Experimental Suppression of Phytoplankton Growth Employing Bactericidal Effects of Copper Ions under Eutrophic Conditions

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Abstract

Eutrophication of closed bodies of freshwater such as lakes and reservoirs has increased in recent times, leading to deterioration of aquatic environments due to hypertrophication and occurrence of cyanobacteria blooms over the entire water surface during summer. This study quantitatively assessed methods to control algal growth using the bactericidal properties of copper ions (Cu^{2+}) as a specific anti-eutrophication measure for aquatic environment restoration in a hypertrophic waterbody. In particular, this study examined the feasibility and effectiveness of controlling phytoplankton growth using bactericidal effects of copper ions under standardized laboratory conditions. Our results indicated that inhibitory effects of Cu^{2+}strongly depend on phytoplankton abundance and nutrient concentrations. Furthermore, Cu^{2+} concentrations of about 400 μg/L successfully suppressed Ch1.a production by all algae including cyanobacteria to 10 μg/L or less, which may help prevent algal blooms.

Keywords: *Organic pollution, Chl.a, DO, Water quality experiment*

Introduction

Abnormal proliferation of phytoplankton associated with eutrophication is an important problem in closed bodies of freshwater such as lakes and reservoirs, and methods to suppress phytoplankton overgrowth are needed for conservation and restoration of eutrophic aquatic environments. The loss of copper ions (Cu^{2+}) due to the formation of complexes with suspended organic materials was suggested as one important causal factor of phytoplankton proliferation in organically polluted waterbodies (Nanjo *et al.* 2000). Cu^{2+} is known to possess bactericidal properties, however, it occurs in natural waterbodies where it is an essential trace element for numerous organisms. This study investigated a potential means of control of algal growth by employing the bactericidal effects of Cu^{2+} for the countermeasure to prevent eutrophication. The purpose of this study was to assess the relationship of Cu^{2+} concentrations algal growth while also considering other environmental factors that facilitate phytoplankton proliferation. Specifically, this study quantitatively examined the feasibility and effectiveness of controlling phytoplankton overgrowth by employing the bactericidal effects of Cu^{2+} under hypertrophic conditions. For this, we conducted laboratory experiments on a beaker scale to test the effects of high and low Cu^{2+} concentrations at high water temperatures and high nutrient concentrations.

Fig. 1 Eutrophic agricultural reservoir where water was sampled.

Materials and Methods

Laboratory testing was conducted as follows, using water sampled at a eutrophic agricultural reservoir (**Fig. 1**): firstly, the sampled water was aliquoted to several tall beakers of 500 mL and adjusted to specific concentrations of NH_4-N (0.50 mg/L) and PO_4-P (0.02 mg/L). After this, the beakers were placed in a dark thermostatic chamber at 30 °C (**Fig. 2**). A fluorescent lamp used to grow ornamental plants was placed directly above the tanks, and a 12:12 h light/dark cycle was applied. Water quality was monitored twice per week over the first four weeks by measuring the concentrations of Cu^{2+}, chlorophyll a (Chl.a) produced by each algae class, total Chl.a, inorganic nitrogen, phosphate-phosphorus, and organic carbon. Phytoplankton proliferation was estimated by measuring dissolved oxygen (DO) concentrations. Three initial Cu^{2+} concentrations were used: a low concentration treatment (56 µg/L), and high concentration treatment (715 µg/L), and a control treatment with no Cu^{2+}. Copper is not referenced in the environmental quality standards for water pollution in Japan, therefore Cu^{2+} levels were set according to the Japanese agricultural water standards (≤ 20 µg/L) and drinking water quality standards (≤ 1000 µg/L). Nine replicates were used for each condition.

Results and Discussion

Figure 3 shows the measurements of Chl.a and DO under high temperature conditions. **Figure 4** shows the changes over time after adding Cu^{2+}. In the control treatment, Chl.a increased from a eutrophic level of about 40 µg/L to about 70 µg/L, which indicates hypertrophy. Furthermore, Ch1.a production by each algal class indicated the continuous occurrence of cyanobacteria which can cause algal blooms. DO gradually increased to a maximum of 5.2 mg/L, indicating substantial proliferation of phytoplankton.

In the low-Cu^{2+} treatment, the Chl.a concentration reached a maximum of 63 µg/L, which was within the range of that of the control treatment. However, cyanobacterial Chl.a was almost zero throughout the experiment. The rate of Ch1.a increase in the low-Cu^{2+} treatment was 0.5 µg/(L d), which was slightly below the 0.8 µg/(L d) observed in the control treatment. DO reached a maximum of 5.1 mg/L on day 6 of the experiment but decreased thereafter to 2.4 mg/L, which was about half the concentration observed in the control treatment. The fact that the DO maximum remained at 3.7 mg/L on the last day of monitoring

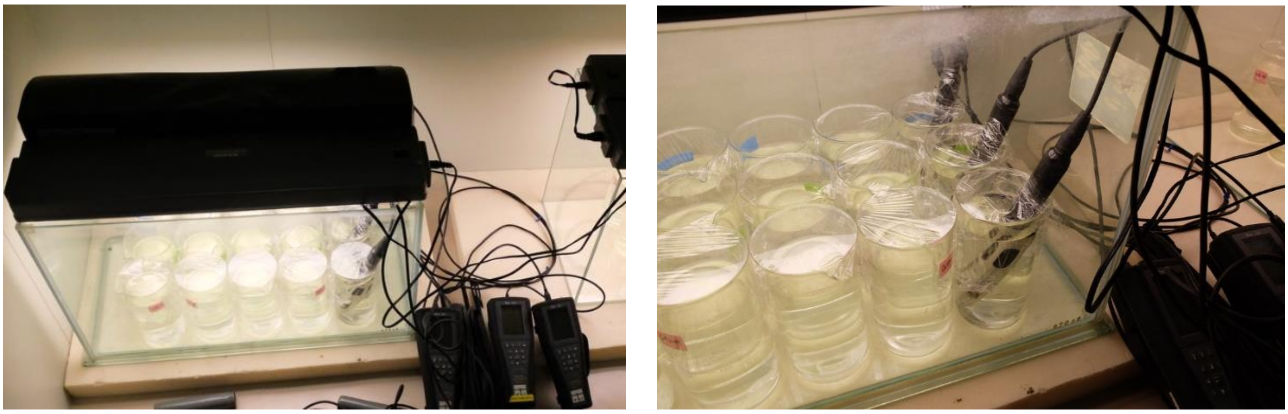

Fig. 2 Experimental setup in the thermostatic chamber.

indicated that the inhibitory effect of Cu^{2+} occurred after about one week. In a previous study, Chl.a concentrations ranged from 20 to 40 μg/L in the control treatment, and addition of about 20 μg/L Cu^{2+} produced a marked inhibitory effect on phytoplankton growth (Kawara *et al.* 2018). However, in the current study, no decrease in Ch1.a was observed even after one week of Cu^{2+} concentrations of about 20 μg/L. The initially high levels of nutrient salts likely increased phytoplankton growth which may explain the lack of an antimicrobial effect at Cu^{2+} concentrations of about 20 μg/L. In addition, Cu^{2+} concentrations decreased to about 10 μg/L due to the formation of complexes with organic matter, including phytoplankton. Therefore, while growth inhibition was detected one week after the start of the experiment, Chl.a production likely remained high because Cu^{2+} concentrations continued to decrease to levels of about 10 μg/L.

In the high-Cu^{2+} treatment, Chl.a decreased considerably to 8 μg/L on day 4 after the start of the experiment and decreased to 1 μg/L on day 28, which suppressed growth of all algae, including cyanobacteria. Throughout the experiment, DO concentrations were low (0.1 mg/L), indicating a growth-inhibiting effect. Due to the initially high level of Chl.a, Cu^{2+} declined from 715 μg/L to about 450 μg/L (roughly half the initial concentration). However, this treatment produced a strong inhibitory effect, even at high water temperature and high nutrient salt levels.

Conclusions

This study was conducted to examine inhibitory effects of Cu^{2+} on algal growth. The results showed that growth inhibition strongly depends on phytoplankton abundance and nutrient concentrations. Furthermore, Cu^{2+} at about 400 μg/L effectively suppressed Ch1.a production from all algae including cyanobacteria to 10 μg/L or less, which may help prevent algal blooms.

Acknowledgements

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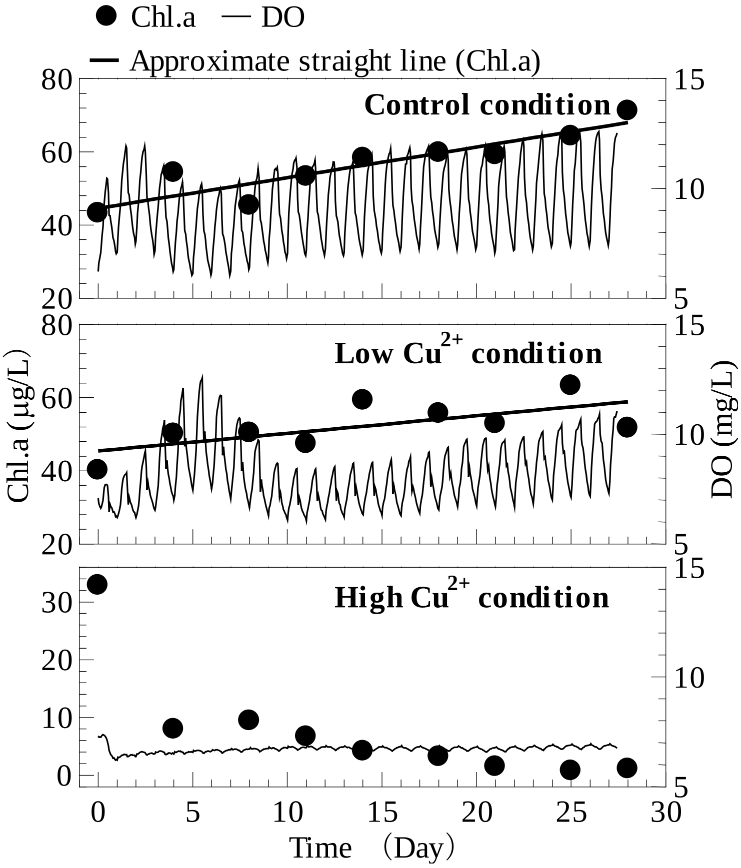

Fig. 3 DO and Chl.a changes over time.

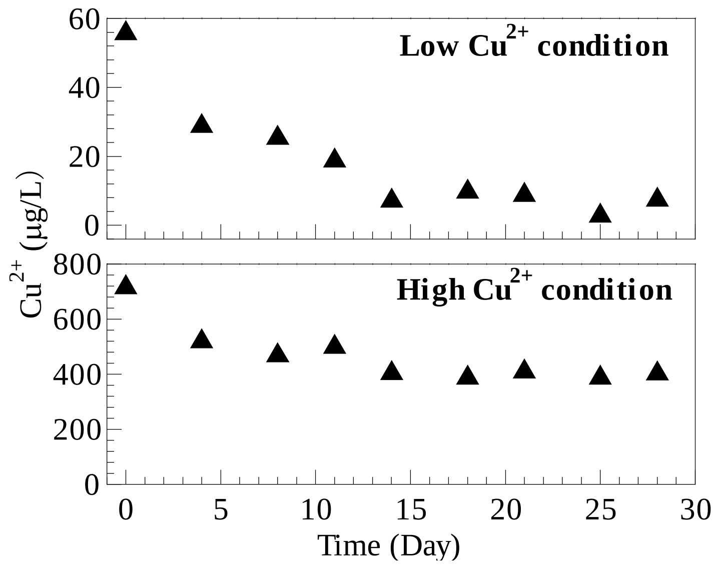

Fig. 4 Copper ion concentration changes over time.

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