

Combining artificial aeration and biological zeolite mulch for nitrogen removal from eutrophic water bodies

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Abstract

Endogenous nitrogen pollution can be a major cause of eutrophication. Nitrogen species release from sediments can be reduced by biologically-enforced zeolite mulch or water column aeration. This study was about their combined effects. Water and surface sediment samples from the Yangzhou ancient canal were aerated and biozeolite mulching was applied separately and in combination for 81 days, while the nitrogen species removal rate was recorded. The combination of aeration and biozeolite mulching removed >95% of the ammoniacal-nitrogen in 15 days. This was better than either the blank control or biozeolite mulch without aeration. The ammoniacal-nitrogen concentration was lowered faster by combined treatment than by aeration alone. Nitrate nitrogen was only detected during aeration between days 10 and 18, and reached lower concentration in the presence of biozeolite. Nitrate was formed during aeration but its concentrations were higher and more variable in the absence of biozeolite; that is, mulching stabilized nitrate formation. The total nitrogen concentration reached its lowest levels after 81 days with biozeolite treatment alone, with 78% total nitrogen removal, whereas combined aeration with biozeolite achieved 41%. This shows that biozeolite mulching can remove nitrogen in eutrophic waters, even without aeration.

Key words: aeration, biological zeolite, coupling effect, nitrogen removal

INTRODUCTION

Eutrophication is an important environment problem caused by excessive nitrogen and phosphorus concentrations arising from both exogenous and endogenous pollution. Exogenous pollution (mostly agriculture and sewage related) can be controlled by regulation, but, even with complete compliance, the problem of endogenous nutrient release from sediments remains. Various methods have been adopted to avoid or remediate endogenous pollution or the subsequent eutrophication resulting in anoxic waters, including sediment dredging, chemical flocculation, artificial aeration or covering of the sediment. Some of these solutions have serious disadvantages: dredging is expensive, disturbs aquatic ecosystems and can cause secondary pollution, while chemical flocculation can introduce new pollutants. Therefore, artificial aeration and sediment coverage have received most attention (Foyle & Norton 2007; Pan *et al.* 2019).

Artificial aeration can restore dissolved oxygen concentrations in the water column and has been studied to overcome the effects of endogenous nitrogen and phosphorus release from sediments (Pan *et al.* 2019). Restoring dissolved oxygen sufficiently can limit nitrogen release, as ammonia, from sediments but can also lead to the release of more ammoniacal and total nitrogen. The aeration

technology is crucial, as, for instance, micro-nano aeration results in a higher total volume and oxygen mass transfer coefficient than blast microporous aeration (Sun *et al.* 2018). Aeration depth is also important in relation to total nitrogen release and the type of nitrogen species released, as river sediment studies have shown (Yu *et al.* 2019). Different aeration modes have been studied in heavily polluted rivers, where it was found that the removal rate of total nitrogen (TN) was significantly higher with intermittent rather than continuous aeration (Seonki *et al.* 2019; Zheng *et al.* 2019). The effect of artificial aeration depends on microbial activity. The composition of the microbial community involved in endogenous nitrogen transformation under intermittent aeration in a river has been investigated and the organisms' functional groups identified (Liu *et al.* 2019).

Aeration technology can also be combined with other control measures; for example, technology stimulating biofilm formation (Clifford *et al.* 2010; Ding *et al.* 2010; Chen *et al.* 2013) or bioremediation (Chang *et al.* 2013; Sun *et al.* 2018). Moreover, it was found that the unique micro-environment formed by the root systems of floating water plants can be an important factor responsible for enhanced aeration and denitrification (Sun *et al.* 2018). The progress in microbial remediation combined with aeration technology contrasts with the lack of effort to couple this to sediment coverage methodologies. As a stand-alone measure, sediment coverage has been proved effective in controlling endogenous nitrogen release.

Sediment coverage has been performed with various adsorbents including bioactive materials like ferric and aluminum salts, and precipitants like quicklime (Liao *et al.* 2017; Sun *et al.* 2019). Various phosphorus passivating agents modified by iron salts (Gibbs & Özkundakci 2011; Meis *et al.* 2012), and natural or modified zeolites (Lin *et al.* 2011; Sun *et al.* 2011; Xiong *et al.* 2018) have also been applied. In one study, the effect of combining zeolite and calcite to inhibit nitrogen and phosphorus release from sediments into water under anaerobic conditions was investigated (Sun *et al.* 2011). In another, the effect of covering sediments with modified zeolite in eutrophic lakes was described (Gibbs & Özkundakci 2011). Both showed that the technology can control nitrogen and phosphorus release from sediment effectively, reducing their concentrations in the overlying water.

Biologically enforced zeolite has also been used as covering material to reduce endogenous nitrogen release (Huang *et al.* 2011). Zeolite can be regenerated to exchange ammonium ions by biological nitrification, and this methodology can remove nitrogen by synchronous nitrification and denitrification. It appears, however, that the combination of artificial aeration and biologically enforced zeolite (biozeolite) mulching has not been tested to control nitrogen pollutant release from sediments.

The aim of this study was to investigate the coupling effect of artificial aeration and biozeolite mulching to control endogenous nitrogen pollution in an experimental setup, to provide theoretical support and technical reference for the control of actual endogenous nitrogen pollution in natural waters.

MATERIALS AND METHODS

Sampling site, sediment and water characteristics

The sediment and overlying water used in this study were derived from a section of the centuries-old Yangzhou Sanwan canal, in Yangzhou city, Jiangsu Province, China (latitude 32.36, longitude 119.43) (Figure 1). The overlying water was collected in 50 L plastic barrels, and the sediment obtained by a grab dredger and transported directly to the laboratory. The raw water was passed through 45 µm filters and sterilized at 120 °C for 30 minutes within 2 hours of sampling. Coarse particles and plant residues were removed from the sediment prior to use. *In situ*, the water temperature was 18 °C and its pH 8.12 and it contained 3.03 mg/L ammoniacal-nitrogen, 0.72 mg/L nitrate-nitrogen, 0.1 mg/L nitrite-nitrogen and 5 mg/L total nitrogen (TN). The total phosphorus concentration was



Figure 1 | The Yangzhou Sanwan section of the ancient canal used as the sampling site. The map shows part of Yangzhou District and the arrow marks the sampling site, shown in the photograph.

0.7 mg/L including 0.26 mg/L soluble phosphorus. The chemical oxygen demand by potassium permanganate (COD_{Mn}) was 5.76 mg/L and the total organic carbon (TOC) 8.13 mg/L.

Biologically enforced zeolite

The natural zeolite used (Figure 2) came from Bayan Nur in Inner Mongolia, China. The particle size is about 1–2 mm, with average pore size 6.75 nm, bulk density 1.01 g/cm³, and true density 2.29 g/cm³ (Huang *et al.* 2011). It was inoculated with bacteria to produce the biozeolite.



Figure 2 | Biozeolite used in the study.

Pure cultures of the heterotrophic nitrifying *Bacillus* sp. strains WGX10 and WGX18, and the two aerobic denitrifying *Acinetobacter* sp. strains HF3 and HF7 described and characterized by others (Zhou 2013) were inoculated into 100 mL of medium in 150 mL conical flasks and incubated, for 2–3 days at 30 °C while being rotated (120 rpm). The cultures were used when $\text{OD}_{600 \text{ nm}}$ reached 0.1 when they were mixed at equal volume and added to the filtered and sterilized raw water at a ratio of 1:9. This mixture was put into a container with the zeolite so that it was covered with at least 5 cm of liquid. The dissolved oxygen concentration was controlled to remain above 2–3 mg/L by continuous aeration for 3 days, during which time the bacteria formed a biofilm on the zeolite surface. On day 3, unattached bacteria were removed by washing under continuous water flow, to obtain biofilm-covered biozeolite. (Zhou 2013).

Test equipment

Figure 3 is a schematic of the experimental setup. Four Plexiglas columns (1 m high, 200 mm diameter) were each filled with a 20 cm layer of sediment overlain with 60 cm of filtered and sterilized river water. In two columns, the sediment was covered with a biozeolite layer approximately 2 mm thick (about 2 kg/m²). Two of the columns were aerated. The blank control (column 1) contained no biozeolite and was not aerated. The aeration control (column 2) was also without biozeolite but was aerated continuously with pure oxygen. Column 3 contained biozeolite but was not aerated, while column 4 contained biozeolite and was aerated (like column 2). Ports 42 cm above the bottom were used to sample the water. Each column was covered in black plastic to exclude light and they were all incubated for 81 days. The water temperature, and concentrations of dissolved oxygen (DO) and nitrogen species were monitored periodically using approximately 40 mL samples to measure the concentrations of TN, ammoniacal nitrogen (NH₄⁺-N), and nitrite (NO₂⁻-N) and nitrate nitrogen (NO₃⁻-N). After sampling, the water column was returned to its original level with additional river water.

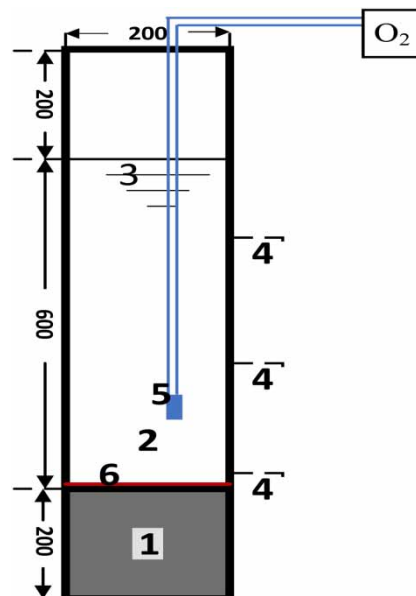


Figure 3 | Schematic experimental column setup. 1: sediment layer, 2: overlying water, surface level 60 cm above sediment (3), 4: sampling outlets, 5: aeration (columns 2 and 4 only), and 6: biozeolite (columns 3 and 4 only). Dimensions in mm.

Analytical methods

Water temperature and DO were monitored with an HQ30d portable dissolved oxygen meter (HACH, Loveland, CO, USA). TN was monitored by UV spectrophotometry after potassium sulfate oxidation, NH₄⁺-N after treatment with Nascar reagent, NO₂⁻-N after treatment with N-(1-naphthyl) ethylenediamine, and NO₃⁻-N by direct ultraviolet spectrophotometry at 200 nm (Du *et al.* 2003).

The removal rate, P , of each species was calculated using Equation (1):

$$P = \frac{C_{0i} - C_{1i}}{C_{0i}} \times 100\% \quad (1)$$

where C_{1i} is the concentration (mg/L) in the overlying water in each column (except the blank) at the time of sampling, C_{0i} the concentration (mg/L) in the overlying water in the blank control at the time of sampling, and i is the number of samples.

RESULTS AND DISCUSSION

The DO concentration in the water columns over time is shown in Figure 4(a). The concentration decreased rapidly in the blank control to levels around 1 mg/L and remained relatively stable after day 10. This was also observed in column 3, where the sediment was covered by biozeolite, indicating that the water became anoxic in these two columns. As expected, the aeration in columns 2 and 4 resulted in much higher DO levels. The concentration rose to approximately 10 mg/L immediately but, due to nitrifying and denitrifying bacterial activity, was not stable until around day 45, when it became relatively stable at about 7–10 mg/L. During the experiment, the water temperature increased from 18 °C initially to 27 °C (Figure 4(b)). The temperature was the same in all four columns.

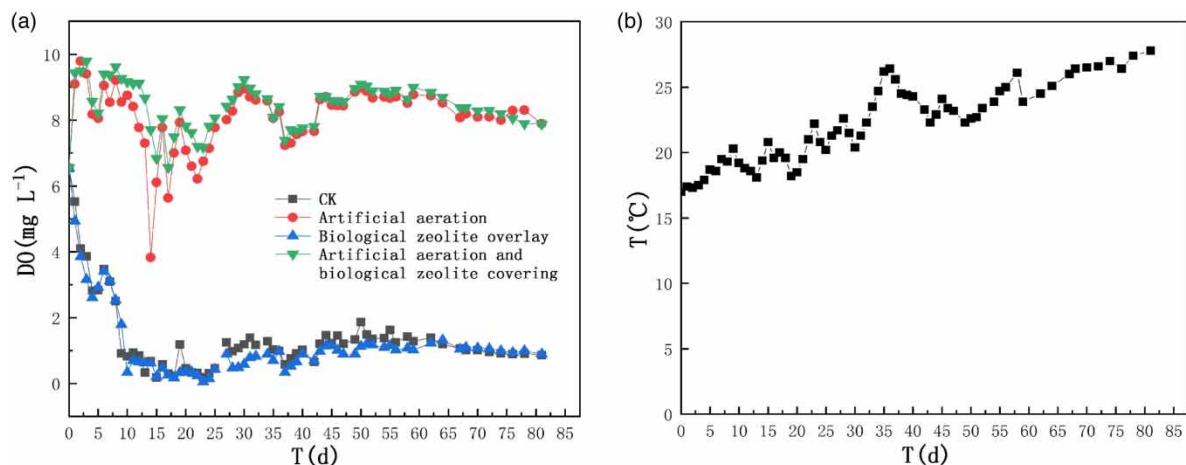


Figure 4 | DO concentration (a), and water temperature (b) in the columns over time.

The ammonium concentration in all columns was 2.8 mg-N/L initially. It increased in the blank control (column 1) during the first 30 days to around 15 mg/L, then fell slowly from day 45 onwards to reach 7 mg/L on day 81 (Figure 5(a)). When biozeolite covered the sediment (column 3), the ammonium concentration was fairly constant at between 3 and 5 mg-N/L for 50 days, falling slowly thereafter to levels close to zero (0.11 mg-N/L) at day 81. With aeration but no biozeolite (column 2), the $\text{NH}_4^+\text{-N}$ concentration increased rapidly to about 9 mg-N/L during the first 10 days; that is, higher than in the blank control (column 1) before decreasing sharply to levels close to zero around day 15, giving removal of 96% and remaining at that level thereafter. The column operating with the combination of aeration and biozeolite (column 4) also achieved maximum ammonia removal on day 15, but the initial increase; that is, during the first 10 days, was weaker than when aeration was performed in the absence of biozeolite (column 2) (Figure 5(a)).

Nitrite concentrations are shown in Figure 5(b). In the blank control (column 1) and the column with biozeolite covering alone (column 3), they remained close to zero throughout. A strong increase, 9.17 mg-N/L, was seen in the aeration-only column (column 2) between days 10 and 18, after which it fell rapidly to zero again. A weaker increase was observed during the same time period in the column with both aeration and biozeolite (column 4) (Figure 5(b)).

Figure 5(c) shows the nitrate concentrations. These were low and relatively stable in the blank control (column 1) and the unaerated column with biozeolite covering (column 3), increasing only slightly after day 40. Aeration (column 2) resulted in highly variable nitrate concentrations; it increased to around 8.0 mg/L, then fell intermittently from day 24 onwards to reach 4.1 mg/L on day 81, when applied on its own, but with more moderate peaks when the sediment was covered with biozeolite, and the nitrate concentration increased to about 3.5 mg-N/L on day 24, falling slowly thereafter to 2.2 mg-N/L at day 81 (column 4) (Figure 5(c)).

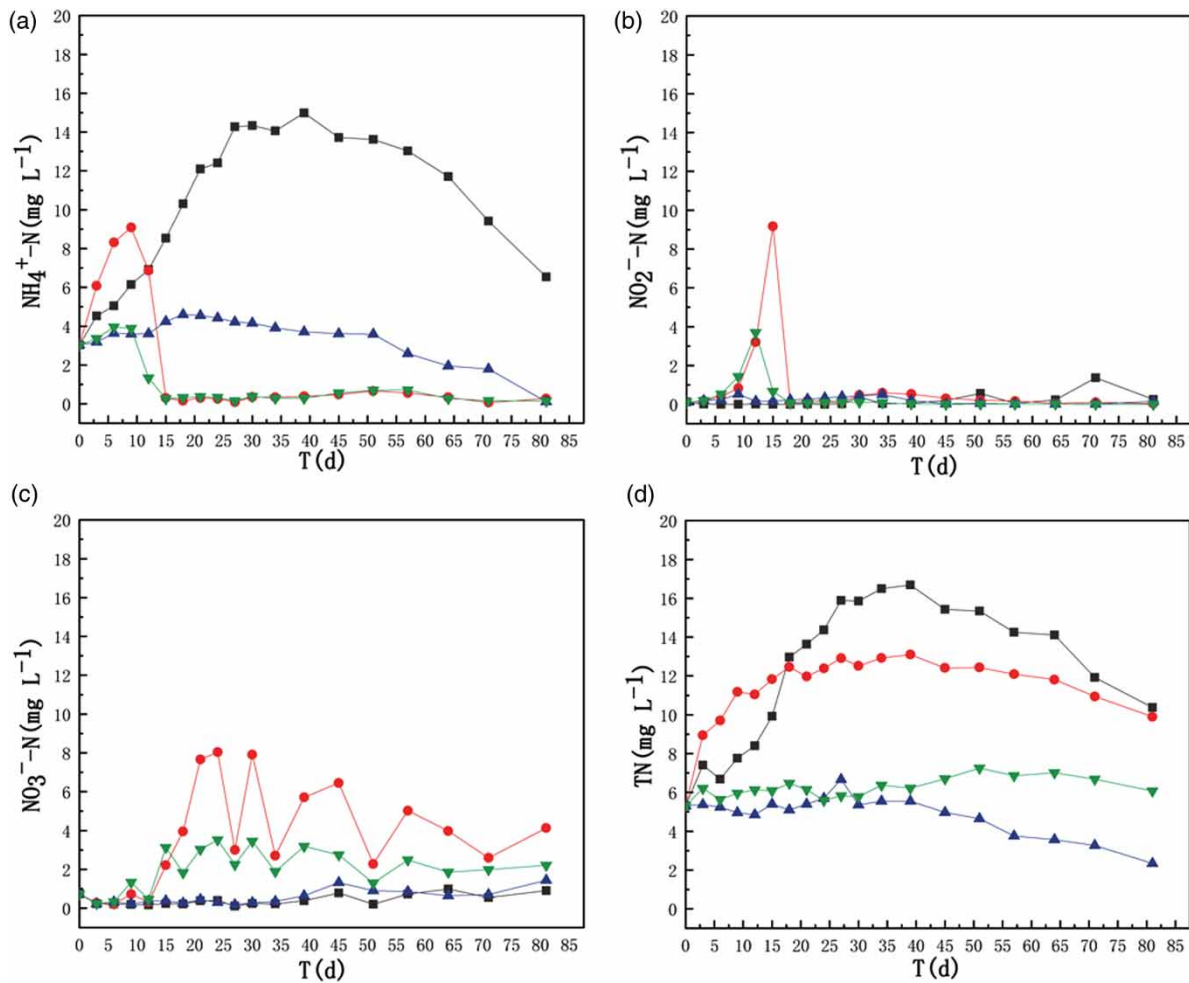


Figure 5 | The concentrations of $\text{NH}_4^+\text{-N}$ (a), $\text{NO}_2^-\text{-N}$ (b), $\text{NO}_3^-\text{-N}$ (c), and TN-N (d) in the water above the sediment sample over time. —■—: blank control (column 1); —●—: aeration only (column 2); —▲—: biozeolite only (column 3); —▼—: green triangles: aeration plus biozeolite (column 4).

Lastly, the TN concentrations are shown in Figure 5(d). In the blank control (column 1) these followed the ammoniacal-nitrogen trends 10.38 mg-N/L on day 81, as the other nitrogen species were mostly absent. In the column with aeration alone (column 2), the TN concentration increased during the first 20 days to a plateau at around 13 mg-N/L, and decreased slowly from day 40 to 9.89 mg-N/L on day 81. Lower levels were found in the unaerated column where the sediment was covered by biozeolite, with the TN concentration decreasing to 2.34 mg-N/L, a removal rate of 78% (column 3). Only about 42% removal was achieved when biozeolite was combined with aeration (column 4) (Figure 5(d)).

The variation in nitrogen species found was clearly due to microbial activity. Early in the experiment, biozeolite, when present, adsorbed the NH_4^+ generated by denitrifying bacteria. This adsorption could have been passive, or have arisen from ion exchange or other physical effects. It has been suggested that $\text{NH}_4^+\text{-N}$ release decreases rapidly when DO exceeds 2.8 mg/L (Liu *et al.* 2011), which fits with the observations made in column (2) (aeration-only) (see Figure 5(a)). Nitrite was formed only between days 9 and 15, and in columns 2 and 4, as a result of nitrifying bacteria activity – they require oxygen, which explains why it is not formed in the absence of aeration. However, once the nitrifying bacteria population had increased sufficiently, any nitrite formed would quickly be converted to nitrate, hence the nitrate concentration increase from day 10 on.

Li (2014) reported that when the DO concentration exceeds 7 mg/L, nitrogen is released from sediments mainly as nitrate, while at DO concentrations below 1 mg/L, most nitrogen is released as ammonia. This again fits the observations in this study, as ammonia concentrations were higher in

the blank control and biozeolite-only columns (1 and 3), where DO-levels were mostly below 1 mg/L. The waters in these columns also contained very little nitrate. On the other hand, nitrate concentrations were higher in the aerated columns (2 and 4), which had high DO levels.

The most relevant nitrogen parameter to consider is the TN concentration. This was lower in the aerated column (2) than the blank control (column 1), from day 18, but reached lower levels still when biozeolite was present (column 3 and 4). On the one hand, the aerobic environment is likely to have promoted the activity of both the nitrifying and facultative nitrification-denitrifying bacteria, and thus played a decisive role; on the other hand, as suggested by others (Wang 2008), nitrification and denitrification can be carried out simultaneously when a system contains both aerobic and anoxic microenvironments. The latter is provided in the sediment, where denitrification is promoted. However, TN removal was more efficient in the absence of aeration (column 3), suggesting that anaerobic bacteria were largely responsible for nitrogen removal. It is noted in this context that the water volume in the columns was rather small, which could have hampered the formation of truly anoxic regions in the presence of aeration and might explain why biozeolite removed more TN when not combined with aeration than when the two were combined.

It has been shown that the combined action of aeration with biozeolite coverage of sediment (column 4) can lead to an $\text{NH}_4^+\text{-N}$ removal rate exceeding 95% after 15 days, under the experimental conditions applied. This is higher than that obtained with either the blank control (column 1) or with biozeolite sediment cover alone (column 3). Although the DO concentration determines the $\text{NH}_4^+\text{-N}$ removal rate, the combined treatment reached maximum ammonium removal rates slightly more quickly than aeration alone; that is, without biozeolite (column 2). It is thought likely that, in the initial phase, the biozeolite adsorbs some of the ammonium formed and, combined with aeration, favors the activity of denitrification bacteria.

Under the combined action of aeration and biozeolite, TN removal was good, but the best nitrogen removal occurred when the sediment was covered with biozeolite but there was no aeration. The species and functions of the microorganisms mainly responsible for what was observed need further study.

CONCLUSIONS

Under the combined action of aeration and biozeolite coverage, the $\text{NH}_4^+\text{-N}$ removal rate remained above 95% after 15 days, This is significantly better than in the blank control and biozeolite-only columns (1 and 3), indicating that the DO concentration determines the $\text{NH}_4^+\text{-N}$ removal rate and that removal is faster than with aeration alone. $\text{NH}_4^+\text{-N}$ removal by dual treatment is clearly better than with either aeration or biozeolite mulching alone. For ammoniacal nitrogen removal, biozeolite combined with aeration (column 4) outperformed aeration alone (column 2), while coverage with biozeolite alone (column 3) had the least effect.

The combined action of aeration and biozeolite (column 4) produced a maximum $\text{NO}_2^-\text{-N}$ concentration of 3.70 mg-N/L after 12 days. This was less than the maximum 9.17 mg/L obtained under aeration (column 2). At 24 days (column 4), the maximum $\text{NO}_3^-\text{-N}$ concentration was 3.52 mg-N/L, which was also below the 8.04 mg/L obtained with aeration alone (column 2). This arose because: (1) biologically enforced zeolite could store $\text{NH}_4^+\text{-N}$, and (2) denitrification by denitrifying bacteria could occur under aerobic conditions. In addition, the time needed to achieve maximum $\text{NO}_2^-\text{-N}$ concentrations was less, due to the presence of nitrifying bacteria on the biozeolite.

Total nitrogen removal by the combined action of aeration and biologically enforced zeolite (biozeolite) (column 4) was good. In terms of TN removal efficiency, biozeolite coverage (column 3)

worked best, while combining it with aeration led to some reduction in performance, making it second-best. Aeration alone (column 2) yielded the least efficient removal.

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