Lignin phenols in sediment cores and its indications of degradation and organic carbon sources of the Yantan Reservoir, China

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Abstract

Three sediment cores collected from the Yantan Reservoir, located in the Pearl River, southwest China, were analyzed for lignin phenols, elemental and stable carbon isotopic composition to investigate the variation patterns, vegetation sources, degradation stage, and relative proportions of terrestrial sedimentary organic carbon (OC). Significant temporal and spatial heterogeneity in terrestrial OC burial was indicated by the changes of lignin contents at different depths in different sampling sites: the inlet zone, the central reservoir zone in front of dam and the reservoir bay. The interception impact of upstream dam, the influence of artificial regulation, as well as the role of interzonal recharge made the terrestrial OC burial remains complex in the reservoir. The oxidized lignin signatures showed spatial heterogeneity suggesting active oxidative degradation and demethylation/demethoxy degradation of sedimentary lignin during deposition, especially in the inlet zone. An angiosperm herbaceous tissue and gymnosperm woody tissue contributed the sedimentary lignin. A soil-plankton-plant three-end-member mixing model revealed that soil-derived OC dominated before impoundment and at the early stage of reservoir operation, while the contribution of autochthonous OC began to dominate after gradually aging and eutrophicating of the reservoir. Our study of lignin evolution in reservoir highlights important temporal and spatial reservoir carbon components and their contribution to sedimentary carbon pools, providing new insights into the estimation of organic carbon burial in reservoirs.

Introduction

Inland water has been regarded as a potentially important part of the terrestrial carbon (C) at global scales, although it takes a small proportion of the Earth's surface area (Hwang et al. 2015; Raquel et al. 2017; Voss et al. 2017; Park et al. 2018). The organic carbon (OC) deposited in inland water is either mineralized to CO$_2$ or CH$_4$ released into the atmosphere or buried in the sediment (Abril et al. 2015; Feng et al. 2015; Ding et al. 2017; Voss et al. 2017). The OC burial flux in lakes and reservoirs is estimated to correspond to ~ 20% of the C emission, indicating that they represent a remarkable long-term C sink in the organic geochemical C cycle (Cole et al. 2007; Vonk et al. 2015). Moreover, it has manifested that inland water OC burial rates increased hugely over the last century and are up to five times greater than previous estimates, as a result of deforestation soil erosion, eutrophication and river damming (Sebastian et al. 2004; Heathcote et al. 2015; Raquel et al. 2017). Lake and reservoir are more efficiently burying OC on account of higher sedimentation rates, lower oxygen availability, and higher proportion of terrestrial organic matter (OM) (Marynowski et al. 2007; Sebastian et al. 2009; Winterfeld et al. 2015).

Driven by the demand for green energy and water resources, countries around the world are building hydroelectric dams at an unprecedented rate, resulting in continuous climbing in river damming (Verpoorter et al. 2014; Zarfl et al. 2014; Messager et al. 2016). To date, there are more than 16 million dams worldwide, and the number of reservoirs in China has reached the top of the world (Li et al. 2018). The continuity of natural rivers and the law of flood pulsation have been significantly disrupted by damming (Verpoorter et al. 2014; Holgerson. et al. 2016). The extension of hydraulic retention time makes the terrestrial OC easier to be intercepted and deposit in reservoirs. Meanwhile, the nutrient retention
increases the primary productivity and generates more autochthonous OC in reservoirs (Wang et al. 2009; Deemer et al. 2016; Rowe et al. 2018), which is considered as a new C sink (Raquel et al. 2017). The frequent and significant input of allochthonous OM in many lake sediments contributed the high OC burial efficiency (Sebastian et al. 2009; Gudasz et al. 2012; Huang et al. 2017). However, the variation patterns and relative proportions of allochthonous OC in reservoir sediments are poorly understood, which are key to the study of the impacts of reservoir C sequestration and its response and feedback of current environmental evolution and global climate change.

Lignin-derived phenols have been widely used as biomarkers for terrigenous OC because these complex compounds only exist in vascular plants tissues with relative strong resistance to degradation (Jex et al. 2014). Through analysis of these terrestrial OC signals, it is possible to glimpse the effect of human activity and climate change on the sources and decomposition stage of sedimentary OC in reservoirs (Dittmar and Lara 2001; West et al. 2016; Salim and Pattiaratchi 2020). The Pearl River has been extensively developed for hydroelectricity generation due to its ample water resource, with a large number of reservoirs. The Yantan Reservoir with an average water depth more than 200 m located in the Pearl River, southwest China, was selected as the research object in this study. Three sediment cores were collected from the Yantan Reservoir and analyzed for lignin-derived phenols, bulk elemental and stable C isotopic composition. The objectives of this study were: a) to trace the variation patterns in the intensity of terrestrial OC input, vegetation evolution and degradation stage of terrestrial OC in reservoir sediment cores, and b) to assess the contributions of allochthonous and autochthonous processes to OC burial in reservoir.

**Materials And Methods**

**Study Area and Sampling**

The Yantan Hydropower Station is the second largest hydropower station built in 1992 in Guangxi province, southwest China. As an annual regulation reservoir, the Yantan Reservoir located in the Hongshui River (the upper reaches of the Pearl River) experiences perennial seasonal thermal stratification, with a catchment area of $1.065\times10^5$ km$^2$ (Zhang et al. 2015).

Three sediment cores about 40 cm in length were collected from the Yantan Reservoir with a gravity columnar sampler lined with a 50-mm diameter polyvinyl chloride core tube on April, 2019 (Fig. 1). A total of 80 samples sliced at 1 or 2 cm depth intervals were sealed in plastic bags, and transported on ice to the laboratory and stored at -20°C until further treatment and analysis.

**Chemical Analysis**

Total organic carbon (TOC) and total nitrogen (TN) were determined by combustion using an elemental analyzer (FLASH 2000 HT, Thermo Fisher Scientific, MA, US), while the stable isotopic analysis of OC ($\delta^{13}$C) was carried out applying an elemental analyzer equipped with isotope ratio mass spectrometer (EA-IRMS, FLASH 2000 HT, Thermo Fisher Scientific, MA, US).
Molecular analysis of lignin-derived phenols, were performed according to the alkaline CuO oxidation method in previous literature (Miguel and Shelagh 2000). Briefly, freeze-dried, ground sediment samples (0.25 g) mixed with CuO (0.25 g) were digested with ferrous ammonium sulfate [Fe(NH₄)₂(SO₄)₂·6H₂O] and N₂-pressurized NaOH (2 M) in a teflon-lined vessel for 1.5 h, 150°C, in the WX-6000 Microwave Digestion System (PreeKem, Shanghai, China). After the oxidation, an internal standards mixture (ethyl vanillin and trans-cinnamic acid) was spiked for recovery, and pH was adjusted to 1 with HCl (6 M). After centrifugation (5000 r/min for 15 min), supernatants were loaded on a Cleanert PEP cartridge (150 mg/6 mL, Bonna-Agela Technologies, CA, US) for solid phase extraction. The lignin oxidation products (LOPs) were eluted by 5 mL of ethyl acetate. The eluents were concentrated to near dryness under a gentle nitrogen stream and reconstituted with 200 µL of acetonitrile for further analysis. LOPs and native standards were derivatized with 99% bis-trimethylsilyl triuoroacetamide (BSTFA) and 1% trimethylchlorosilane (TMCS) for 10 min at 70°C.

Quantification of LOP molecules was by Gas chromatography coupled with the mass spectrometer (GCMS-QP2020, Shimazu, Kyoto, Japan). Eleven lignin-derived phenols and two internal standards were separated by a fused capillary column (DB-1, 30 m × 0.32 mm × 0.25 µm; Agilent, CA, US). Aliquots of 1 µL of extract were injected in splitless mode at 300°C. The column oven temperature program was set from 100°C to 170°C at a rate of 8°C/min and held for 5 min, and then raised to 300°C at 10°C/min, held for 4 min for LOP analysis. The temperature of the ion source was set to 230°C, and that of the interface was maintained at 300°C. The mass spectrometer was operated in the electron ionization (EI, 70 eV) and selected ion monitoring (SIM) modes.

Data Analysis

There are 11 individual lignin-derived phenols produced by alkaline CuO oxidation which are divided into vanillyl, syringyl, cinnamyl (C; p-coumaric, and ferulic acids), and p-hydroxy (P; including p-hydroxybenzaldehyde, p-hydroxyacetophenone, and p-hydroxybenzoic acid) series (John and Richard 1995). Due to the different types of vegetation and degrees of degradation, the ratios between these lignin monomers can have certain biogeochemical indication significance. The values of Σ8 and Λ8 represent carbon- and sediment-normalized lignin yields respectively, indicating the relative contributions of vascular plant material to the total samples and to the TOC, which were calculated as the sum of V, S and C series in the units of mg/10 g sediment and the units of mg/100 mg OC.

Three-end-member Mixing Model

A three-end-member mixing model was applied using δ¹³C and Λ8 as markers to quantify the contributions of three sources (Wang et al. 2004) (soil, terrestrial vascular plant, and plankton, denoted by the following equations):
\[ \Lambda \text{8}_{\text{sample}} = \Lambda \text{8}_{\text{soil}} \times f_{\text{soil}} + \Lambda \text{8}_{\text{plkt}} \times f_{\text{plkt}} + \Lambda \text{8}_{\text{plant}} \times f_{\text{plant}} \] (1)

\[ \delta^{13}C_{\text{sample}} = \delta^{13}C_{\text{soil}} \times f_{\text{soil}} + \delta^{13}C_{\text{plkt}} \times f_{\text{plkt}} + \delta^{13}C_{\text{plant}} \times f_{\text{plant}} \] (2)

\[ 1 = f_{\text{soil}} + f_{\text{plkt}} + f_{\text{plant}} \] (3)

Where \( f_{\text{soil}}, f_{\text{plkt}}, f_{\text{plant}} \) represent the relative proportion of OC from soil, freshwater plankton and terrestrial vascular plants to TOC respectively.

The assumptions used are as follows: \( \Lambda \text{8}_{\text{soil}} = 0.5 \text{ mg/100 mg OC}, \Lambda \text{8}_{\text{plant}} = 15 \pm 5 \text{ mg/100 mg OC} \) (Lima et al. 2007), and it is generally believed that freshwater plankton does not contain \( \Lambda \text{8}; \delta^{13}C_{\text{soil}} = -21.4\%o \) (Raymond et al. 2013), \( \delta^{13}C_{\text{plkt}} = -31.2\%o \) (Chai et al. 2006), \( \delta^{13}C_{\text{plant}} = -15.2\%o \) (Raymond et al. 2013). Monte Carlo (MC) simulation was employed to estimate the relative proportions of each end member in the present study.

**Results**

**Lignin phenols, elemental and stable carbon isotopic composition**

The values of lignin in core YT1 varied from 0.65 to 4.18 mg/10 g for \( \Sigma 8 \), and from 0.42 to 2.45 mg/100 mg OC for \( \Lambda 8 \) (Fig. 2). There was a significant increase in values at -26 cm depth. However, between -14 cm and -26 cm depth, the variation of \( \Sigma 8 \) and \( \Lambda 8 \) values showed an "M" shaped fluctuation. At depths above -14 cm, the values of \( \Sigma 8 \) and \( \Lambda 8 \) decreased significantly and maintained a steady trend. In core YT2 and core YT3, the \( \Lambda 8 \) and \( \Sigma 8 \) values showed an increasing trend at depths above about -14 cm.

Ranges of S/V and C/V ratios were similar in three sediment cores from Yantan Reservoir. The S/V ratios of samples ranged from 0.51 to 1.37, and C/V ratios ranged from 0.08 to 0.57 (Fig. 3). The ratios of (Ad/Al)v and (Ad/Al)s had a similar vertical distribution trend (Fig. 4). The (Ad/Al)v ratios in core YT1 varied from 0.05 to 0.80, and (Ad/Al)s ratios varied from 0.14 to 1. Between the depths of -8 cm and -24 cm, the ratios of Ad/Al showed some fluctuations, and stabilized at depths above -8 cm. The ratios of (Ad/Al)v in core YT2 ranged from 0.01–0.87, and the ratios of (Ad/Al)s ranged from 0.14–1.24, which were slightly higher than those in core YT1. The core YT3 located in front of the dam had more stable trend with depth for the ratios of Ad/Al, only increased slightly near the surface sediment. The ratios of Pon/P and P/(V + S) changes relatively steadily with depth in core YT2 and YT3. For core YT1, the ratios of Pon/P decreased and the ratios of P/(V + S) gradually increased above a depth of -16 cm (Fig. 4).
The TOC values had a range of 2.02%-1.19%, 1.97%-1.06%, and 2.89%-1.44% for core YT1, YT2, and YT3, respectively, which showed a trend of increasing with decreasing depth. The overall trend of the C/N ratio decreased with depth decreasing, with a range of 10-6.23, 11.5–8.92, and 15.6–6.87 for core YT1, YT2, and YT3, respectively. Similarity, the $\delta^{13}\text{C}$ values for TOM in the samples decreased as depth, range from $-30.60^\text{‰}$ to $-22.05^\text{‰}$, with a mean value of $-24.56^\text{‰}$ (Fig. S1 and S2).

**The Three-end-member Mixing Model Analysis**

The three-end-member mixing model estimated that the contribution of soil to the TOC ranges from 91.2–0.56%, with an average of 52.8% (Fig. 5). A decreasing trend of soil was observed from the lower to upper layer, which provides evidence that soil organic material is the dominant source of OC in sediment cores in Yantan Reservoir, followed by the contribution of plankton, ranges from 7.73–95.7%, with an average of 42.5%, then the plant, ranges from 1.09–14.1%, with an average of 5.49%. The contribution of plankton to TOC gradually increases with the decrease of depth and the variation trend of contribution of terrestrial vascular plants to TOC did not change too much.

**Discussion**

**The pattern of lignin in sediment cores of Yantan Reservoir.**

For core YT1 located in the inlet zone of the reservoir, the significant increase in values at the depth of -26 cm likely indicated a shift in terrestrial OM inputs with increased proportions of lignin-rich constituents in sediments accumulated during the initial period of impoundment. Obviously, human activities after impoundment might have a local effect on terrestrial OM inputs to the reservoir aquatic system as reflected by some fluctuations between ~14 cm and ~26 cm depth in the lignin level. The construction and impoundment of the upstream dam (Longtan Dam) may have a greater impact on the input of terrestrial OC (Yao and Lu 2011), shown by the change in values above the depth of -14 cm. The upstream incoming water mass as plug flow in this region was mainly the bottom water in front of the Longtan Dam, which was built several years later than the Yantan Dam, with low nutrient loading. In addition to terrestrial biogenic elements, particulate matters were similarly intercepted by the upstream dam, as evidenced by the reduction in particle size of particulate matters in core YT1 (3.78 µm on average for upper layer, and 4.63 µm on average for lower layer), so the terrestrial OC carried by particulate matters was also reduced accordingly. Different from core YT1, the lignin in core YT2 located in reservoir bay showed a slight increase with decreasing depth, indicating that the contribution of land-derived OM gradually increased. There were certain differences between the reservoir bay and the central reservoir area in terms of flow velocity, water depth, nutrient load, turbidity, and incoming water. Uncompacted shallow sediments in reservoir bay were more susceptible to interzonal recharges such as surface runoff and rainfall, resulting in changes of OM input (Nunes et al. 2011;Aagaard and Jensen 2013). Studies have shown that changes in rainfall caused by climate change have a great impact on the lignin content in reservoir sediments (Sánchez et al. 2009;Rezende et al. 2010;Qiu et al. 2014). Moreover, the sampling site of YT2 is located at the confluence of the Hongshui River into the reservoir, where the main stream of the Hongshui River contributed fresh terrestrial OM, reflected in the relative higher lignin level in core YT2.
than that in the other two sediment cores. In core YT3 collected in front of the Yantan Dam, the lignin showed some fluctuations above the depth of -20 cm. Although there appeared to be a relatively slight accumulation of lignin in front of dam, there were fewer sources of fresh lignin due to the bottom discharge water from the upstream reservoir with lower temperature and lower nutrient levels. Reservoirs always have a stable thermal stratification close to the dam, resulting in a significant decrease in mass transfer between the upper and lower layers (Chen et al. 2018). This would not be conducive to the accumulation of terrestrial OM, reflecting the influence of artificial regulation. Furthermore, the decreased hydraulic retention time induced by the discharge of bottom water would make more dissolved oxygen in bottom sediment, which might accelerate the degradation rate of OM and nutrients in sediment (Ali W et al. 2013). From the above comparison, it can be found that there is significant spatial heterogeneity in terrestrial OC burial in the sediment cores from this artificially regulated reservoir.

**Vegetation Information Recorded By Lignin Parameters**

The ratios of S/V were used to distinguish lignin from angiosperm or gymnosperm origin, while the ratios of C/V were used to distinguish lignin from herbaceous or woody tissue (Miguel and Shelagh 2000). These ratios, together with the relatively depleted $\delta^{13}C$ values, implied that the lignin in the Yantan Reservoir was derived from angiosperm herb tissue $C_3$ plant sources, and mixed with very small amount of woody tissue, which was consistent with the forest vegetation and main crops in the Pearl River Basin, such as rice, sugar cane, and corn (Loh et al. 2012). The ratio of Pon/P is used to determine whether it is a terrestrial vascular plant source (Miguel et al. 2000). The pattern of Pon/P ratios reflected that the contribution of vascular plants to OM was decreasing with the depth decreased in core YT1. The ratios of Pon/P in core YT2 were relative higher than those in the other two sediment cores, demonstrating the contribution of interval recharge to fresh land-derived OM. However, it is worth noting that the analysis of parent vegetation information in sediment obtained by the ratios of Pon/P sometimes are subject to certain biases due to the degradation degree of each monomer and the content of degradation products (Rezende et al. 2010;Chen et al. 2017). Due to the short time scale, the ratios of S/V and C/V in the Yantan Reservoir did not reflect a significant change about the parent vegetation information.

**Degradation Of Sedimentary Organic Carbon**

Oxidative degradation of lignin side-chains caused by white-rot fungi often produces Ad/Al ratios $> 0.4$ (Shi et al. 2020). A specific lignin degradation pathway due to brown-rot fungi leading to demethylation of methoxylated vanillyl and syringyl constituents could mark an increase in the P/(V + S) ratio (Feng et al. 2016). The ratios of Ad/Al and P/(V + S) showed relative higher values in the middle layer of core YT1. It indicated a moderate degree of side chain oxidation and demethylation/demethoxy degradation for lignin at this stage, which might be related to the construction and water storage of the upstream Longtan Reservoir. The oxidation and denitrification of OM due to the reduction of inflow water enhanced the degradation of lignin in upstream damming period. The patterns of Ad/Al and P/(V + S) ratios in core YT2 may imply the input of degraded or fresh terrestrial OM washed from the watershed soil and other interval supplies. For core YT3, the discharge of bottom water resulted in less retention time of water flow,
and slowed down the decrease of dissolved oxygen concentration. As a result, the ratios of Ad/Al showed that the degradation of lignin maintained at a low level in the lower sediment, while slightly increased near the surface layer.

**Lignin tracers of terrestrial OC burial in the Yantan Reservoir sediment cores**

To better understand the role of reservoirs in the global C cycle, it is important to delineate the origins of OC supply and burial. The simulation results of the three-end-member showed that terrestrial plants contributed the least to OC for the sediment cores (less than 20%). Research indicated that vegetation located in karst terrains had low aboveground biomass and land degradation that reduces vegetation biomass (Ni et al. 2015). In addition to the harsh and fragile natural habitat on karst terrain, human activity also induced the degradation of vegetation. All three sediment cores showed a trend of decreasing soil contribution with depth and increasing phytoplankton contribution with depth. For core YT1, before the dam was built, OC was dominated by soil contribution, accounting for 80%. After the impoundment, the water level began to rise, and the soil layer in the submerged area of reservoir became unstable, shown by the fluctuation of contributions of soil-derived OC, ranging from 60–90%. The release of nutrients in the submerged area and the retention of nutrients in the incoming water make the river section of the reservoir area higher in soil-derived nutrient load (Osisele and Beck 2004). When the reservoir operated for a period of time, the relative contribution of soil-derived OC decreased as the soil layer in the submerged area of reservoir stabilized and the upstream dam interception effect. Meanwhile, a large amount of nutrients promoted productivity of phytoplankton due to the increase of reservoir storage and the stimulation of extra nutrient salts in domestic sewage (Liu et al. 2011). Eutrophication allows more CO₂ to be fixed in inland water and increases the amount of OC buried in inland water sediments (Huang et al. 2017). Autochthonous OC began to dominate, ranging from 50–95%. At the sampling site YT2, OC was strongly influenced by interval recharge. A large fluctuation at -20 cm depth may be the result of an extreme event that exacerbated erosion and the soil-derived OC was deposited in large quantities in sediment, with the contribution of soil increasing to 90%. In front of the dam (YT3), the contribution of soil was relatively stable, probably due to the bottom drainage of the reservoir, which does not facilitate algal growth. It was not until near the surface sediment that the contribution of algae began to increase from 40–70%, indicating that the reservoir was undergoing aging or eutrophicating gradually. Lacustrine made the interception and retention of nutrients including terrestrial OC stimulate the growth of algae, so the contribution of algae to OC in the sediment core began to rise. An evidence was a report of an survey on the water quality of this reservoir from 2011 to 2012, which showed that the water quality was moderate to heavy pollution (Liu et al. 2011).

**Conclusions**

In the present study, lignin-derived phenols were used as biomarkers to trace the terrigenous OC in sediment. As one member of the cascade reservoirs in the Pearl River, the sediment cores collected from different sampling sites in the Yantan Reservoir showed the impact of the upstream dam interception, the role of interzonal recharge, as well as the influence of artificial regulation, resulting in the significant
temporal and spatial heterogeneity in terrestrial OC burial in this artificially regulated reservoir. Most of the terrigenous OC exported by the river appears to be a moderate degree of degradation. Several factors, including the variable compositions of particles and their hydrodynamic sorting following initial depositions, pose important challenges to the construction of OC budgets for the reservoir using three-end-member mixing model. In spite of these factors, we have been able to ascertain that soil-derived OC dominated before impoundment. After water storage, the hydraulic retention time was extended and the level of primary productivity was increased, the proportion of autochthonous OC began to dominate.

**Declarations**

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**Authors' contributions**

The authors contribute according to their research specialty: XL: sample collection, sample and data analysis, and writing the original manuscript. LG, JQH: sampling analysis. JF: funding instruments for data testing. JM, MY, FSW: reviewing, editing and supervision. All authors read and approved the final manuscript.

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**Declarations**

**Ethics approval** This manuscript has not been published or presented elsewhere in part or in entirety.

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**Consent for publication** All authors agreed to publish this article in Environmental Science and Pollution Research.

**Competing interests** The authors declare no competing interests

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