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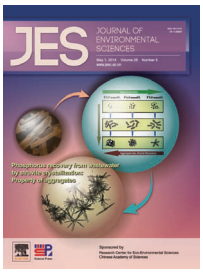

A restoration-promoting integrated floating bed and its experimental performance in eutrophication remediation

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ABSTRACT

Numerous studies on eutrophication remediation have mainly focused on purifying water first, then restoring submerged macrophytes. A restoration-promoting integrated floating bed (RPIFB) was designed to combine the processes of water purification and macrophyte restoration simultaneously. Two outdoor experiments were conducted to evaluate the ecological functions of the RPIFB. Trial 1 was conducted to compare the eutrophication purification among floating bed, gradual-submerging bed (GSB) and RPIFB technologies. The results illustrated that RPIFB has the best purification capacity. Removal efficiencies of RPIFB for TN, TP, NH_4^+-N, NO_3^--N, COD_{Cr}, Chlorophyll-*a* and turbidity were 74.45%, 98.31%, 74.71%, 88.81%, 71.42%, 90.17% and 85%, respectively. In trial 2, influences of depth of GSB and photic area in RPIFB on biota were investigated. When the depth of GSB decreased and the photic area of RPIFB grew, the height of *Potamogeton crispus* Linn. increased, but the biomass of *Canna indica* Linn. was reduced. The mortalities of *Misgurnus anguillicaudatus* and *Bellamya aeruginosa* in each group were all less than 7%. All results indicated that when the RPIFB was embedded into the eutrophic water, the regime shift from phytoplankton-dominated to macrophyte-dominated state could be promoted. Thus, the RPIFB is a promising remediation technology for eutrophication and submerged macrophyte restoration.

Introduction

The increasing concentrations of nitrogen and phosphorus provoke serious environmental problems in surface waters, which not only lead to eutrophication but also disturb the biodiversity of organisms (Mitsch and Jørgensen, 2003; Gurkana et al., 2006). Widespread occurrence of water eutrophication results in loss of ecological integrity, decrease of aquatic biodiversity, vanishing of submerged vegetation, potential production of toxins, etc. (Geurts et al., 2009; Estrada et al., 2011). Many conventional and novel methods with physical, chemical and biological processes have been applied to treat the negative effects of eutrophication over the past decades (Benndorf, 1995; Deppe at al., 1999; Wang et al., 2012). Among those methods, eco-technologies such as artificial floating bed (AFB) have been applied world-wide due to their advantages such as low cost and simple maintenance (Chen et al., 2013). In particular, the selected aquatic or terrestrial plants can not only remove pollutants from water, but also bring economic benefits (Keskinkan et al., 2004; Lesley et al., 2008; Bal et al., 2011). Therefore, the eco-technologies have received increasing public attention in recent years (Shan et al., 2009). In order to enhance the pollutant carrying capacity and self-purification capacity of an aquatic eco-system, it is necessary to optimize its ecological structure. Submerged macrophyte restoration is

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crucial to ecological structure (Scheffer et al., 1993). There are many important eco-functions of submerged macrophytes in aquatic eco-systems. Firstly, they can compete for light and nutrients with phytoplankton; secondly, they provide the structure for periphyton, shelter for organisms and invertebrates, and spatial refuge for zooplankton and small fish; thirdly, they reduce the mixing of the water column and resuspension; additionally, they provide food for aquatic animals; finally, they can inhibit the growth of phytoplankton by releasing allelochemicals (Van Donk and Van de Bund, 2002; Li et al., 2008; Taguchi and Nakata, 2009). However, there are many environmental conditions that can affect the restoration of submerged macrophytes. These environmental conditions include hydrodynamic factors such as waves, water depth, sediment properties, fish communities, nutrient loading and periphyton (Qin, 2009; Lloret and Marín, 2009).

Although many researchers have investigated the application of AFB and the purification capacity of submerged plants, few papers have focused on how to restore the submerged plants and maintain a stable ecosystem based on AFB technology (Lloret and Marín, 2009; Wu et al., 2010). Previous studies have generally divided the eutrophication remediation into two steps. The first one is to purify eutrophic water and the second one is to restore the submerged macrophytes (Qin, 2009). In this study, a restoration-promoting integrated floating bed (RPIFB) was designed to combine the processes of water purification and macrophyte restoration simultaneously. The RPIFB can help submerged macrophytes to overcome various constraints in the eutrophic water. It can not only reduce nutrients in the eutrophic water efficiently, but can also help submerged macrophytes to reach the photosynthetic light compensation point by adjusting the depth of the gradual-submerging bed (GSB). Then the ecological function of submerged plants can be effectively performed, and the habitat for aquatic animals can be ameliorated. The regime shift from phytoplankton-dominated to macrophyte-dominated state can be promoted.

Outdoor experiments were carried out to (1) evaluate the water purification efficiency of the RPIFB; (2) verify whether the biotic components can maintain health in the RPIFB; (3) analyze the restoration-promoting capabilities of the RPIFB on eutrophication.

1 Materials and methods

1.1 Materials and biota of RPIFB

All plants and animals used in the RPIFB were collected from the Xiangjiang River Basin in Changsha, China. Aquatic animals such as *Misgurnus anguillicaudatus* (body length 6.0 ± 1.2 cm), *Bellamya aeruginosa* (weight 4.0 ± 1.3 g) and plants such as *Canna indica* Linn. (plant height 35.0 ± 2.6 cm), *Potamogeton crispus* Linn. (plant length 25.0 ± 1.2 cm) were all tamed and cultured with experimental water for a month.

Natural zeolite was obtained from Jinyun, Zhejiang Province, China. Its typical unit cell composition is $Na_6[(AlO_2)_6(SiO_2)_{30}] \cdot 24H_2O$ with a density of 2.16 g/cm^3 and particle size of (4.0 ± 1.2) mm, moisture content of 7.0% to 14.0%, pore diameter of 3.4 to 4.0 Å, and specific area of 230 to 320 m^2/g.

The experimental water was pumped from a pool which is in severe-eutrophic state in the Xiangjiang River Basin. Discharge of domestic wastewater was the main nutrient source of the pool, which is rich in freshwater blue-green algae and protozoa. The initial water-quality parameters such as total phosphorus (TP, 3.36 ± 0.01 mg/L), total nitrogen (TN, 10.20 ± 0.10 mg/L), ammonia-nitrogen (NH_4^+-N, 2.65 ± 0.15 mg/L), nitrate-nitrogen (NO_3^--N, 1.10 ± 0.02 mg/L), chemical oxygen demand (COD_{Cr}, 88.26 ± 9.61 mg/L), Chlorophyll-*a* (Chl-*a*, 15.15 ± 1.65 μg/L) and water turbidity (85.00 ± 5.00 NTU) were analyzed (Jin and Tu, 1995; NEPA, 2002).

All the tubular structural skeleton elements of RPIFB were made of ethylene vinyl acetate, and the cage that coated the RPIFB was an oxidation-resistant polyvinyl chloride (PVC) net.

1.2 Design of RPIFB

The perspective drawing (**Fig. 1a**) shows that the RPIFB was comprised of a floating bed (FB) and GSB. The FB contained three isosceles trapezoid interposed baskets (ITIBs), which could hold aquatic plants above the water. The GSB with an inner orthohexagonal interposed basket (OIB) could hold submerged plants under the water. Buoyancy of the RPIFB was largely provided by three inflatable cuboid air chambers (ICACs), and partly by the tubular structural skeletons. The proportions between plant growing area and total hexagon surface area (G/T) and the dimensions of the main components in the RPIFB are shown in **Table 1**.

An ample photic zone in the ITBs was designed to

Table 1 Dimensions and flat surface area ratio of main components in restoration-promoting integrated floating bed (RPIFB)[a]

Component	Plane area	Edge length	G/T (%)
FB	S	$6 \times l$	37.5
GSB	S	$6 \times l$	100.0
ITIB	$S/8$	$l + 3 \times l/2$	12.5
OIB	$S/4$	$6 \times (l/2)$	25.0

[a] FB: floating bed; GSB: gradual-submerging bed; ITIB: isosceles trapezoid interposed basket; OIB: orthohexagonal interposed basket; G/T: the proportions between plant growing area and total hexagon surface area; S: the plane area of external orthohexagonal skeletons for FB and GSB, S = 3182.6 cm^2; l: the edge length of orthohexagonal skeletons in FB and GSB, l = 35.0 cm.

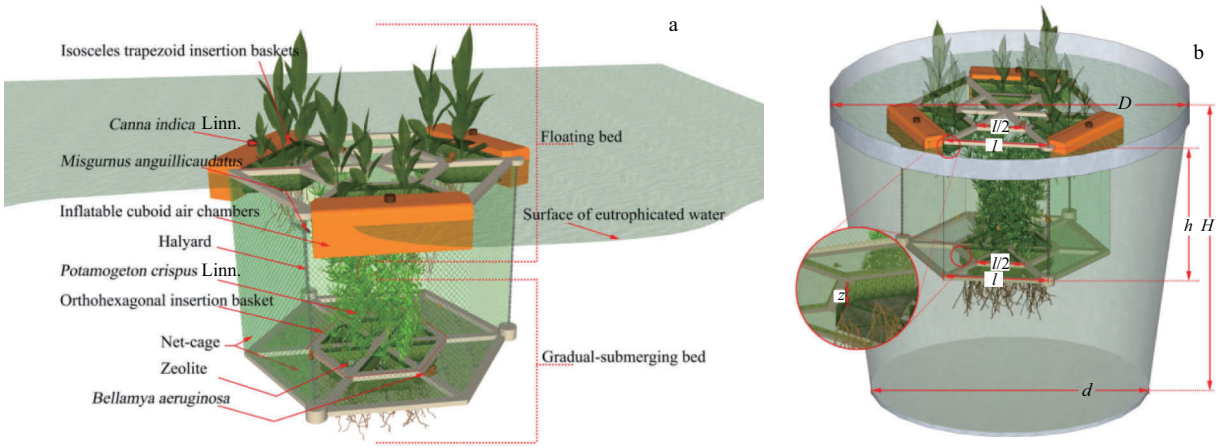

Fig. 1 Schematic and parametric diagram for restoration-promoting integrated floating bed (RPIFB). (a) Schematic diagram of RPIFM; (b) Parametric diagram of the experimental facilities.

guarantee enough light for the *P. crispus*. For the same reason, the OIB was set centrally within the GSB. As an auxiliary, three halyards were set to adjust the depth of *P. crispus*. To avoid environmental perturbations, the whole underwater part of RPIFB was covered with PVC net (pore diameter 1.5 mm × 1.5 mm). The PVC net could provide an extensive surface area for periphyton, which might help enhance the purification ability of the RPIFB system (Wu et al., 2010). **Figure 1b** shows the dimensions of the experimental facilities. The detailed values were: *D*, 82 cm; *d*, 64 cm; *H*, 90 cm; *h*, 30 to 70 cm; *z*, 6 cm.

1.3 Experimental setup

The experiment processes consisted of 2 trials with 14 groups (3 parallel tests in each group). Detailed settings of the two trials are shown in **Table 2**. Trial 1 was conducted to compare the water purification efficiencies of FB, GSB and RPIFB. In addition, the influence of light condition on GSB's function could also be testified through the results of the groups A2 and A3. To compare the purification performance of FB, GSB and RPIFB, concentrations of pollutants (TP, TN, NH_4^+-N and NO_3^--N) and three other water-quality indicators (COD_{Cr}, Chl-*a*, water turbidity) were chosen as the evaluation parameters.

Trial 2 was conducted to investigate whether the RPIFB could provide suitable environmental conditions for biota. Physiological responses of *C. indica*, *P. crispus* and aquatic animals to the depth of GSB and photic area in RPIFB were measured, which included the height of *P. crispus*, the weight of *C. indica*, and the mortality of *B. aeruginosa* and *M. anguillicaudatus*.

All tests were conducted in tanks, which were each filled with 380 L of experimental water (**Fig. 1b**). Basic compositions of FB, GSB and RPIFB are shown in **Table 3**. After 60 days incubation, when growth characteristics of organisms stabilized, FB, GSB and RPIFB were then put into the experimental tanks. To exclude rainfall and ensure natural ventilation as well as light, the experiments were performed at a horticultural shelter without walls.

1.4 Sampling and analysis

To compensate for the loss by evaporation and transpiration, distilled water was added daily to keep the

Table 3 Main components of FB, GSB and RPIFB

Subsystem	Component				
	C. indica	*P. crispus*	*B. aeruginosa*	*M. anguillicaudatus*	Natural zeolite (kg)
FB	9[a]	0	40	0	2.1
GSB	0	48	40	30	1.4
RPIFB	9	48	80	30	3.5

[a]Number of components or mass of natural zeolite.

Table 2 Settings of trial 1 and trial 2[a]

Trial	Trial 1					Trial 2								
	A1	A2	A3	A4	C	B1	B2	B3	B4	B5	B6	B7	B8	B9
System	F	G	G	R	–	R	R	R	R	R	R	R	R	R
P/*W* (%)	77	100	77	77	100	40	40	40	60	60	60	85	85	85
DG (cm)	–	40	40	40	–	30	50	70	30	50	70	30	50	70

[a]*P*/*W*: the proportion of photic area and total water surface area; DG: the depth of GSB; –: no setting; F, G, and R stand for FB, GSB and RPIFB, respectively; A and B stand for serial number of groups in trial 1 and trail 2; C: control group in trial 1.

experimental water volume stabilized. Twenty days' data was collected after FB, GSB and RPIFB were put into the experimental water.

Water samples were collected at the depths of 15, 45 and 75 cm at 10:30 and 15:00 every five days (for water turbidity every 2 days). The samples were mixed before being analyzed. Parameters of water samples were determined according to Standard Methods of the China National EPA (NEPA, 2002).

Plant height of *P. crispus* was measured every five days. At the end of the experiment, *C. indica* plants were carefully removed from the RPIFB and gently washed with tap water, and then blotted with absorbing paper, and finally the fresh weights were measured and recorded. The mortality rate of aquatic animals was assessed by counting their numbers (dead individuals were picked out from the water once found).

All the compositional analyses were performed in triplicate, and the data were expressed as mean ± standard errors. The removal rates (R, %) within each group were analyzed according to the following formulas.

$$R = 100 \times (C_r - C_i)/C_i \quad (1)$$

$$R_i = R_t - R_c \quad (2)$$

where, C_r (mg/L) represents the average residual concentration of nutrients in each tank; C_i is the initial concentration (mg/L); R_i (%) is the individual actual average removal rate; R_t (%) is the total ultimate average removal rate in each group; R_c (%) represents the average removal rate of the control (group C).

2 Results and discussion

2.1 Comparison of phosphorus removal in FB, GSB and RPIFB

As a major nutrient for aquatic ecology, excess phosphorus can lead to eutrophication of the receiving water. As shown in **Fig. 2**, the R_i of TP in trial 1 was: A4 > A1 > A2 > A3 > C. C_r of TP in group C increased from (2.38 ± 0.01) to (2.45 ± 0.11) mg/L (R = –2.84% ± 4.42%), while it reduced dramatically from (2.38 ± 0.01) to (0.11 ± 0.01) mg/L (R_i = 98.31% ± 4.25%) in group A4. The R_i of TP in A1, A2 and A3 were all higher than that of group C, but lower than that of the group A4. The results also showed that the RPIFB is applicable to remove phosphorus when

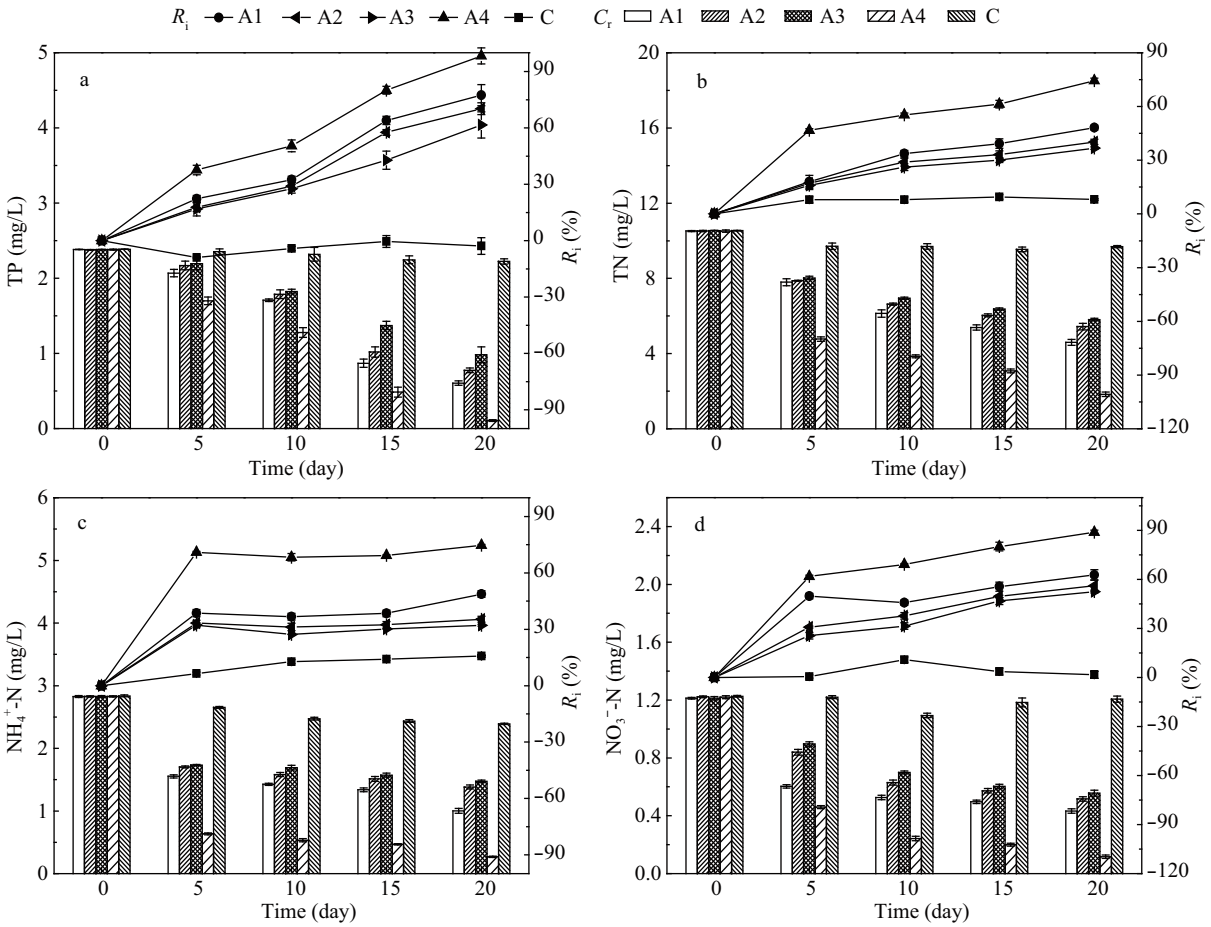

Fig. 2 Variations of nutrients concentration and homologous removal rate in trial 1. (a) concentration and removal rate of TP in trial 1; (b) concentration and removal rate of TN in trial 1; (c) concentration and removal rate of NH_4^+-N in trial 1; (d) concentration and removal rate of NO_3^--N in trial 1.

TP ranges from (0.11 ± 0.01) to (3.36 ± 0.01) mg/L.

Several mechanisms may explain the results. C_r of TP in group C increased first and then stabilized. That might be due to the equilibrium between the physiological activities of phosphorus-decomposing bacteria (PDB) and phosphorus-concentrating bacteria (PCB). PDB can help release endogenous phosphorus (Chuai et al., 2011). Without any macrophytes, group C maintained an anaerobic aquatic environment, which might have accelerated the physiological activities of PDB and increased phosphorus concentration. When phosphorus assimilation was less than the release of phosphorous, the TP concentration would increase. Meteorological conditions can impact nutrient removal via affecting the physiological activities of the biota (Chang et al., 2012). Equilibrium between PDB and PCB may be affected by the meteorological conditions, because moderate temperature and lighting conditions can promote the proliferation of the microorganisms in different ways. Detailed meteorological conditions during the experiment are illustrated in **Fig. S1** (supplementary data).

With the RPIFB in the group A4, phosphorus was removed efficiently. The reasons may be as follows. Firstly, phosphorus concentration was decreased by the assimilation by *C. indica* and *P. crispus*. Secondly, TP removal was enhanced by the coupling effects of microorganisms and plants in the rhizosphere micro surroundings. Thirdly, soluble inorganic phosphorus could be partly precipitated under the promotion of $A1^{3+}$, Fe^{3+}, Ca^{2+} in zeolite (Xiao et al., 2009). Besides, phosphorus release of the organic detritus could be reduced by the grazing effects of *B. aeruginosa* and *M. anguillicaudatus*.

The order of R_i of TP of groups A1, A2 and A3 were A1 > A2 > A3, which might be due to the diverse experimental set-ups as shown in **Tables 2** and **3**. The difference between A2 and A3 was in the proportions of photic area and total water surface area (P/W). Therefore, the experiment results proved that the larger the P/W was, the better the purification effect of the GSB would be. Meanwhile, it also proved that better illumination can promote the functioning of *P. crispus* in the GSB. The result showed that R_i of TP of A2 and A3 were lower than that of A1, which proved that the phosphorus absorption of FB was greater than that of GSB. The reason might be that with its flourishing roots and enormous aboveground biomass, *C. indica* has greater phosphorus absorption than *P. crispus*. Another reason may be that the biomass of *C. indica* increases faster than that of *P. crispus* (Zhang et al., 2009).

2.2 Comparison of Nitrogen removal in FB, GSB and RPIFB

Nitrogen is also a major cause of eutrophication. The removal efficiencies of TN, NH_4^+-N and NO_3^--N can be seen in **Fig. 2**. In group C, the C_r of nitrogen varied little (TN, 10.54 ± 0.02 to 9.69 ± 0.06 mg/L; NH_4^+-N, 2.84 ± 0.02 to 2.39 ± 0.01 mg/L; NO_3^--N, 1.23 ± 0.01 to 1.21 ± 0.02 mg/L). In group A4, three forms of nitrogen were efficiently eliminated (TN, (10.52 ± 0.06) to (1.84 ± 0.12) mg/L; NH_4^+-N, (2.83 ± 0.01) to (0.27 ± 0.01) mg/L; NO_3^--N, (1.22 ± 0.01) to (0.12 ± 0.01) mg/L). The R_i of all three forms of nitrogen also varied in the order A4 > A1 > A2 > A3 > C. The results verified that the RPIFB can play its role to remove nitrogen.

C_r of nitrogen varied little in group C, since there was no other component besides algae, which demonstrated that a simple ecosystem structure and composition are not good for water purification. The RPIFB can be an independent ecosystem, in which different levels of dissolved oxygen can be formed at the rhizosphere or surface of leaves. Thus, nitrifying bacteria and denitrifying bacteria can coexist in a system and achieve a prominent denitrification effect (International Water Association, 2000). Multiple mechanisms, such as volatilization, ammonification, nitrification/denitrification, plant uptake (*C. indica*, *P. crispus*), matrix adsorption (zeolite) and predation (*B. aeruginosa*, *M. anguillicaudatus*), can explain the result of nitrogen removal in the group A4 (Saeed and Sun, 2012). There are some common characteristics shared by groups A1, A2, A3 and A4. For example, firstly, with the same matrix (zeolite), NH_4^+ would be absorbed rapidly (especially in the first 5 days) through ion exchange, and the absorbed NH_4^+ could be further recycled through microbial decomposition or be absorbed by aquatic macrophytes (Zheng et al., 2008). Secondly, plant residues in FB, GSB and RPIFB could serve as food for *M. anguillicaudatus* and *B. aeruginosa*, which would prevent a further endogenous nitrogen release. Thirdly, synergy of biota interactions and absorption by the matrix and microorganisms could enhance the nutrient removal.

2.3 Comparison of COD_{Cr}, Chl-*a*, and water turbidity removal in FB, GSB and RPIFB

The R_i and C_r of COD_{Cr} and Chl-*a* showed a similar pattern to that of nutrients. The most notable difference was in group C, in that its turbidity and C_r of Chl-*a* experienced a significant negative growth (turbidity, from (87.01 ± 0.77) to (143.73 ± 1.49) NTU; Chl-*a*, from (15.37 ± 0.15) to (26.20 ± 0.26) μg/L). In group C, R_i of turbidity attained (–65.21 ± 1.11)%, and R_i of Chl-*a* reached (–70.51 ± 2.85)%, thus taking them as the basis, R_i of Chl-*a* in group A4 reached (160.69 ± 2.37)% and R_i of turbidity reached (150.21 ± 4.82)%. All results indicated that the RPIFB is suitable for reducing COD_{Cr} (from (22.60 ± 0.50) to (88.26 ± 9.61) mg/L), Chl-*a* (from (1.50 ± 0.10) to (15.15 ± 1.65) μg/L) and water turbidity (from (1.70 ± 0.20) to (85.00 ± 5.00) NTU).

COD is an important indicator of organic substances in water and an indirect manifestation of water pollution. As shown in **Fig. 3**, the removal of COD_{Cr} was the best in group A4 (R_i = 71.42% ± 2.45%). There are two

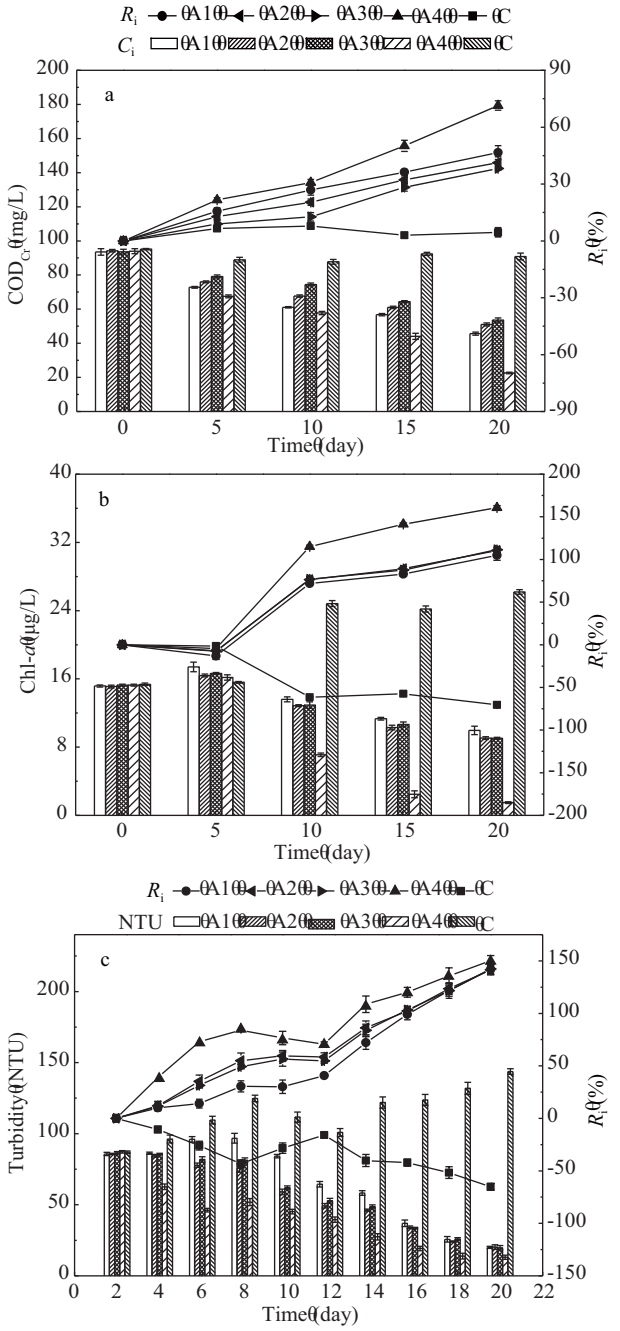

Fig. 3 Cr and R_i variations in trial 1. C_r represents the average residuary concentration of nutrients in each tank, R_i is the individual actual average removal rate. (a) C_r and R_i variations of COD_{Cr} in trial 1; (b) C_r and R_i variations of Chl-*a* in trial 1; (c) C_r and R_i variations of water turbidity in trial 1.

main reasons for this result. Firstly, organic degradation of microorganisms in the root zone could be assimilated by *C. indica* and *P. crispus*. Secondly, the COD_{Cr} removal could be enhanced by the predation of *M. anguillicaudatus* and *B. aeruginosa*. Without *C. indica* and *M. anguillicaudatus*, groups A1–A3 showed relatively weaker removal capacity. The COD_{Cr} removal of A2 (R_i = 41.26% ± 3.14%) was better than that of A3 (R_i = 38.16% ± 0.83%), which was probably due to their different *P*/*W* (A2, 100%; A3, 70%) settings. The result proved that the COD removal efficiency in the RPIFB is better than that in both the FB and GSB. It also proved that the less the depth of GSB is, the better *R* of COD_{Cr} will be.

As an important indicator to evaluate the level of eutrophication, Chl-*a* is significantly correlated with the biomass of phytoplankton. As shown in **Fig. 3**, with sufficient nutrients and proper illumination, C_r of Chl-*a* in group C experienced a significant negative growth (from (15.37 ± 0.15) to (26.20 ± 0.26) μg/L). In particular, when the temperature increased during day 5 to 10, the C_r of Chl-*a* increased rapidly and finally reached (24.83 ± 0.35) μg/L. Results of A4 showed that the growth of algae can be inhibited by the RPIFB. The R_i of Chl-*a* in the RPIFB finally achieved 160.69% ± 2.36%. Mechanisms may involve nutrient limitation due to zeolite and competition for nutrients and light among *C. indica*, *P. crispus* and periphyton. Allelopathic interactions between emerged plants and submerged macrophytes might also play an important role in Chl-*a* reduction in groups A1–A4 (Mulderij et al., 2007).

Turbidity can increase the optical attenuation coefficient and therefore decrease water transparency. The turbidity of eutrophic water can be reduced in FB, GSB and RPIFB, respectively. As shown in **Fig. 3c**, changes of water turbidity in each group fluctuated fiercely, especially in group C. Turbidity in groups A1–A4 decreased, from (85.73 ± 0.44) to about (13.09 ± 1.54) NTU, while group C increased from (87.01 ± 0.77) to (143.73 ± 1.94) NTU due to the proliferation of algae. The turbidity removal of A1-A4 could be promoted by the sedimentation of suspended particulate substances via root adsorption by *C. indica* and *P. crispus*, grazing of herbivorous zooplankton and matrix adsorption, etc. The results demonstrated that the turbidity of water can be increased by the growth of algae, and can be affected by weather conditions via influencing the growth of algae.

2.4 Effect of depth of GSB and photic area in RPIFB on the biota

The DG-*P*/*W*-Height three-dimensional graph (**Fig. 4a**) reflects the growth of *P. crispus* at different depths of GSB and photic area in RPIFB. **Figure 4b** shows the growth of *C. indica* based on the experimental setting (**Table 2**).

As shown in **Fig. 4a**, the larger the DG value was, the lower the height of *P. crispus* would be. And the larger the *P*/*W* was, the higher the *P. crispus* would grow. That is because DG and *P*/*W* are quite relevant to the light intensity, which is an important factor for the growth of *P. crispus* (Carter et al., 1996). When DG was smaller, the leaves of *P. crispus* would be much closer to the water surface to obtain more light. When *P*/*W* increased, more light could get through the water to accelerate the photosynthesis of *P. crispus*. In group BT, with the depth

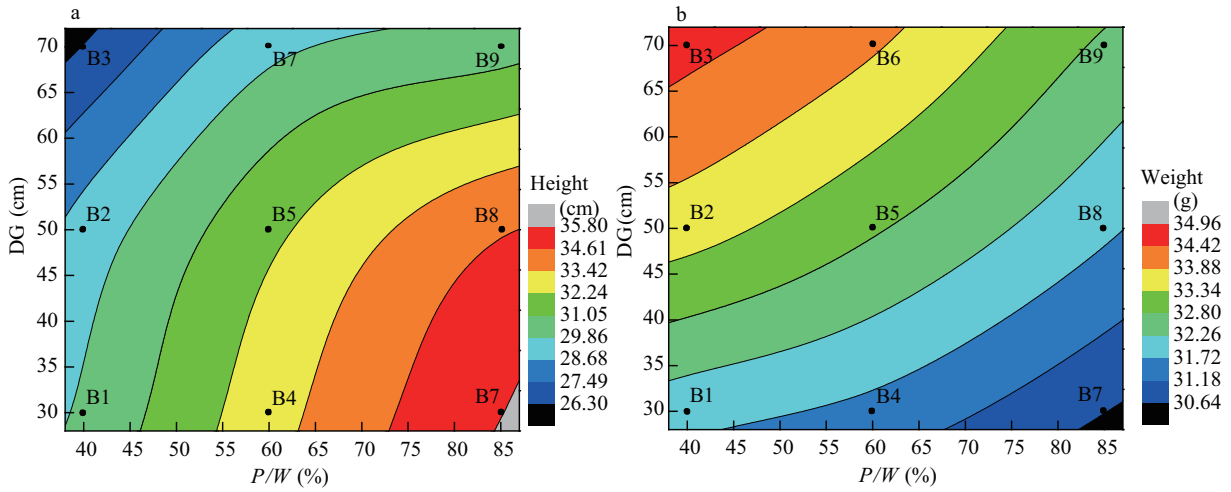

Fig. 4 Growth of aquatic plants in trial 2. (a) height variation of *P. crispus* affected by depth of GSB (DG) and the proportion of photic area and total water surface area (P/W) in trial 2; (b) weight variation of *C. indica* affected by DG and P/W in trial 2.

of 30 cm and P/W of 85% for GSB, *P. crispus* achieved the greatest height increment. Therefore, this proved that the RPIFB could effectively promote the restoration of *P. crispus* by adjusting the depth of GSB to guarantee the light demand of *P. crispus*. Besides, the purifying function of RPIFB may also play an important role in promoting the *P. crispus* restoration.

The growth rhythm of the biomass (wet weight) of *C. indica* was contrary to that of *P. crispus* as shown in **Fig. 4b**. The height increment of *P. crispus* in Group B3 minimized, while the biomass growth of *C. indica* maximized. The results indicated that factors such as the negative phototropism of the root system, nutritional competition of *P. crispus*, interaction of plants, and grazing of *M. anguillicaudatus* and *B. aeruginosa* can affect the growth of *C. indica* (Han et al., 2010). Therefore, it is necessary to keep the RPIFB covered to guard the roots of *C. indica* from damage by phytophagic fish, etc.

The mortality of *M. anguillicaudatus* and *B. aeruginosa* is shown in **Table 4**. There was a significantly lower mortality of the two species within the groups B4, B5, B7, B8 and B9. The main reason might be that the DG and P/W in those groups could well meet the light compensation point of *P. crispus*, thus, aquatic animals could obtain enough oxygen produced by *P. crispus*. Oxygen excreted by the roots of *C. indica* might also have helped the survival of aquatic animals. Therefore, the experiment implied that the RPIFB can effectively improve the survival condition for aquatic animals.

Table 4 Ultimate mortality of aquatic animals of RPIFB in trial 2

Group	*M. anguillicaudatus*			*B. aeruginosa*		
	Initial	Residual	Mortality (%)	Initial	Residual (m, n)[b]	Mortality (%)
B1	30[a]	30	0.00	80	78 (0, 2)	2.50
B2	30	29	3.33	80	76 (1, 3)	5.00
B3	30	28	6.67	80	75 (1, 4)	6.25
B4	30	30	0.00	80	79 (0, 1)	1.25
B5	30	30	0.00	80	79 (0, 1)	1.25
B6	30	29	3.33	80	78 (1, 1)	2.50
B7	30	30	0.00	80	80 (0, 0)	0.00
B8	30	30	0.00	80	80 (0, 0)	0.00
B9	30	30	0.00	80	79 (0, 1)	1.25

[a]Number of aquatic animals; [b]m stands for the number of dead aquatic animals in FB, n stands for the number of dead aquatic animals in GSB.

2.5 Role of RPIFB in the eutrophication restoration process

Waters often maintain either a phytoplankton-dominated or macrophyte-dominated state. The regime shift mechanism of the two phases is shown in **Fig. 5** (Scheffer et al., 2001). If the water state maintains F_0 to F_2, the water must be dominated by macrophytes. When the state is close to the bifurcation point F_2, slight incremental changes such as waves, intake of nutrients, and higher air temperature may take it beyond the bifurcation point (F_2), inducing a catastrophic shift to the algae-dominated state F_1 to F_3. To restore the macrophyte-dominated state via reversal of the conditions, the system may show hysteresis (from F_3 to F_1). Conditions such as nutrient concentration and biodiversity would need to be reversed far enough to reach the other bifurcation point F_1 ($F_1 < F_2$). Then the regime shift from algae-dominated to macrophyte-dominated state occurred. Traditionally, it was hard for submerged macrophytes to be restored, because the growth of submerged macrophytes would be inhibited by the algae-dominated state (Scheffer et al., 2001).

However, the RPIFB brings new thinking to overcome those traditional limitations in this study. Two prominent features of the RPIFB as proved in this study were that it can reduce the water nutrients efficiently and can promote the restoration of submerged plants effectively. As shown in **Figs. 2** and **3**, the RPIFB has decreased the nutrition concentration from eutrophic status to at least mesotrophic status, and also reduced the phytoplankton biomass (Jin

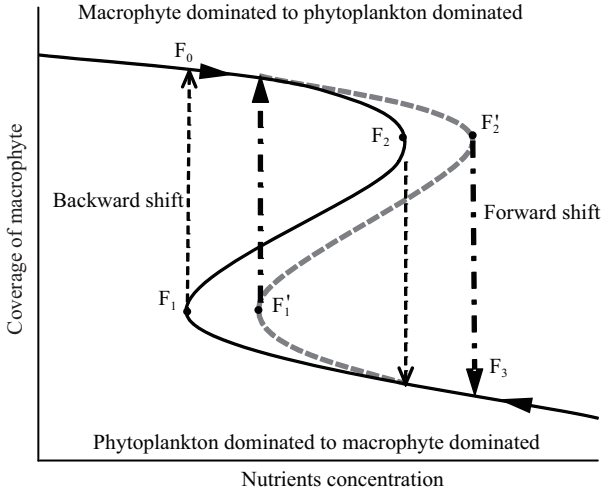

Fig. 5 Role of RPIFM in regime shift between phytoplankton-dominated and macrophyte-dominated. F_0 and F_3 stand for water status, F_1, F_1', F_2, F_2' stand for water state bifurcation point.

and Tu, 1995). When the RPIFB was embedded into the water, it maintained a macrophyte-dominated state (**Fig. 5**, F_0 to F_2), and state stability in water would be enhanced and expanded (**Fig. 5**, F_0 to F_2'). The RPIFB can continuously purify water and help the submerged plants overcome various restrictions. For example, it can enhance illumination compensation via adjusting its halyards to help the submerged plants overcome high turbidity and high concentration of nutrients, etc. When embedded into the water, the RPIFB could maintain the algae-dominated state (**Fig. 5**, between F_3 and F_1), and it could make the backward bifurcation happen ahead of schedule (**Fig. 5**, from F_1 to F_1'). There were two main reasons as follows: On one hand, all the biota compositions of the RPIFB were those species that could well endure the eutrophic water (see introduction in Supplementary Data). On the other hand, the nutrient removal ability and condition-improving capacity of biota in the RPIFB could be the restoring force for the reverse regime shift.

3 Conclusions

The RPIFB showed excellent restoration over deteriorated eutrophic water in this study. There are some main conclusions that can be drawn as follows. (1) Under optimal conditions, the removal rates of TN, TP, NH_4^+-N, NO_3^--N and COD_{Cr} can reach (74.45 ± 1.44)%, (98.31 ± 4.25)%, (74.71 ± 0.47)%, (88.81 ± 1.26)%, and (71.42 ± 2.45)%, respectively. Chl-*a* and water turbidity decreased to 10% of their initial concentrations. (2) purification efficiencies of RPIFB, FB and GSB in this experiment were in the order: RPIFB > FB > GSB. (3) RPIFB can improve water transparency quickly and purify eutrophic water continuously, and can help submerged plants survive in difficult circumstances in turbid eutrophic shallow waters. (4) RPIFB can purify the domestic wastewater and promote the regime shift from phytoplankton-dominated to macrophyte-dominated states.

In short, RPIFB is a promising ecological technology for eutrophication remediation and submerged macrophyte restoration.

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REFERENCES

Bal, K., Struyf, E., Vereecken, H., Viaene, P., De Doncker, L., de Deckere E. et al., 2011. How do macrophyte distribution patterns affect hydraulic resistances? Ecol. Eng. 37(3), 529–533.

Benndorf, J., 1995. Possibilities and limits for controlling eutrophication by biomanipulation. Hydrobiology 80(4), 519–534.

Carter, V., Rybicki, N. B., Turtora, M., 1996. Effect of increasing photon irradiance on the growth of *Vallisneria americana* in the tidal Potomac River. Aquat. Bot. 54(4), 337–345.

Chang, J. J., Wu, S. Q., Dai, Y. R., Liang, W., Wu, Z. B., 2012. Treatment performance of integrated vertical-flow constructed wetland plots for domestic wastewater. Ecol. Eng. 44, 152–159.

Chen, C. J., Zhang, R., Wang, L., Wu, W. X., Chen, Y. X., 2013. Removal of nitrogen from wastewater with perennial ryegrass/artificial aquatic mats biofilm combined system. J. Environ. Sci. 25(4), 670–676.

Chuai, X. M., Ding, W., Chen, X. F., Wang, X. L., Miao, A. J., Xi, B. D. et al., 2011. Phosphorus release from cyanobacterial blooms in Meiliang Bay of Lake Taihu. China. Ecol. Eng. 37(6), 842–849.

Deppe, T., Ockenfeld, K., Meybohm, A., Opitz, M., Benndorf, J., 1999. Reduction of Microcystis blooms in a hypertrophic reservoir by a combined ecotechnological strategy. Hydrobiology 408-409, 31–38.

Estrada, V., Di Maggio, J., Diaz, M. S., 2011. Water sustainability: A systems engineering approach to restoration of eutrophic Lakes. Comput. Chem. Eng. 35(8), 1598–1613.

Geurts, J. J. M., Sarneel, J. M., Willers, B. J. C., Roelofs, J. G. M., Verhoeven, J. T. A., Lamers, L. P. M., 2009. Interacting effects of sulphate pollution, sulphide toxicity and eutrophication on vegetation development in fens: A mesocosm experiment. Environ. Pollut. 157(7), 2072–2081.

Gurkana, Z., Zhang, J. J., Jørgensenc, S. E., 2006. Development of a structurally dynamic model for forecasting the effects of restoration of Lake Fure, Denmark. Ecol. Model. 197(1-2), 89–102.

Han, S., Yan, S. H., Chen, K. N., Zhang, Z. H., Zed, R., Zhang, J. Q. et al., 2010.^{15}N isotope fractionation in an aquatic food chain: *Bellamya aeruginosa* (Reeve) as an algal control agent. J. Environ. Sci. 22(2), 242–247.

International Water Association, 2000. Constructed Wetlands for Pollution Control, Processes, Design, and Operation, IWA Publishing, London.

Jin, X. C., Tu, Q. Y., 1995. Specification for Investigation of Lake Eu-

trophication (2nd ed). China Environmental Science Press, Beijing. 271–281.

Keskinkan, O., Goksu, M. Z. L., Basibuyuk, M., Forster, C. F., 2004. Heavy metal adsorption properties of a submerged aquaticplant (*Ceratophyllum demersum*). Bioresour. Technol. 92(2), 197–200.

Lesley, B., Daniel, H., Paul, Y., 2008. Iron and manganese removal in wetland treatment systems: rates, processes and implications for management. Sci. Total Environ. 394(1), 1–8.

Li, E. H., Li, W., Liu, G. H., Yuan, L. Y., 2008. The effect of different submerged macrophyte species and biomass on sediment resuspension in a shallow freshwater lake. Aquat. Bot. 88(2), 121–126.

Lloret, J., Marín, A., 2009. The role of benthic macrophytes and their associated macroinvertebrate community in coastal lagoon resistance to eutrophication. Mar. Pollut. Bull. 58(12), 1827–1834.

Mitsch, W. J., Jørgensen, S. E., 2003. Ecological Engineering and Ecosystem Restoration. John Wiley and Sons, New York.

Mulderij, G., Van Nes, E. H., Van Donk, E., 2007. Macrophyte-phytoplankton interactions: The relative importance of allelopathy versus other factors. Ecol. Model. 204(1-2), 85–92.

NEPA, 2002. Standard Methods for Monitoring and Analysis of Water and Wastewater (4th ed.). China Environmental Science Press, Beijing.

Qin, B. Q., 2009. Lake eutrophication: Control countermeasures and recycling exploitation. Ecol. Eng. 35(11), 1569–1573.

Saeed, T., Sun, G. Z., 2012. A review on nitrogen and organics removal mechanisms in subsurface flow constructed wetlands: Dependency on environmental parameters, operating conditions and supporting media. J. Environ. Manag. 112, 429–448.

Scheffer, M., Carpenter, S., Foley, J. A., Folke, C., Walkerk, B., 2001. Catastrophic shifts in ecosystems. Nature 413(6856), 591–596.

Scheffer, M., Hosper, S. H., Meijer, M. L., Jeppesen, E., Moss, B., 1993. Alternative equilibria in shallow lakes. Trends Ecol. Evol. 8(8), 275–279.

Shan, M. J., Wang, Y. Q., Shen, X., 2009. Study on bioremediation of eutrophic lake. J. Environ. Sci. 21(S1): S16–S18.

Taguchi, K., Nakata, K., 2009. Evaluation of biological water purification functions of inland lakes using an aquatic ecosystem model. Ecol. Model. 220(18), 2255–2271.

Van Donk, E., Van de Bund, W. J., 2002. Impact of submerged macrophytes including charophytes on phyto- and zooplankton communities: allelopathy versus other mechanisms. Aquat. Bot. 72(3-4), 261–274.

Wang, J., Zhang, L. Y., Lu, S. Y., Jin, X. C., Gan, S., 2012. Contaminant removal from low-concentration polluted river water by the bio-rack wetlands. J. Environ. Sci. 24(6), 1006–1013.

Wu, Y. G., Zhang, S. Q., Zhao, H. J., Yang, L. Z., 2010. Environmentally benign periphyton bioreactors for controlling cyanobacterial growth. Bioresour. Technol. 101(24), 9681–9687.

Wu, Y. H., Hu, Z. Y., Yang, L. Z., 2010. Hierarchical eco-restoration: A systematical approach to removal of COD and dissolved nutrients from an intensive agricultural area. Environ. Pollut. 158(10), 3123–3129.

Xiao, G. L., Li, T. X., Zhang, X. Z., Yu, H. Y., Huang, H. G., Gupta, D. K., 2009. Uptake and accumulation of phosphorus by dominant plant species growing in a phosphorus mining area. J. Hazard. Mater. 171(1-3), 542–550.

Zhang, Z. H., Rengel, Z., Meney, K., 2009. Kinetics of ammonium, nitrate and phosphorus uptake by *Canna indica* and *Schoenoplectus validus*. Aquat. Bot. 91(2), 71–74.

Zheng, H., Han, L. J., Ma, H. W., Zheng, Y., Zhang, H. M., Liu, D. H. et al., 2008. Adsorption characteristics of ammonium ion by zeolite 13X. J. Hazard. Mater. 158(2-3), 577–584.