 = 0.56$, $p < 0.01$), where X is $SUVA_{254}$ and Y is fluorescence index)

of the carbon skeleton and the source. It was also suggested that $f_{450/500}$ could be used to distinguish fulvic acids derived from microbial sources of organic matter from those derived from terrestrial sources of organic matter (McKnight et al., 2001): an $f_{450/500}$ of ~1.9 would suggest the fulvic acids were microbially derived and an $f_{450/500}$ of ~1.4 the terrestrially derived. In addition to $f_{450/500}$, $SUVA_{254}$ was found to strongly correlate with percent aromaticity as determined by ^{13}C-NMR for organic matter isolates obtained from a variety of aquatic environments (Weishaar et al. 2003).

As shown in Fig. 6, there was a strong and negative relationship between $SUVA_{254}$ and $f_{450/500}$ for the porewater DOM from Lake Erhai, suggesting that both $SUVA_{254}$ and $f_{450/500}$ were useful to estimate the aromaticity of DOM. $f_{450/500}$ ranged from 1.46 to 1.59 with the lowest values at the 6–9 cm depth (Fig. 2E) while $SUVA_{254}$ from 0.45 to 2.98 with the highest at 6–10 cm depth (Fig. 2F). Below approximately 10 cm depth, both $f_{450/500}$ and $SUVA_{254}$ remained essentially constant, $f_{450/500}$ ranged from 1.53 to 1.57 and $SUVA_{254}$ from 0.45 to 0.70. It is interesting to note that both the dramatic decrease in $f_{450/500}$ and increase in $SUVA_{254}$ occurred near the oxic–anoxic interface, indicating a significant increase in the aromaticity of porewater DOM across the oxic–anoxic interface (Fig. 2E, F).

Molecular weight of porewater DOM

All porewater DOM samples from Lake Erhai exhibited multimodal HPSEC chromatograms with sharp and well resolved peaks (Fig. 7). The chromatograms differed significantly from those of porewater DOM from Green Bay of Lake Michigan, USA, which exhibited as broad, unimodal distributions with poorly resolved shoulders and minor trailing peaks (O'Loughlin and Chin 2004). But they are similar to those of porewater DOM from sediments of the northern basin of Lake Michigan (Chin et al. 1994).

The majority of the porewater DOM from Lake Erhai was of relatively low molecular weight (<3000 Da). M_w ranged from 1462 to 1953 Da, and M_n from 547 Da to 900 Da, which agreed well with the ranges reported for the

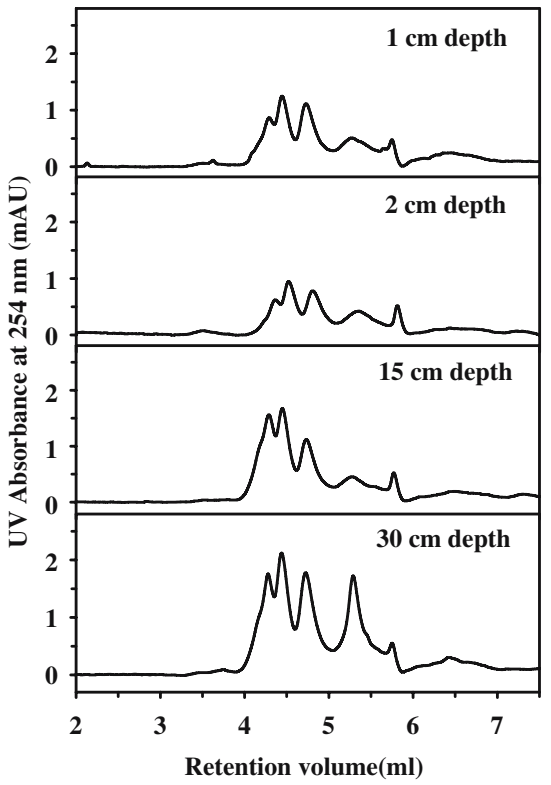

Fig. 7 HPSEC chromatograms of some porewater samples from Lake Erhai

porewater DOM from Lake Michigan (O'Loughlin and Chin 2004). Although they fluctuated considerably, there seemed to be a slight increase in M_w and M_n with depth (Fig. 8). It was reported that DOM in the oxic sediments was composed of relatively small organic compounds while DOM in suboxic sediments exhibited a much larger fulvic-like peak (O'Loughlin and Chin 2004).

The polydispersity (ratio of M_w to M_n) of the porewater DOM was relatively low, ranging from 2.0 to 3.1 (Fig. 8), suggesting that the DOM consisted of compounds with a narrow range of molecular weights (O'Loughlin and Chin 2004). The polydispersity was around 2.8 near the sediment–water interface, decreased to 2.0 at a depth of 4 cm, and then increased to 3.1 at a depth of 14 cm. From there downwards, the polydispersity decreased slightly with depth (Fig. 8).

Conclusions

Four major fluorescence peaks were observed in porewaters: two humic-like fluorescence peaks (A and C) and two protein-like fluorescence peaks (B and D). Strong correlation between fluorescence and DOC indicates that the proportion of CDOM in the porewaters in Lake Erhai relative to DOC was constant. It is well accepted that humic material are products of early diagenesis (Rashid, 1985). The coherent depth trends of humic-like and protein-like fluorescence suggest that there existed an intrinsic relationship between the origins of protein-like and humic-like fluorescence material in lake sediments, and indicate that porewater protein-like fluorescence may be associated with amino acids incorporated into humic-like structure.

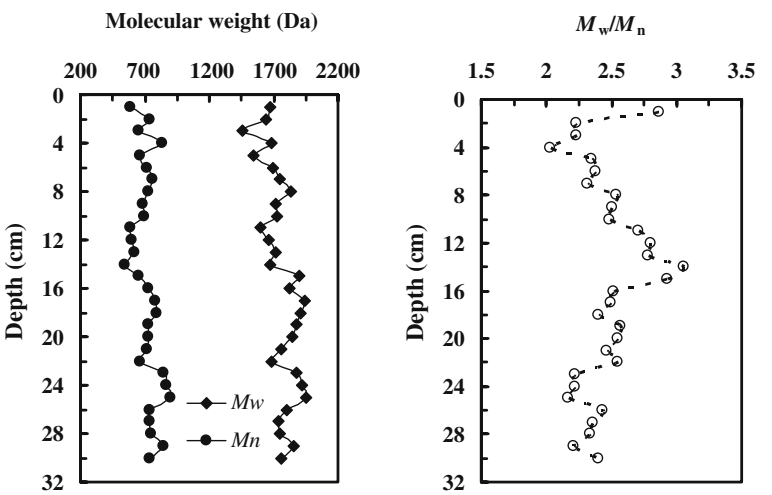

Fig. 8 Depth profiles of DOM molecular weight and polydispersity in the porewaters

For vertical distributions of $SUVA_{254}$, $f_{450/500}$, fluorescence, DOC, and molecular weight in sediment porewaters, together with the DNA profile in the sediments, major changes were found at approximately 7 cm sediment depth, where great change in redox chemistry occurred. In the upper oxic layer, because of the existence of aerobic bacteria and oxygen, organic matter was more efficiently decomposed, while in the anoxic layer of the sediment, organic matter was already partially decomposed before burial in anoxic layer, and thus was less labile (Canfield 1994). Therefore, the enhanced DOC preservation was observed in the deeper layer. These results offer insight into the evolution, composition, and reactivity of porewater DOM and its influence on the biogeochemical cycling of environmentally relevant compounds (such as heavy metals or organic pollutants) in lake environments.

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References

Alperin MJ, Albert DB, Martens CS (1994) Seasonal variations in production and consumption rates of dissolved organic carbon in an organic-rich coastal sediment. Geochim Cosmochim Acta 58:4909–4929

Alperin MJ, Martens CS, Albert DB, Suayah IB, Benninger LK, Blair NE, Jahnke RA (1999) Benthic fluxes and porewater concentration profiles of dissolved organic carbon in sediments from the North Carolina continental slope. Geochim Cosmochim Acta 63:427–448

Baker A (2001) Fluorescence excitation-emission matrix characterization of some sewage-impacted rivers. Environ Sci Technol 35:948–953

Baker A, Spencer RGM (2004) Characterization of dissolve organic matter from source to sea using fluorescence and absorbance spectroscopy. Sci Tot Environ 333:217–232

Battin TJ (1998) Dissolved organic matter in a blackwater tributary of the upper Orinoco River, Venezuela. Org Geochem 28:561–569

Burdige DJ, Gardner KG (1998) Molecular weight distribution of dissolved organic carbon in marine sediment pore waters. Mar Chem 62:45–64

Burdige DJ (2001) Dissolved organic matter in Chesapeake Bay sediment pore waters. Org Geochem 32:487–505

Burdige DJ (2002) Sediment pore waters. In: Hansell DA, Carlson CA (eds) Biogeochemistry of marine dissolved organic matter. Academic Press, London, pp 611–663

Burdige DJ, Kline SW, Chen W (2004) Fluorescent dissolved organic matter in marine sediment pore waters. Mar Chem 89:289–311

Cammack WKL, Kalff J, Prairie YT, Smith EM (2004) Fluorescent dissolved organic matter in lakes: Relationships with heterotrophic metabolism. Limnol Oceanogr 49:2034–2045

Canfield DE (1994) Factors influencing organic carbon preservation in marine sediments. Chem Geol 114:315–329

Chen RF, Bada JL (1989) Seawater and porewater fluorescence in the Santa Barbara basin. Geophys Res Lett 16:687–690

Chen RF, Bada JL, Suzuki Y (1993) The relationship between dissolved organic carbon (DOC) and fluorescence in anoxic marine porewaters: Implications for estimating benthic DOC fluxes. Geochim Cosmochim Acta 57:2149–2153

Chen RF, Bada JL (1994) The fluorescence of dissolved organic matter in pore waters of marine-sediments. Mar Chem 45:31–42

Chen W, Westerhoff P, Leenheer JA, Booksh K (2003) Fluorescence excitation–emission matrix regional integration to quantify spectra for dissolved organic matter. Environ Sci Technol 37:5701–5710

Chin Y-P, Gschwend PM (1991) The abundance, distribution, and configuration of porewater organic colloids in recent sediments. Geochim Cosmochim Acta 55:1309–1317

Chin Y-P, Aiken GR, O'Loughlin E (1994) Molecular weight, polydispersity, and spectroscopic properties of aquatic humic substances. Environ Sci Technol 28:1853–1858

Chin Y-P, Traina SJ, Swank CR (1998) Abundance and properties of dissolved organic matter in pore waters of a freshwater wetland. Limnol Oceanogr 43: 1287–1296

Coble PG, Green S, Blough NV, Gagosian RB (1990) Characterization of dissolved organic matter in the Black Sea by fluorescence spectroscopy. Nature 348:432–435

Coble PG (1996) Characterization of marine and terrestrial DOM in seawater using excitation emission matrix spectroscopy. Mar Chem 51:325–346

Hedges JI, Keil RG (1995) Sedimentary organic matter preservation:an assessment and speculative synthesis. Mar Chem 49:81–115

Kalle K (1966) The problem of gelbstoff in the sea. Oceanogr Mar Biol Annu Rev 4:91–104

Krom MD, Sholkovitz ER (1977) Nature and reactions of dissolved organic matter in the interstitial waters of marine sediments. Geochim Cosmochim Acta 41:1565–1573

Leenheer JA, Croué JP (2003) Characterizing aquatic dissolved organic matter. Environ Sci Technol 37(1):19A–26A

Luo SS, Wan GJ, Huang RG (2000) Characteristics of distribution and removal of Fe and Mn at the sediment–water interface of Erhai Lake, Yuannan province. Environmental Science in Chongqing 22(6): 19–21 (in Chinese with English abstract)

Mathews BJH, Jones AC, Theodorou NK, Tudhope AW (1996) Excitation–emission-matrix fluorescence spectroscopy applied to humic acid bands in coral reefs. Mar Chem 55:317–332

Mayer LM, Schick LL, Loder III TC (1999) Dissolved protein fluorescence in two Maine estuaries. Mar Chem 64:171–179

McKnight DM, Boyer EW, Westerhoff PK, Doran PT, Kulbe T, Andersen DT (2001) Spectrofluorometric characterization of dissolved organic matter for indication of precursor organic material and aromaticity. Limnol Oceanogr 46:38–48

Mopper K, Schultz CA (1993) Fluorescence as a possible tool for studying the nature and water column distribution of DOC components. Mar Chem 41:229–238

O'Loughlin EJ, Chin YP (2004) Quantification and characterization of dissolved organic carbon and iron in sedimentary porewater from Green Bay, WI, USA. Biogeochem 71:371–386

Orem WH, Gaudette HE (1984 Organic matter in anoxic marine pore water: oxidation effects. Org Geochem 5(4):175–181

Orem WH, Hatcher PG, Spiker EC, Szeverenyi NM, Maciel GE (1986) Dissolved organic matter in anoxic pore waters from Mangrove Lake, Bermuda. Geochim Cosmochim Acta 50:609–618

Orem WH, Hatcher PG (1987) Solid-state ^{13} NMR studies of dissolved organic matter in pore water from different depositional environments. Org Geochem 11(2):73–82

Patel-Sorrentino N, Mounier S, Benaim JY (2002) Excitation–emission fluorescence matrix to study pH influence on organic matter fluorescence in the Amazon basin rivers. Wat Res 36:2571–2581

Rashid MA (1985) Geochemistry of marine humic compounds. Springer Verlag, New York

Senesi N (1990) Molecular and quantitative aspects of the chemistry of fulvic acid and its interactions with metal ions and organic chemicals: Part II. The fluorescence spectroscopy approach. Anal Chim Acta 232:77–106

Sierra MMD, Donard OFX, Etcheber H, Soriano-Sierra EJ, Ewald M (2001) Fluorescence and DOC contents of pore waters from coastal and deep-sea sediments in the Gulf of Biscay. Org Geochem 32:1319–1328

Skoog A, Hall POJ, Hulth S, Paxéus N, van der Loeff MR, Westerlund S (1996) Early diagenetic production and sediment–water exchange of fluorescent dissolved organic matter in the coastal environment. Geochim Cosmochim Acta 60:3619–3629

Simoneit BRT, Sparrow MA (2002) Dissolved organic carbon in interstitial waters from sediments of Middle Valley and Escanaba Trough, Northeast Pacific, ODP Legs 139 and 169. Appl Geochem 17:1495–1502

Stewart AJ, Wetzel RG (1981) Asymmetrical relationships between absorbance, fluorescence, and dissolved organic carbon. Limnol Oceanogr 26:590–597

Traina SJ, Novak J, Smeck NE (1990) An ultraviolet absorbance method of estimating the percent aromatic carbon content of humic acids. J Environ Qual 19:151–153

Wan GJ, Bai ZG, Qing H, Mather JD, Huang RG, Wang HR, Tang DG, Xiao BH (2003) Geochemical records in recent sediments of Lake Erhai: implications for environmental changes in a low latitude-high altitude lake in southwest China. J Asian Earth Sci 21:489–502

Wei ZQ, Liu CQ, Liang XB, Wang FS (2004) Characteristics of organic matter and DNA in sediments of Lake Erhai. Acta Sedimentol Sinica 22:672–675 (in Chinese with English abstract)

Wei Z, Liu C, Liang X, Wang F, Wang S (2005) Degradation of organic matter in the sediments of Hongfeng Reservoir. Chin Sci Bull 50:2377–2380

Weishaar JL, Aiken GR, Bergamaschi BA, Fram MS, Fujii R, Mopper K (2003) Evaluation of specific ultraviolet absorbance as an indicator of the chemical composition and reactivity of dissolved organic carbon. Environ Sci Technol 37:4702–4708

Wolfe AP, Kaushal SS, Fulton JR, McKnight DM (2002) Spectrofluorescence of sediment humic substances and historical changes of lacustrine organic matter provenance in response to atmospheric nutrient enrichment. Environ Sci Technol 36:3217–3223

Wu FC, Tanoue E (2001) Isolation and partial characterization of dissolved copper-complexing ligands in streamwaters. Environ Sci Technol 35:3646–3652

Wu FC, Tanoue E (2002) Tryptophan in the sediments of lakes from Southwestern China Plateau. Chem Geol 184:139–149

Wu FC, Tanoue E, Liu CQ (2003) Fluorescence and amino acid characteristics of molecular size fractions of DOM in the waters of Lake Biwa. Biogeochem 65:245–257

Yamashita Y, Tanoue E (2003) Chemical characterization of protein-like fluorophores in DOM in relation to aromatic amino acids. Mar Chem 82:255–271

Yelverton GF, Hackney CT (1986) Flux of dissolved organic carbon and pore water through the substrate of a *Spartina alterniflora* marsh in North Carolina. Estuar Coast Shelf Sci 22:255–267

Zhou Q, Cabaniss SE, Maurice PA (2000) Considerations in the use of high-pressure size exclusion chromatography for determining molecular weights of aquatic humic substances. Water Res 34:3505–3514