



Purification Efficiency of Eutrophic Water by Three Submerged Plants

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ABSTRACT

The increase of nitrogen and phosphorus causes eutrophication in water bodies. Using submerged plants to decrease the pollution from water bodies is an effective way. In this research, three common submerged plants (*Vallisneria natans*, *Hydrilla verticillata*, and *Ceratophyllum demersum*) and their combinations were used to purify eutrophic water. The control treatment did not contain any plants. The removal effects and dynamic regulations of the three plants with their combinations of nutrients (such as nitrogen and phosphorus) in water were analyzed. All three species and their combinations above could grow in the eutrophic water and efficiently remove aquatic nutrients. All the treatment groups had a higher pollutant removal rate for total nitrogen (TN) and total phosphorus (TP) than that of the blank control. In these treatment groups, treatment F (50 g *Vallisneria natans* plus 50 g *Ceratophyllum demersum*) had the highest removal rate of TP at 57.53%; treatment B (100 g *Vallisneria natans*) had the best removal rate of TN at 92.04 %. Among these plants and their combinations, *Vallisneria natans* and *Ceratophyllum demersum* showed better purification ability; the combination of these two submerged plants and the combination of three submerged plants were more applicable for the restoration of eutrophic water.

INTRODUCTION

Under the dual influence of the rapid development of modern society and human activities, water environmental problems caused by eutrophication are becoming more and more serious (Feng et al. 2020). In 2007, massive algal blooms occurred in Taihu Lake, Wuxi, causing drinking water problems for hundreds of thousands of people there and serious social impact and economic losses (Zhao et al. 2004). Therefore, there is an urgent need for a solution to the eutrophication of lake water. Besides that, most rivers, lakes, and partial landscape waters in China have experienced pollution from organic pesticides and algal blooms.

As the primary producers of a freshwater ecosystem, submerged plants can provide many kinds of food, habitat, and breeding sites for various aquatic organisms (Zhou et al. 2012). At the same time, submerged plants can improve the dissolved oxygen and light conditions of the water body, increase the spatial ecological niche of the water body, and play an important role in the matter and energy cycles in the water ecosystem (Li et al. 2010). The results show that submerged plants can improve the content of nutrient salts and planktonic algae in eutrophication water, thus improving the water quality (Wang et al. 2014). Therefore, submerged vegetation is often used for the ecological restoration of eutrophic lake water.

Submerged plant restoration is an effective means to improve the water quality and ecological environment of urban eutrophic lakes (Huang et al. 2018). Without dredging, submerged plant restoration can still effectively control the endogenous load, reduce the content of nitrogen and phosphorus in lake water, inhibit the growth of algae, and reduce the level of water eutrophication. Through the absorption of nutrients, submerged plants slow down the increase of nutrient content in the water body, conduct photosynthesis at the same time, increase the dissolved oxygen in the water body, and provide oxygen for the respiration of animals and aerobic microorganisms in the water body (Fan et al. 2007). The physiological and biochemical reactions of submerged plants absorb nitrogen and phosphorus in the water as nutrients and form nucleic acids in their own structure, so as to effectively reduce the possibility of eutrophication and control the outbreak of algal blooms (Zhang et al. 2012). The growth cycle of submerged plants is also longer, which can more effectively improve the oxidation degree of a water body than floating plants and significantly affect the content of dissolved oxygen, pH, and inorganic carbon in the water body (Zhang et al. 2009). The oxygen released from submerged plants in eutrophic waters will be rapidly consumed. The formation of the rhizosphere oxidation zone reduces the diffusion of phosphorus in sediment from sediment to the water body by

the forming iron-phosphorus complex, so as to reduce the efflux of phosphorus to a certain extent and purify the water quality (Wang et al. 2019). Different submerged plants have different removal rates of total nitrogen, ammonia nitrogen, nitrate nitrogen, and total phosphorus.

China has relatively abundant natural resources. Suzhou, located in east China, is one of the most important central cities in the Yangtze River Delta and a scenic tourist city. It has a complex water system and abundant rainfall. Meanwhile, during the industrial process, some river and lake waters have been polluted, which destroys the ecological structure and leads to the loss of the original function of the water ecosystem. Therefore, considering the characteristics of the climate and environment in Suzhou, three submerged plants (*Vallisneria natans*, *Hydrilla verticillata*, and *Ceratophyllum demersum*) and their combinations were adopted to study their effects on the purification of eutrophic water. In this study, the purification effect of them in eutrophication water was explored through controlled experiments, providing a certain theoretical basis for improving the water environment by submerged plants.

MATERIALS AND METHODS

Plant material: Seedlings of *Vallisneria natans*, *Hydrilla verticillata*, and *Ceratophyllum demersum* were bought from the flowers and seedlings base in Suzhou. The selected aquatic plants were common species in Jiangsu Province. The bought seedlings were cultured indoors for 14 days to adapt to the experimental environment, and seedlings of similar growth in each species were chosen for the experiment.

Water of treatment and experimental location: The water was tap water from the laboratory after exposure to the sun to remove chlorine. Compared with *Environmental Quality Standards for Surface Water (GB 3838-2002)*, the pH of treatment water was 7.80, and TN and TP were 5.50 and 0.45 mg·L⁻¹, respectively. The experimental location was on the platform of the second floor of the Gold Mantis School of Architecture at Soochow University. The plants were exposed to the natural sunlight fully, and there was no effect of precipitation.

Experimental method: The experiment period was July–August 2021. The temperature during the experiment was maintained at 30 ± 2°C. The plants were planted in 60-liter plastic barrels with a radius of 37 cm and a water depth of 40 cm. And the control planting barrels contained no plants. Before the experiment, seedlings of the same species and sizes were selected, and the bodies were washed carefully using tap water to avoid the adhesion of soil and other factors. The quartz sand at the bottom of the plastic barrels used to fix the plants in place was rinsed with water after purchase. The

plant combinations follow: A was the blank control with no plants; B was 100 g *Vallisneria natans*; C was 100 g *Hydrilla verticillata*; D was 100 g *Ceratophyllum demersum*; E was 50 g *Vallisneria natans* plus 50 g *Hydrilla verticillata*; F was 50 g *Vallisneria natans* plus 50 g *Ceratophyllum demersum*; G was 50 g *Hydrilla verticillata* plus 50 g *Ceratophyllum demersum* and H consisted of 33.3 g of each of the three plants mentioned above. Except for the blank control, the same initial fresh plant weight was guaranteed in each barrel. Each treatment had three repetitions.

Each water sample was taken at 7 a.m. (GMT +8) from 20 cm below the water surface in the control and treatment barrels. PH, TP, TN, NH₄⁺-N and NO₃⁻-N were analyzed each time. After the last sampling, the fresh weight and dry weight of the plant would be counted. Water lost through evaporation was added back to its original state over time.

Chemical analysis: The pH of water samples was measured with a pH meter FE20 (Mettler-Toledo, Shanghai, China). The TN was measured with the potassium persulfate digestion UV spectrophotometric method (*HJ 636-2012*), and the TP was measured with the ammonium molybdate spectrophotometric method (*GB11893-89*). The NH₄⁺-N and NO₃⁻-N were analyzed by Nessler's reagent spectrophotometry (*HJ 535-2009*) and the spectrophotometric method with phenol disulfonic acid (*GB 7480-87*).

Data processing: The removal rate was calculated using the method of Lei et al. (2015):

$$\text{Removal rate} = (C_0 - C_i) / C_0 \times 100\% \quad \dots(1)$$

C₀ and C_i are pollutant concentrations at the beginning of the experiment and on the day i, respectively.

RESULTS

Plant growth and development: The three species of submerged plants grew well during the experiment period. The water in the blank barrels remained clear and odorless. Plants all survived in the treatment environment and developed normally. They grew appreciably in length, width, and weight. Among the three species, *Vallisneria natans* grew the most in fresh weight and *Ceratophyllum demersum* in length and branch number (Fig. 1). *Ceratophyllum demersum* changed the least in its dry weight (Fig. 2). During the treatment period, the changes in fresh weight, dry weight, and moisture content of three submerged plants showed statistically significant differences. In this process, the moisture content of all three plants decreased, and the most was *Hydrilla verticillata*, from 96.73% to 95.26%. *Vallisneria natans* and *Ceratophyllum demersum* were from 96.23% to 94.02% and 96.84 % to 96.66%, respectively.

Changes in pH: The initial pH of the treatment water was

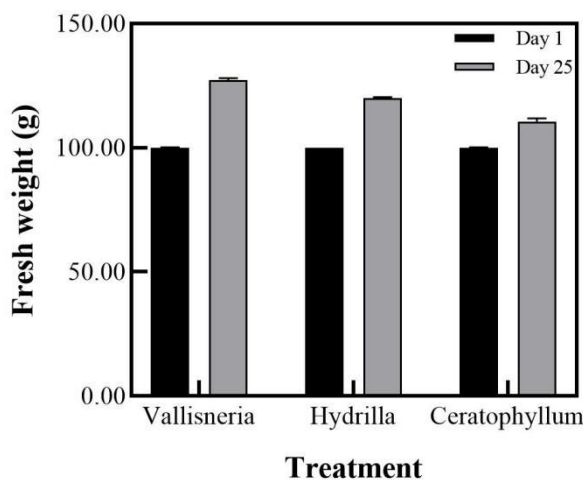


Fig. 1: Fresh weight changes of the three submerged plants on the 1st and 25th day.

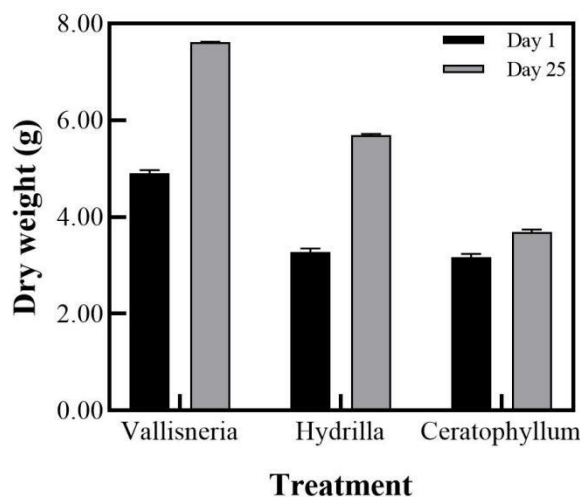


Fig. 2: Dry weight changes of the three submerged plants on the 1st and 25th day.

7.80, and there were differences in the changes in pH among the different treatments (Fig. 3). The differences between the pH of the blank control and the pH of different treatments, including three submerged plants and their combinations, were used to analyze the pH changes of eutrophic water. At the beginning of the five days, the differences between the pH of the blank control and that of all treatments increased slightly between 0.00 and 0.60. From the 5th to the 15th, the differences between the pH of the blank control and that of treatments B, C, and H continued to decrease; the differences between the pH of the blank control and that of treatments D and F increased after the first decrease, while the other two treatments decreased after the first increase. After the 15th, except for C and D, other treatments all increased slightly. And during the experiment period, the

pH in all the samples remained slightly alkaline. At the beginning of the experiment, there were no statistically significant differences between the different treatments ($p < 0.05$). On the 5th, treatments B and C had significant differences from the control groups, respectively. Over the period, only treatments E and F had significantly different results from each other on the last sampling day. The pH differences between the blank control and the other treatments varied greatly at first but eventually became stable.

TP concentration reduction and removal rate: The TP content in each treatment decreased with treatment time. Comprehensive analysis showed that the TP content of the water decreased during the sampling days. After 25 days of treatment, the removal rates of TP content in eutrophic water

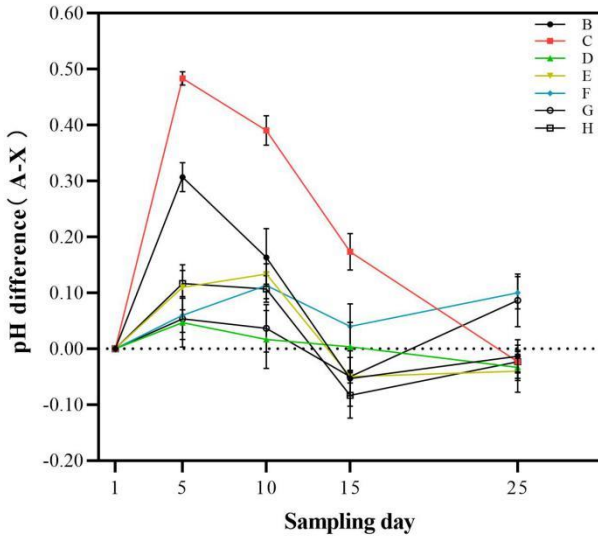


Fig. 3: Effects of different treatments on pH of eutrophic water.

by different submerged plant combinations were analyzed (Figs. 4 and 5).

The average TP content in the blank control decreased from 0.45 mg·L⁻¹ to 0.32 mg·L⁻¹. And corresponding values for treatment B were from 0.45 mg·L⁻¹ to 0.22 mg·L⁻¹; for treatment, C were from 0.45 mg·L⁻¹ to 0.26 mg·L⁻¹; for treatment, D were from 0.45 mg·L⁻¹ to 0.23 mg·L⁻¹; for treatment, E were from 0.45 mg·L⁻¹ to 0.21 mg·L⁻¹; for treatment, F were from 0.45 mg·L⁻¹ to 0.19 mg·L⁻¹; for treatment, G were from 0.45 mg·L⁻¹ to 0.24 mg·L⁻¹; and for treatment H were from 0.45 mg·L⁻¹ to 0.20 mg·L⁻¹.

At the end of the experiment, the differences between them and the blank control were significant ($P < 0.05$). On the 25th day, the removal rate of the blank control was 28.46%. Among all the treatments, treatment F had the highest removal rate at 57.53%, which was slightly higher than that of treatment H (55.41%). The removal rates of TP by treatments E, B, D, and G were 54.2%, 52.08%, 49.36%, and 46.93%, respectively. Treatment C had the lowest removal rate of TP at 42.7%. During the experiment, the variation trends of TP residues within treatments in water were similar. In the first 5 days of the experiment, the concentration decreased in a wide range and the removal rates were high, but they slowed down in the later period. After 25 days of treatments, treatment F showed only 0.19 mg·L⁻¹. In this experiment, the TP contents were removed from the water by 42.70-57.53% in the plant treatments compared with the blank control.

TN concentration reduction and removal rate: In general, the TN content of water showed a decreasing trend over time,

and the control group was no exception. The TN removal rates of plant treatments were higher than those of the blank control (Figs. 6 and 7).

The TN content of each treatment decreased with the extension of treatment time to different degrees. The average TN content in the blank control decreased from 5.50 mg·L⁻¹ to 1.69 mg·L⁻¹. And corresponding values for treatment B were from 5.50 mg·L⁻¹ to 0.44 mg·L⁻¹; for treatment C, from 5.50 mg·L⁻¹ to 1.06 mg·L⁻¹; for treatment D, from 5.50 mg·L⁻¹ to 0.54 mg·L⁻¹; for treatment E, from 5.50 mg·L⁻¹ to 0.61 mg·L⁻¹; for treatment F, from 5.50 mg·L⁻¹ to 0.51 mg·L⁻¹;

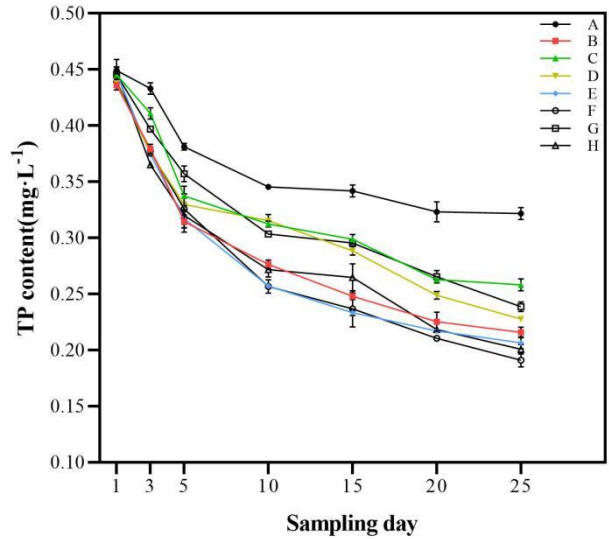
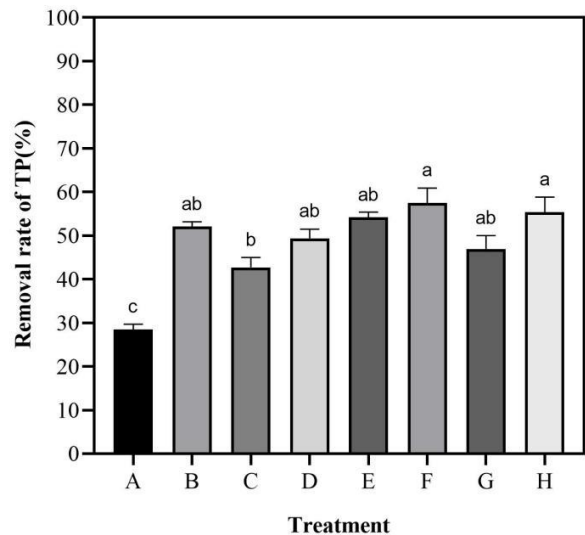


Fig. 4: Effects of different treatments on TP content of eutrophic water.



*Different low case letters above columns indicate statistical differences at $P < 0.05$.

Fig. 5: Removal rate of TP by different treatments.

for treatment G, from $5.50 \text{ mg}\cdot\text{L}^{-1}$ to $1.02 \text{ mg}\cdot\text{L}^{-1}$; and for treatment H, from $5.50 \text{ mg}\cdot\text{L}^{-1}$ to $0.47 \text{ mg}\cdot\text{L}^{-1}$.

The TN removal rates varied with plant combinations and treatment time. In our study, the TN removal rates of the treatment group containing plants were higher than those of the blank control, and the differences between them and the control group were significant ($P < 0.05$). On the 25th day, the removal rate of the blank control was 69.31%. Treatment B had the best removal rate of TN at 92.04%. The removal rates of TP by treatments H, F, D, and E were 91.38%, 90.76%, 90.24%, and 88.90%, respectively. Treatment C

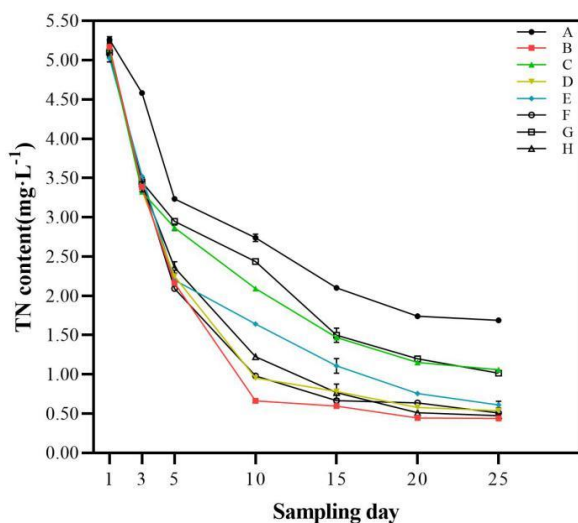
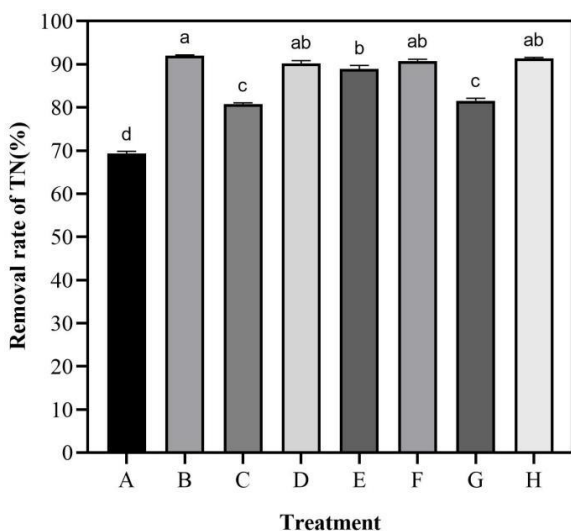


Fig. 6: Effects of different treatments on TN content of eutrophic water.



*Different low case letters above columns indicate statistical differences at $P < 0.05$.

Fig. 7: Removal rate of TN by different treatments.

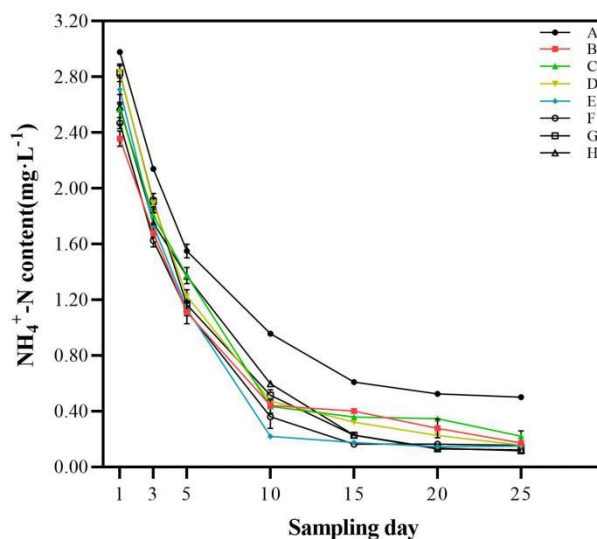


Fig. 8: Effects of different treatments on $\text{NH}_4^+\text{-N}$ content of eutrophic water.

had the lowest removal rate at 80.74%, a little lower than that of treatment G (81.53%).

$\text{NH}_4^+\text{-N}$ concentration reduction and removal rate:

During the experiment, all treatment groups had a certain removal effect on $\text{NH}_4^+\text{-N}$ in water, and the removal trends were almost the same (Fig. 8). At the end of day 25, the content of the blank control was $0.50 \text{ mg}\cdot\text{L}^{-1}$, and the final contents of all treatments were lower than $0.23 \text{ mg}\cdot\text{L}^{-1}$. There was little difference in the removal effect of $\text{NH}_4^+\text{-N}$ in different treatment groups. Among them, treatment G had the best effect, and the content of $\text{NH}_4^+\text{-N}$ in the final water was only $0.12 \text{ mg}\cdot\text{L}^{-1}$, and the removal rate was slightly higher than treatment H. Treatment B had the lowest removal effect, which was $0.22 \text{ mg}\cdot\text{L}^{-1}$.

$\text{NO}_3^-\text{-N}$ concentration reduction and removal rate:

At the first five days of the experiment, the $\text{NO}_3^-\text{-N}$ content in each treatment decreased rapidly, then the trend became gentle (Fig. 9). At the end of day 25, the content of the blank control was $1.13 \text{ mg}\cdot\text{L}^{-1}$, and the final contents of all treatments were lower than $0.55 \text{ mg}\cdot\text{L}^{-1}$. Treatment C had the best removal effect, with a final water content of less than $0.12 \text{ mg}\cdot\text{L}^{-1}$, followed by treatment B. In the process of detecting the content of this substance in water by submerged plants, the following conclusions were drawn by analyzing the experimental data. The removal effects of these three submerged plant monomers were better than those of the combined plant combinations under the same fresh weight.

DISCUSSION

Water eutrophication is one key cause of water pollution

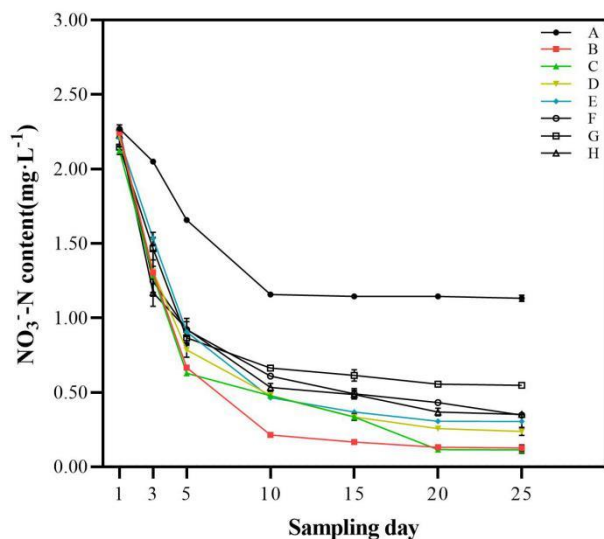


Fig. 9: Effects of different treatments on NO₃⁻-N content of eutrophic water.

(Dai et al. 2016) and a difficult problem of environmental pollution control (Quan et al. 2003). At present, there are many management methods for eutrophic water bodies, both at home and abroad. Whether it is to reduce the source of exogenous nutrients or take various measures to eliminate algae, existing methods cannot achieve a lasting and ideal effect (Gu et al. 2019). Effective control of nutrient elements in water bodies is considered to be the basic step in the management of water eutrophication and its impact on the environment (Rezania et al. 2021). Phytoremediation is a potentially suitable technology method to absorb large amounts of nitrogen, phosphorus, and plant substances during plant growth so as to avoid water eutrophication (Su et al. 2019). Submerged plants also have good ecological effects on water restoration.

The results showed that three submerged plants and their four combinations had played a significant role in purifying eutrophic water within 25 days. Generally speaking, the biomass of the studied plants increased with time during the test period, indicating that they all grew normally in the test environment and had strong ecological adaptability to nitrogen and phosphorus (Su et al. 2019). The pH differences between each treatment group and the blank control also changed from large to small, and the pH changes gradually tended to be stable. In this regard, the intervention of submerged plants also had a certain balancing effect on the pH of the water. With the change in treatment time, the pH of each group had been kept slightly alkaline, which was consistent with other results (Wang et al. 2019). After 25 days of the experiment, the concentration of total nitrogen, total phosphorus, ammonia nitrogen, and nitrate nitrogen in

each group showed a downward trend, which was also the same as some previous research results (Yang et al. 2018).

However, this research also had limitations. Many studies have used artificial sewage (Yu et al. 2019) with different initial concentrations of nitrogen and phosphorus. This research only used one concentration combination in water. This led to a lack of comprehensive analysis of the purification capacity of three submerged plants. This experiment did not simulate the living environment of submerged plants under natural conditions and had no impact from natural precipitation. The biological experiment amount was on a small scale, which would also lead to the deviation of the research results.

Although there were some limitations in this experiment, three common submerged plants in Jiangsu Province were selected for this research. The experimental results applied to the ecological environment of this region and could provide a theoretical basis and practical experience for the ecological protection of this region. Moreover, in the current research on the role of aquatic plants in the remediation of water eutrophication, more researchers have studied emergent plants and floating plants, as well as their combinations. Therefore, this experiment provided new ideas and experience for further study of submerged plants and their combinations for aquatic ecological restoration.

CONCLUSIONS

The results of this experiment showed that, compared with the blank control without submerged plants, the submerged plants and their combinations could improve the removal effect of nitrogen and phosphorus in eutrophic water and reduce the degree of eutrophication in the water. Among the three submerged plants and four combinations selected in this experiment, each treatment had a purification effect. Both *Vallisneria natans* and *Ceratophyllum demersum* had a higher capacity for monomer purification in submerged plants. Their removal rates of TP and TN from water were 92.04%, 52.08%, and 90.24%, 49.36%, respectively. *Vallisneria natans* showed better purification ability, which was consistent with many existing research results. From the aspect of combinations of submerged plants, treatments F (50 g *Vallisneria natans* plus 50 g *Ceratophyllum demersum*) and H (33.3 g of each of the three plants) were better. These two combinations removed more nitrogen and phosphorus from eutrophic water. Meanwhile, *Hydrilla verticillata* had a poorer purification capacity than others.

In this experiment, the same initial biomass of submerged plants in each treatment was used as a unified standard. The microbial community is often studied as an important part of the water ecosystem, which is of great significance

to water restoration. However, no differences in microbial communities in water before and after the experiment were found in this study. In addition, photosynthetic indexes in submerged plants were not detected in this experiment, so more comprehensive discussions and comparisons could not be carried out. The interaction between submerged plants will be carried out in future experiments.

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