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The ecological risks of hydrogen peroxide as a cyanocide: its effect on the community structure of bacterioplankton*

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Abstract *Microcystis* blooms are an environmental and ecological concern that has received a serious attention. Hydrogen peroxide (H_2O_2) is an environment-friendly cyanocide that is commonly used to control *Microcystis* blooms. While the ecological safety of H_2O_2 has been previously studied, its influence on bacterioplankton has not been investigated to date. In this study, we used mesocosm experiments to determine the influence of H_2O_2 on the dynamic changes of the community structure of bacterioplankton. By using deep-sequencing and metagenomics strategy we determined the community structures of phytoplankton and bacterioplankton assemblages that were dominated by *Microcystis* at a highly eutrophic Dianchi Lake, China. The results showed that *Microcystis* was more sensitive to H_2O_2 than other eukaryotic algae. More interestingly, application of H_2O_2 changed the community structure of bacterioplankton, evidenced by the emergence of *Firmicutes* as the dominant species in place of *Bacteroidetes* and *Proteobacteria*. The H_2O_2 treatment resulted in the community of bacterioplankton that was primarily dominated by *Exiguobacterium* and *Planomicrobium*. Our results show that the abundance changed and the bacterioplankton diversity did not recover even after the concentration of H_2O_2 reached to the background level. Thus, the response of bacterioplankton must be considered when assessing the ecological risks of using H_2O_2 to control *Microcystis* blooms, because bacterioplankton is the key player that forms the basis of food web of aquatic environment.

Keyword: hydrogen peroxide; *Microcystis* bloom; ecological risks; bacterioplankton

1 INTRODUCTION

Because of heavy eutrophication and global warming, cyanobacteria blooms have been rapidly rising recently throughout the world (Paerl and Huisman, 2009; Wagner and Adrian, 2009). *Microcystis* blooms have been reported in more than 108 countries and have resulted in negative economic and societal impacts including drinking water crises (Qin et al., 2007; Harke et al., 2016). Harmful cyanobacterial bloom has been a scientific interest and a societal concern, because cyanobacteria such as *Microcystis* can produce toxins or odorous substances, including microcystins, β-N-methylamino-L-alanine (BMAA), paralytic shellfish poison (PSP), anatoxins, and 2-Methylisoborneol (MIB) (Bishop et al., 1959; Cox and Sacks, 2002; Sant'Anna et al., 2011; Wang et al., 2015b; Zhang et al., 2016).

Because of serious environmental concerns, a wide array of strategies have been tested to control or mitigate cyanobacteria blooms (Hamilton et al., 2016; Triest et al., 2016; Matthijs et al., 2016; Visser et al., 2016). Hydrogen peroxide (H_2O_2) were often used to control cyanobacterial blooms (Drábková et al., 2007), because it is particularly effective against cyanobacteria and is considered environmentally friendly. Compared to eukaryotic algae, cyanobacteria are highly susceptible to reactive oxygen species

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(ROS)-generating chemicals such as H_2O_2. Cyanobateria do not have the complete biochemical pathway of Mehler reaction that produces superoxide anion (Mehler, 1951; Helman et al., 2003, 2005). Furthermore, different species of cyanobacteria have different tolerances to ROS. Although *Microcystis* is more resistant to H_2O_2 than other cyanobacetria because of its ability to form colonies and richness of extracellular polymeric substances (Lürling et al., 2014; Gao et al., 2015), *Microcystis* bloom can be still controlled by H_2O_2.

The ecological risks associated with applying H_2O_2 and its effects on macrofauna, fishes, macro-invertebrates, and zooplankton have been studied (Matthijs et al., 2012; Reichwaldt et al., 2012; Burson et al., 2014). However, there is no published report on the effects of H_2O_2 on bacterioplankton and merits an investigation, because bacterioplankton plays major role in the natural food web of aquatic ecosystems. In aquatic ecosystems, the background concentrations of H_2O_2 can reach 1 μmol/L (Häkkinen et al., 2004). Hydrogen peroxide at environmentally relevant concentrations could influence the microbial activity and composition of bacterial communities (Glaeser et al., 2014). Bacterioplanktons play an important role in nitrogen, carbon, phosphorus, and sulfur cycling in aquatic ecosystems (Simon et al., 2002; Zeng et al., 2007), and directly influence the dynamic of cyanobacteria (Canfield and Des Marais, 1993; Stuart et al., 2016). Some bacteria capable of algicidal effect are associated with the decline of *Microcystis* bloom (Manage et al., 2001; Zhang et al., 2012; Su et al., 2016). On the other hand, the decomposition of *Microcystis* bloom at natural environment influences the structure of bacterial communities (Shao et al., 2013). Additionally, the algicidal nanosilver particles can synergize the effect of antibiotics against gamma-negative fish pathogenic bacteria (Satapathy et al., 2017). Therefore, we speculated that the application of H_2O_2 on cyanobacteria bloom can influence population dynamics and the community structures of bacteriplanktons.

Dianchi Lake is one of the most eutrophic lakes in China and accumulates a massive amount of *Microcystis* biomass throughout the year (Wu et al., 2014, 2016). In this study, we carried out mesocosm experiments at Dianchi Lake (latitude: 24°51′N, longitude: 102°42′E, altitude: 1 887 m a.s.l.) to evaluate how the application of H_2O_2, a *Microcystis* bloom control measure, affects the overall population dynamics, composition and structures of bactrioplanktons. Our objectives were to determine (1) the dose that are lethal to algae and cyanobacteria, (2) whether H_2O_2 would affect bacterioplankton at the dose that can control *Microcystis* blooms, and (3) the ecological risks that the use of H_2O_2 causes to the community of bacterioplanktons in aquatic ecosystems.

2 MATERIAL AND METHOD

2.1 Mesocosm experiment

Dianchi Lake, the sixth largest freshwater lake in China, has been suffering from harmful algal blooms over recent decades. The concentration of Chl *a* is generally higher in the northern part of Dianchi Lake than any other areas in the Lake and has been reported to exceed 100 μg/L even in December (Wu et al., 2014, 2016). We first transferred the lake water from the water surface into ten plastic barrels and placed the barrels on an experimental platform on the lake. Each barrel contained 150 L the lake water. We then added H_2O_2 to these barrels at concentrations of 2, 4, 8, and 12 mg/L. The original temperature, Chl *a* concentration, dissolved oxygen, and pH in the bulk samples were 10.2°C, 84 μg/L, 9.9 mg/L, and 8.8, respectively. We performed the experiment during the period of 23 December 2013 and 3 January 2014. At the end of the experiment, the H_2O_2 concentrations in all of the treatment groups were below the detection limit. We analyzed the H_2O_2 concentrations as described by Drábková et al. (2007) and conducted the experiment in duplicates.

2.2 Bacterioplankton and phytoplankton abundance

The phytoplankton assemblages were microscopically identified as described by Wu et al. (2016). Water samples (1 L) were fixed with 10 mL of acidic Lugol iodine solution. The samples were concentrated to 30 mL and examined via microscopy (Olympus CS31, Japan). To calculate the bacterial cell density, 8 mL of the water samples were collected into 10 mL axenic tubes and fixed with 0.3 mL of 25% glutaraldehyde solution. Samples were kept in the dark at 4°C. After staining with DAPI (4’,6-diamidino-2-phenylindole), samples were filtered onto a 0.22-μm nuclepore membrane (Whatman, 110656, UK). The bacterial abundance was then determined manually using epifluorescence microscopy (Olympus BX51, Japan) (Porter and Feig, 1980).

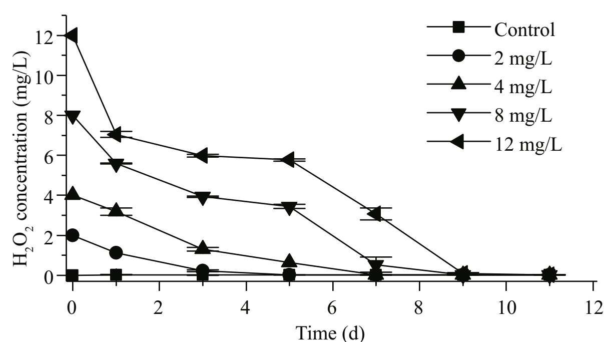

Fig.1 Reduction of H_2O_2 concentrations during experimental period

2.3 The community structures of bacterioplanktons and phytoplanktons community

2.3.1 Sample collection

In order to prepare samples for analysis of the community structures of the bacterioplankton and phytoplankton by deep sequencing, water samples were filtered through a 0.22-μm pore-size polycarbonate membrane (XinYa factory, Shanghai). The membranes with planktons were kept at -20°C until DNA extraction.

2.3.2 DNA extraction and Illumina sequencing

Genomic DNAs were extracted with a DNA kit (MP Fast DNA SPIN Kit for soil, 116560-200, USA). Illumina sequencing was performed at RIBOBIO Co., Ltd. (Guangzhou, China) by Illumina MiSeq using primers that targeted the V3 and V4 regions of the 16S rDNA (Klindworth et al., 2013) (forward: 5′ TCGTCGGCAGCGTCAGATGTGTATAAGAGAC-AG; reverse: 5′ GTCTCGTGGGCTCGGAGATGTG-TATAAGAGACAG). Paired-end reads were stitched together into Unique Tags with FLASH (Magoč and Salzberg, 2011) and Mothur (V 1.27.0) (Schloss et al., 2009). Unique Tags were assigned into OTUs with a cutoff of 97%. We discarded OTUs that were identified only once. The representative sequences that were selected from each of the OTUs were aligned with the Greengene (http://greengenes.lbl.gov/) database GG 13.5 to obtain the taxonomic framework.

Illumina sequencing yielded a total of 2 331 945 tags. An overall average of 72 873 sequences (between 3 971 and 110 259 sequences) were obtained for each sample. A total of 15 709 OTUs were aligned, 99.1%, 99.1%, 98.3%, 74.4%, and 52.5% of which were classified at the phylum, class, order, family, and genus levels, respectively. The 16S rDNA amplicons were separated into three parts: 1) cyanobacteria phylum were used to analyze the *Cyanophyta* community; 2) chloroplasts were used to analyze the eukaryote phytoplankton community, and 3) the non-cyanobacteria 16S rDNA amplicons were used to analyze the community structures of bacterioplankton. The OTUs of prokaryote and eukaryote phytoplanktons, and the non-cyanobacteria were re-sampled to achieve the minimum sample sizes of 104, 75, and 1 828, respectively. Given that the total cyanobacteria tags were below 32 after the 11-day treatment at doses of 8 and 12 mg/L, respectively, we did not analyze the 16S rDNA amplicons of cyanobacteria in those experimental groups.

2.4 Statistical analysis

Distance-based ANOVA (dbANOVA), calculation of Shannon-Wiener index and rarefaction curve were performed with vegan Package in R 3.1.2 (http://www.r-project.org/). ANOVA was performed in SPSS 17.0.

3 RESULT

3.1 Effect of hydrogen peroxide on phytoplankton

The H_2O_2 concentrations decreased during the experiment. On the third day, the H_2O_2 concentrations in the 2, 4, 8, and 12 mg/L groups decreased to 0.2, 1.3, 3.9, and 6.0 mg/L, respectively. The concentration of the control group remained below 0.05 mg/L during the experiment. On day 9 and 11, the H_2O_2 concentrations in all treatments were reduced to the level that was similar to that of the control group (Fig.1).

The abundance of *Microcystis* decreased when H_2O_2 was applied at doses of 4 mg/L and above, but the cell density of *Microcystis* did not decrease after 11 days when the used H_2O_2 dose was 2 mg/L (ANOVA, P>0.05). At higher concentrations of H_2O_2 (>4 mg/L), the more rapid decrease in the abundance of *Microcystis* was observed. The cell density of *Microcystis* was 1.6×10^7 cells/L at the beginning (Fig.2a). After 1 day, the abundance of *Microcystis* in 12 mg/L group was only 25% of that of the control group. After 3 days, *Microcystis* of 8 and 12 mg/L groups were less than 20% of that of the control group. After 11 days, the cell densities of *Microcystis* in the treatment groups with H_2O_2 doses of 4, 8, and 12 mg/L were less than half of the density of the control group.

As well as examining the effects on *Microcystis*, we also evaluated the algicidal effects of H_2O_2 on other algae, namely *Chlorophyta*, diatoms, *Cyanophyta*,

Table 1 ANOVA results showing the influence of hydrogen peroxide on phytoplankton and bacterioplankton

	Day	F	P		Day	F	P
Wet weightof phytoplankton	1	6.452	0.033*	*Dinoflagellates*	1	0.423	0.790
	3	23.611	0.002**		3	2.315	0.105
	11	6.604	0.031*		11	4.475	0.066
Bacterial cell density	0	2.38	0.184	*Microcystis*	1	3.087	0.124
	1	3.496	0.101		3	4.175	0.075
	3	6.729	0.030*		11	11.572	0.010*
	11	6.344	0.034*	*Cyanophyta*	1	2.753	0.148
Cryptophyta	1	151.938	0.000**		3	5.834	0.040*
	3	12.22	0.009**		11	13.556	0.007**
	11	1	0.486	*Chlorophyta*	1	0.827	0.561
Diatom	1	2.067	0.233		3	0.457	0.766
	3	1.026	0.475		11	9.588	0.015*
	11	22.738	0.002**				

*: P<0.05; **: P<0.01.

Dinoflagellates and *Cryptophyta* (Table 1, Figs.2, 3), and found that the effects were variable. *Cyanophyta* and *Cryptophyta* were more sensitive to H_2O_2 than *Chlorophyta*, diatoms, and *Dinoflagellates*. The structure of phytoplankton communities (Fig.4) changed during the experimental period. The proportions of *Cyanophyta* and *Cryptophyta* decreased when H_2O_2 was added at concentrations of 4, 8, and 12 mg/L, while the proportions of diatoms and *Chlorophyta* increased when treated with H_2O_2 at concentrations of 8 or 12 mg/L.

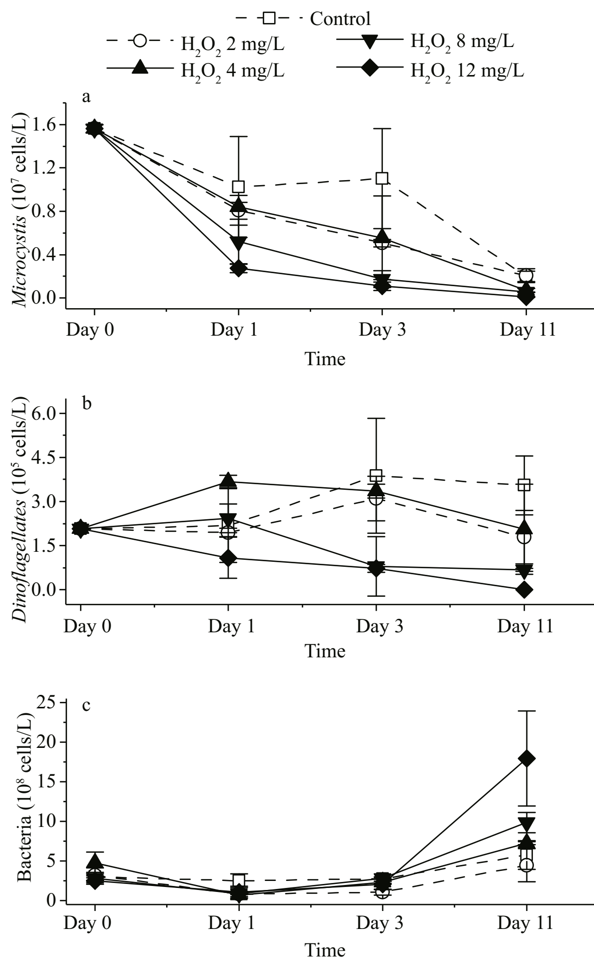

Fig.2 Change of cell density of *Microcystis* (a), *Dinoflagellates* (b) and bacteria (c) after the application of H_2O_2

We also examined the phytoplankton community composition by analyzing the 16S rDNA amplicons. The rarefaction curves were shown in Fig.5. The results of Illumina sequencing were consistent with those from the microscopic examinations (Fig.6). At the beginning of the experiment, the eukaryote phytoplankton community was dominated by *Stramenopiles* and *Cryptophyta*. There were less *Cryptophyta*, and the proportion of *Chlorophyta* increased after H_2O_2 was added. The prokaryotic phytoplankton community was dominated by *Microcystis* at the start of the experiment. The *Microcystis* proportion increased three days after the H_2O_2 treatment and was higher than that of the control group. However, after 11 days the *Microcystis* proportion was declined in the H_2O_2-treated groups and lower than that in the control groups.

Microcystis and *Cryptophyta* were more sensitive to H_2O_2 than *Chlorophyta*, diatoms, and *Dinoflagellates*. Applications of H_2O_2 appeared to selectively control the abundance of *Microcystis*.

3.2 Effect of hydrogen peroxide on bacterioplanktons

The structures of the bacterioplankton communities

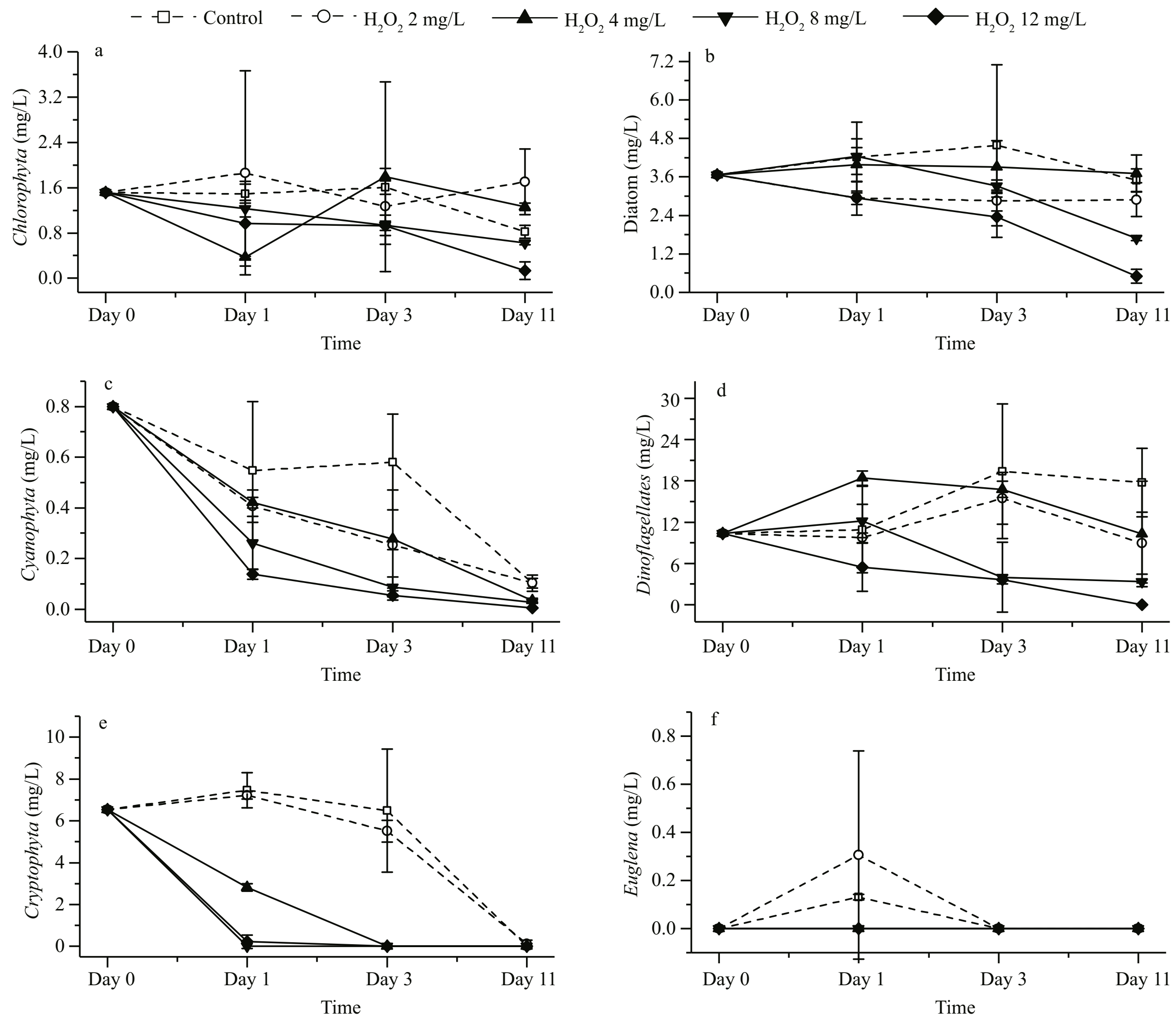

Fig.3 Changes of wet weight after the application of H_2O_2

a. *Chlorophyta*; b. diatoms; c. *Cyanophyta*; d. *Dinoflagellates*; e. *Cryptophyta*; f. *Euglena*.

Table 2 Distance-based ANOVA analysis results of the influence of hydrogen peroxide on the composition of phytoplankton and bacterioplankton communities (Bray-Curtis distance)

	Day	*F*. model	*P*		Day	*F*. model	*P*
Phytoplankton community[a]	1	3.873	0.002**	Bacterioplankton community[b]	1	2.925 2	0.010 9*
	3	4.268 1	0.012*		3	6.891	0.001 6**
	11	6.604 3	0.016*		11	12.11	0.000 8**

[a] phytoplankton community at phylum level via microscopic examination; [b] bacterial community based on 16S rDNA amplicons. *: $P<0.05$; **: $P<0.01$.

changed and became significantly different after additions of H_2O_2 (Table 2, $P<0.05$, Fig.7). The effects of H_2O_2 on bacterioplankton varied depending on the H_2O_2 doses. At the beginning of the experiment, the bacterioplankton community was dominated by *Actinobacteria*, *Flavobacteria*, *Alphaproteobacteria*, *Betaproteobacteria*, and *Gammaproteobacteria* (Fig.6).

Flavobacterium was the dominant genus at the beginning (Fig.6) of the experiment and maintained its population at the level of ~10% in the control group. The proportion of *Flavobacterium* decreased in the experimental group that was treated with H_2O_2 (2 mg/L), but recovered after 3 days when the concentration of H_2O_2 decreased. On day 11, the proportion of *Flavobacterium* in the 2 mg/L group was higher than that in the control group and accounted for 25% of the bacterioplankton community. In contrast, the proportions of *Flavobacterium* in the 4, 8, and 12 mg/L H_2O_2 groups decreased, and

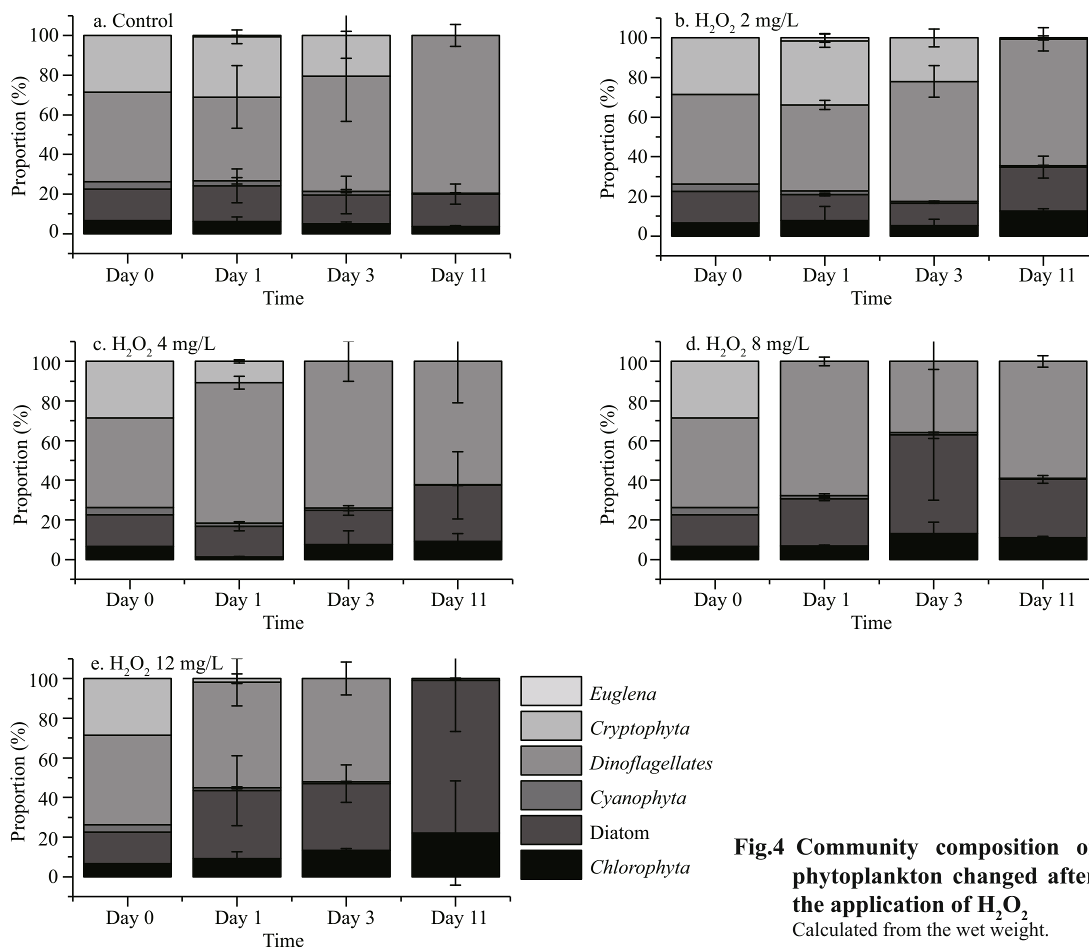

Fig.4 Community composition of phytoplankton changed after the application of H_2O_2
Calculated from the wet weight.

represented only 1.5%, 1%, and 1% of the bacterial communities after 11 days, respectively. While the concentration of H_2O_2 was less than 0.05 mg/L on days 9 and 11, the proportion of *Flavobacterium* did not recover when the doses of H_2O_2 were 4 mg/L and above.

The proportions of *Exiguobacterium*, *Planomicrobium*, and *Sphingomonas* were higher in the H_2O_2 treated groups compared to the control group. After the 11-day H_2O_2 treatment, *Exiguobacterium* occupied 59% and 29% of the bacterioplankton communities in the experimental groups that were treated with 4 and 8 mg/L H_2O_2, respectively. *Planomicrobium* in the control group represented less than 1% during the experiment, but was high in the H_2O_2-treated groups. As the concentration of the H_2O_2 treatment increased, the proportion of *Planomicrobium* also increased, and accounted for 2.1%, 12.5%, 50%, and 62.6% of the bacterioplankton communities after 11 days in the 2, 4, 8, and 12 mg/L H_2O_2 groups, respectively. After 11 days, *Sphingomonas* was higher in the 4, 8, and 12 mg/L groups than in the control, where it accounted for 1.6%, 2.3%, and 4.1% of the total, respectively.

The H_2O_2 applications also influenced the abundance of bacterioplankton (Fig.2c). On day 1, the cell densities of bacteria in all of the H_2O_2 treated groups were lower than that in the control group. On day 11, the cell densities of bacteria that were treated with 2, 4, and 8 g/L of H_2O_2 and the control group were similar, while the cell density in the 12 mg/L group was twice of that in the control group.

3.3 The influence of H_2O_2 on the alpha-diversity of phytoplankton and bacterioplankton

The Shannon-Wiener Index of phytoplankton at the phylum level decreased in all groups, and was lowest in the 12 mg/L group after the 11-day H_2O_2 treatment (Fig.8). Meanwhile, the Shannon-Wiener Index of bacterioplankton at the genus level was stable for the control group, but was low in the groups treated with H_2O_2 at concentrations of 4, 8, and 12 mg/L. These results indicate that the α-diversity of phytoplankton and bacterioplankton decreased after being treated by H_2O_2.

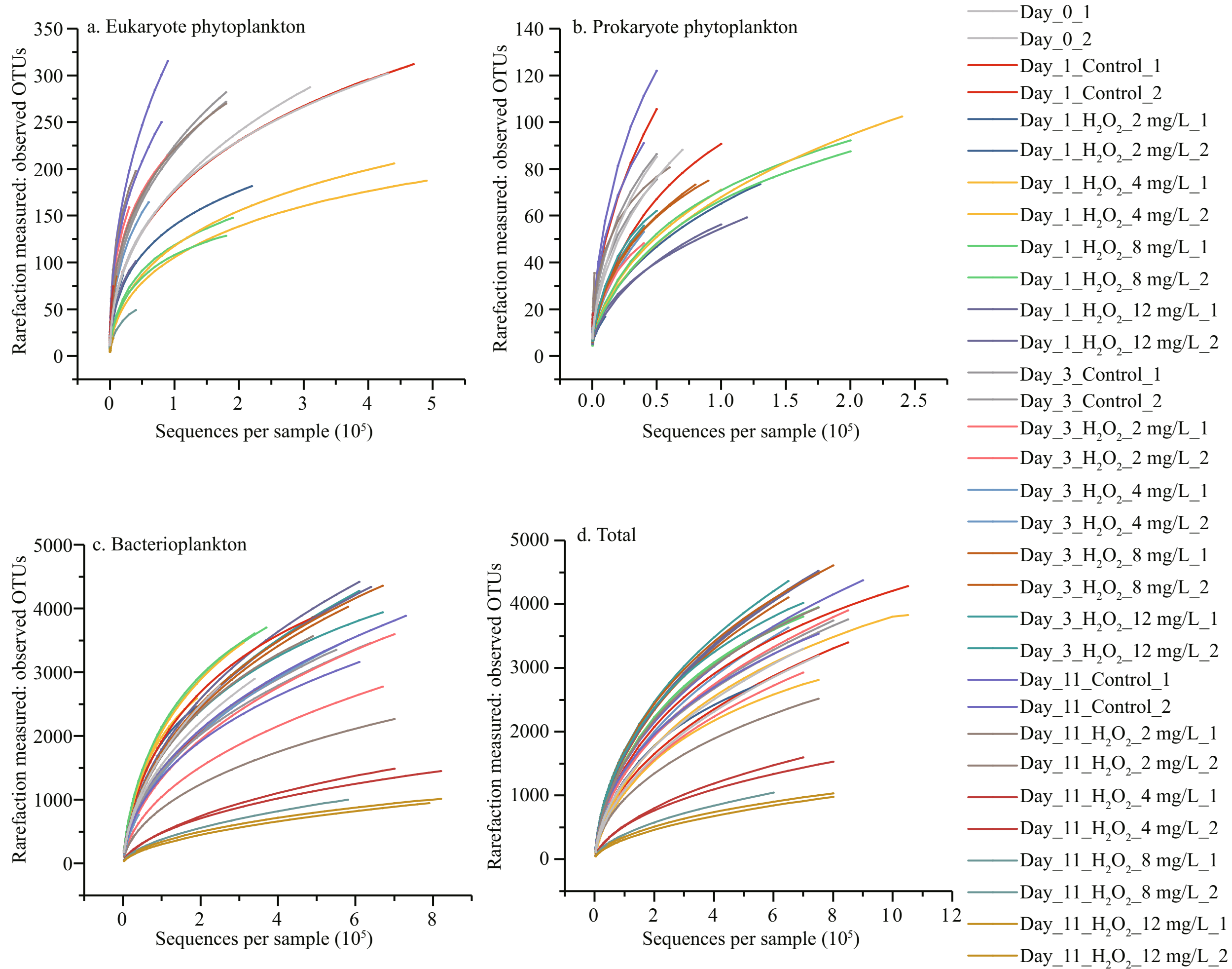

Fig.5 Rarefaction plots for Illumina sequencing results of samples

OTUs were assigned with a 97% similarity cut-off.

4 DISCUSSION

4.1 The selectivity of hydrogen peroxide on different phytoplankton

Producing no harmful secondary pollutants, H_2O_2 is regarded as an environmental friendly cyanocide and has been used to control cyanobacteria blooms (Matthijs et al., 2012, 2016; Burson et al., 2014). The selectivity of H_2O_2 has been reported previously; for example, cyanobacteria have been reported to be more sensitive to H_2O_2 than diatoms and *Chlorophyta* (Barroin and Feuillade, 1986; Drábková et al., 2007; Barrington and Ghadouani, 2008). This study showed that the decrease in the abundance of phytoplankton was often associated with the increase of *Chlorophyta*, diatoms and *Dinoflagellates* upon the application of H_2O_2, indicating that eukaryote phytoplankton were more tolerant to H_2O_2 as previously shown (Barroin and Feuillade, 1986; Drábková et al., 2007; Barrington and Ghadouani, 2008).

The responses to H_2O_2 by different bloom-forming cyanobacteria also varied. Two previous studies reported that 2 mg/L of H_2O_2 could effectively control *Planktothrix* bloom in an entire lake, but 5 mg/L of H_2O_2 were needed to successfully control *Microcystis* blooms (Matthijs et al., 2012, 2016). When H_2O_2 was added to our mesocosm systems in which *Planktothrix* and *Microcystis* co-existed, *Planktothrix* and *Microcystis* responded differently (Figs.2a, 6). While H_2O_2 doses of 4 mg/L and above resulted in reduced abundance of *Microcystis*, the proportion of *Microcystis* in cyanophyta increased as the proportion of *Planktothrix* decreased, indicating that *Microcystis* was more tolerant to H_2O_2 than *Planktothrix*. The strong tolerance of *Microcystis* may be attributed to the formation of *Microcystis* colonies in the field. *Microcystis* colonies are capsuled with extracellular polymeric substances that may help guard them against the H_2O_2 treatment (Gao et al., 2015). The strong tolerance of *Microcystis* colonies was also

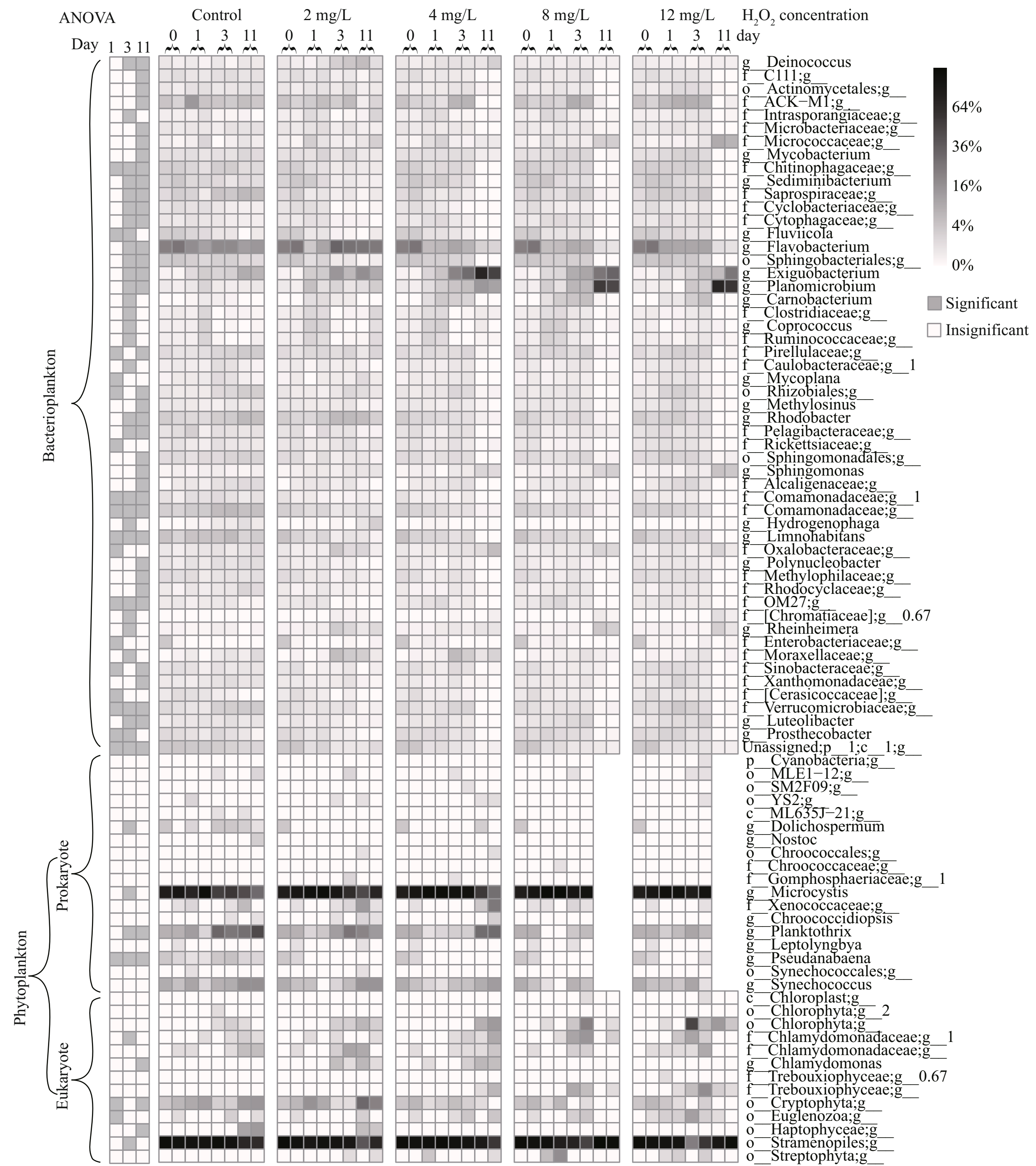

Fig.6 Community structure of bacterioplankton and phytoplankton based on the proportion of 16S rDNA amplicons

The bacterioplankton genus that occupied less than 1% in all samples, or were insignificant, based on ANOVA, were ignored.

correlated with the tolerance levels of colonial or unicellular *Microcystis* to other algicides. For example, unicellular *Microcystis* was reported to be more sensitive to copper sulfate than colonial *Microcystis* (Wu et al., 2007). Different strains of unicellular *Microcystis* also responded differently to copper sulfate (Wu et al., 2017). Researchers have suggested that H_2O_2 could effectively inhibit the growth of *Microcystis* or mitigate cyanobacteria blooms at doses between 0.4 and 6 mg/L (Drábková et al., 2007; Lürling et al., 2014; Gao et al., 2015; Wang et al., 2015a). Therefore, understanding of the cyanobacteria composition and the *Microcystis* characteristics is necessary before the application of H_2O_2.

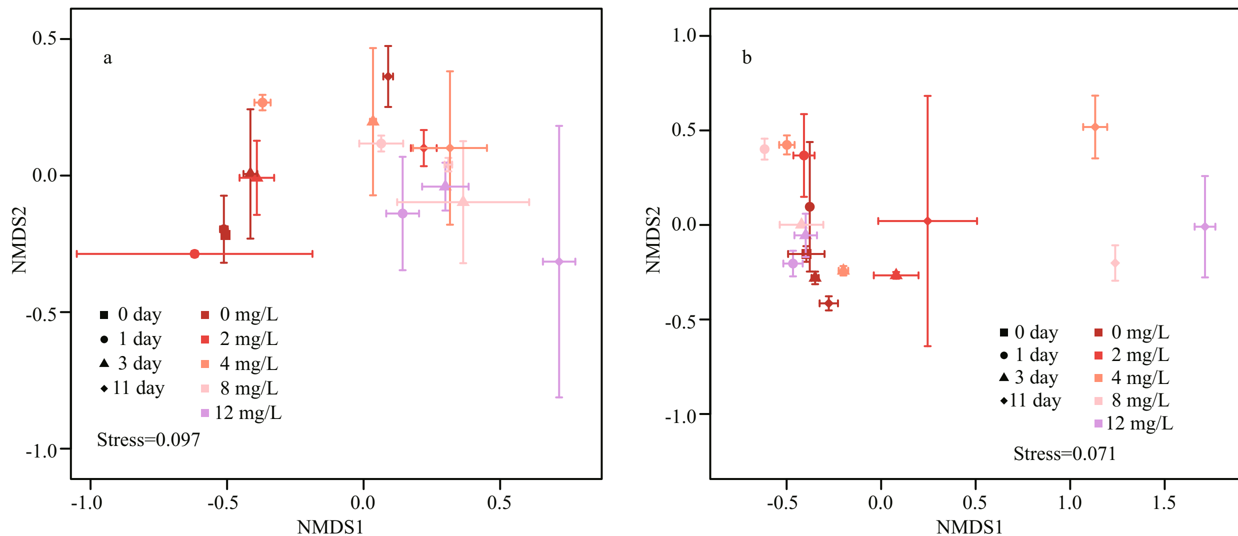

Fig.7 NMDS results of phytoplankton communities (a) and bacterioplankton communities (b) based on Bray-Curtis distance

Phytoplankton communities were calculated via microscopic examination. Bacterioplankton communities were calculated via Illumina sequencing at genus level.

4.2 The influence of hydrogen peroxide on bacterioplankton at the doses that effectively control *Microcystis* blooms

The effects of H_2O_2 on untargeted organisms, including zooplankton, macroinvertebrates, aquatic plants, and fishes, have been studied (Matthijs et al., 2012; Reichwaldt et al., 2012; Burson et al., 2014). Applications of H_2O_2 at the doses that are necessary to control cyanobacteria blooms are regarded safe for macroinvertebrates, aquatic plants, and fishes. While we know that some zooplankton are sensitive to H_2O_2 at doses of 5 mg/L (Matthijs et al., 2012; Reichwaldt et al., 2012), the influence of H_2O_2 on bacterioplankton at cyanocidal doses has not been studied. In this study, we found that the composition and abundance of bacterioplankton communities were significantly affected by H_2O_2 treatments at the doses that are commonly used for cyanobloom control (Figs.2, 6), indicating that bacterioplankton was sensitive to H_2O_2.

The effect of H_2O_2 on bacterioplankton appears to be dose dependent, and become more severe as the doses of H_2O_2 increase. The cell density of bacterioplankton increased 2-fold after being treated for 11 days with H_2O_2 at a concentration of 12 mg/L. The composition of bacterioplankton communities cannot recover at doses of 4 mg/L and above, which is the dose needed to control *Microcystis* blooms. Upon treatment, the proportion of *Flavobacterium* decreased, while *Exiguobacterium* and *Planomicrobium* emerged as the dominants and accounted for more than half of the bacterioplankton community. Some *Exiguobacterium* species could produce highly active oxidoreductive enzymes and tolerant to H_2O_2 (Takebe et al., 2007; Lee et al., 2009; Anbu et al., 2013). This might be one of the reasons why *Exiguobacterium* could survive after applications of H_2O_2. Additionally, phytoplankton might also influence the community structure and abundance of bacterioplankton (Casamatta and Wickstrom, 2000; Bagatini et al., 2014). In addition to the direct effect of H_2O_2 to bacterioplankton populations, the influence of H_2O_2 to phytoplankton might also contribute to the changes of the community structure of bacterioplankton that are subjected to H_2O_2.

Bacterioplankton is the major player of the freshwater ecosystem. For example, heterotrophic bacteria play an important role in the food chain between *Microcystis* and zooplankton (de Kluijver et al., 2012). In Dianchi Lake, the high rates of nutrients cycling in the microbial loop of the food web contribute to the outbreak and persistence of cyanobacteria blooms (Shan et al., 2014). The introduction of hydrogen peroxide might disturb the community structure of bacterioplankton which in turn might change the structure and function of freshwater ecosystem. Therefore, the structure and dynamics of bacterioplankton community can be used as an indicator to assess the risks of hydrogen peroxide treatment in the lakes with cyanobloom.

5 CONCLUSION

Our results showed that bacterioplanktons were more sensitive to H_2O_2 than most eukaryote phytoplankton. Hydrogen peroxide at doses of 4 mg/L

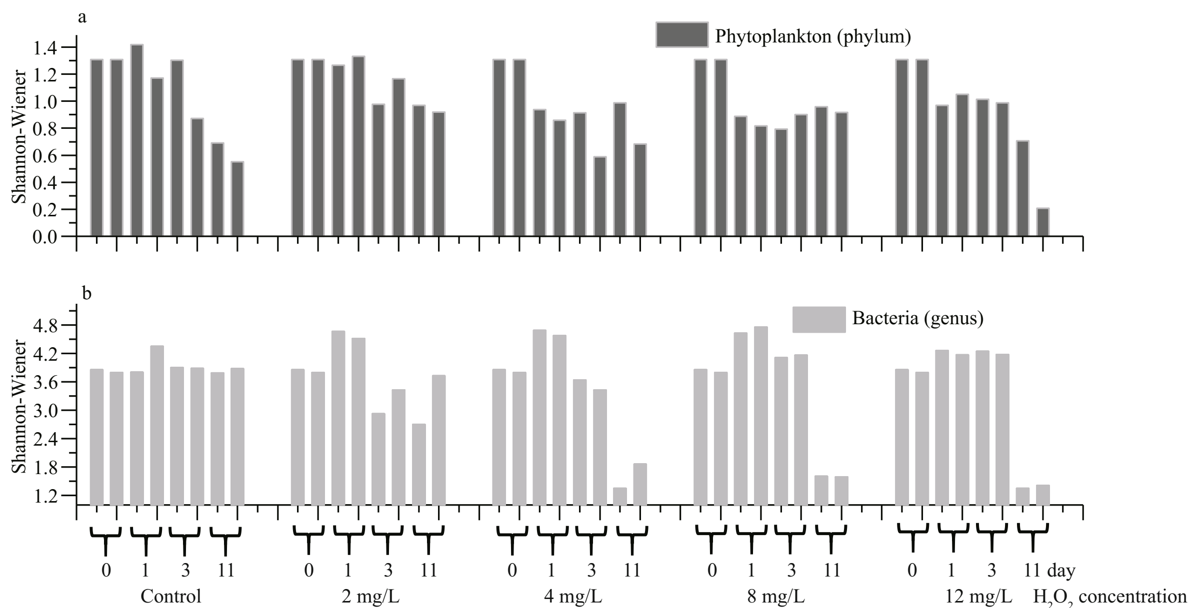

Fig.8 Changes of Shannon-Wiener index of bacterioplankton and phytoplankton during 11 days experiment

a. phytoplankton community structure based on the microscope examination; b. bacterioplankton community structure based on the proportion of 16S rDNA amplicons at the genus level.

and above can change the abundance and community structure of bacterioplanktons. *Firmicutes*, instead of *Bacteroidetes* and *Proteobacteria*, emerged as the dominant bacteria. High H_2O_2 doses can have a negative impact on the lake ecosystem by eliminating sensitive bacteria, mainly gram negative, which are predominant in aquatic environments. High concentration of H_2O_2 can cause "water disinfection" in which surviving gram-positive bacteria including *Exiguobacterium* and *Planomicrobium* might become dominant species in H_2O_2 treated body of water. This type of unintended changes in community structures of bacterioplankton could have a short and long term consequences to aquatic ecosystems. Thus, we suggest that bacterioplankton is a useful index for aquatic ecosystems when H_2O_2 was used to control cyanobacteria blooms.

6 DATA AVAILABILITY STATEMENT

The datasets during and/or analyzed during the current study available from the corresponding author on reasonable request.

7 ACKNOWLEDGEMENT

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