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ORIGINAL ARTICLE

Phosphorus speciation in surface sediments of a hypertrophic lake, Southwestern China: insights from fractionation and ^{31}P NMR

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Abstract Phosphorus (P) species in surface sediments from a shallow, hypertrophic lake, Lake Dianchi, China, were investigated by P fractionation and ^{31}P nuclear magnetic resonance (NMR) spectroscopy during a regional algal bloom. In addition, their potential contributions to the overlying water were also evaluated. Labile fractions of P extracted by NH_4Cl, bicarbonate dithionite and NaOH ranged from 340.6 to 1,725.8 mg kg^{-1}, accounting for 20.5 %–67.2 % of the total P. A two-step extraction method refinement of P recovery was performed before the ^{31}P NMR analysis. Recovery rates of sedimentary TP and organic P, by combining EDTA pretreatment and NaOH or mixed reagents, ranged from 31.8 % to 69.3 % and from 19.8 % to 51.7 %. ^{31}P NMR results showed that ortho-P and monoester-P were the most abundant P components in the sediment extractable P of sediments, followed by diester-P and pyro-P. Spatial distribution of the sum of ortho-P, diester-P and pyro-P detected by ^{31}P NMR corresponded well with the labile P concentration determined by fractionation. Both exhibited a significant positive correlation with the total P in the water column, suggesting that internal loading may be an important source of P for the lake ecosystem. Biogenic P other than ortho-P may contribute to phytoplankton growth, with the relative proportion being 4.4 %–18.7 %. The release of labile P fractions fueled algal bloom, and the decay of organic matter, following the bloom events, consumed oxygen and elevated the pH value. This co-dependence might lead to a vicious cycle. Transformation mechanisms of various P species remain ambiguous and are worthy of further investigation.

Keywords Sediment · Phosphorus · Bioavailability · Eutrophication · ^{31}P NMR

1 Introduction

Eutrophication, characterized by excessive nutrient loading and harmful algal blooms, is a global problem in lakes (Smith et al. 1999). The supply of phosphorus (P), which in most cases is the growth-limiting nutrient, initiates phytoplankton bloom as well as the development of cyanobacteria, and thus severely deteriorates the water quality (Schelske 2009). Exogenous particulate P accumulates in sediments, from which, under certain condition, a reactive fraction can be released into the overlying water. The reservoir of sedimentary P can exceed the external inputs from rivers, and may delay the recovery of water quality in shallow lakes (Jeppesen et al. 2007). However, not all P fractions are labile and can be released into the overlying water. The bioavailability of P mainly depends on their form and composition in the sediment (Ahlgren et al. 2005). Therefore, a deeper understanding of the long-term behavior of sedimentary P requires detailed species information.

Fractionation schemes make it possible to operationally define P forms that bind to metals and organic matters in lake sediments (e.g., Hupfer et al. 1995; Ruban et al. 2001;

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Lukkari et al. 2007), providing valuable parameters for predicting future internal P-loading (Rydin 2000). Many studies have focused on the bioavailability of inorganic P, including water-soluble P, readily de-sorbable P and redox-sensitive P (Zhou et al. 2001; Kaiserli et al. 2002). Organic P was treated as the refractory part of P pool due to limitations in previous analytical methods (Oluyedun et al. 1991). However, it was recently shown that it contains some labile species, which play an important role in sustaining eutrophication (Zhang et al. 2008).

Nuclear magnetic resonance spectroscopy (NMR) is another efficient way of assessing the P composition in sediment, and offers an accurate method to identify P bound as orthophosphate (ortho-P), orthophosphate monoesters (monoester-P), orthophosphate diesters (diester-P), pyrophosphates (pyro-P) and phosphonates. Due to the low natural abundance of sedimentary P, pretreatment and extraction are two key steps for solution ^{31}P NMR spectroscopy (Ahlgren et al. 2007). ^{31}P NMR analysis of a sediment sample may lead to different results depending on the sediment's properties and the extraction procedure. Extreme low TP recovery attenuated the practical application of ^{31}P NMR. For instance, 10 %–19 % of the P recoveries from Lake Taihu sediments were seen in an earlier study (Bai et al. 2009). This should be considered when drawing conclusions based on specific procedures (Ding et al. 2010).

Lake Dianchi, with an area of 309 km^2 and an average depth of 4.4 m, is the sixth largest lake in China, and is located in the Yunnan Province, (Fig. 1). This lake serves many social and economic purposes for the Kunming City, the capital of the Yunnan Province, which has 2.68 million residents in this basin, and a yearly burden of 216 million m^3 of household waste water and 47.6 million m^3 of industrial waste water (Yang et al. 2010). Presently, the lake water is heavily polluted and suffers from eutrophication. P is the uppermost limiting agent of the primary productivity in Lake Dianchi (Gao et al. 2005); the atomic ratio of N: P exceeds 31 in the water column (Hu et al. 2009). A recent survey of emission inventories has shown that the total P emissions approximated 542 t in 2008, and the municipal sewage and non-point source pollution accounted for 42.8 % and 29.4 %, respectively, of the P loading into Lake Dianchi (Gao et al. 2013). The average annual accumulation amount in the entire lake corresponded to 60 % of the exogenous P input (Zhang et al. 2005). Thus, high-nutrients accumulated sediments have been taken into account in previous restoration projects. Much effort has been focused on sedimentary inorganic P (Gao et al. 2005; Hu et al. 2007), whereas few studies have addressed the organic P cycle (Zhu et al. 2013), although it accounted for 21 %–30 % of total P in sediments from Lake Dianchi. In this study, the spatial distribution of P species, especially organic P in surface sediments of Lake Dianchi, was investigated by P fractionation and ^{31}P NMR. In addition, the potential effect of internal P loading on the water quality was also evaluated.

2 Methods

2.1 Study area

Lake Dianchi (24°28′–25°28′N, 102°29′–103°01′E) was divided in 1996 by the Haigeng Dam into two parts, Caohai and Waihai, covering, respectively, 2.7 % and 97.3 % of the total area (Fig. 1). As a result, Caohai became a dead zone with a low water flow and high pollutant concentration. The total drainage basin area of Dianchi Lake is approximately 2,920 km^2, which includes 29 main inflow rivers and the Haikou River as its only output. The lake has a relatively long residence time of 2–3 years, which would exacerbate retention and the accumulation of nutrients.

2.2 Sampling

Sampling was carried out in August 2011 during a heavy algal bloom in the north Waihai basin. 16 sites representing four lake regions were selected (Fig. 1), based on different geographical locations and sources of nutrients (Hu et al. 2007). Caohai is a submerged macrophyte dominant zone, with 2,707 t biomass and a coverage of 29.4 %. In contrast, Waihai is dominated by algal, and algal blooms have occurred frequently, especially in the Haigeng Bay, every year from April to November for the last two decades (Yang et al. 2010). The central zone of the lake is impacted by few anthropogenic influences, except some agricultural patches of rice, vegetables and flowers around its central-east zone. Phosphate rock and phosphoric chemical enterprises exist in the southern area.

For each site, three sediment cores were sampled using a gravity core sampler. The top 3 cm of the sediment of each core were sliced. This segment corresponds to the sediment which accumulated over the last 10 years and is prone to re-suspension (Zhang et al. 2005). Samples from each site were pooled and homogenized, in order to ensure the representativeness of the sediment input within the various catchments of the lake. All sediments were lyophilized at −80 °C, sieved to pass through a 100-mesh sieve, and then stored until analysis. Lake water was also collected approximately 0.5 m below the surface using a water sampler. Water samples were filtered using a 0.45 μm membrane, and preserved in acid washed polypropylene bottles. Unfiltered water samples were also collected and similarly stored.

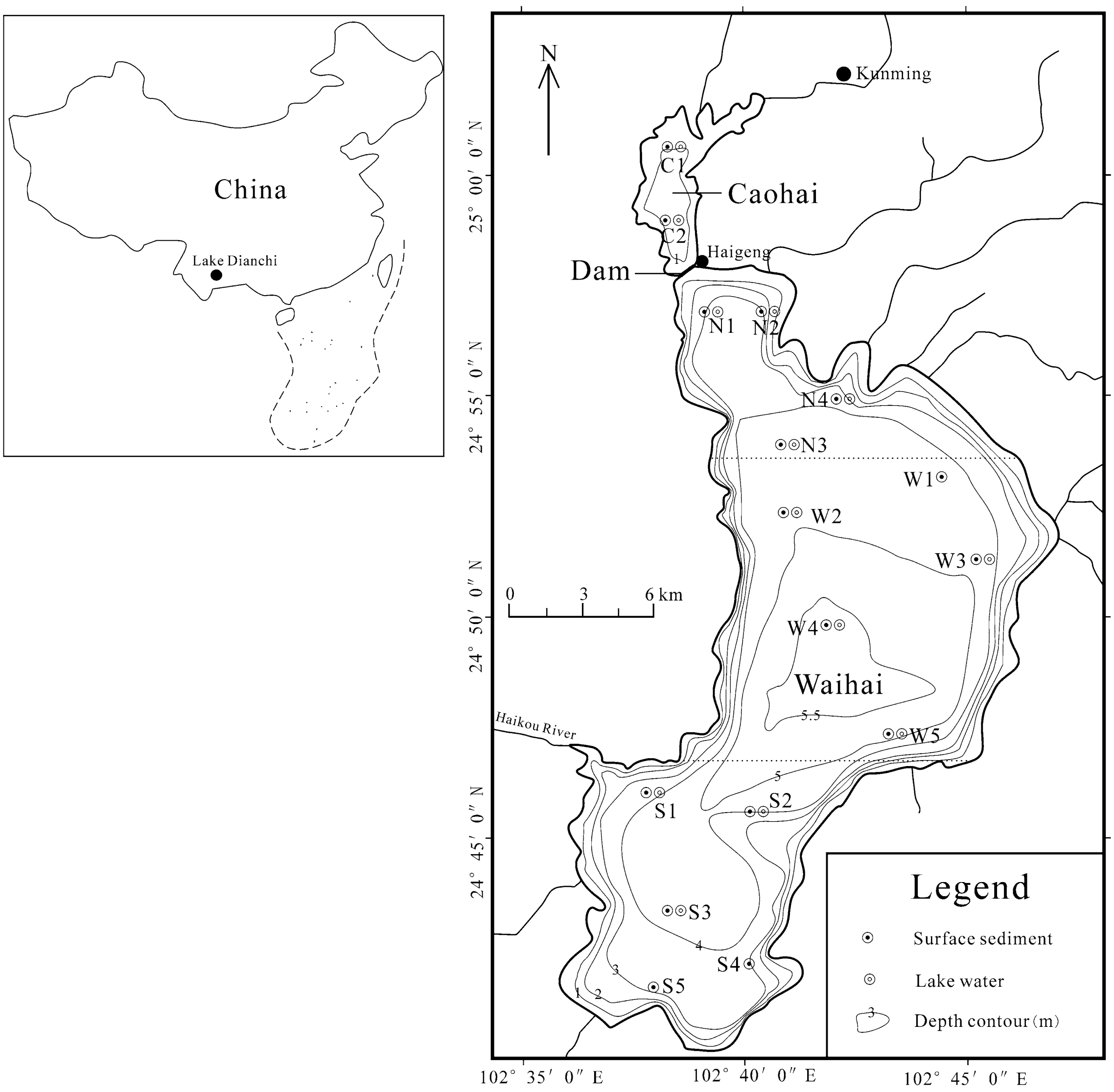

Fig. 1 A sketch map of the sampling sites in Lake Dianchi

2.3 Analysis

The total dissolved solids (TDS), dissolved O_2 (DO), pH and temperature (WT) of the water samples and chlorophyll a (chl a) were measured on site using portable multimeters (YSI Model 556MPS, USA). Total P (TP) in the water was determined after acid-persulphate digestion (Rowland and Haygarth 1997). Total contents of Fe, Al and Ca in sediments were measured with inductively coupled plasma-optical emission spectroscopy (ICP-OES) after digestion with HNO_3-HF-$HClO_4$. Sediment TP and organic P were determined according to the protocol of Ruban et al. (2001).

Sedimentary P fractionation was performed following the scheme of Hupfer et al. (1995), with sequential extraction by NH_4Cl, bicarbonate dithionite (BD), NaOH and HCl. These extracts were centrifuged and the soluble reactive P (SRP) in the supernatants was analyzed by the molybdenum-blue method (Drummond and Maher 1995). Additionally, TP in the NaOH extracts was determined as SRP after digestion in the same manner as the water sample treatment. This scheme classifies sediment P into loosely sorbed P (NH_4Cl-P), redox-sensitive P (BD-P), NaOH extracted P (NaOH-P) and Ca bound P (HCl-P). Residual-P is given by the difference between sediment TP and the sum of the other four P fractions. The calculation accuracy was checked by TP analysis on the residue with a recovery of over 90 %. All analyses were done in replicates (n = 3) and the mean value was reported.

To design an optimum procedure for sediment extraction for ^{31}P NMR analysis, three samples from other lakes were processed along with two sediments previously collected in

Lake Dianchi (Table 1). Three extraction procedures were compared including a single-step extraction with a mixed solution of 0.25 M NaOH and 0.05 M EDTA (Zhang et al. 2009), a 0.1 M NaOH extraction which followed a pre-extraction with a 0.067 M EDTA (Ahlgren et al. 2007), and a two-step extraction starting with 0.05 M EDTA and following with NaOH–EDTA (Ding et al. 2010). Extractions were made on 1 g of dry sediment, with the sediment to extractant ratio, 1:30 w/v. Samples were extracted at room temperature for 0.5 h for the pre-extraction, and 16 h for the principal extraction. Total P and SRP in the all extracts were analyzed spectrophotometrically after dilution of at least 25-fold to avoid EDTA interferences.

Based on P recovery using the above methods, sediments from sites C1, C2, N1, N2, W2, and S1–S5 with high Ca/(Fe + Al) were extracted by Ding et al. (2010), while the others were carried out according to Ahlgren et al. (2007). After centrifugation, supernatant in the principal extractant was concentrated 20 times by rotary evaporation at 30 °C, and was frozen at −20 °C until analysis. Before ^{31}P NMR measurement, 5 % (vol.) of BD solution was added in order to reduce interference from paramagnetic ions such as Fe and Mn, and 20 % (vol.) of D_2O was also added to obtain a stable signal. Samples were vortexed for 10 min and equilibrated for 5 min.

Solution ^{31}P NMR spectra were performed at 202.5 MHz on a JEOL ECX 500 MHz spectrometer (Japan) at 25 °C. Spectra were recorded using a 90° observe pulse and a total acquisition time 5.6 s, with a broadband proton decoupling and around 30,000 scans (Zhang et al. 2009). Chemical shifts were indirectly referenced to external 85 % H_3PO_4 standard. The spectra were processed using the JEOL software, Delta 5.0.1 (JEOL Ltd., Japan) for peak-picking and spectra integration. The contribution of individual P compound group was calculated relative to TP based on its peak area.

3 Results

3.1 Lake water

During this sampling campaign, WT showed little change, ranging from 21.9 to 23.2 °C (Table 2). The lake water was alkaline. Average pH value in Waihai basin was higher (9.87 ± 0.24) than that in Caohai basin (8.21 ± 0.23). Because of the shallow water body with intensive hydrodynamic disturbance, the water is oxic as a whole. Concentrations of TDS in the open Waihai basin were three folds of that from the Caohai basin. Among all sites, the highest content of chl a occured in the Haigeng Bay. Total P in the water exhibited a discernable downward trend from the Caohai to Waihai basin.

3.2 Surface sediment

Concentrations of P and several metal elements in surface sediments are listed in Table 3. Total P in sediments ranged from 1,330.0 to 3,711.4 mg kg^{-1}. Organic P in sediments ranged from 352.6 to 725.8 mg kg^{-1}, accounting for 19.6 %–33.8 % of total P. Total Ca ranged from 55.38 to 132.94 mg g^{-1}, Al from 37.70 to 79.14 mg g^{-1}, and Fe from 26.51 to 74.96. Concentration ratios of Ca/(Fe + Al) in sediments ranged from 0.38 to 2.03, with high values present in the macrophyte-dominated region and low values mostly in the central zone of Lake Dianchi. Ratios of Fe/P varied in the range of 10.3–52.3.

P fractionation results are shown in Fig. 2. In Caohai, the rank order of P fractions were NH$_4$Cl-P (23.4–51.5 mg kg^{-1}) < BD-P (114.2–278.6 mg kg^{-1}) < Residual-P (289.8–297.8 mg kg^{-1}) < HCl-P (538.4–552.8 mg kg^{-1}) < NaOH-P (1,338.5–1,395.7 mg kg^{-1}). NaOH-P was also dominant in the north tip of Waihai, with its relative contribution of 23.5 %–45.1 %. In contrast, Residual-P was the main fraction in the sediments from the central basin. HCl-P dominated in the south region, varying from 687.9 to 1,820.0 mg kg^{-1}, and contributing 33.5 %–49.0 % to total P.

3.3 Comparison of total P recovery from three different extraction procedures

Total P in the single-step extraction using NaOH–EDTA ranged from 239.3 to 764.6 mg kg^{-1} for the 5 sediment samples tested, with recovery rates from 25.6 % to 38.5 % (Fig. 3). The yield of P pre-extracted with 0.067 M EDTA

Table 1 Limnological and chemical properties of selected lake sediments

Name	Lake		Sediment				
	Water area km^2	Trophic status	TP mg kg^{-1}	Ca mg g^{-1}	Al	Fe	Ca/(Fe + Al)[a]
Caohai	8.34	Heavily eutrophic	1,985.9	138.03	34.77	27.42	2.22
Waihai	300.6		2,774.7	79.16	64.31	40.17	0.76
Lake Daihai	249.8	Moderately eutrophic	894.6	53.72	70.58	49.43	0.45
Lake Erhai	133.5	Mesotrophic	836.6	16.87	76.72	47.90	0.14
Lake Fuxian	216.6	Oligotrophic	1,031.5	68.24	73.59	27.86	0.67

[a] Concentration ratios of Ca to the sum of Fe and Al in sediments, calculated as weight basis

Table 2 Summary of water parameters in Lake Dianchi in August 2011

Basin	WT (°C)	pH	DO (mg L^{-1})	TDS (mg L^{-1})	Total P (mg L^{-1})	chl a (mg L^{-1})
Chaohai	22.2 ± 0.2 (22.0–22.3)	8.21 ± 0.23 (8.04–8.37)	2.66 ± 0.84 (2.06–3.25)	38.4 ± 12.4 (29.6–47.1)	0.90 ± 0.12 (0.82–0.98)	0.32 ± 0.12 (0.23–0.4)
Waihai						
North	22.9 ± 0.3 (22.5–23.2)	9.85 ± 0.26 (9.60–10.12)	7.32 ± 0.20 (7.05–7.56)	113.2 ± 9.1 (103.4–124.8)	0.29 ± 0.05 (0.22–0.33)	0.72 ± 0.15 (0.58–0.88)
Central	22.8 ± 0.2 (22.6–23.0)	9.98 ± 0.09 (9.84–10.06)	8.39 ± 0.47 (7.77–9.07)	123.3 ± 6.9 (113.8–133.1)	0.20 ± 0.06 (0.15–0.27)	0.48 ± 0.05 (0.41–0.53)
South	22.3 ± 0.3 (21.9–22.6)	9.78 ± 0.33 (9.24–10.05)	7.71 ± 0.20 (7.46–7.95)	112.2 ± 3.5 (107.9–115.6)	0.17 ± 0.02 (0.16–0.20)	0.32 ± 0.06 (0.26–0.37)

was significantly higher than that of 0.05 M EDTA, accounting for 9.1 %–17.0 % and 4.4 %–14.0 %, respectively. In the pre-extracts, most of the P (66 %–95 %) was inorganic. In the latter two-step treatment, 36.6 %–52.0 % of total P was effectively extracted by the principal extractants. The extraction yield was significantly higher for sediments with higher concentration ratios of Ca/(Fe + Al) using the Ding et al. (2010).

3.4 Sediment extraction and ^{31}P NMR analysis

As presented in Table 4, total sediment P pre-extracted with EDTA ranged from 110.7 to 452.8 mg kg^{-1} in Lake Didanchi, with the corresponding recovery from 6.9 % to 20.2 %. Inorganic P (an average of 97.1 %) was dominant in the pre-extracts. In the principal extraction step, NaOH and mixed reagents extracted TP ranged from 513.6 to 1,747.4 mg kg^{-1}, and the recovery rate ranged from 31.8 % to 69.3 %. Of extracted P, between 95.5 and 346.8 mg kg^{-1} P was present as organic P, accounting for 12.9 %–30.7 % of the principal extraction, and representing recoveries of 19.8 %–51.7 % (mean 39.4 %) of total organic P.

Four predominant P-containing groups were detected by ^{31}P NMR in the principal extracts, including ortho-P (5.5–7 ppm), monoester-P (3–5.5 ppm), diester-P (0 to −1 ppm) and pyro-P (−4 to −5) ppm (Fig. 4). Weak peaks in the range of 1–3 ppm have been identified as trace amount of P lipids (0.9 and 1.8 ppm) or teichoic acid P (2.3 ppm) (Cade-Menun 2005), which were also observed in a few sediments from sites C2, N1, N3 and S4. Ortho-P was the dominant P form in all sediment extracts, with its relative contribution of 68.2 %–87.7 %. Monoester-P ranged from 41.3 to 206.4 mg kg^{-1}. DNA-P was similar in abundance to pyro-P, with their corresponding proportions of 1.8 %–7.9 % and 0.8 %–8.2 %. In general, the relative proportion of organic P determined by ^{31}P NMR spectroscopy was 9.4 %–24.2 % (Table 4). Relationships between these P compounds in the sediments and TP in the overlying water were shown in Fig. 5.

4 Discussion

In Waihai, the water column showed a distinctly elevated pH due to the strong influence of the algal bloom. Lower DO was exhibited in the north part, compared to other regions (Table 2). The DO value was the lowest in the Caohai basin. Recent modeling results based on statistical analysis suggested that 6.8 % of the Lake Dianchi's volume is affected by hyproxia (DO $\leq$ 2.0 mg L^{-1}) under the current loading (Liu et al. 2014). As a whole, a notable decrease in total P appeared along a latitudinal gradient

Table 3 Selected chemical properties of sediments in Lake Dianchi

Site	Total P mg kg^{-1}	Organic P	Ca mg g^{-1}	Al	Fe	Ca/(Fe + Al)[a]	Fe/P[b]
C1	2,568.4	570.3	132.94	38.96	26.51	2.03	10.3
C2	2,312.4	590.4	122.32	37.70	26.66	1.90	11.5
N1	1,860.5	548.8	100.39	62.52	45.32	0.93	24.4
N2	2,198.3	670.8	97.93	64.14	47.18	0.88	21.5
N3	1,849.0	619.7	85.75	71.14	54.51	0.68	29.5
N4	1,330.0	449.9	84.63	74.01	69.54	0.59	52.3
W1	1,717.0	483.2	55.38	71.87	74.96	0.38	43.7
W2	2,223.3	622.4	83.23	66.25	48.74	0.72	21.9
W3	2,239.7	688.8	73.66	65.26	48.81	0.65	21.8
W4	2,202.7	620.3	64.65	79.14	55.97	0.48	25.4
W5	1,734.3	352.6	55.52	68.06	43.86	0.50	25.3
S1	2,054.7	625.5	90.92	69.73	47.41	0.78	23.1
S2	2,300.6	576.8	83.60	59.96	41.80	0.82	18.2
S3	2,203.6	535.3	84.50	58.58	41.26	0.85	18.7
S4	2,366.0	637.9	85.06	63.59	42.14	0.80	17.8
S5	3,711.4	725.8	80.16	63.21	40.17	0.78	10.8

[a] Concentration ratios of Ca to the sum of Fe and Al in sediments, calculated as weight basis

[b] Concentration ratios of Fe to total P in sediments, calculated as weight basis

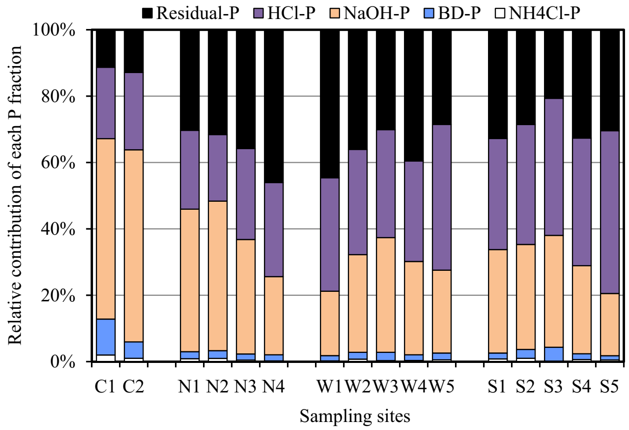

Fig. 2 Relative contributions of various P fractions in surface sediments

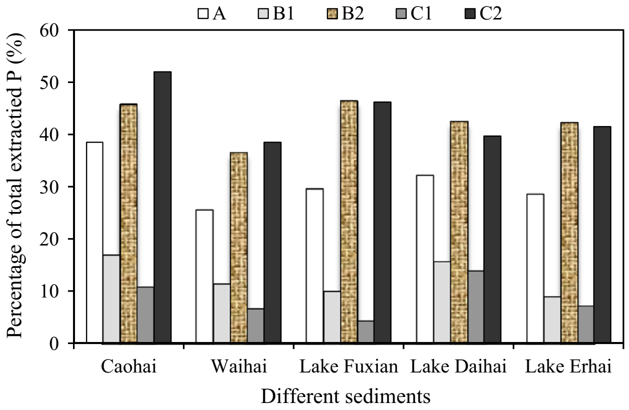

Fig. 3 Total extracted-P yield in given sediments by three different procedures. A represents the single-step 0.25 M NaOH-0.05 M EDTA, B1 and C1 denote the pretreatment with 0.067 M and 0.05 M EDTA, B2 and C2 represent the principal extraction using 0.1 M NaOH and NaOH–EDTA, respectively

with distance from Kunming City, likely in response to a reduction of external inputs. Noticeably, Caohai basin received more than 45 % of the total pollution loading from Kunming City (Gao et al. 2013).

Speciation data may shed further light on the processes that control transport, distribution of P and its lability in lake sediments (Fig. 2). NH_4Cl-P mainly means loosely adsorbed P on the surface of Fe and $CaCO_3$, soluble reactive P in interstitial water and leached P from deposited phytodetrital aggregates (Pettersson 2001). It was readily released from sediments and therefore represents the immediately available P for accumulation by phytoplankton (Rydin 2000). This fraction accounted for 0.2 %–2.0 % of the total sediment P, which is comparable to previous results from Lake Courtille (Hullebusch et al. 2003) and Lake Erken (Ahlgren et al. 2005).

BD-P and the molybdenum reactive NaOH-P (NaOH-rP) represent P bound to metal oxides, Fe or Mn, and Al, and are more informative for estimating the P load in the anoxic sediment layer. The relative contribution (1.3 %–10.8 %) of BD-P to TP is low in comparison with the values reported for other eutrophic lakes, such as Erken (16.7 %), Courtille (24.0 %), and Haussee (22.8 %, Gonsiorczyk et al. 1998). High pH and suboxic condition help the release of these P fractions from sediment. P amount released from sediment was several hundred times higher (27.7 mg) at pH 10.5 than that (0.16 mg) at pH 8.5 (Gao et al. 2005). Furthermore, redox potential (*E*h) in the oxic surface's 2 cm layer (>200 mV) rapidly declines with sediment depth, and sediments become

Table 4 Total P pre-extracted by Na_2-EDTA, and total P, organic P and P speciation mainly extracted by 0.1 M NaOH or 0.25 M NaOH-0.05 MNa_2-EDTA (mg kg^{-1})

Site	Total P in pre-extracts	Total P in principal extracts	Organic P in principal extracts	Ortho-P[c]	Monoester-P	Diester-P	Pyro-P
C1	389.0(15.1)[a]	1,747.4(68.0)[b]	224.6	1,434.7	81.3	88.4	143.0
C2	342.4(14.8)	1,603.2(69.3)	288.4	1,271.1	181.6	74.1	76.4
N1	139.3(7.5)	1,097.1(59.0)	272.3	748.3	206.4	59.0	83.4
N2	214.8(9.8)	1,309.6(59.6)	346.8	1,020.5	198.6	47.2	43.4
N3	204.3(11.0)	849.0(45.9)	260.6	632.6	90.8	67.1	58.5
N4	110.7(8.3)	513.6(38.6)	107.3	405.3	57.8	30.8	19.7
W1	119.8(7.0)	613.2(35.7)	95.5	537.6	41.3	25.6	8.7
W2	201.7(9.1)	1,012.6(45.5)	213.3	884.9	76.9	18.7	32.1
W3	452.8(20.2)	932.6(41.6)	250.1	729.0	105.6	63.5	34.5
W4	243.3(11.0)	905.5(41.1)	190.4	754.3	88.8	55.6	6.8
W5	243.4(14.0)	681.4(39.3)	175.4	518.3	88.7	50.3	24.1
S1	186.1(9.1)	936.9(45.6)	273.5	723.9	117.8	53.6	41.6
S2	273.2(11.9)	1,051.7(45.7)	267.6	818.0	151.9	45.4	36.4
S3	272.1(12.3)	1,009.4(45.8)	213.3	738.7	165.0	51.6	54.1
S4	162.7(6.9)	1,026.8(43.4)	284.7	748.6	173.0	58.5	46.7
S5	286.4(7.7)	1,181.1(31.8)	220.0	956.5	163.8	44.9	15.9

[a] The percentage of total P pre-extracted with Na_2-EDTA

[b] The percentage of total P mainly extracted by 0.1 M NaOH or 0.25 M NaOH-0.05 M Na_2-EDTA

[c] Detected by ^{31}P NMR

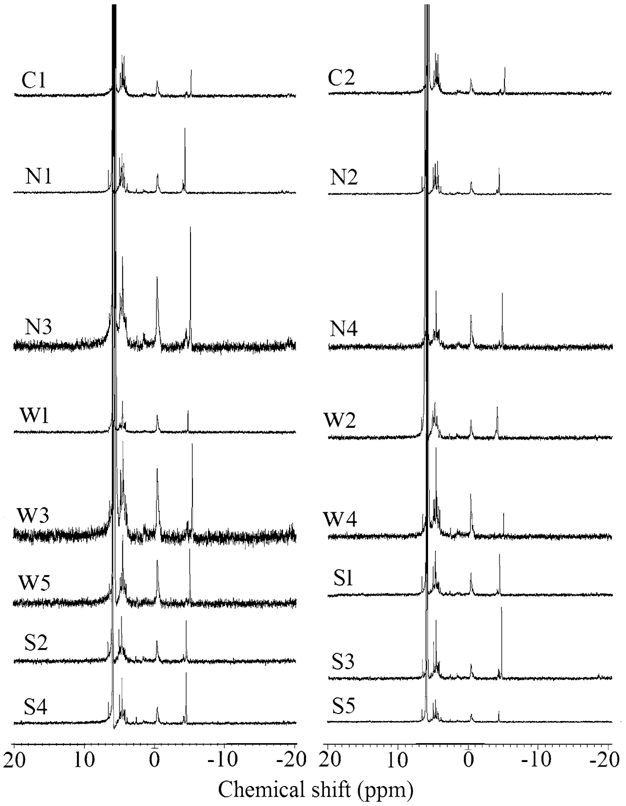

Fig. 4 ^{31}P NMR spectra of the principal extracts from Lake Dianchi

anoxic below 10 cm depth (*E*h < −200 mV) in Lake Dianchi. It's worth mentioning that there were remarkable differences of phosphate diffusive flux between, the layer under the oxic condition (−0.2 to 12.5 μmol m^{-2} d^{-1}) and the anoxic condition (1.8–96.2 μmol m^{-2} d^{-1}), through nature sediment core incubation from Lake Dainchi (Wang et al. 2010).

The Fe/P ratio is also an indicator of phosphate release from the sediments in shallow lakes. It may be possible to control internal P-loading by keeping the surface sediment oxidized, if the Fe/P ratio is above 15 (Jensen et al. 1992). The Fe/P ratio of the Waihai basin (10.8–52.3) was much higher than that of the Caohai basin (10.3–11.5) (Table 3), indicating the low BD-P or NaOH-rP release from the sediments. This finding is consistent with the diffusive flux of phosphate in Waihai (25.1 μmol m^{-2} d^{-1}) and in Caohai (234.3 μmol m^{-2} d^{-1}) calculated from the Fick's first law (Wang et al. 2010). The non-molybdenum reactive NaOH-P (NaOH-nrP) mainly comprised of polyphosphate and organic P bound in humic matrix. This fraction indirectly affects the availability levels of dissolved P for primary production (Hupfer et al. 1995). Specifically, mobilization of the recently sedimentary labile organic P could be the driving force of P release from this fraction (Gonsiorczyk et al. 1998).

HCl-P is assumed to consist mainly of apatite P, and is relatively stable in the lake under alkaline condition and

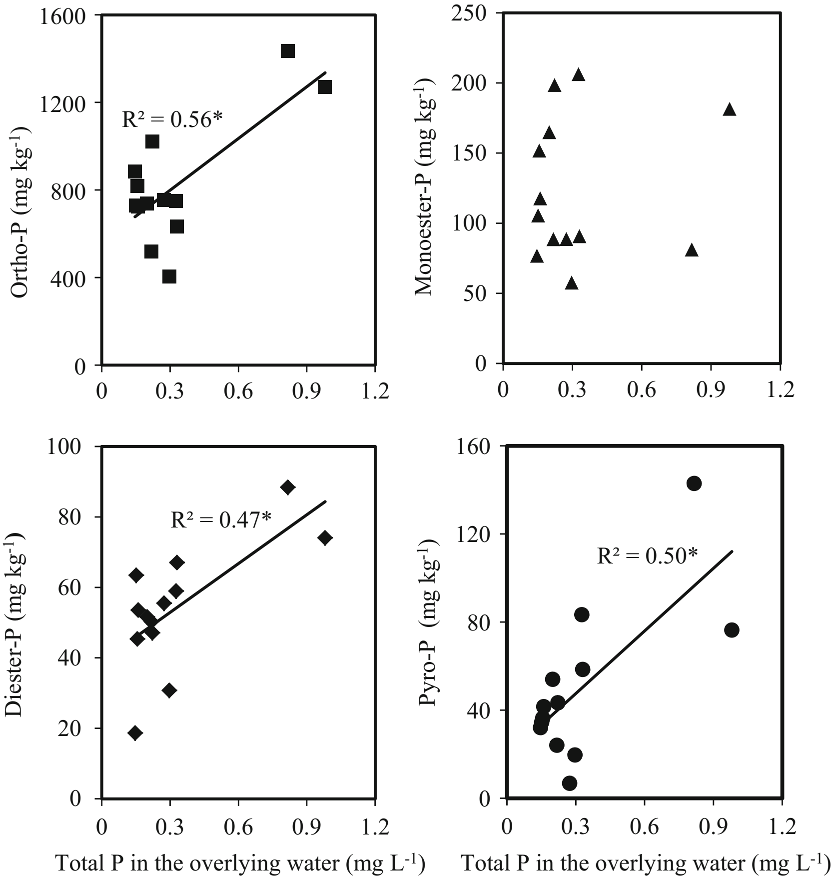

Fig. 5 Relationships between individual P compound concentrations in the principal extracts detected by ^{31}P NMR and total P in the water column (n = 13, $*P < 0.05$)

contributes to a permanent burial of P in sediments. A large amount of phosphate rock enriched in calcium of the early Cambrian Age exists in the south catchment (Gao et al. 2005). In addition, a fertilizer factory was built in the 1950s in this region. High HCl-P concentration in surface sediments depended partially on the co-precipitation of calcite with the gypsum and superphosphate in phosphate fertilizer manufacturing.

Residual-P is the most stable phase in sediment, and is hardly utilized by algal. In the central and deep water region of Lake Dianchi, allochthonous particulate organic matter underwent relatively weak hydrodynamic effect, and substantial recalcitrant fraction reached the bottom sediments and buried as this residual P fraction.

Labile P in sediments is a good index to estimate the potential contribution of sediment P to the overlying water. NH_4Cl-P, BD-P and NaOH-P are generally regarded as the most labile phases among P fractions (Rydin 2000). There is a positive correlation of labile P fractions in the sediments to TP in the water column ($R = 0.79$, $P < 0.01$, not shown). Hu et al. (2007) suggested that concentrations of labile P were reduced significantly during cyanobacterial blooms in Lake Diacnhi. When cyanobacterial blooms occur, it is expected that the concentration of NH_4Cl-P will be kept low due to algal rapid assimilation. The death and decomposition of cyanobacteria in turn makes the sediment more reductive. High temperatures in August enhanced the biological activity, which also lead to a lower of redox potential and accelerated the release of BD-P and NaOH-P to the overlying water.

In this study, the yield of TP was obviously higher than 10 %–19 % of Lake Taihu sediments (Bai et al. 2009) and slightly higher than the values (2.0 %–51.0 %) of Ding et al. (2010). Recovery of total organic P with NaOH–EDTA in Lake Dianchi was also higher than that extracted with pure NaOH (27 %–44 %) from Swedish lakes (Carman et al. 2000). Undoubtedly, the chelating ability of EDTA increased the efficiency of single NaOH extraction by breaking P-containing organo-metal complexes and causing less hydrolysis of P compounds (Ahlgren et al. 2007).

Unlike monoester-P, the other three P compounds in the sediments present strong correlations to TP in the water column (Fig. 5). It is interesting to note that concentrations of labile P factions determined by the traditional fractionation procedure correlated significantly with the sum of these three P compounds by ^{31}P NMR analysis (Fig. 6), indicating their important roles in sustaining lake eutrophication.

Ortho-P is the simplest P compounds, and exists in sediments as Fe-, Al- and Ca-bound P. When soluble reactive P in the overlying water column is low, sedimentary ortho-P

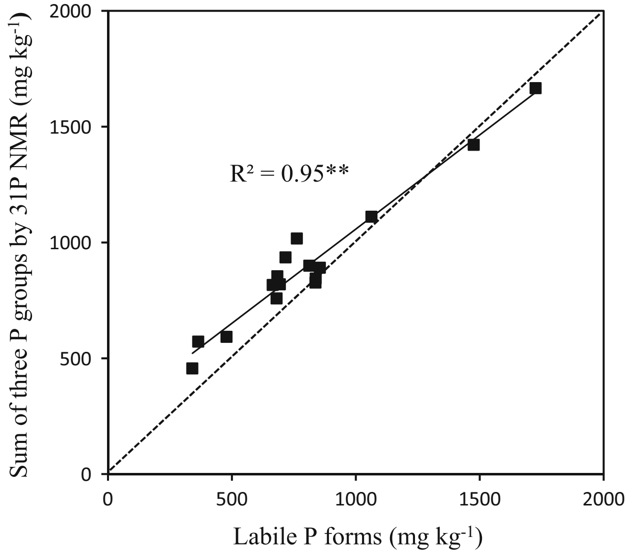

Fig. 6 Correlation of labile P factions by fractionation with sum of ortho-P, diester-P and pyro-P detected by ^{31}P NMR (n = 16,**P<0.01, the *dotted line* represents the 1:1 slope)

would be released and serve as a source of nutrients for algae and phytoplankton utilization. A small proportion of Pyro-P is in agreement with previous reports (Ding et al. 2010). Ahlgren et al. (2005) showed that pyro-P was only discernable in the top 4 cm of the sediments. The proportion of pyro-P in sediments was lower than in suspended particles from Lake Kasumigaura, suggesting that pyro-P could be released during or after sedimentation (Shinohara et al. 2012). Cade-Menun (2005) claimed that pyro-P concentration increases with urbanization degree. Relative high abundance of pyrop-P in Caohai and north Waihi agreed with the rapid expansion of the city in the corresponding basin.

Diester-P generally consists of DNA-P, P lipids and teichoic-P. Organic P compounds between 0 and −1 ppm were often assigned to DNA-P (Cade-Menun 2005). The source of DNA-P is bacterial DNA and phytoplankton decomposition (Zhang et al. 2009). It is conspicuous that this compound is present in higher abundance in the summer (47.9 %) than in the autumn (33.2 %) or spring (18.9 %) in the relevant study of Goczałkowice Reservoir (Młynarczyk et al. 2013). This difference may be attributed to algal blooms and the decomposition of phytoplankton.

Monoester-P comprises of sugar phosphates, mononucleotides and phytic acid. Intensive signals at 4.0–5.5 ppm in all extracts might indicate high concentrations of phytic acid (Cade-Menun 2005). Phytic acid is considered to be a major component in many types of sediment (Carman et al. 2000; Zhang et al. 2008). Due to the higher charge density, it could form strong complexes with metal cations and be resistant during acid and base hydrolysis (Zhang et al. 2009). Hence, these clustered monoesters-P species are less labile to decomposition.

Mono- and diester-P, and pyro-P can be considered biogenic. These biogenic P fractions collectively contributed 12.3%–31.8 % to TP in the NaOH-EDTA extracts and are close to 22.6 %–30.0 % in Lake Kasumigaura (Shinohara et al. 2012). Rydin (2000) claimed that 50 % of the non-molybdenum reactive NaOH-P in the surface sediment of Lake Erken is biogenic and can be degraded and eventually recycled to the overlying water. In Lovejoy Pond, the decrease of 8.2 mmol m^{-2} of the sediment biogenic P in the top 2 cm was comparable to the 6.5 mmol m^{-2} increase of ortho-P during the summer season, implying that some of the mineralized organic P re-partitioned to the solid-phase (Amirbahman et al. 2013).

Organic P can partially become inorganic due to some mechanisms, such as phosphatase hydrolysis, bacterial degradation and photolysis (Zhang et al. 2008). The sum of hydrolysable organic P in the H_2O, $NaHCO_3$ and NaOH fractions using Hedley's procedure ranged from 62.5 to 147.0 mg kg^{-1}, and accounted for 12.1 %–27.2 % of the total sedimentary organic P in Lake Dianchi by enzymatic hydrolysis (Zhu et al. 2013). Because ortho-P predominates in the studied sediments, further investigation is needed to clarify the transformation mechanisms of biogenic P. These studies could include bioavailability evaluation of various P species using the enzymes activities and oxygen isotopic tracing of phosphate (Liang and Blake 2009; Zhu et al. 2013).

5 Conclusion

P composition in surface sediments and its release to the water column contributes to the deterioration of water quality in Lake Dianchi. Several P fractions including NH_4Cl-P, BD-P and NaOH-P are potentially released from sediments and constitute the main source for the eutrophication of Lake Dianchi. These labile P fractions ranged from 340.6 to 1,725.8 mg kg^{-1}, accounting for 20.5 %–67.2 % of the total P. Organic P species also supplied P to the water column, which contributed 19.6 %–33.8 % of the total P. The high pH of lake water and the formation of hyproxia following algal death further accelerated the P release. The release of labile P and appearance of the algal bloom were stimulated reciprocally.

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