Recycling of rural abandoned constructed wetlands: mariculture wastewater treatment

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ABSTRACT

This study aimed to investigate the behavioral shifts of constructed wetland (CW) when the treated water was changed from domestic wastewater to mariculture wastewater. The results showed that the average removal efficiencies of ammonium nitrogen (NH_4^+-N), total nitrogen and chemical oxygen demand (COD) were 29.54, 46.07 and 57.15% in mariculture wastewater, respectively, which were significantly lower than those in domestic wastewater (71.35, 66.34 and 74.98%, respectively). While there was no significant difference in the removal efficiency of nitrate and phosphate ($P > 0.05$) between the two systems. Based on the analysis of bacterial community and adsorption properties, the results further indicated that the removal mechanism of NH_4^+-N between both systems was mainly due to substrate adsorption: the maximum adsorption capacity of NH_4^+-N on the substrate in mariculture wastewater was 5,432 mg kg^{-1}, whereas that in domestic wastewater was 18,033 mg kg^{-1}. In terms of bacterial communities, the dominant bacteria at the family level were *Victivallaceae* (18.63%) in domestic wastewater and *Porphyromonadaceae* (18.37%) in mariculture wastewater, which showed the significant alteration to the bacterial community. In conclusion, this study showed that conventional CW could be used for treating wastewater from land-based marine aquaculture, while the operating conditions needed to be optimized in the process of application.

Key words │ bacteria community, metabolism pathways, removal efficiency, substrate adsorption

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HIGHLIGHTS

- We addressed changes in the reuse of constructed wetlands for treating mariculture wastewater.
- Significant inhibitive effects on NH_4^+-N, total nitrogen and COD removal occurred during the wastewater transformation.
- Substrate adsorption negatively affected NH_4^+-N removal efficiency in constructed wetland-treatment of mariculture wastewater.

INTRODUCTION

Recently, there has been growing interest in the development of intensive land-based marine aquaculture that

could bring economic benefits to coastal areas with environmental sustainability (Martins *et al.* 2010). In a study on land-based Atlantic salmonin farming in China, Sun *et al.* (2016) reported that only 36.5–47.8% nitrogen (N) and 20.4–38.6% phosphorus (P) in the feed were digested by fish and the residual nutrients were dissolved in drainage.

Without proper treatment, nutrient enrichment or eutrophication in aquatic environments can cause an increase in algae growth and a decrease in dissolved oxygen, which further destroys the ecological balance and structure (Liang *et al.* 2017). Therefore, the treatment of wastewater from marine aquaculture in an environmentally friendly way is crucial for sustainable intensification of aquaculture.

Many technologies have been widely employed for purification of mariculture wastewater including biological technologies and physicochemical equipment (Munavalli & Pise 2012; Zhao *et al.* 2016; Cao *et al.* 2019), while these approaches lead to some problems such as capital investment, energy consumption, maintenance requirements, secondary pollution and low removal efficiency (He *et al.* 2017). Apart from these methods, constructed wetland (CW) as a lower cost, environment friendly, work efficient and energy-saving technology with no production of secondary pollutants, have shown potential for the mariculture wastewater treatment (Teresa 2018; Wang *et al.* 2020a). In addition, with the development of the urban ecological civilization construction and the progress of rural domestic wastewater interception, many CWs that were originally used for domestic wastewater treatment in rural areas are now idled. Therefore, these CWs had the feasibility of being reused to treat mariculture wastewater. However, during the transformation of the wastewater, understanding of the key purification mechanisms of CWs for specific contaminants in mariculture wastewater remains unclear.

Therefore, the current study was conducted to (1) investigate and compare the removal efficiency of pollutants and substrate bacterial community of CW in treating domestic and mariculture wastewater and (2) clarify the influence mechanisms that cause and influence the changes in performance of pollutant removal during the transformation of the wastewater.

METHODOLOGY

Experiment design and operation

Experimental wetland system

In this study, two horizontal flow structure CW systems were constructed to treat simulated domestic wastewater (R1) and mariculture wastewater (R2). The volume of each CW was 100 L, and the inlet and outlet were arranged at the bottom and upper parts of the device, respectively. Each CW tank was filled with zeolite ($\varphi = 4$–14 mm; porosity $= 0.43$), which was taken from the CW used to treat domestic wastewater in our research group. During the experimental period, the hydraulic loading rate of each system was set to 20 cm d^{-1}.

Artificial wastewater was used in this study: mariculture wastewater was made of chemical oxygen demand (COD) (glucose) 100 mg L^{-1}, NH$_4^+$-N 25 mg L^{-1}, phosphate 10 mg L^{-1}, nitrate 30 mg L^{-1} and nitrite 5 mg L^{-1}, which were mixed with seawater; domestic wastewater used tap water and pollutants were consistent with mariculture wastewater. Water samples were collected at a fixed time from the inlet and outlet of each CW system. At the end of the experiment, substrate samples were collected from each CW system to analyze the bacterial community.

Adsorption properties of the substrate

A quantitative sterilized substrate of CW was placed in 250 mL conical flasks. 200 mL 30 mg L^{-1} ammonia solution and 200 mL 30 mg L^{-1} ammonia solution with 3% NaCl were added to two separate conical flasks, which were placed in a thermostatic oscillator at 50 r min^{-1}, 25 °C. Samples were taken at 0, 10, 30, 60, 120, 240, 360 and 480 min. Finally, the adsorption capacity of the substrate to NH$_4^+$-N was calculated according to the residual concentration of the solution in each conical flask. Based on the experimental results, the quasi first-order model, quasi second-order model and intramolecular diffusion model were used to fit the adsorption process of the substrate for ammonium nitrogen. The three models are shown as follows:

Quasi first-order model: $\ln(C_e - C_t) = -K_1 t + \ln C_e$ (1)

Quasi second-order model: $\dfrac{t}{C_t} = \dfrac{1}{k_2 C_e^2} + \dfrac{1}{C_e} t$ (2)

Intramolecular diffusion model: $C_t = K_p t^{0.5}$ (3)

where C_e is the amount of pollutants adsorbed by the substrate at adsorption equilibrium (mg kg^{-1}); C_t is the amount of pollutants adsorbed by the substrate at t (mg kg^{-1}); K_1 is

Quasi first-order model adsorption kinetic constant (min^{-1}); K_2 is Quasi second-order model adsorption kinetic constant ($kg\,mg^{-1}\,min^{-1}$) and K_p is the diffusion rate constant ($mg\,kg^{-1}\,min^{1/2}$).

A quantitative sterilized substrate of CW was added to 250 mL conical flasks and ammonia solution at 10, 15, 25, 30 and 35 $mg\,L^{-1}$ with and without 3% NaCl was added to two equal sets of these flasks. Then, the conical flasks were placed in a thermostatic oscillator at 50 $r\,min^{-1}$, 25 °C and equilibrated 10 h. Finally, the adsorption capacity of the substrate to ammonium nitrogen was calculated according to the residual concentration of the solution in each conical flask. Combined with the experimental results, the Langmuir, Freundlich equation and Dubinin–Radushkevich (D-R) equation can be used to describe the relationship between the adsorption quantities of the substrate material and the equilibrium concentration of the solution. The three models are shown as follows:

$$\text{Langmuir equation: } \frac{C_w}{C_s} = \frac{C_w}{Q_{max}} + \frac{1}{K_L} \tag{4}$$

$$\text{Freundlich equation: } \lg C_s = \lg K_f + \frac{1}{n}\lg C_w \tag{5}$$

$$\text{Dubinin–Radushkevich equation: } \ln C_s = \ln Q_{max} - \beta\varepsilon^2$$
$$\varepsilon = RT\ln\left(1 + \frac{1}{C_w}\right)$$
$$E = \frac{1}{(2\beta)^{1/2}} \tag{6}$$

where C_w is the concentration of pollutants in the solution at adsorption equilibrium ($mg\,L^{-1}$); C_s is the adsorption capacity per unit substrate material ($mg\,kg^{-1}$); Q_{max} is the maximum adsorption capacity per unit substrate material ($mg\,kg^{-1}$); K_L is the Langmuir equilibrium constant, the larger the value, the stronger the adsorption capacity ($L\,kg^{-1}$); K_f is the Freundlich coefficient, which represents the adsorptive capacity of the substrate ($L\,kg^{-1}$) and n is a constant of the equation, which can represent the adsorptive strength of the substrate. β is the D-R adsorption constant; ε is the Polanyi potential; R is the ideal gas constant, 8.314 $J\,mol^{-1}\,K^{-1}$; T is the absolute temperature (K) and E is the average adsorption energy ($kJ\,mol^{-1}$).

Sample analysis

Samples were analyzed for NH_4^+-N, nitrate, nitrite, total nitrogen, COD and phosphate. In R1, all pollutants were measured based on the national standard method (China E.P.A. 2002). In R2, considering the character of mariculture wastewater, the NH_4^+-N was measured by indophenol blue spectrophotometry, COD was measured by the potassium permanganate method and other pollutants were measured based on the national standard method. In addition, as nitrite can interfere with the use of COD as an indicator of organic matter content; thus, in this study, COD correction data were applied to characterize the removal of organic matter.

Substrate samples were extracted by shaking each sample in 100 mL of sterile physiological saline with 100 μL of Tween 80 detergent solution using a vortex mixer for 10 min. Then, the solution was filtered through a 0.22 μm polycarbonate filter (Millipore, MA, USA) to collect the microorganisms. All the processed samples were stored at −80 °C until microbial DNA extraction. The total DNA on the filter paper was extracted with E.Z.N.A.® Water DNA Kit (Omega Bio-Tek, Norcross, GA, USA) according to the manufacturer's protocol. Thereafter, the extracted DNA was separated using agarose gel (1.0%) at 150 V for 20 min to examine the quality of DNA. DNA purity and quantity were determined using a NanoDrop spectrophotometer (NanoDrop Technologies Inc., Wilmington, DE, USA). Extracted DNAs was diluted to 1 $ng\cdot(l\,μL)^{-1}$ with sterile water in a centrifuge tube and stored at −80 °C prior to high-throughput sequencing. Deep sequencing of the 16S rRNA gene amplicons from the R1 and R2 samples was performed using the Illumina HiSeq 2000. Polymerase chain reaction (PCR) was carried out using Phusion® High-Fidelity PCR Master Mix with GC Buffer (New England Biolabs), the 1 $ng\cdot(l\,μL)^{-1}$ of genomic DNA, the specific primer with Barcode and high-fidelity enzyme. The primer pair used for PCR was 341F (5′-CCTAYGGGRBGCASCAG-3′) and 806R (5′-GGAC-TACNNGGGTAT CTAAT-3′), targeting the V3-V4 hypervariable region of the bacterial 16S rRNA gene. The PCR products were separated by electrophoresis on a 2% agarose gel. Then, Uparse software was used to cluster all effective tags of all samples, obtain operational taxonomic

units (OTUs) and conduct annotated analysis of species with the GreenGene database. Finally, PICRUSt function prediction analysis was used to clarify the variations in metabolic pathways at R1 and R2 at the gene level. After obtaining KEGG Orthology (KO) information according to OTU, the abundances of relevant functional enzyme-encoding genes involved in the metabolism of nitrogen and carbon were calculated based on OTU abundance and KO database.

Statistical analysis

In our experiment, the statistical analysis was performed using SPSS. One-way analysis of variance (ANOVA) was used to compare mean values between different treatments with the least significant difference (LSD) test at the significance level of 0.05.

RESULTS

Pollutant removal

The pollutant removal efficiencies in R1 and R2 are shown in Figure 1. During the transformation of the wastewater, there were significant differences ($P < 0.05$) in the removal efficiencies of NH_4^+-N, total nitrogen and COD in CW. In R1, the average removal efficiencies of NH_4^+-N, total

Figure 1 | Pollutant removal efficiency in both systems.

nitrogen and COD were 71.35, 66.34 and 74.98%, respectively. In R2, the average removal efficiency of NH_4^+-N, total nitrogen and COD were all significantly lower than R1 ($P < 0.05$) at 29.54, 46.07 and 57.15%, respectively. However, the removal efficiency of phosphate and nitrate was not statistically significant between the two systems ($P > 0.05$). In R1, the average removal efficiencies of these two indices were 77.78 and 16.44%, and in R2, they were 75.31 and 16.78%, respectively.

Bacterial community

The relative abundances of the dominant bacterial community (>1%) at phylum, class, order and family levels in substrate samples of each respective CW system are presented in Figure 2. During the transformation of the CW treatment process, the bacterial community retained high similarity at the phylum level. The dominant phylum in R1 and R2 was *Proteobacteria*, with relative abundances of 26.35 and 48.35%, respectively. However, there were several differences between these two systems at other taxonomic levels during the treatment of mariculture wastewater in CW. For example, in R1, *Lentisphaeria*, *Victivallales* and *Victivallaceae* were the dominant bacteria at class, order and family levels, with relative abundances of 18.66, 18.63 and 18.63%, respectively, whereas these bacteria were no longer predominant in R2, and their relative abundances were all reduced to 0.07%. In R2, *Bacteroidia*, *Bacteroidales* and *Porphyromonadaceae* had become the dominant bacteria at class, order and family levels and their relative abundances were 27.76, 27.76 and 18.37%, respectively. In addition, we found that the microorganisms in CW were not uniformly distributed on the surface of zeolite, and they mainly existed in the cracks of zeolite.

Carbon and nitrogen metabolism pathways in CW

To reveal the changes in the degradation pathways of nitrogen and carbon in different CW systems, relevant functional enzyme-encoding genes from R1 and R2 were analyzed based on function prediction analysis (Figures 3 and 4). The nitrogen metabolism pathways in R1 and R2 were the same and included four reduction pathways of nitrification, assimilation nitrate reduction, dissimilation nitrate
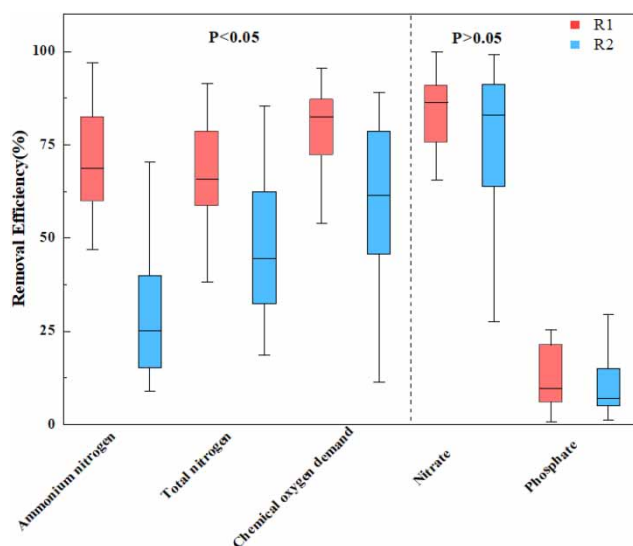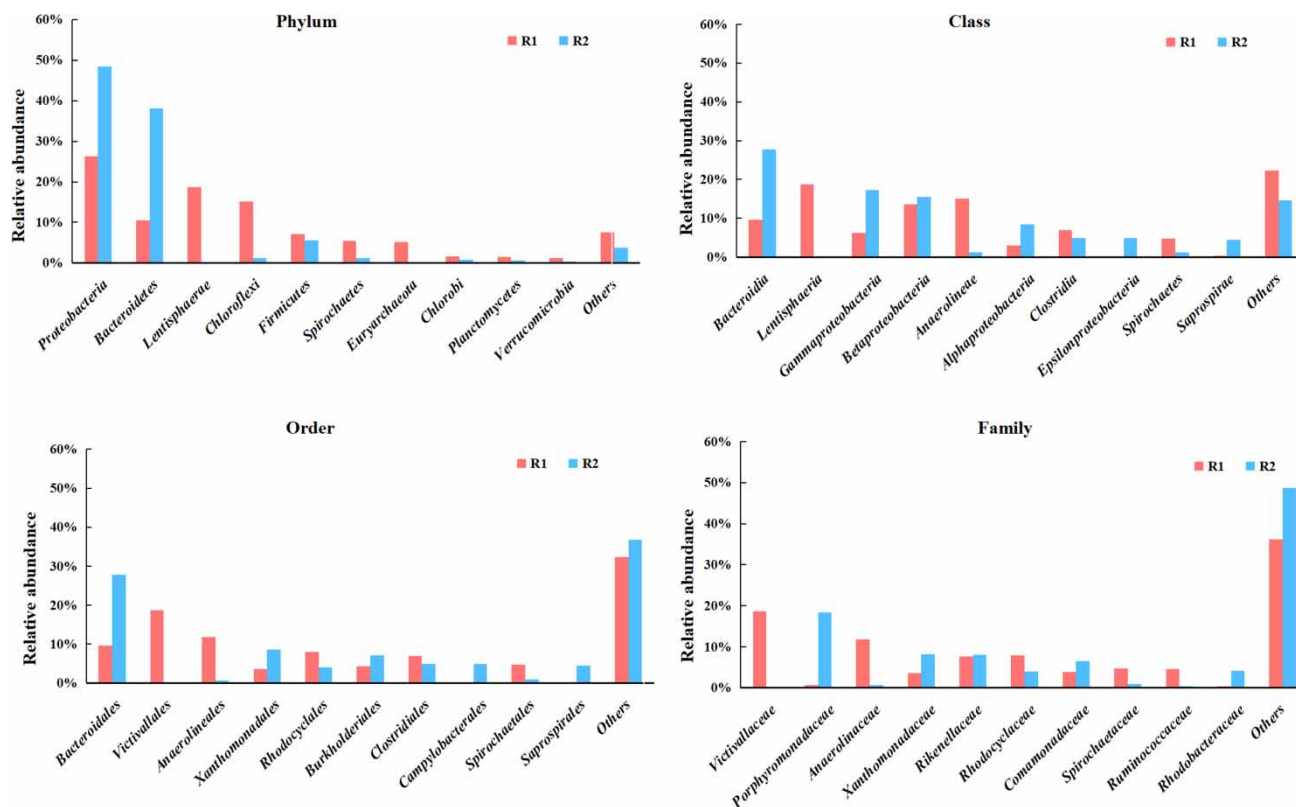
Figure 2 │ Taxonomic affiliation of dominant bacterial diversity at phylum, class, order and family levels in different substrates.
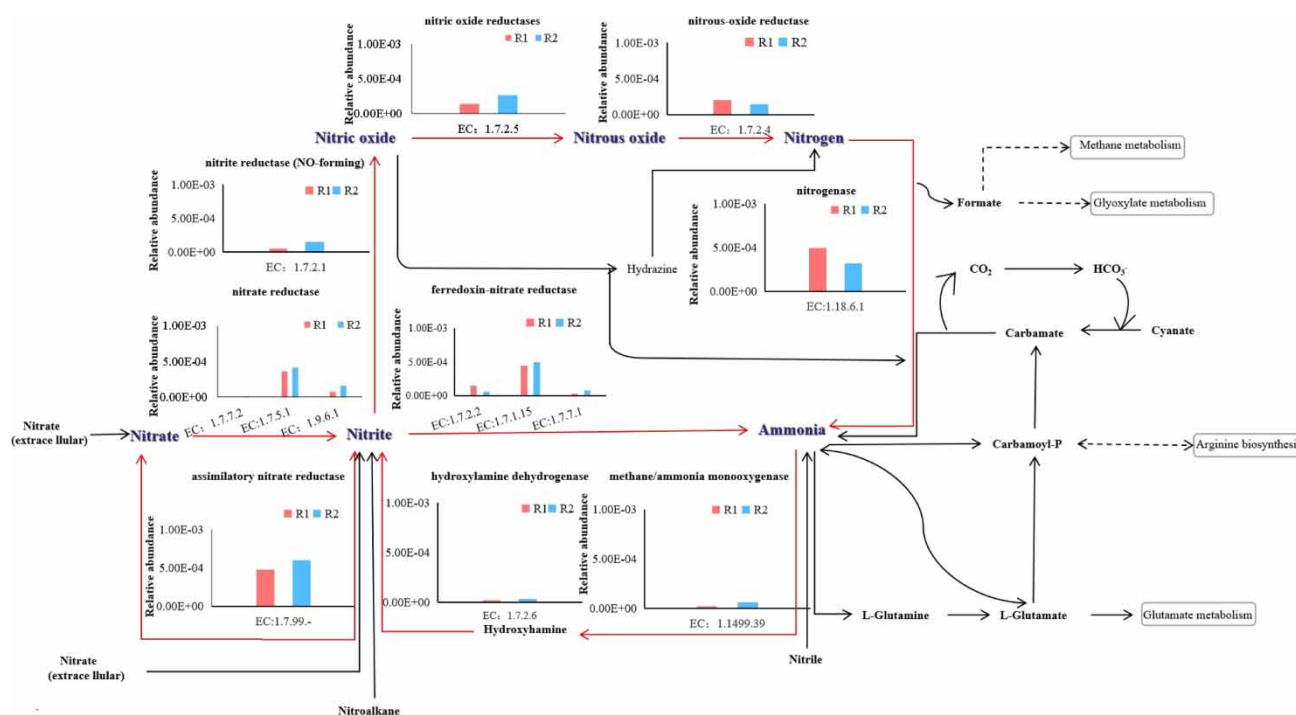

Figure 3 │ Nitrogen metabolic pathways and relative abundance of enzyme-encoding genes in two CWs.
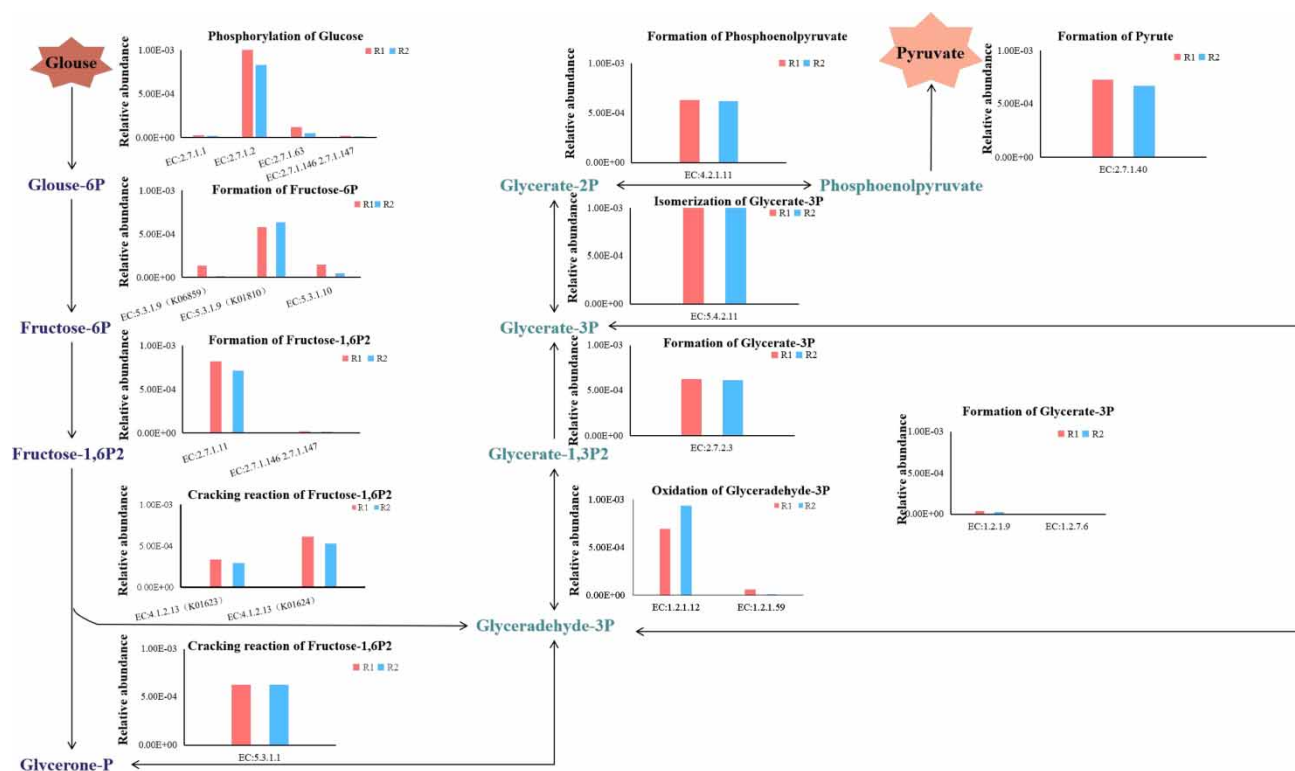
Figure 4 | Carbon metabolic pathways and relative abundance of enzyme-encoding genes in two CWs.

reduction and denitrification, and two oxidation pathways of nitrification and complete nitrification comammox, while the relative abundance of functional enzyme-encoding genes in different pathways were different. The relative abundance of encoding genes of nitrogenase (EC: 1.18.6.1) participating in nitrification was slightly higher in R1 (4.93×10^{-4}) than that in R2 (3.21×10^{-4}). During nitrification and complete nitrification comammox, the relative abundance of encoding genes of methane/ammonia monooxygenase, hydroxylamine dehydrogenase and assimilatory nitrate reductase (EC: 1.1499.39, EC: 1.7.2.6 and EC: 1.7.99, respectively) in R1 $(2.27 \times 10^{-5}, 2.16 \times 10^{-5}$ and 4.78×10^{-4}, respectively) were slightly lower than in R2 $(6.22 \times 10^{-5}, 3.46 \times 10^{-5}$ and 5.97×10^{-4}, respectively), and this phenomenon also existed in the process of assimilation nitrate reduction. At dissimilation nitrate reduction, the relative abundance of encoding genes of functional enzymes in R1 and R2 varied in different reaction stages. For example, at the nitrate to nitrite stage, the proportion of encoding genes of nitrate reductase/nitrite oxidoreductase (EC: 1.7.5.1) in R2 (4.09×10^{-4}) was higher than that in R1

(3.61×10^{-4}); however, at the nitrite to ammonium stage, the proportion of encoding genes of ferredoxin–nitrate reductase (EC: 1.7.2.2) in R2 (5.84×10^{-5}) was obviously lower than that in R1 (1.4×10^{-4}). For carbon metabolism pathways, we only focused on the Embden–Meyerhof pathway of glycolysis which was closely related to carbon source. The Embden–Meyerhof pathway begins with phosphorylation of glucose and ends in pyruvate formation. In this stage, the encoding genes of major functional enzymes were only marginally more abundant in R1 (9.20×10^{-3}) than in R2 (8.02×10^{-3}). For example, the relative abundance of encoding genes of glucokinase was obviously higher in R1 (1.05×10^{-3}) than in R2 (8.32×10^{-4}). Notably, this phenomenon implied that the removal efficiency of COD in R1 was higher than in R2.

Adsorption properties of NH_4^+-N

Since there was a greater significant difference in the NH_4^+-N removal efficiencies compared with that of the other pollutant removal efficiencies between the different

CW systems, the removal mechanism of NH_4^+-N was further analyzed by substrate the adsorption properties (Table 1). Adsorption kinetics are primarily used to describe the rate at which adsorbents adsorb solutes. The results shown in Table 1 indicated that in simulated wastewater, three models all could fit the adsorption process of NH_4^+-N on zeolite ($R^2 > 0.9$, $P < 0.05$). However, in wastewater with 3% NaCl, the quasi second-order model could fit the adsorption process of NH_4^+-N on zeolite ($R^2 = 0.91$) better than the intramolecular diffusion model ($R^2 = 0.81$) and the quasi first-order model ($R^2 = 0.84$). Therefore, we selected the quasi second-order model to analyze the difference in adsorption between the different systems. Based on these parameters, it could find that the adsorption rate of zeolite to NH_4^+-N in the wastewater (7.04×10^{-5} kg mg^{-1} min^{-1}) was lower than that in the wastewater with 3% NaCl (1.90×10^{-4} kg mg^{-1} min^{-1}). In addition, the quasi second-order model assumed that the adsorption rate was controlled by chemisorption (Liu *et al.* 2014), thereby the result also indirectly explained that the adsorption process of NH_4^+-N on zeolite in the different wastewater was chemisorption.

Table 1 | Parameters of adsorption properties for NH_4^+-N

	NH_4^+-N solution	NH_4^+-N solution with 3% NaCl
Quasi first-order model		
K_1 (min^{-1})	1.17×10^{-2}	9.2×10^{-3}
R^2	0.91	0.81
Quasi second-order model		
K_2 (kg mg^{-1} min^{-1})	7.04×10^{-5}	1.90×10^{-4}
R^2	0.95	0.91
Intramolecular diffusion model		
K_p (mg kg^{-1} min$^{1/2}$)	10.69	2.52
R^2	0.93	0.84
Freundlich		
K_f (L kg^{-1})	23.59	0.037
n	1.05	0.45
R^2	0.95	0.83
D-R		
Q_{max} (mg kg^{-1})	18,033	5,432
E (kJ mol^{-1})	10	8.3
R^2	0.80	0.94

Adsorption isotherm describes the solute molecule concentration between the liquid phase and the solid phase when the adsorption processes of the molecules are in equilibrium at a given temperature. In this experiment, the Langmuir equation was unable to fit the data ($R^2 < 0.3$, $P > 0.05$), so we selected the Freundlich equation and D-R equation to analyze the difference in adsorption among the different systems ($R^2 > 0.8$, $P < 0.05$). As shown in Table 1, the Freundlich equation and D-R equation fitted the NH_4^+-N data in simulated wastewater and wastewater with 3% NaCl ($R^2 > 0.9$, $P < 0.05$), respectively, and the respective K_f values, n values and Q_{max} values were ranked as follows: wastewater with 3% NaCl ($K_f = 0.037$ L kg^{-1}, $n = 0.45$, $Q_{max} = 5,432$ mg kg^{-1}) < wastewater ($K_f = 23.59$ L kg^{-1}, $n = 1.05$, $Q_{max} = 18,033$ mg kg^{-1}). The n values of the Freundlich model were used to describe the adsorption strength for NH_4^+-N were 1.05 in wastewater and 0.45 in wastewater with 3% NaCl. Based on these results, NH_4^+-N had a higher theoretical saturation capacity and adsorption strength in wastewater than in wastewater containing 3% NaCl. In addition, the E values of the D-R model indirectly explained the adsorption mechanism and the values lie within the range expected for chemical adsorption and physical adsorption (8–16 and <8 kJ mol^{-1}, respectively) (Fan *et al.* 2016). From Table 1, the values of E were 10 kJ mol^{-1} in wastewater and 8.3 kJ mol^{-1} in simulated wastewater with 3% NaCl, respectively. Thus, in our experiment, the adsorption mechanism of the zeolite for NH_4^+-N was chemisorption, which was consistent with the results of the adsorption kinetics simulation.

DISCUSSION

In this study, the inhibitive effects on NH_4^+-N and total nitrogen removal were observed during treatment transformation with inhibition ratios of 41.81 and 20.27%, respectively, which are consistent with the previous studies (Liang *et al.* 2017; Fu *et al.* 2019). However, there was no significant removal difference in phosphate and nitrate which was different from other studies. For example, Gao *et al.* (2015) found that the salt in mariculture wastewater could negatively influence phosphate removal in CW. Moreover, there are limited literature reviews on COD during CW for

treating mariculture wastewater. While in our experiment, COD removal was also inhibited, and the inhibition ratio was 21.67%.

Generally, microbes play a key role in the nitrification and denitrification processes, which are widely considered as the major N removal mechanism in CWs (Vymazal 2007). Besides, substrate adsorption was another major way for N removal in CWs due to zeolite as the substrate in this experiment. From Figure 3, in nitrification, the relative abundance encoding genes of enzymes involved in mariculture wastewater (9.7×10^{-5}) were more abundant than those in domestic wastewater (4.5×10^{-5}). In contrast, the removal efficiency of NH_4^+-N in mariculture wastewater was significantly lower than that of domestic wastewater. Combining the results of adsorption properties, we believed the reason for this phenomenon was the adsorption property of NH_4^+-N on the substrate. Natural zeolite is a kind of silicate with silica-oxygen tetrahedron and alumina-oxygen tetrahedron as its skeleton and it has large specific surface area (SSA), strong ion exchange capacity and high static adsorption capacity for NH_4^+-N (Inglezakis et al. 2018; Wang et al. 2018, 2020b). However, the adsorption process is affected by environmental factors, such as organic matter, PH and salinity. Mariculture wastewater is a form of saline wastewater, and when sodium ions are present in the water, they can displace the cations on the zeolite surface (Behin et al. 2019), thereby reducing the steric hindrance and increasing the adsorption rate of zeolite for ammonium ions. Simultaneously, sodium ions would compete with the ammonium ions for the adsorption sites on the zeolite surface, so that the adsorption capacity of NH_4^+-N on zeolite would decrease during the transformation of wastewater as shown in Table 1.

Compared with NH_4^+-N, nitrate removal efficiency did not significantly differ between the two systems. In our research, we thought that there were two main reasons for this phenomenon: (1) low substrate adsorption capacity and (2) insufficient carbon compounds in the denitrification process. A previous study concluded that the affinity for adsorption of nitrate onto Al oxides was weak (Zhou & Haynes 2010). Although there was a salinity difference between the two systems, this did not affect the adsorption for nitrate on the substrate. In denitrification, theoretically, the reduction 1.0 g nitrate to nitrogen requires

approximately 4.4 g of organic matter (COD). However, the organic content in both systems could not meet the requirements of denitrification. Therefore, the carbon source became a limiting factor in the denitrification process which limited the significant difference in nitrate removal efficiency between different systems. Combining these two factors, there was no difference in the nitrate removal efficiency during the transformation of the wastewater. This conclusion also could be supported by the analysis results of the bacterial community and nitrogen metabolism pathway. As shown in Figures 2 and 3, in denitrification, the relative abundance of encoding genes of enzymes in mariculture wastewater was not inhibited; several genes even had elevated expression. For example, the relative abundance of nirK, which is a key functional gene in denitrification (Jones & Hallin 2010) in R2 (1.4×10^{-4}) was greater than that in R1 (5.2×10^{-5}). This result indicated that there was no effect on denitrification after the transformation of the wastewater objective. In terms of total nitrogen, we thought that the NH_4^+-N removal efficiency in mariculture wastewater was significantly lower than that in domestic wastewater, which directly led to lower total nitrogen removal efficiency in mariculture wastewater.

The removal of COD in CW mainly depends on the metabolic activities of microorganisms. As shown in Figure 2, the relative abundances of the bacterial community at the family level in each CW were significantly different. The dominant bacteria changed from Victivallaceae to Porphyromonadaceae during the transformation of the wastewater. Previous studies have shown that the growth of Victivallis is chemoorganotrophic and restricted to a variety of sugars (Plugge & Zoetendal 2014). The CW microbes were originally used for domestic wastewater treatment, Victivallis was abundant in domestic wastewater, and significantly improved the removal efficiency of COD in domestic wastewater compared with that in mariculture wastewater. In addition, as shown in Figure 4, relevant functional enzyme-encoding genes involved in glycolysis were more abundant in domestic wastewater than that in mariculture wastewater, which also implied that the removal efficiency of COD may be higher in domestic wastewater than that in mariculture wastewater, and this result was consistent with COD removal.

With regards to phosphate, CWs can effectively remove phosphorus through filtration, adsorption, co-precipitation,

ion exchange, plant absorption and microbial decomposition by taking advantage of the physical, chemical and biological coordination (Vymazal 2007). However, in our experiment, we did not add the plants to the CW, and the most important point was that there was no biofilm-enhanced phosphorus removal mechanism in our systems because the environmental conditions were not suitable for the growth of phosphorus accumulating organisms. Thus, in our study, the phosphate removal mechanism mainly depended on the biosynthetic utilization and substrate adsorption. In general, the maximum phosphate uptake rate of bacteria was achieved under a higher concentration ratio of organic carbon to phosphate in the wastewater. However, the organic content in both systems was all insufficient. In addition, Zhou & Haynes (2010) found that the relative affinities for adsorption of anions onto Al oxides often follow the order: phosphate > chloride ion. Therefore, chlorine ions in mariculture wastewater did not affect the adsorption of phosphate onto the substrate. Considering these two factors, there was no significant difference in the phosphate removal efficiency between the two systems. Besides, zeolites are a class of crystalline aluminosilicates that have a net negative charge due to the substitution of Si (IV) by Al (III) in the Si tetrahedral (Nikolakis 2005). Therefore, adsorption of anions, such as phosphate, by zeolites is generally lower than that of cations (Haynes 2015).

CONCLUSION

Conventional CW can be applied in mariculture wastewater treatment, while the process was affected by wastewater characteristic, and it had negative effects on NH_4^+-N, total nitrogen and COD removal efficiency. These negative effects on COD were mainly caused by salts inhibiting the function of microorganisms in the substrate which will further change the community structures; meanwhile, salts would weaken NH_4^+-N removal efficiency by the substrate due to the competition for uptake into ion. This study provides both a new strategy for reuse of abandoned CW in rural areas and a new understanding of the removal mechanism of mariculture wastewater in CW.

DATA AVAILABILITY STATEMENT

All relevant data are included in the paper or its Supplementary Information.

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