Contents lists available at ScienceDirect







journal homepage: www.elsevier.com/locate/jhazmat

### Facilitation of phosphorus adsorption onto sediment by aquatic plant debris

### S.T. Du<sup>a,b</sup>, J.L. Shentu<sup>b</sup>, B.F. Luo<sup>a</sup>, I.H. Shamsi<sup>a</sup>, X.Y. Lin<sup>a</sup>, Y.S. Zhang<sup>a</sup>, C.W. Jin<sup>a,\*</sup>

<sup>a</sup> MOE Key Laboratory of Environment Remediation and Ecosystem Health, College of Environmental and Resource Science, Zhejiang University, Hangzhou 310058, China <sup>b</sup> College of Environmental Science and Engineering, Zhejiang Gongshang University, Hangzhou 310035, China

#### ARTICLE INFO

Article history: Received 4 September 2010 Received in revised form 11 April 2011 Accepted 16 April 2011 Available online 22 April 2011

Keywords: Cellulose Glucose Microorganism Sediment Phosphorus Plant debris

#### ABSTRACT

Aquatic plant debris in lakes or rivers may affect phosphorus flux in water-sediment systems. In this study, either aquatic plant debris or typical plant components (cellulose or glucose), were added into a system of sediment (50 g) and overlying water (2 L) with different initial SRP (soluble reactive phosphorus) concentrations to investigate the impact. After 18 days of treatment with 4 g of plant debris, the SRP in the overlying water for 0.5 and  $2 \text{ mg L}^{-1}$  initial SRP tests at 30 °C decreased by 41 and 53%, respectively, compared to the treatments without plant debris. Cellulose and glucose treatments gave similar results as plant debris treatment. When the water-sediment system was sterilized, the cellulose-or glucose-facilitated decrease in SRP vanished. Additionally, in the non-sterilized system, the glucose treatment significantly increased both the microbial biomass carbon and the microbial biomass phosphorus in the sediment. Although total phosphorous in the sediment increased with glucose treatment, its water soluble and iron associated inorganic fractions, two labile phosphorus fractions, were clearly reduced. Our results suggest that the short-term retention of plant debris in water systems facilitates a decrease in overlying water SRP through microbe-mediated mechanisms of phosphorus adsorption and stabilization in sediment.

© 2011 Elsevier B.V. All rights reserved.

#### 1. Introduction

Water eutrophication has become a serious environmental problem worldwide [1,2]. Severe algal blooms due to eutrophication can not only lead to unpleasant taste and odor of water, but also cause water supply filtration problems and subsequent public health concerns [3]. Consequently, the factors that promote water eutrophication have been extensively studied in recent decades. Available phosphorus (P) in the water is considered one of the most important factors responsible for eutrophication [4–6]. The amount of available P in the overlying water is mainly determined by the following two processes: external phosphorous loading and phosphorous exchange between the sediment and the overlying water. The latter process is particularly interesting to many researchers, because the sediment can function either as an internal P load or as P sink for its overlying water [7]. Internal P loading facilitates algal blooms, while internal P sinking improves water quality [8]. Therefore, many researchers have focused on investigating the factors affecting P behavior at the sediment/water interface [9–11]. Traditionally, the cycling of P at the sediment/water interface was considered an abiotic process [12]. Recently, there has been evidence that biotic processes mediated by microorganisms are also involved in P cycling [13–15]. Therefore, any factors affecting the growth of microorganisms should also be expected to influence P cycling at the sediment/water interface.

Most lakes and rivers contain large distributions of aquatic plants. For example, according to an investigation of Gu et al. [16], the area of aquatic plants in East Taihu Lake in China accounted for more than 70% of the total water area. Because of the purification properties associated with aquatic plants in their acquisition of nutrients from the water column, restoration of aquatic plants is encouraged in many cases. This restoration is encouraged particularly in shallow lakes or rivers, where the area covered by aquatic plants is more than 50% of the total water area [17]. Most of the aquatic plants are herbaceous, and thus their life cycles are usually completed within months. Harvesting of aquatic plants at their maximum biomass stages in these controlled lakes and rivers can remove the nutrients that were previously acquired by plants. However, in uncontrolled lakes and rivers (which are prevalent in China), aquatic plants generally remain in water systems after death and produce small organic compounds during decay [18,19]. Formation of these small organic compounds can serve as a carbon (C) source for the growth of microorganisms. Therefore, the plant debris remaining in water systems may alter the behaviors of microorganism-mediated phosphorous fluxes between the sediment and its overlying water. At the moment limited information is available to support this hypothesis.

<sup>\*</sup> Corresponding author. Tel.: +86 571 88982478. *E-mail address:* jincw@zju.edu.cn (C.W. Jin).

<sup>0304-3894/\$ -</sup> see front matter © 2011 Elsevier B.V. All rights reserved. doi:10.1016/j.jhazmat.2011.04.067

Table 1
The basic physico-chemical properties of testing sediments

рН	7.64	
$TP(mg kg^{-1})$	555	
TN (g kg <sup>-1</sup> )	1.91	
LOI (%)	3.02	
$CEC (cmol kg^{-1})$	23.02	

In this study, sediment taken from a lake covered with a large quantity of aquatic plants was used to investigate the effect of plant debris on P flux between sediment and its overlying water. Whether the microorganisms mediated this behavior was also examined.

#### 2. Materials and methods

#### 2.1. Sediment collection and characterization

Taihu Lake (119°54′-120°36′N, 30°56′-31°33′E) is located in the Yangtze River Delta and has a surface area of 2338 km<sup>2</sup> [20]. The sediment used in the present study was collected from the Gonghu region in the northeast part of Taihu Lake, which has a surface area of 176 km<sup>2</sup> and a mean depth of 1.8 m. Water in this region is frequently agitated because of wind and water transport vessels, which create waves. These factors allow oxygen to be easily dissolved into the water [21]. The sediment sample was collected in May 2008. A sampling location covered with dense aquatic plants was selected. The aquatic plants mainly include Alternanthera philoxeroides Grise, Potamogeton malaianus, Nymaphoides peltata and Myriophyllum spicatum L. The sediment sample was collected and prepared as described by Jin et al. [21]. The surface sediment (0-10 cm) was collected using a Petersen grab sampler, immediately stored in iceboxes (<4°C) and taken to the laboratory. The sediment was then freeze-dried and ground, passed through a 1.8mm nylon sieve and stored at 4 °C for subsequent experiments. All experiments in this study were conducted under aerobic conditions.

The pH of the sediment was measured in 1:2.5 (w/v) solid/water suspensions with a pH meter (Sartorious, PB-10, Germany). The concentrations of total nitrogen (TN) and total phosphorus (TP) in sediment were measured according to the methods of Müller et al. [22]. The total organic matter content was estimated by loss on ignition (LOI) at 550 °C [23]. The cation exchange capacity (CEC) of the sediment was analyzed using the EDTA-NH<sub>4</sub><sup>+</sup> method [21]. Selected properties of the sediment are summarized in Table 1.

#### 2.2. Incubation experiments

*A. philoxeroides* Griseb, a typical aquatic plant, was used to investigate the effect of aquatic plant debris on P flux between sediment and overlying water. The fresh plants were cut into short pieces. Fifty grams of sediment and 2 L of overlying water with an initial SRP (soluble reactive phosphorous) concentration at 0.5 or 2 mg L<sup>-1</sup> were mixed into a polyethylene container. The initial SRP in the overlying water was added in the form of KH<sub>2</sub>PO<sub>4</sub>–P. Then 4 g of plant pieces were added into the water–sediment mixture and, along with a control solution (i.e. no plant debris added), were incubated in an orbital shaker at 90 rpm in dark conditions at 10 or 30 °C. The two temperatures used here were to examine whether the plant debris-mediated P flux was temperature dependent or not. The SRP in the overlying water was monitored every 6 days with the ammonium molybdate–ascorbic acid method as described by Murphy and Riley [24].

Because cellulose comprises a large portion of plant anatomy (about 40–50% of the biomass by dry weight [25]), we used this macro-molecule carbohydrate instead of actual plant debris for further confirmation of the function of aquatic plant debris. The water-sediment mixtures were prepared as above. Then 0, 50 or 100 mg of cellulose was added into the mixtures, which were then incubated in an orbital shaker at 90 rpm in dark conditions at  $30 \,^{\circ}$ C. The SRP in the overlying water was also monitored every 6 days.

Glucose is another important plant component, as it is the basic unit of macro-molecule carbohydrates. In turn, macro-molecule carbohydrates can also be degraded into glucose in microbeinhabited environments [18]. Therefore, the effect of glucose on P flux in a water-sediment system is another focus of this study. Glucose was also used to study the possible mechanisms of plant debris affecting P flux in water-sediment system. To begin, 50 g of sediment was mixed into 2L of overlying water with an initial SRP concentration of 0.5, 1, 2 or 8 mg L<sup>-1</sup>. Then, glucose was added into the mixtures at a concentration of 0, 50, 100 or  $200 \text{ mg L}^{-1}$ , and incubated in an orbital shaker at 90 rpm in dark conditions at 30 °C. Soluble reactive phosphorous in the overlying water was monitored after 0, 6, 12, 24, 48 and 72 h of incubation. In order to more accurately evaluate the variations of P fractions, microbial biomass C and microbial biomass P in the sediment, the sediment with 8 mg L<sup>-1</sup> initial SRP concentration was collected after 72 h of incubation for subsequent analysis.

#### 2.3. Sterilization and inoculation experiments

The role of microorganisms in mediating the effect of cellulose and glucose on the P flux between sediment and its overlying water was investigated with sterilized and non-sterilized sediments. For the sterilized treatment, 1 g of sediment with 39 mL of deionized water in 50-mL centrifuge tubes was first autoclaved at 121 °C for 30 min. Then, 1 mL of pre-sterilized KH<sub>2</sub>PO<sub>4</sub> and cellulose powder or glucose solution was mixed into the overlying water under axenic conditions, with the final concentration of phosphorous at  $2 \text{ mg L}^{-1}$  and cellulose or glucose at 0 or  $100 \text{ mg L}^{-1}$ , respectively. For non-sterilized treatments, sterilized sediment solution was first mixed with the KH<sub>2</sub>PO<sub>4</sub> and cellulose or glucose as described above. Then the mixture was re-inoculated with 100 µL of sediment suspension inoculums. The suspension inoculums were prepared by mixing 1 g unsterilized sediment with 30 mL deionized water and cultivating the mixture for weeks at room temperature. Both the sterilized and non-sterilized treatments were incubated in an orbital shaker at 90 rpm in dark conditions at 30 °C. The SRP in the overlying water was measured after 18 days for cellulose treatment and 3 days for glucose treatment, respectively.

#### 2.4. Microbial biomass C and microbial biomass P

Microbial biomass C and microbial biomass P concentrations were determined in 1g samples by extracting chloroformfumigated (24-h fumigation) and non-fumigated samples for 30 min, with 30 mL of 0.5 M K<sub>2</sub>SO<sub>4</sub> for biomass C (BC) and 0.5 M NaHCO<sub>3</sub> (pH 8.5) for biomass P (BP), respectively [26,27]. Carbon in K<sub>2</sub>SO<sub>4</sub> and phosphorus in NaHCO<sub>3</sub> extracts were determined by a Shimadzu TOC-5000 Analyzer and the ammonium molybdate–ascorbic acid method described by Murphy and Riley [24], respectively. Biomass C and biomass P were then estimated by the equations BC = EC × 2.64 [26] and BP = EP × 2.64 [28], where EC and EP are the differences between organic C and NaHCO<sub>3</sub>–P<sub>i</sub> extracted from the fumigated and non-fumigated sediments.

#### 2.5. Measurement of P fractions

Because the microbial activity was thought necessary for glucose to affect P flux in a water–sediment system, the changes in organic P fractions in sediment after glucose treatment could be more significant than those of inorganic P fractions without glucose treatment. Therefore, in this study, we used the P fractionation



Fig. 1. Schematic representation of the sequential P fractionation scheme. P<sub>i</sub> and P<sub>o</sub> denote orthophosphate and organic phosphate, respectively.

method of McDowell and Koopmans [29] (Fig. 1), which allowed us to analyze several organic P fractions in sediment. In this procedure, 1 g of sediment was sequentially extracted with 30 mL of deionized water, 0.05 M Ca–EDTA (+1% Na-dithionite, pH 7.8), 0.1 M Na–EDTA (pH 4.5), 0.5 M H<sub>2</sub>SO<sub>4</sub>, cold 0.5 M trichloroacetic acid (TCA; 0 °C), hot 0.5 M TCA (95 °C), 2 M NaOH (90 °C), and finally digested by K<sub>2</sub>S<sub>2</sub>O<sub>8</sub>. These fractions represent, in sequential order, water soluble P (H<sub>2</sub>O–P), Fe associated P (Fe–P), Ca–Al associated P (Ca–Al–P), acid soluble organic P (ASOP), sugar bound organic P (P<sub>0</sub>), nucleic acid P<sub>0</sub> and polyphosphate, humic bound P<sub>0</sub> and phytate, and residual P. Following extraction, each sediment-suspension was centrifuged (5000 × g) for 10 min and decanted, and then an aliquot was taken for P determination. For Fe–P and Ca–Al–P fractions, the organic P fraction was defined as the difference between P detected before and after digestion by K<sub>2</sub>S<sub>2</sub>O<sub>8</sub>.

#### 2.6. Statistical analysis

All treatments in this study were repeated four times. Values presented in this manuscript were the means  $\pm$  standard error of four replicated experiments. The overall significance of differences between treatments was determined using ANOVA followed by Dunnett's Test (*P*<0.05). All statistical analyses were conducted with SAS software (SAS Institute, Cary, NC, USA).

#### 3. Results

## 3.1. Effects of plant debris, cellulose and glucose on P flux between sediment and overlying water

At both 0.5 and  $2 \text{ mg L}^{-1}$  initial SRP tests, the SRP concentration in the control overlying water (i.e. no plant debris added) decreased with an increase in incubation time at both 10 and 30 °C (Table 2). The addition of plant debris significantly accelerated a reduction in SRP, with the more dramatic decreases seen at 30 °C. After 18 days of incubation at 10 and 30 °C, the SRP concentration in overlying water with plant debris added decreased by 28 and

41%, respectively for 0.5 mg  $L^{-1}$  initial SRP test, and by 30 and 53%, respectively for  $2 \text{ mg } L^{-1}$  initial SRP test, compared to those in the control overlying water (Table 2).

Cellulose addition had effects similar to the plant debris on SRP decrease at 30 °C (Fig. 2). On the 18th day of incubation, cellulose treatments of 50 and 100 mg L<sup>-1</sup> decreased SRP concentration in the overlying water by about 30 and 40%, respectively at 0.5 mg L<sup>-1</sup> initial SRP test, and by about 25 and 35%, respectively at 2 mg L<sup>-1</sup> initial SRP test, compared to cellulose-free controls.

Glucose addition also decreased SRP concentrations. The SRP decrease accelerated as more glucose was added (Fig. 3) and was prominent at tests of initial SRP lower than  $2 \text{ mg L}^{-1}$  (Fig. 3a–c). In these initial SRP tests, the SRP concentration of overlying water in the 200 mg L<sup>-1</sup> treatment was less than 50% of the SRP concentration in the glucose-free treatment after 72 h of incubation.

# 3.2. Role of microorganisms in mediating cellulose-enhanced and glucose-enhanced P reduction in overlying water

Since the cellulose and glucose had similar effects on P behavior with plant debris, these two carbohydrates were used to investigate how plant debris facilitated P reduction in overlying water. In the sterilized system, the addition of both 100 mg L<sup>-1</sup> cellulose and 100 mg L<sup>-1</sup> glucose did not affect the SRP concentration in overlying water, whereas in the non-sterilized system, the SRP concentration was significantly lowered by the same treatments (Fig. 4). The results demonstrate that microorganisms play a central role in organic matter-facilitated SRP decrease in overlying water.

# 3.3. Effects of glucose on microbial biomass C, microbial biomass P and P fractions in sediment

After 72 h of incubation, the microbial biomass C in the sediment was increased by 27%, 68% and 89% in 50, 100 and 200 mg L<sup>-1</sup> glucose treatments, respectively, compared to the glucose-free treatments (Fig. 5a). Both microbial biomass P and C/P ratios also increased with an increase in glucose concentration (Fig. 5b and c).



**Fig. 2.** Effects of cellulose addition on SRP flux between sediment and overlying water. (a and c) Initial P concentration at 0.5 and  $2 \text{ mg L}^{-1}$ , respectively, depicts the SRP concentration in overlying water with or without cellulose addition. Error bars represent the S.E. (n = 4). (b) Initial P concentration at 0.5 mg L<sup>-1</sup> and (d) initial P concentration at 2 mg L<sup>-1</sup> depicts the percentage of SRP concentration in cellulose added overlying water relative to that in no cellulose added overlying water.



**Fig. 3.** Effects of glucose addition on SRP flux between sediment and overlying water. The initial P concentration in overlying water were 0.5 (a and e), 1 (b and f), 2 (c and g) and 8 (d and h) mg L<sup>-1</sup>, respectively. (a–d) The SRP concentration in overlying water with glucose addition. (e–h) The percentage of SRP concentration in glucose added overlying water relative to that in no glucose added overlying water. Error bars represent the S.E. (*n*=4).

### 216 **Table 2**

Effect of plant debris addition on SRP decrease in overlying water. RP denotes the percentage of SRP concentration in plant debris added overlying water relative to that in control (i.e. no plant debris added) overlying water.

Initial SRP concentration (mg L <sup>-1</sup> )	Incubation time (d)	10°C			30 °C			
		Control	Plant debris addition	RP (%)	Control	Plant debris addition	RP (%)	
		SRP concentration (mg L <sup>-1</sup> )			SRP concentration (mg L <sup>-1</sup> )			
0.5	0	$0.50\pm0.02$	$0.53\pm0.02$	105	$0.51\pm0.02$	$0.54\pm0.02$	105	
	6	$0.34\pm0.02$	$0.32\pm0.01$	96	$0.26\pm0.02$	$0.25 \pm 0.01$	94	
	12	$0.19\pm0.01$	$0.16 \pm 0.01^{*}$	84	$0.14\pm0.01$	$0.11\pm0.00^{*}$	78	
	18	$0.11\pm0.00$	$0.08\pm0.01^{*}$	72	$0.09\pm0.00$	$0.05 \pm 0.01^{*}$	59	
2	0	$2.10\pm0.04$	$2.18\pm0.05$	104	$1.94\pm0.03$	$2.01\pm0.04$	104	
	6	$1.11\pm0.09$	$1.09\pm0.01$	98	$0.73\pm0.02$	$0.64 \pm 0.02^{*}$	87	
	12	$0.65\pm0.03$	$0.58 \pm 0.01^{*}$	89	$0.32\pm0.03$	$0.19 \pm 0.03^{*}$	59	
	18	$0.54 \pm 0.01$	$0.38\pm0.04^{*}$	70	$0.25\pm0.01$	$0.12\pm0.00^{*}$	47	

Values represent the means  $\pm$  S.E. (n = 4).

\* Significant difference (*P*<0.05) between the control and plant debris addition treatment.



**Fig. 4.** Effect of sterilization on the cellulose (a) and glucose (b) mediated SRP flux between sediment and overlying water (initial P at  $2 \text{ mg L}^{-1}$ ). The SRP in the overlying water was measured after 18 days for cellulose treatment and 3 days for glucose treatment, respectively. Error bars represent the S.E. (*n*=4). An asterisk denotes significant difference (*P*<0.05) between 0 and 100 mg L<sup>-1</sup> cellulose or glucose treatments.



Fig. 5. Effects of glucose addition on the microbial biomass C (a), P (b) and C/P (c) of sediment. Error bars represent the S.E. (*n*=4). Different letters indicate significant differences (*P*<0.05) among different glucose treatments.

Table 3

Mean P forms (mg kg<sup>-1</sup>) in the sediment after 72 h of treatment with the glucose at 0, 50, 100 and 200 mg  $L^{-1}$ .

Glucose concentration (mg L <sup>-1</sup> )	Н <sub>2</sub> О-Р	Fe-P <sub>i</sub>	Fe-P <sub>o</sub>	Ca-Al-P <sub>i</sub>	Ca-Al-P <sub>o</sub>	ASOP	Sugar bound P <sub>o</sub>	Nucleic acid P <sub>o</sub> and polyphos- phate	Humic bound P <sub>o</sub> and phytate	Residual P
0	$34.2\pm0.3a$	$197\pm4.8a$	$42.4\pm3.2c$	$169\pm8.2a$	$82.0\pm4.8a$	$153\pm14.1c$	$54.1 \pm 5.9c$	$10.8\pm0.7b$	$56.3 \pm 3.9c$	$89.7\pm6.3a$
50	$20.5\pm0.5b$	$176\pm5.6b$	$58.1\pm2.8b$	$166\pm7.3a$	$79.2\pm5.1a$	$229\pm10.2b$	$89.7\pm4.4b$	$14.7\pm3.8ab$	$75.1\pm4.1b$	$91.8\pm5.9a$
100	$13.2\pm0.4c$	$154\pm6.2c$	$77.4 \pm 3.1a$	$165 \pm 3.1a$	$82.1\pm6.7a$	$254 \pm 11.8a$	$92.8\pm6.3ab$	$16.2 \pm 1.2a$	$84.2\pm6.2b$	$91.8 \pm 4.5 a$
200	$12.7 \pm 0.4 c$	$146\pm7.9c$	$76.6\pm1.6a$	$162\pm 6.5a$	$76.3\pm4.5a$	$248 \pm 7.2a$	$99.8\pm3.7a$	$19.2\pm3.9a$	$97.6\pm2.9a$	$90.2\pm4.3a$

Values represent the means  $\pm$  S.E. (n = 4). Different letters indicate significant differences (P < 0.05) among different concentrations of glucose treatments. P<sub>i</sub> and P<sub>o</sub> denote inorganic and organic P forms, respectively.

For P fractions, both  $H_2O-P$  and Fe-associated inorganic  $P(Fe-P_i)$  decreased as glucose concentration increased. For a 200 mg L<sup>-1</sup> glucose treatment,  $H_2O-P$  and Fe- $P_i$  decreased by 63 and 26%, respectively, compared to those in glucose-free treatment (Table 3). In contrast, elevation of glucose concentrations increased Fe associated organic P (Fe- $P_0$ ), ASOP, sugar bound  $P_0$ , nucleic acid  $P_0$  and polyphosphate, and humic bound  $P_0$  (Table 3).

#### 4. Discussion

By using short-term incubation experiments, we have demonstrated in this study that plant debris remaining in water systems could contribute to an SRP decrease in overlying water (Table 2). The plant debris itself contains a certain amount of elemental P that is released into the overlying water during decomposition. Therefore, the actual contribution of plant debris to the decrease in SRP concentration in the overlying water may be underestimated in this study. During decomposition, aquatic plants are physically leached of soluble organic material (e.g., glucose) at the early phase [30], and then the relatively stable components, such as cellulose, are gradually degraded into small molecular forms of organic compounds in the microbe-inhabited environment [18]. We found that both cellulose and glucose clearly facilitated the SRP decrease in overlying water (Figs. 2 and 3). The results further confirm the function of plant debris. In recent decades, sediment dredging has been regarded as a useful strategy to reduce P in lakes and rivers [31,32]. Thus, if plant debris is not causing harm to the water system, we recommend that before, sediment dredging, the plant debris should be retained in the water ecosystems for few days, which may increase the efficacy of sediment dredging for removing P from the water.

It has been demonstrated that microbial processes are involved in the P flux between sediment and overlying water [13-15]. In addition, Boström et al. [33] found that microbial biomass P accounts for up to 13% of total P in the surface sediment of Lake Vallentunasjön. Therefore, changing P flux by plant debris may be achieved through microbial action. Our results show that the effect of plant debris on the SRP decrease in overlying water was more prominent at 30 °C than at 10 °C (Table 2), which may be attributed to the fact that higher temperatures are preferable for microbial growth [34]. In addition, glucose, a more available plant component to microbes than macro molecule carbohydrates [35], functions faster than cellulose in reducing SRP in overlying water (Figs. 2 and 3). All of these results support the above deduction. More importantly, in the sterilized water-sediment system, cellulose/glucose addition did not affect the SRP flux between sediment and overlying water, whereas cellulose/glucose did show an effect on the SRP flux in the non-sterilized water-sediment system (Fig. 4). These results confirm the microbial role in plant debris enhancing P behavior.

The question of how the microbes coordinate SRP transfer between sediment and overlying water was also posed. To answer this, we used glucose to investigate the mechanisms behind P behavior in this process. Theoretically, microbial function in SRP flux should be related to the population of microorganisms in the sediment. The microbial biomass C is usually used to evaluate the total microbial population in environments [36]. Elevating the glucose concentration in overlying water clearly increased the microbial biomass C of sediment (Fig. 4a), indicating an increase in the population of sedimentary microbes. This overpopulation would likely drive microbes to acquire more SRP from overlying water to sustain their growth. Consequently, the microbial biomass P in the sediment increased proportionally as glucose conditions were elevated (Fig. 4b). Interestingly, the ratio of microbial biomass C/P also increased with higher glucose concentrations, with ratios greater than 140:1 in all glucose treatments (Fig. 4c). It was reported that bacteria have a higher relative P content than algae and plants, with the C/P ratio being typically about 20:1 [37]. A high microbial biomass C/P ratio would encourage the microbes to access more soluble P from their environment [38]. Therefore, although the microbes in the sediment with more glucose had adsorbed more SRP, they are still expected to have a stronger capacity for absorbing SRP from overlying water than those with little or no glucose treatment. In all, these results suggested that the direct absorption of P by microbes should be a mechanism of microbe-mediated plant debris-enhanced P decrease in overlying water.

When the decrease in total overlying water SRP and the increase in total microbial biomass P in the sediment are calculated from Figs. 3d and 5b, respectively, we found that the increase in microbial biomass P only accounted for 4.6-7.4% of the SRP decrease. In addition to the direct absorption actions of microbes, other important microbe-mediated processes should be considered in plant debris-mediated P flux between sediment and overlying water. By analyzing the P fractions in the sediment, we found that the H<sub>2</sub>O-P and Fe-P<sub>i</sub> fractions decreased as incubated glucose concentrations increased, whereas the opposite was true for Fe-Po, ASOP, sugar bound Po, nucleic acid Po and polyphosphate, and humic bound Po and phytate (Table 3). These are five relatively stable fractions [39], particularly the ASOP, sugar bound Po, nucleic acid P<sub>o</sub> and polyphosphate, and humic bound P<sub>o</sub> and phytate fractions which are poorly accessible and hardly involved in exchange processes with the interstitial or overlying water [40]. H<sub>2</sub>O-P is a labile fraction that is directly available for algal growth. Fe-P<sub>i</sub>, although insoluble under certain conditions, has the potential to be easily released into the overlying water when the redox potential in the sediment is lower [41]. Hence, decreases in H<sub>2</sub>O-P and Fe-P<sub>i</sub> in sediment by glucose addition seem to reduce the risk of phosphorus utilization by alga. For both the Ca-Al-P<sub>i</sub> and Ca-Al-P<sub>o</sub> fractions in the experiment, no significant changes occurred during the glucose-incubations (Table 3), implying that Ca-Al-P<sub>i</sub> and Ca-Al-Po are not utilized by microbes in the P cycle. Combining the above results, we suggest that increased P stabilization in sediment after absorption/adsorption from overlying water is another mechanism of microbe-mediated, plant debris-enhanced P decrease in overlying water.

In summary, the present study provides evidence that shortterm treatment with plant debris could improve the SRP decrease in overlying water through the microbe-mediated mechanisms of P absorption and stabilization in sediment. In addition, we cannot rule out P adsorption by the sediment itself, in which the microbes may initially alter the physical-chemical properties of sediment, thus enhancing its P adsorption. It is worth noting that all investigations in this study were performed with short-term lab experiments, and, therefore, the long-term effect of plant debris on P flux between sediment and overlying water under in situ lake conditions is still unclear and warrants further investigation.

#### 5. Conclusions

Short-term treatment of plant debris, glucose or cellulose significantly reduced SRP concentration in overlying water. However, in the sterilized water–sediment system, glucose treatment did not affect the SRP concentration in overlying water. In the unsterilized system, the microbial biomass C, microbial biomass P and microbial biomass C/P in the sediment with glucose treatment all clearly increased compared with those without treatment of glucose. By analyzing the P fractions in sediment, we found that H<sub>2</sub>O–P and Fe–P<sub>i</sub> decreased along with the increase in incubated glucose concentrations, whereas the opposite was true for Fe–P<sub>o</sub>, ASOP, sugar bound P<sub>o</sub>, nucleic acid P<sub>o</sub> and polyphosphate, and humic bound P<sub>o</sub> and phytate, five relatively stable fractions. In conclusion these results suggest that aquatic plant debris remaining in water systems could facilitate a reduction in overlying water SRP through mechanisms of direct phosphorus absorption by microbes and microbial stabilization of P in the sediment.

#### Acknowledgments

This work was financially supported by the Zhejiang Province Natural Science Foundation (No. Y5090106), the Natural Science Foundation of China (NSFC, Nos. 30900170, 30900920) and Major Special Technological Program of Water Pollution Control and Management (2009ZX07106 -002).

#### References

- X. Jin, Q. Xu, C. Huang, Current status and future tendency of lake eutrophication in China, Sci. China Ser. C 48 (2005) 948–954.
- [2] I. Kagaloua, E. Papastergiadoub, I. Leonardosa, Long term changes in the eutrophication process in a shallow Mediterranean lake ecosystem of W. Greece: response after the reduction of external load, J. Environ. Manage. 87 (2008) 497–506.
- [3] V.H. Smith, G.D. Tilman, J.C. Nekol, Eutrophication: impacts of excess nutrient inputs on freshwater, marine, and terrestrial ecosystems, Environ. Pollut. 100 (1999) 179–196.
- [4] K. Ramm, V. Scheps, Phosphorus balance of a polytrophic shallow lake with the consideration of phosphorus release, Hydrobiologia 342/343 (1997) 43– 53.
- [5] J. Ahlgren, L. Tranvik, A. Gogoll, M. Waldebäck, K. Markides, E. Rydin, Sediment depth attenuation of biogenic phosphorus compounds measured by <sup>31</sup>P NMR, Environ. Sci. Technol. 39 (2005) 867–872.
- [6] D.J. Conley, H.W. Paerl, R.W. Howarth, D.F. Boesch, S.P. Seitzinger, K.E. Havens, C. Lancelot, G.E. Likens, Controlling eutrophication: nitrogen and phosphorus, Science 323 (2009) 1014–1015.
- [7] H.K. Pant, K.R. Reddy, Phosphorus sorption characteristics of estuarine sediments under different redox conditions, J. Environ. Qual. 30 (2001) 1474–1480.
- [8] W. Scharf, Restoration of the highly eutrophic lingese reservoir, Hydrobiologia 416 (1999) 85–96.
- [9] M. Søndergaard, J.P. Jensen, E. Jeppesen, Role of sediment and internal loading of phosphorus in shallow lakes, Hydrobiologia 506/509 (2003) 135–145.
- [10] C.S. Reynolds, Phosphorus recycling in lakes: evidence from large limnetic enclosures for the importance of shallow sediments, Freshwater Biol. 35 (2008) 623–645.
- [11] W.C. An, X.M. Li, Phosphate adsorption characteristics at the sediment–water interface and phosphorus fractions in Nansi Lake, China, and its main inflow rivers, Environ. Monit. Assess. 148 (2009) 1–4.
- [12] C.J. Redshaw, C.F. Mason, C.R. Hayes, R.D. Roberts, Factors influencing phosphate exchange across the sediment-water interface of eutrophic reservoirs, Hydrobiologia 192 (1990) 233–245.
- [13] R. Gächter, J.S. Meyer, A. Mares, Contribution of bacteria to release and fixation of phosphorus in lake sediments, Limnol. Oceanogr. 33 (1988) 1542–1558.
- [14] X.C. Jin, X. Jiang, Y. Yao, L.H. Li, F.C. Wu, Effects of organisms on the release of phosphorus at the interface between sediment and water, Water Environ. Res. 79 (2007) 2253-2259.
- [15] L.D. Huang, S.T. Du, L. Fan, X.Y. Lin, H.L. Wang, Y.S. Zhang, Microbial activity facilitates phosphorus adsorption to shallow lake sediment, J. Soils Sediments 11 (2011) 185–193.
- [16] X.H. Gu, S.Z. Zhang, X.L. Bai, W.P. Hu, Y.H. Hu, X.L. Wang, Evolution of community structure of aquatic macrophytes in East Taihu Lake and it wetlands, Acta Ecol. Sin. 25 (2005) 1541–1548 (in Chinese).

- [17] F. Shen, D.B. Kuang, Remote sensing investigation and analysis for water resources utilization and its dynamic change of representing mid- or small lake groups in Taihu drainage area, J. Remote Sens. 7 (2003) 221–227 (in Chinese).
- [18] B. Berg, C. McClaugherty, Plant Litter, Decomposition, Humus Formation, Carbon Sequestration, Springer, Berlin, 2003.
- [19] B.D. Lindahl, K. Ihrmark, J. Boberg, S.E. Trumbore, P. Högberg, J. Stenlid, R.D. Finlay, Spatial separation of litter decomposition and mycorrhizal nitrogen uptake in a boreal forest, New Phytol. 173 (2007) 611–620.
- [20] B.Q. Qin, W.P. Hu, W.M. Chen, Process and Mechanism of Environment Changes of the Lake Taihu, Science Press, Beijing, 2004.
- [21] X.C. Jin, S.R. Wang, Y. Pang, F.C. Wu, Phosphorus fractions and the effect of pH on the phosphorus release of the sediments from different trophic areas in Taihu Lake, China, Environ. Pollut. 139 (2006) 288–295.
- [22] B. Müller, A.F. Lotter, M. Sturm, A. Ammann, Influence of catchment quality and altitude on the water and sediment composition of 68 small lakes in Central Europe, Aquat. Sci. 60 (1998) 316–337.
- [23] W.J.F. Standring, D.H. Oughton, B. Salbu, Remobilization of 109Cd, 65Zn and 54Mn from freshwater-labelled river sediments when mixed with seawater, Environ. Int. 26 (2002) 185–195.
- [24] J. Murphy, J.P. Riley, A modified single solution method for the determination of phosphate in natural water, Anal. Chim. Acta 27 (1962) 31–36.
- [25] P. McKendry, Energy production from biomass (part 1): overview of biomass, Bioresour. Technol. 83 (2002) 37–46.
- [26] E.D. Vance, P.C. Brookes, D.S. Jenkinson, An extraction method for measuring microbial biomass C, Soil Biol. Biochem. 19 (1987) 697–702.
- [27] B.G. Zhang, G.T. Li, T.S. Shen, J.K. Wang, Z. Sun, Changes in microbial biomass C, N, and P and enzyme activities in soil incubated with the earthworms *Metaphire* guillelmi or Eisenia fetida, Soil Biol. Biochem. 32 (2000) 2055–2062.
- [28] P.C. Brookes, D.S. Powlson, D.S. Jenkinson, Measurement of microbial biomass phosphorus in soils, Soil Biol. Biochem. 14 (1982) 319–321.
- [29] R.W. McDowell, G.F. Koopmans, Assessing the bioavailability of dissolved organic phosphorus in pasture and cultivated soils treated with different rates of nitrogen fertilizer, Soil Biol. Biochem. 38 (2006) 61–70.
- [30] J.R. Webster, E.F. Benfield, Vascular plant breakdown in freshwater ecosystems, Annu. Rev. Ecol. Syst. 17 (1986) 567–594.
- [31] S.O. Ryding, Lake Trehörningen restoration project. Changes in water quality after sediment dredging, Hydrobiologia 91/92 (1982) 549–558.
- [32] J.C. Zhong, B.S. You, C.X. Fan, B. Li, L. Zhang, S.M. Ding, Influence of sediment dredging on chemical forms and release of phosphorus, Pedosphere 18 (2008) 34-44.
- [33] B. Boström, J.M. Andersen, S. Fleischer, M. Jansson, Exchange of phosphorus across the sediment–water interface, Hydrobiologia 170 (1988) 229–244.
- [34] K. Hunter, A.H. Rose, Influence of growth temperature on the composition and physiology of microorganisms, J. Appl. Chem. Biotechnol. 22 (1972) 527–540.
- [35] U. Münster, R.J. Chróst, Origin, composition, and microbial utilization of dissolved organic matter, in: J. Overbeck, R.J. Christ (Eds.), Aquatic Microbial Ecology Biochemical and Molecular Approaches, Springer, New York, 1990.
- [36] A.J. King, A.F. Meyer, S.K. Schmidt, High levels of microbial biomass and activity in unvegetated tropical and temperate alpine soils, Soil Biol. Biochem. 40 (2008) 2605–2610.
- [37] T. Fenchel, T.H. Blackburn, Bacteria and Mineral Cycling, Academic Press, London, 1979.
- [38] Z.L.He, J. Wu, A.G. O'Donnell, J.K. Syers, Seasonal responses in microbial biomass carbon, phosphorus, and sulphur in soils under pasture, Biol. Fert. Soils 24 (1997) 421–428.
- [39] C.J. De Groot, H.L. Golterman, On the presence of organic phosphate in some Camargue sediments: evidence for the importance of phytate, Hydrobiologia 252 (1993) 117–126.
- [40] S. Gerhardt, K. Boos, B. Schink, Uptake and release of phosphate by littoral sediment of a freshwater lake under the influence of light or mechanical perturbation, J. Limnol. 69 (2010) 54–63.
- [41] V. Mesnage, B. Picot, The distribution of phosphate in sediments and its relation with eutrophication of a Mediterranean coastal lagoon, Hydrobiologia 197 (1995) 29–41.