Nutrient removal of a floating plant system receiving low-pollution wastewater: Effects of plant species and influent concentration

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Abstract. Plant floating bed was adopted in this study to compare the purification effect of four plant species (Oenanthe javanica, Ipomoea aquatica, Hydrocotyle vulgaris, and Iris sibirica) receiving high and low treated domestic sewage. The experiment was conducted for eight months during the low temperature season. The results indicated that the average removal rates of TN and NH₄⁺-N in I. aquatica floating bed were relatively high both under high and low influent concentration during the first stage of the experiment. During the second stage, H. vulgaris showed the best performance for nitrogen treatment, and the average removal rates of TN were 70.7% and 87.7% under high and low influent concentration, while the average removal rates of NH₄⁺-N were as high as 98.9% and 98.9%, accordingly. Moreover, H. vulgaris contributed most for plant assimilation to nitrogen removal among different plant floating systems. It was also found that the existence of hydrophytes effectively controlled the rise of water pH value and algae growth and reproduction, which helped to improve the aquatic environment. The results provide engineering parameters for the future design of an ecological remediation technology for low-pollution wastewater purification.

1. Introduction
As the rapid development of the economy, the problem of agricultural non-point pollution is getting increasingly serious. Runoffs from the farmland, containing nitrogen, phosphorus and other nutrients, go into ditches firstly. Ditches are the bond of farmland and rivers or lakes. In order to reduce the risk of farmland nutrients into water bodies, Yang set forward an innovative ecological ditch including a variety of plants planted in the side wall and bottom, and other auxiliary engineering facilities like interception dam and interception box [1]. The promising ecological ditch could promote the sedimentation of particulate materials and intercept nitrogen and phosphorus in drainage, which is an effective ecological treatment technology for the control of non-point pollution [1, 2]. However, the stereoscopic space of the ecological ditch was not fully utilized, and the upper space of it could be used efficiently.

Floating beds were often used on the surface of water for polluted river water, eutrophic water and domestic wastewater treatment [3–5]. It can be used in the ecological ditch to purify the upper space of the drainage. Little hydrophytes, such as Oenanthe javanica, Ipomoea aquatica, Iris sibirica, were appropriate to grow on floating bed system.

Floating bed systems exhibit high removal rates of nitrogen and phosphorus during the growing season of the plant [6, 7]. However, when cold weather arrives, the performance of floating beds is
often getting worse because of the weakness of the macrophytes. Few macrophytes, like \textit{O. javanica} and \textit{I. sibirica}, could grow well in cold weather, and had purification effect on polluted river water [8, 9]. But the performance of these plants for nutrient removal at different influent concentrations was not fully studied.

In the present study, we investigated the purification effect of four plant species at two influent concentrations.

2. Materials and methods

2.1. Plant material
\textit{O. javanica}, \textit{I. aquatica}, \textit{H. vulgaris} and \textit{I. sibirica} were selected as the plant materials which were commonly planted in the Yangtze River Delta. In this study, hydrophytes except \textit{H. vulgaris} were got from a garden centre in Suqian, Jiangsu province, China; \textit{H. vulgaris} was obtained from a pond around Tai Lake in Yixing, Wuxi city, China. The plantlets were washed with distilled water and preincubated in experimental water for about 3 weeks. When the plantlets were about 16 cm high, uniform and healthy ones were chosen for the experiment.

2.2. Experimental water
In this study, treated domestic wastewater (DW), which was pumped from a septic tank in the Jiangsu Academy of Agricultural Science (Nanjing, China), was used as the experimental water. After ultraviolet sterilizing, DW was diluted to high or low concentration by tap water before use (namely, H groups and L groups, respectively). The wastewater used in the experiment was a kind of low-pollution wastewater (LPW), which met the Chinese Farm Irrigation Water Standard [10]. The characteristics of the experimental water were exhibited in Table 1.

\begin{table}[h]
\centering
\begin{tabular}{|c|c|c|c|c|c|c|c|}
\hline
Treatment & TN (mg\textcdot L\textsuperscript{-1}) & NH\textsubscript{4}\textsuperscript{+}-N (mg\textcdot L\textsuperscript{-1}) & NO\textsubscript{3}\textsuperscript{-}-N (mg\textcdot L\textsuperscript{-1}) & TP (mg\textcdot L\textsuperscript{-1}) & COD\textsuperscript{a} (mg\textcdot L\textsuperscript{-1}) & BOD\textsuperscript{b} (mg\textcdot L\textsuperscript{-1}) & FC\textsuperscript{c} \text{/}100 mL & C:N\textsuperscript{d} \\
\hline
H groups & 20.25 & 17.23 & 1.19 & 1.62 & 40.0 & 14.19 & 4900 & 2.0 \\
& ±7.73 & ±5.95 & ±1.49 & ±0.89 & ±17.3 & ±1.34 \\
L groups & 10.90 & 9.90 & 1.15 & 0.94 & 27.4 & 10.74 & 3400 & 2.5 \\
& ±6.18 & ±7.00 & ±0.79 & ±0.63 & ±10.8 & ±1.14 \\
\hline
\end{tabular}
\caption{Characteristics of the domestic wastewater during the experiment.}
\end{table}

Note: H groups and L groups represented treatment groups receiving domestic wastewater of high and low concentration.
\textsuperscript{a}Chemical oxygen demand
\textsuperscript{b}5-day biochemical oxygen demand
\textsuperscript{c}Fecal Coliform
\textsuperscript{d}C and N were measured as COD and TN in C:N ratio

2.3. Design of the floating plant system
Floating bed was used to receive DW, which was made of a 20 mm diameter PVC pipe (65.0 cm length × 50.0 cm width) with grilling (4.0 cm × 4.0 cm) binding on it. Plastic tanks (internal dimensions of 74.8 cm × 52.0 cm × 48.5 cm) were used as the container.

\textit{O. javanica}, \textit{I. aquatica}, and \textit{I. sibirica} were planted on the floating bed receiving high and low concentration LPW to form six treatment groups (H1, H2, H3, L1, L2, and L3); Control groups were set receiving the two kinds of LPW (Control 1 and Control 2). Each treatment had three replicates. \textit{I. aquatica} was harvested at the end of December, and then \textit{H. vulgaris} was planted in H2 from January to May. Other treatments remained unchanged.

Apart from \textit{H. vulgaris}, hydrophyte seedlings were planted into the nutritive cups on the holes of the grilling and were held stably with a sponge. \textit{H. vulgaris} of a considerable weight with other
hydrophytes, was placed directly on the surface of the floating bed (Figure 1). Aerator was added in each tanks to provide enough dissolved oxygen to the system. The plastic tanks were filled with 145 L of DW at the beginning of the experiment. The experiment lasted from October 2, 2014, to May 9, 2015.

**Figure 1.** Composition of the floating plant system used in the present study.

2.4. **Sampling and analysis**

About 50 mL of water samples in the plastic tanks were collected every week at 9:00 am, stored in a refrigerator of 4°C, and measured immediately. Flow injection analysis was used to analyse TN, TP, NH$_4$$^+$-N, and NO$_3$$^-$-N on a Skalar San$^+$+ System (Skalar Co., The Netherlands). 2 mL of water samples was added into a tube before measuring Chemical oxygen demand, and COD analyser (DR1010 COD, HACH, China) was used. Temperature, pH, and dissolved oxygen (DO) were monitored using a multi-parameter water quality monitor (HI9828100, HANNA) before water sampling. Chlorophyll a in water was measured with ethanol-non-grinding method [11].

Plant height and biomass were measured before and after the experiment. The relative growth rate (RGR) of the plant was calculated based on $W_i$ and $W_0$, defined as:

$$RGR (%) = 100 \times \frac{(W_i - W_0)}{W_0}$$  \hspace{1cm} (1)

where $W_i$ is the average fresh weight of plants on day $i$ of the experiment and $W_0$ is the initial average fresh weight of the plants.

At the end of the study, plants were harvested and removed from the floating plant system. Plant samples were prepared and analysed for N concentration via the Kjeldahl method [12]. N accumulations were estimated by multiplying the N concentration by the plant biomass [13].

Nitrate, free amino acid, and soluble sugar of the leaves were analysed according to standard methods [14].

2.5. **Statistical analysis**

One-way analysis of variance (ANOVA) was performed to test the effects of different hydrophytes and DW concentrations on plant growth situation and nitrogen removal. Statistical analyses were conducted with SPSS13.0. Differences were considered statistically significant when $p < 0.05$. 

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3. Results and discussion

3.1. Changes in water quality

In the present study, pH values of plant floating beds were lower than in control groups, and pH values in H groups were lower than in L groups during the growth period of I. aquatica and H. vulgaris (Table 2). DO concentrations were higher during the growth period of I. aquatica. Except for H1, DO concentrations in other treatments met the grade one of the Surface Water Quality Standard (GB 3838-2002). The relatively low DO values in plant floating beds than control groups might ascribed to plant’s residues consuming oxygen through decomposition in low temperature season.

As one of the water quality indexes, DO concentration reflects the pollution degree of water bodies and physical, chemical and biological characteristics [5]. Hydrophytes can transport O₂ to the root through the internal aerenchyma, and providing oxygen to the underground part [15]. In addition, biofilm can be formed on the surface of the plant roots, providing enough oxygen to heterotrophic bacteria and nitrobacteria [16].

Table 2. Physical parameters in different treatment groups.

| Growth stages                  | Treatment group | pH        | Water temperature(℃) | DO(mg•L⁻¹)        |
|-------------------------------|-----------------|-----------|-----------------------|------------------|
|                               | H1              | 7.84(6.90-9.71) | 16.72 (7.70-23.60)          | 6.92 (0.13-13.80) |
|                               | H2              | 7.96(6.86-9.70) | 16.56 (7.40-22.70)          | 7.57 (0.10-13.56) |
|                               | H3              | 8.21(6.57-9.64) | 16.72 (7.70-23.20)          | 8.27 (0.08-16.6) |
|                               | Control 1       | 8.40(6.92-10.39)| 16.66 (7.80-23.10)          | 9.12 (0.04-16.00) |
|                               | L1              | 8.57(7.26-9.83) | 17.42 (7.70-23.20)          | 9.06 (0.83-14.50) |
|                               | L2              | 8.53(7.19-10.33)| 16.96 (7.80-23.3)           | 9.65 (1.32-1.95) |
|                               | L3              | 8.54(7.33-9.72) | 16.98 (7.70-23.20)          | 9.36 (1.78-15.30) |
|                               | Control 2       | 9.02 (7.26-10.76)| 17.07 (8.20-22.80)          | 12.66 (2.15-22.42) |
|                               | H1              | 7.48 (6.86-8.55)| 15.12 (6.10-23.28)          | 3.95 (0.13-12.37) |
|                               | H2              | 7.63 (6.45-8.97)| 14.91 (5.80-22.90)          | 4.28 (3.18-13.47) |
|                               | H3              | 8.23 (7.09-9.50)| 15.15 (6.10-24.17)          | 4.38 (2.03-13.29) |
|                               | Control 1       | 9.04(7.41-11.27)| 15.06 (6.00-25.80)          | 4.72 (0.55-21.23) |
|                               | L1              | 8.69(6.99-10.70)| 15.49 (6.10-23.06)          | 4.51 (0.79-15.61) |
|                               | L2              | 8.10 (6.42-10.32)| 15.22 (6.10-23.32)          | 4.61 (1.46-15.76) |
|                               | L3              | 8.42 (7.22-10.15)| 15.41 (6.10-24.08)          | 4.47 (0.77-15.89) |
|                               | Control 2       | 10.11 (7.29-11.70)| 15.42 (6.10-25.80)          | 5.96 (1.80-26.87) |

Note: Data in the above table are average value (minimal value-maximal value).

The nitrogen removal of plant floating beds did not have obvious advantage compared to control groups during the growth period of I. aquatica(Figure 2-4). The average removal rates of TN and NH₄⁺-N in I. aquatica floating bed were relatively high both under high and low concentration of LPW. With the raise of the temperature in spring, the removal efficiency of Nitrogen increased in plant floating beds (Figure 5-7). Among the hydrophytes in the experiment, H. vulgaris showed the best performance. The average removal rates of TN were 70.7% and 87.7% for high and low LPW treatment in H. vulgaris floating beds, while the average removal rates of NH₄⁺-N were as high as 98.9% and 98.9%, accordingly.

The chlorophyll a concentrations in plant floating beds were significantly lower than in control groups (Table 8), showing higher algae growth without hydrophytes in the low pollution aquatic water.
Bu and Xu reported that *Canna indica* and *Accords calamus* and other two floating bed plants all had the algae inhibiting effect, which confirmed to our study [5].

**Figure 2.** Changes of TN concentration in H groups (a) and L groups during the growth of *I. aquatica*. Note: arrows represent water exchanging. The same below.

**Figure 3.** Changes of NH$_4^+$-N concentration in H groups (a) and L groups during the growth of *I. aquatica*.

**Figure 4.** Changes of NO$_3^-$-N concentration in H groups (a) and L groups during the growth of *I. aquatica*. 
Figure 5. Changes of TN concentration in H groups (a) and L groups during the growth of *H. vulgaris*.

Figure 6. Changes of NH$_4^+$-N concentration in H groups (a) and L groups during the growth of *H. vulgaris*.

Figure 7. Changes of NO$_3^-$-N concentration in H groups (a) and L groups during the growth of *H. vulgaris*.
Figure 8. The concentration of chlorophyll a in the outflow of each treatment group.

3.2. Plant growth and nutrient accumulation

Although the biomass and average plant height of *I. sibirica* were larger, the same indexes were not the highest among the four plants at harvest. The biomass of *H. vulgaris* was the highest under high or low concentration of LPW treatments, and *O. javanica* and *I. aquatica* were larger in H treatment groups. At the same time, the RGR of *H. vulgaris* was the highest under both of the influent concentration; and the average root length of *I. sibirica* was the highest (Table 3). TN assimilation of *H. vulgaris* was the highest of all during the experiment, and the ratio of plant assimilation to TN input were 60.1% and 108.0% in H2 and L2 groups during the growth of *H. vulgaris*, respectively (Table 4).

As a edible plant, *I. aquatica* favors high temperature and humid environment, and distributed widely in China and India [17]. *I. aquatica* has a high economic value, compared with other hydrophytes. It has been used for heavy metals and organic pollutants adsorption, and agricultural wastewater treatment [17-19]. In the present study, the removal rates of TN and NH$_4^+$-N in *I. aquatica* floating bed was relatively high receiving high and low LPW. Free amino acids and soluble sugar were higher in *I. aquatica* than in *O. javanica* both in H groups and L groups at harvest by quality determination (Table 5). The NO$_3^-$-N in stem and leaf were 296.7 and 173.8 mg·kg$^{-1}$ in *O. javanica*, and the corresponding values were 188.0 and 187.3 mg·kg$^{-1}$ in *I. aquatica*. The NO$_3^-$-N concentrations were at a low level among leafy vegetables which can be safe to eat [20, 21].

Table 3. Growth situation of plants.

| Treatment groups | Plants      | Initial planting | Harvesting |
|------------------|-------------|------------------|------------|
|                  |             | Biomass (g)      | Average plant height (cm) | Biomass (g) | Average plant height (cm) | Average root length (cm) | Relative growth rate RGR (%) |
| H 1              | *O. javanica* | 7.0±0.2          | 15.5±1.4    | 153.0±27.3 | 64.2±9.9           | 17.2±5.2           | 2085.7                       |
| H 2              | *I. aquatica* | 7.7±0.2          | 17.1±2.1    | 44.3±7.4    | 18.9±4.2           | 17.3±4.8           | 475.3                       |
|                  | *H. vulgaris* | 6.5±0.0          | 3.5±3.1     | 398.3±35.5 | 51.1±7.5           | 13.5±2.3           | 6027.7                      |
| H 3              | *I. sibirica* | 21.5±0.3         | 29.3±5.8    | 89.0±19.4  | 30.4±4.7           | 21.4±7.0           | 314.0                       |
| L 1              | *O. javanica* | 7.0±0.1          | 16.3±1.4    | 48.1±9.4    | 30.2±15.5          | 20.6±4.5           | 587.1                       |
| L 2              | *I. aquatica* | 7.9±0.0          | 17.9±1.8    | 27.5±0.7    | 15.4±2.4           | 13.1±4.5           | 248.1                       |
|                  | *H. vulgaris* | 6.5±0.0          | 4.7±5.1     | 353.5±31.2 | 52.9±5.9           | 13.4±2.3           | 5338.5                      |
| L 3              | *I. sibirica* | 22.5±1.2         | 27.0±8.3    | 98.7±22.3  | 30.4±5.7           | 23.6±4.7           | 338.7                       |
Table 4. Contribution of plant assimilation to TN removal in different plant floating system.

| Treatment group | Input amounts of TN (g•m⁻²) | Removal amounts of TN (g•m⁻²) | TN assimilation (g•m⁻²) | Ratio of assimilation to input (%) | Ratio of assimilation to removal (%) |
|-----------------|------------------------------|------------------------------|-------------------------|-----------------------------------|-------------------------------------|
| H 1             | 50.27                        | 27.08                        | 9.85                    | 19.6                              | 36.4                                |
| H 2 - I. aquatica | 16.32                        | 10.14                        | 2.81                    | 17.20                             | 27.68                               |
| H 2 - H. vulgaris | 33.96                        | 24.50                        | 23.45                   | 69.05                             | 95.70                               |
| H 3             | 50.27                        | 35.15                        | 3.41                    | 6.8                               | 9.7                                 |
| L 1             | 37.39                        | 22.72                        | 2.70                    | 7.23                              | 11.9                                |
| L 2 - I. aquatica | 18.70                        | 8.11                         | 1.17                    | 6.25                              | 14.40                               |
| L 2 - H. vulgaris | 18.69                        | 15.45                        | 20.18                   | 107.98                            | 130.61                              |
| L 3             | 37.39                        | 19.51                        | 3.17                    | 8.5                               | 16.3                                |

Table 5. Quality determination of O. javanica and I. aquatica.

| Treatment group | Plant   | Tissue | NO₃⁻-N (mg•kg⁻¹) | Free amino acids (mg•kg⁻¹) | Soluble sugar (%) |
|-----------------|---------|--------|------------------|---------------------------|-------------------|
| H 1             | O. javanica | leaf   | 173.8±57.1       | 322.7±17.1                | 1.56±0.63         |
|                 |         | stem   | 296.7±153.6      | 324.1±116.5               | 0.94±0.69         |
| L 1             | O. javanica | leaf   | -                | 294.8±46.6               | 1.30±0.35         |
|                 |         | stem   | -                | 280.5±67.7               | 1.20±0.45         |
| H 2             | I. aquatica | leaf   | 187.3±24.4       | 1060.0±210.2             | 2.61±1.44         |
|                 |         | stem   | 188.0±53.4       | 1258.3±141.0             | 1.99±0.98         |
| L 2             | I. aquatica | leaf   | -                | -                        | -                  |
|                 |         | stem   | 70.1±10.8        | 1176.7±152.6             | 3.21±1.00         |

4. Conclusions
A floating Plant system was adopted to investigate purification effect of four plant species at high and low influent concentrations in this study. O. javanica and I. aquatica had both the nitrogen absorption effect and economic benefits, and were compared with common H. vulgaris and I. sibirica. Results showed that the average removal rates of TN and NH₄⁺-N in I. aquatica floating bed were relatively high both under high and low concentration of LPW. During the growth of H. vulgaris, H. vulgaris showed the best performance for nitrogen treatment, and nitrogen assimilation of it was the highest of all in the experiment. Quality determination illustrated that the NO₃⁻-N concentrations of stem and leaf in O. javanica and I. aquatica were at a low to medium level among leafy vegetables which can be safe to eat. It was also found that the existence of hydrophytes effectively controlled the rise of water pH value and algae growth and reproduction, which helped to improve the aquatic environment.

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References
[1] Yang L, Zhou X, Wang J, Wang D, Shi W and Shi L 2005 Ecological ditch system with
interception function and its effects on controlling farmland non-point pollution Chinese Journal of Ecology 24 1371

[2] Wang Y, Wang J, Li W, Bo L and Yang L 2010 Initial exploration of mechanism of ecological ditch intercepting nitrogen and phosphorus in drainage from farmland Journal of Ecology and Rural Environment 26 586

[3] Zhang Z, Feng M and Yang L 2007 Nitrogen and phosphorus removal and N2O emission from domestic sewage in floating-bed plant systems Acta Ecologica Sinica 27 4333

[4] Guo Y, Liu Y, Zeng G, Hu X, Li X, Huang D, Liu Y and Yin Y 2014 A restoration-promoting integrated floating bed and its experimental performance in eutrophication remediation J. Environ. Sci. 26 1090

[5] Bu F and Xu X 2013 Planted floating bed performance in treatment of eutrophic river water Environ. Monit. Assess. 185 9651

[6] Zhu L, Li Z and Ketola T 2011 Biomass accumulations and nutrient uptake of plants cultivated on artificial floating beds in chinas rural area Ecol. Eng. 37 1460

[7] Zhang Q, Achal V, Xu Y and Xiang W 2014 Aquaculture wastewater quality improvement by water spinach (Ipomoea aquatica Forsskal) floating bed and ecological benefit assessment in ecological agriculture district Aquac. Eng. 60 48

[8] Xin Z, Li X, Yan Z, Zhou Y, Jia Y, Tang Y, Guo W and Sun Y 2011 Purification effects of different stubble height Oenanthe javanica (Blume) DC floating bed systems in winter in two successive experimental periods Chinese J. Ecol. 30 2745

[9] Gao J, Wang W, Guo X and Zhu S 2014 Nutrient removal capability and growth characteristics of Iris sibirica in subsurface vertical flow constructed wetlands in winter Ecol. Eng. 70 351

[10] Duan J, Feng Y, Yu Y, He S, Xue L and Yang L 2016 Differences in the treatment efficiency of a cold-resistant floating bed plant receiving two types of low-pollution wastewater Environ. Monit. Assess. 188 283

[11] Yang Y, Xia M, Yang J, Wang T and Ma W 2011 The research of methods of measuring chlorophyll a in water Environmental Monitoring in China 27 24

[12] Lu R 2000 Soil agricultural chemical analysis method (Beijing: China Agricultural Science and Technology Press) p 308

[13] Hu G, Zhou M, Hou H, Zhu X and Zhang W 2010 An ecological floating-bed made from dredged lake sludge for purification of eutrophic water Ecol. Eng. 36 1448

[14] Zhang Z and Qu W 2003 Experiment guide for plant physiology (Beijing: Higher Education Press) p 118

[15] Tanner C 2001 Plants as ecosystem engineers in subsurface-flow treatment wetlands Water Sci. Technol. 44 9

[16] Orth H, Lerptocasombut K and Wilderer P 1987 Wastewater treatment for industrial estates in Southeast Asia using water hyacinths Water Sci. Technol. 19 85

[17] Zhang Q, Achal V, Xu Y and Xiang W 2014 Aquaculture wastewater quality improvement by water spinach (Ipomoea aquatica Forsskal) floating bed and ecological benefit assessment in ecological agriculture district Aquac. Eng. 60 48

[18] Yang U, Yoon S, Chung J, Kim Y, Park K, Park T and Shim S 2012 Water spinach (Ipomoea aquatica Forsk) reduced the absorption of heavy metals in an in vitro bio-mimicking model system Food Chem. Toxicol. 50 3862

[19] Chen J, Wang K, Chen H, Lu C, Huang L, Li H, Peng T and Chang S 2010 Phytoremediation of Cr (III) by Ipomoea aquatica (water spinach) from water in the presence of EDTA and chloride: Effects of Cr speciation Bioresour. Technol. 101 3033

[20] Shen M, Zhai B, Dong H and Li J 1982 Studies on nitrate accumulation in vegetable crops Acta Horticulturae Sinica 9 41

[21] Wang C, Zong Z, Li S and Chen B 2002 Nitrate accumulation in vegetables and its residual in vegetable fields Environmental Science 23 79