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Effects of Water Particles in the Jinjiang River Estuary on the Physiological and Biochemical Characteristics of *Microcystit Flos-Aquae*

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Research Article

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Abstract

The effects of different concentrations (100,150,200,250 mg/L) and different particle sizes (0–75μm, 75–120μm, 120–150μm, 150–500μm) on soluble protein content, SOD and CAT activity, MDA content, chlorophyll a content and photosynthetic parameters of *Microcystis flos-aquae* were studied, the mechanism of the effect of suspended particulate matter on the physiology and biochemistry of *Microcystis flos-aquae* was discussed. The results showed that the soluble protein content of *Microcystis flos-aquae* did not change obviously after being stressed by suspended particles of different concentration/diameter. The SOD activity of *Microcystis flos-aquae* increased at first and then decreased with the increase of the concentration of suspended particulate matter. The SOD activity of *Microcystis flos-aquae* reached 28.03 U/mL when the concentration of suspended particulate matter was 100 mg/L. The CAT activity of *Microcystis flos-aquae* increased with the increase of the concentration of suspended particles, and reached the maximum value of 12.45 U/mgprot in the concentration group of 250 mg/L, showing a certain dose-effect. The effect of small particle size on SOD, CAT and MDA of *Microcystis flos-aquae* was more significant than that of large particle size. The larger the concentration and the smaller the particle size, the stronger the attenuation of light and the lower the content of chlorophyll *a*. Both Fv/Fm and Fv/F_0 of *Microcystis flos-aquae* increased at first and then decreased under different concentration/size of suspended particles. The relative electron transfer rate gradually returned to the normal level with the passage of time. There was no significant difference in α value between treatment group and control group, ETRmax and I_k decreased.

1. Introduction

Water particulate matter generally refers to the suspended matter which is more than 0.45μm, including all particles with particle size larger than 1nm, the upper limit can reach tens to hundreds of microns, and widely exists in natural water, with the change of time and environment, migration, transformation and deposition take place continuously. Water particles, including minerals, clay, organic particles, inorganic particles surrounded by organic particles and biological residues, are a kind of special pollutants, which will have an impact on the water environment(Lu &Allen 2006). On the one hand, water particles affect the transparency of water bodies, and the degree to which water particles of different concentrations and sizes affect the transparency of water bodies is also different. On the other hand, water particles have a large specific surface area, it can be used as a carrier to combine with pollutants and thus affect the transfer and transformation of pollutants. In addition, water particles can also regulate the content of nutrients in water bodies. On the one hand, it can be used as a carrier to absorb nutrients in water bodies, settling into a sediment layer inhibits algal bloom, on the other hand it can also desorb nutrients an promote eutrophication of the water body(He et al. 2017, He et al. 2021, Swift et al. 2006) These changes in the water environment will affect the growth of algae in the water body, thus affecting the algal bloom process, so it is important to explore the effect of different concentration/size of water particles on the growth of algae.

As a unique ecosystem, estuaries can form an important pollutant barrier between the sea and the land boundary, and their Biogeochemistry processes have an impact on the regional material cycle. The Jinjiang estuary in Fujian is the third largest river in Fujian and also the river with the largest amount of sand. The water in the estuary area is strongly mixed, and the suspended particulates undergo physical, chemical and biological interactions, which makes the geochemical characteristics of the suspended particulates in the estuary more complicated, so it is important to study the effect of suspended particulate matter on algae growth and its response in the Jinjiang Estuary.

In this experiment, *Microcystis flos-aquae* was chosen as the research object to study the effects of different concentration/particle size of water particles on the physiological and biochemical indexes and photosynthetic activities of *Microcystis flos-aquae* in the estuary of Jinjiang River, and to reveal the mechanism of the effects of particles on the growth of algae, in order to provide a scientific basis for the study of the mechanism of the influence of water particles on the growth of algae.

2. Experimental Materials And Methods

2.1 Algae material and culture condition

Microcystis flos-aquae (FACHB-1028) was provided by the Freshwater Algae Culture Collection of the Institute of Hydrobiology (FACHB-Collection), Wuhan City, China. All the experimental vessels used for algae culture were sterilized at 121°C for 20min. *Microcystis flos-aquae* was grown in 250mL Erlenmeyer flasks containing 100mL BG-11 medium. The Algae were cultivated at 25 ± 1°C, 3000lx-4000lx and 50 $\mu mol \cdot m^{-2} \cdot s^{-2}$ illumination (cool white fluorescent tube) with a 14h/10h (light/dark) cycle. To minimize the effects of small variations in light, shake the lamp manually 3 times a day and rotate it randomly.

2.2 Suspended Particulate Matter treatment

Suspended particulates were collected from the surface sediments of the Jinjiang Estuary Wetland in April 2019. They were air-dried, ground, sifted and placed in a sealed plastic bag for later use. In the logarithmic growth period, the concentration of suspended particulate matter was 0, 100, 150, 200, 250 mg/L, the diameter of suspended particulate matter was CK, 0–75μm, 75–120μm, 120–150μm, 150–500μm. Each experimental concentration and particle size were repeated three times, all operations were carried out under sterile conditions to avoid bacterial contamination. They were cultured in an artificial climate box under the same conditions as before, and were manually shaken three times a day, and then randomly changed positions.

2.3 Determination of physiological and biochemical parameters

Microcystis flos-aquae was exposed to suspended particulate matter for 7 days, 50 mL of each culture medium was centrifuged at 8000 $r \cdot min^{-1}$ and 4°C for 10 min to harvest the cyanobacteria cells. Collected cells were resuspended in 5 mL phosphate buffer (pH 7.8), and then crushed in ice bath with ultrasonic cell crusher (power 500W). At last, 8000R Min-1, centrifuged at 4°C for 10min, the supernatant was used to determine physiological and biochemical indexes.(Nie et al. 2013) The content of soluble protein was determined by Coomassie Brilliant Blue G-250 staining. The activity of SOD was determined by SOD kit (NBT), and the activity of CAT was determined by Catalase kit (molybdic acid visible light colorimetry). Malondialdehyde (MDA) kit (TBA method) was used to determine MDA content.

2.4 Determination of chlorophyll a and photosynthetic parameters

In this study, the chlorophyll *a* (Chla) content and photosynthetic parameters of *Microcystis flos-aquae* were determined by a pulse amplitude-modulated fluorometer (Phyto-PAM Walz, Effeltrich, Germany). The Chlorophyll *a* content and photosynthetic parameters were determined by analysing samples (1mL) of each treatment every day after dark reaction for at least 15 minutes. The maximum quantum yield of PSII (Fv/Fm) and the potential photosynthetic activity of PSII (Fv/F_0) can be obtained by the following calculation (Wang et al. 2017):

$$\frac{F_v}{F_m} = \frac{F_m - F_0}{F_m}$$

$$\frac{F_v}{F_0} = \frac{F_m - F_0}{F_0}$$

A rapid light response curve (RLCS) was then determined. RLC is constructed by exposing the sample to each of 17 increased actinic light levels for 20 seconds. The nonlinear curve was fit using the model of by Platt et al. (Serodio et al. 2013)and applied to estimate the initial slope (alpha), the maximum photo synthetic rate (ETRmax) and the semi-light saturation (I_k). Alpha indicate the solar energy utilization efficiency of algae, the ETRmax shows the maximum electron transport rate and I_k represents the tolerance of algae to strong light(Houliez et al. 2017).

2.5 Statistical analysis

In this study, all data are from three parallel samples and are expressed as mean ± standard deviation. All data are mapped using Origin 2018 and statistical analysis is performed by SPSS 25.0 software package. One-way analysis of variance was used to determine the significant difference between the control group and the treatment. When $p < 0.01$, the difference is extremely significant, when $p < 0.05$, the difference is significant.

3. Results And Analysis

3.1 Effect of suspended particulate matter on soluble protein content of *Microcystis flos-aquae*

The soluble protein contents of *Microcystis flos-aquae* under different concentrations of suspended particulate matter are shown in Fig. 1a. The content of soluble protein of *Microcystis flos-aquae* was 0.51 ~ 0.49 mg/mL. The content of soluble protein of *Microcystis flos-aquae* was lower than that of control group under different concentration of particulate matter stress, but there was no significant difference between them ($p > 0.05$). *Microcystis flos-aquae* was tested for its soluble protein content under the same concentration of suspended particulate matter with different particle sizes, as shown in Fig. 1b. The soluble protein content of *Microcystis flos-aquae* was 0.39 ~ 0.41 mg/mL, and there was no significant difference between the two groups ($p > 0.05$). It can be seen that the concentration and size of water particles have no effect on the soluble protein content of *Microcystis flos-aquae*

3.2 Effects of suspended particulate matter on antioxidant enzymes of *Microcystis flos-aquae*

3.2.1 Effects of suspended particulate matter on SOD activity of *Microcystis flos-aquae*

The changes of SOD activity of *Microcystis flos-aquae* under different concentration/size of suspended particulate matter stress are shown in Fig. 2. As can be seen from Fig. 2a, the SOD activity of different concentration of suspended particulate matter was significantly higher than that of control group ($p < 0.01$), increased by 124.23%, 42.75%, 98.06% and 47.30%, respectively, the SOD activity of *Microcystis flos-aquae* was the highest (28.03 U/mL). The SOD activity of suspended particulate matter with different particle size under the stress of *Microcystis flos-aquae* is shown in Fig. 2b. SOD activity of algae in 0–75μm particle size group was significantly lower than that in control group ($p < 0.05$), which was 16.73% lower than that in control group, and significantly lower than that in 120–150μm particle size group ($p < 0.01$), which was 75.67% lower than that in control group, there was no significant difference in SOD activity between 75–120μm and 150–500μm groups ($p > 0.05$). The SOD activity in 75–120μm group was 9.27% higher than that in control group, and the SOD activity in 150–500μm group was 11.20% lower than that in control group, the activity of SOD was the lowest when the particle size was 120–150μm, which was 11.59 U/mL.

3.2.2 Effects of suspended particulate matter on CAT activity of *Microcystis flos-aquae*

The effects of different concentrations of suspended particulate matter on the CAT activity of *Microcystis flos-aquae* are shown in Fig. 3a. The CAT activity of Microcystis algae in the low concentration treatment group (100mg/L) was not significantly different from that of the control group ($p > 0.05$). With the increase of stress concentration, the activity of CAT increased. When the concentration of suspended particulate matter was 150mg/L, the CAT activity of algae was significantly higher than that of the control group ($p < 0.05$). When the concentration of suspended particulate matter was 200 mg/L and 250 mg/L, the CAT activity of algae was significantly higher than that of the control group ($p < 0.01$), which were 11.61 U/mgprot and 12.45 U/mgprot, respectively. The effects of different particle sizes on the CAT activity of *Microcystis flos-aquae* are shown in Fig. 3b. The CAT activity in the 0–75μm size group was significantly higher than that in the control group ($p < 0.01$), which was 11.32 U/mgprot. The CAT activity in the 75–120μm size group and the 150–500μm size group was significantly lower than that in the control group (51.34% and 62.19%, respectively), there was no significant difference in CAT activity between the 120–150μm size group and the control group ($P > 0.05$).

3.3 Effects of suspended particulate matter on the content of MDA in *Microcystis flos-aquae*

The effect of different concentration/size of suspended particulate matter on the MDA content of *Microcystis flos-aquae* is shown in Fig. 4. The effect of different concentrations of suspended particles on the MDA of Microcystis algae is shown in Fig. 4a. The MDA content is 0.25 ~ 4.11 nmol/mL. There is no significant difference between the 200 mg/L concentration group and the control group ($p > 0.05$). The contents of MDA in *Microcystis flos-aquae* at other concentrations were lower than those in the control group, the concentration of 100mg/L group was significantly lower than that of the control group ($p < 0.01$). Compared with the control group, the concentration of 150 mg/L group was significantly lower than that of 250 mg/L group ($p < 0.05$). Compared with the control group, the concentration of 150 mg/L group was 78.23% and 79.50% lower respectively. The effect of suspended particles of different particle sizes on the MDA of *Microcystis flos-aquae* is shown in Fig. 4b. Its MDA content is 1.39 ~ 6.43 nmol/mL, except that the MDA content of the 0–75μm particle size group is significantly higher than that of the control group ($p < 0.05$), the MDA content of the other particle size groups was lower than the control group. The MDA content of *Microcystis flos-aquae* in the 75–120μm and 150–500μm diameter groups was significantly lower than that in the control group ($p < 0.01$), which decreased by 66.14% and 65.58%, respectively, and by 32.12% in the 120–150μm diameter groups, but there was no significant difference between the two groups ($p > 0.05$).

3.4 Effects of suspended particulate matter on chlorophyll-*a* content of M*icrocystis flos-aquae*

The effect of the same diameter and different concentration of suspended particulate matter on the content of chlorophyll-*a* in *Microcystis flos-aquae* is shown in Fig. 5a. As can be seen from the graph, the content of chlorophyll *a* increased obviously, and the inhibition or promotion effect on algae growth would be different in each experimental group because of the different concentration of suspended particulate matter. The content of chlorophyll *a* of *Microcystis flos-aquae* in the 200mg/L and 250mg/L concentration groups was significantly different from that in the control group at the 3rd and 4th day ($p < 0.05$), and the other concentration groups had no significant difference from that in the control group (p >

0.05). From the 3rd day, the content of chlorophyll a in *Microcystis flos-aquae* was lower in each particle concentration group than that in the control group, and the inhibitory effect of 250mg/L particle concentration group on algae growth was not as good as that of 200mg/L particle concentration group, but the inhibitory effect was better than the other concentration groups. The chlorophyll-*a* contents of *Microcystis flos-aquae* at the same concentration and different particle sizes were shown in Fig. 5b. It can be seen from the diagram that the chlorophyll *a* content of *Microcystis flos-aquae* in the 120–150μm diameter group was significantly lower than that in the control group ($p < 0.01$), and the chlorophyll a content of *Microcystis flos-aquae* in the 0–75μm diameter group was significantly lower than that in the control group ($p < 0.05$). There was no significant difference in the content of chlorophyll-a between the control group and the other size groups ($p > 0.05$), but the content of chlorophyll-*a* in the other size groups was lower than that in the control group.

3.5 Effects of suspended particulate matter on fluorescence parameters of *Microcystis flos-aquae*

3.5.1 Effect of suspended particulate matter on Fv/Fm of *Microcystis flos-aquae*

The maximum quantum yield of PSII, Fv/Fm, of *Microcystis flos-aquae* under the same particle size and different concentration of suspended particles stress is shown in Fig. 6a. It can be seen from the graph that Fv/Fm value of 200mg/L particulate matter on the 6th day was significantly lower than that of the control group ($p < 0.01$), and there was no significant difference between the other groups ($P > 0.05$). The effect of suspended particles of the same concentration and different size on the maximum quantum yield of PSII, Fv/Fm, is shown in Fig. 6b, the Fv/Fm value of *Microcystis flos-aquae* in each particle size group increased firstly and then decreased with time, except the control group, the Fv/Fm values of *Microcystis flos-aquae* in each size group were significantly higher than those of the corresponding size group on the first day of the 5th and 6th day.

3.5.2 Effect of suspended particulate matter on Fv/F_0 of *Microcystis flos-aquae*

The effect of suspended particulate matter with the same size and different concentration on the potential activity of PSII, Fv/F_0, of *Microcystis flos-aquae* was shown in Fig. 7a. There was no significant difference in Fv/F_0 between the control group and the control group on the 5th and 6th day ($p > 0.05$). The effect of suspended particles of the same concentration and different size on the potential activity of PSII, Fv/F_0, of *Microcystis flos-aquae* was shown in Fig. 7b. It can be seen that the Fv/F_0 value of each size group increased firstly with the passage of time, on the 3rd day, it reached its maximum value, which was significantly higher than that of the control group ($p < 0.01$), and then decreased. In a word, under the stress of suspended particulate matter of different concentration/size, *Microcystis flos-aquae* Fv/F_0 recovered to some extent with the passage of time.

3.5.3 Effects of suspended particulate matter on the fast light response curve of *Microcystis flos-aquae*

Figure 8 shows the effect of the same particle size and different concentration of suspended particulate matter on the fast light response curve of *Microcystis flos-aquae* at day 1–6. In the first two days, there was no significant difference in the rETR value of *Microcystis flos-aquae* in each concentration group ($p > 0.05$). On the third day, the rETR value of *Microcystis flos-aquae* in all concentration groups was higher than that in the control group, and the rETR value of *Microcystis flos-aquae* in the 200mg/L concentration group was significantly higher than that in the control group ($p < 0.05$), at the 4th day, except the 100mg/L concentration group was significantly lower than the control group, all the other concentration groups were reduced to the level of the control group. After the 5th day, the 100mg/L concentration group also recovered to the level of the control group.

Figure 9 shows the effect of the same concentration of suspended particulate matter of different size on the fast light response curve of *Microcystis flos-aquae* at day 1–6. On the first day, the rETR value of *Microcystis flos-aquae* in all particle size groups was higher than that in control group, and the rETR value of 0–75μm particle size group and 75–120μm particle size group were significantly higher than that in control group ($p < 0.05$). In the following two days, the rETR values of each particle size group were not significantly different from those of the control group ($p > 0.05$), and both fell to the control group level. On the 4th day, the rETR value of *Microcystis flos-aquae* in all size groups was higher than that in the control group. The rETR values of algae in the 0–75μm and 120–150μm particle size groups were significantly higher than those in the control group ($p < 0.01$), and those in the 75–120μm particle size groups were significantly higher than those in the control group ($p < 0.05$). On the 5th day, the rETR value of 0–75μm and 75–120μm particle size groups was significantly higher than that of control group ($p < 0.05$).

3.5.4 Effects of suspended particulate matter on α, ETRmax and I_k of *Microcystis flos-aquae*

In Fig. 10, a, b, and c are the effects of suspended particles of the same size and different concentrations on the photosynthetic parameters α, ETRmax, I_k of *Microcystis flos-aquae*. The results showed that there was no significant change in α value at all particle concentrations ($p > 0.05$), and the ETRmax and I_k value of *Microcystis flos-aquae* at all particle concentrations were significantly higher than those of the control group on the 6th day ($p < 0.01$). In Fig. 10, d, e, and f are the effects of suspended particles of the same concentrations and different size on the photosynthetic parameters α, ETRmax, I_k of *Microcystis flos-aquae*. Through the analysis, it can be obtained that there is basically no significant change in the α value under the stress of various particle sizes ($p > 0.05$). The ETRmax and I_k values of all the size groups of algae showed a trend of decreasing in the first 3 days, rising sharply on the 4th day and then decreasing again. On the 6th day, the ETRmax of the 150–500μm particle size group was significantly lower than that of the control group ($p < 0.01$), and the ETRmax of the 75–120μm particle size group and the 0–75μm particle size group were significantly higher than the control group ($p < 0.01$), the ETRmax of the 120–150μm group algae was significantly higher than that of the control group ($p < 0.05$). The I_k of the 0–75μm particle size group of *Microcystis flos-aquae* was significantly lower than that of the control group ($p < 0.05$), and the I_k of the other particle size groups was not significantly different from that of the control group ($p > 0.05$).

4. Discussion

4.1 Effects of suspended particles in water on lipid peroxidation and antioxidant capacity of *Microcystis flos-aquae*

Under the stress of different concentration/size of suspended particles, the content of soluble protein did not change obviously, it is suggested that the effect of suspended particulate matter on *Microcystis flos-aquae* is not accomplished by inhibiting the synthesis of soluble protein.

In this study, SOD activity first increased and then decreased with the increase of suspended particulate matter concentration. When *Microcystis flos-aquae* was slightly stressed, SOD activity increased to remove excess $O_2^{-}•$, when the threshold of its action is exceeded, the activity decreases. The same results were found in the study by Wang et al. (Wang et al. 2011), which showed that a great enhancement of SOD activity in the low concentrations of cypermethrin. The CAT activity increased with the increase of the concentration of suspended particulate matter, showing a certain dose-effect. Reactive oxygen species (Ros) are thought to be key regulatory molecules that are essential to cells (Wrzaczek et al. 2013). Under normal conditions, the reactive oxygen species in plant cells can be maintained in a relatively stable dynamic equilibrium. When subjected to external stress, this balance will be disrupted. At this time, plant cells will clear the over-produced reactive oxygen species through enzymatic and non-enzymatic systems. SOD and CAT are important antioxidant stress-related enzymes. SOD is the first line of defense against reactive oxygen species, which can convert $O_2^{-}•$ into O_2 and H_2O_2. CAT is a multifunctional enzyme responsible for scavenging H_2O_2(Gill &Tuteja 2010). The SOD activity of the 0–75μm particle size group was significantly lower than that of the control group, and the CAT activity showed an increasing trend with the decrease of the particle size. This indicates that the small particle size suspended particles have a more significant impact on the antioxidant enzyme system of *Microcystis flos-aquae* than the large particle size.

The MDA content of the 0–75μm particle size group of *Microcystis flos-aquae* was significantly higher than that of the control group, indicating that the stress of suspended particles on the algae caused a large amount of active oxygen in the cells of *Microcystis flos-aquae* to accumulate, which could not be effectively eliminated, resulting in cells lipid peroxidation. MDA is the final product of lipid peroxidation. When the cell is under stress, the cell's antioxidant defense system will act first to remove active oxygen. If the excessively produced active oxygen is not effectively eliminated, it will cause cell lipid peroxidation, and the MDA content will increase. (Lushchak 2011).

4.2 Effects of suspended particulate matter on chlorophyll *a* and photosynthetic parameters of *Microcystis flos-aquae*

Photosynthesis is one of the most important metabolic activities of algae, and chlorophyll is its main pigment. Studies have shown that when algae are stressed by the outside world, the algae cell chlorophyll a will also be affected(Alahverdi &Savabieasfahani 2012, Metzler et al. 2018, Patil et al. 2017). In this study, with the increase of the concentration of particulate matter, the chlorophyll *a* content of *Microcystis flos-aquae* gradually decreased, that is, the higher the concentration of particulate matter, the stronger the attenuation of light by suspended particles. On the 6th day, the chlorophyll a content of the 250mg/L group was higher than that of the 200mg/L group, which may be due to the role of suspended particulate matter in releasing nutrients at this time(He et al. 2017) It was found that the small particle size group (0–75μm) had a greater effect on the content of chlorophyll a in *Microcystis flos-aquae*, this indicates that the attenuation of light is different with the same concentration and different particle size. The smaller the particle size, the more the amount of the particles and the denser the distribution, the stronger the attenuation of light.

Chlorophyll fluorescence parameters can reflect the changes of photosynthesis of algae under external stress. The maximum quantum yield of PSII, Fv/Fm, reflects the intrinsic energy conversion efficiency of PSII reaction center, and the potential activity of PSII, Fv/F_0, are the two most widely used chlorophyll fluorescence parameters(Wang et al. 2008, Wu et al. 2019). Both Fv/Fm and Fv/F_0 showed a rising and then a falling trend, which might be due to the stress response of *Microcystis flos-aquae* after stress, which promoted the efficiency of photoelectron transfer.

The fast light response curve is a photosynthesis curve drawn by ETR, which can provide fast and simple measurement for plant photosynthetic response (Huang et al. 2021). ETR has been found to be associated with O_2 and CO_2 metabolism. On the 4th day, except for the 100mg/L concentration group, which was significantly lower than the control group, all the other concentration groups fell to the control level. On the 5th day, the 100mg/L concentration group also returned to the control level. The 0–75μm particle size group was extremely significantly higher than the control group on the 4th day, and decreased on the 5th day. The relative electron transfer rate of *Microcystis flos-aquae* increases or decreases under the stress of suspended particles. As time goes by, the relative electron transfer rate of *Microcystis flos-aquae* can be restored to the normal level by self-regulation. There was no significant difference in α value between the treatment group and the control group, which indicated that the utilization of light energy by *Microcystis flos-aquae* could not be affected. The decrease of ETRmax and I_k indicates that most of the receptor QA in the PSII reaction center of algae cells cannot be oxidized and is in a reduced state, which hinders the transfer of electrons from PSII to PSI, thus affecting the electron transfer rate of PSII in algae cells(Hong et al. 2008). The decrease of I_k also indicates that the tolerance of algal cells to strong light is reduced.

5. Conclusions

1)The content of soluble protein in *Microcystis flos-aquae* did not change obviously after being stressed by suspended particles of different size/concentration, the contents were 0.51 ~ 0.49 mg/mL (concentration group) and 0.39 ~ 0.41 mg/mL (particle size group), respectively. With the increase of the concentration of suspended particles, the activity of SOD increased at first and then decreased. The activity of SOD reached 28.03 U/mL at the concentration of 100 mg/L. When *Microcystis flos-aquae* was slightly stressed, SOD activity increased to remove excess $O_2^{-}\bullet$, but decreased when the threshold was exceeded. The CAT activity of 250 mg/L group reached a maximum of 12.45 U/mgprot, which showed a dose-effect. The effect of small particle size on SOD, CAT and MDA of *Microcystis flos-aquae* was more significant than that of large particle size.

2)The larger the concentration and the smaller the diameter of suspended particles, the stronger the attenuation of light and the lower the content of chlorophyll *a*. Both Fv/Fm and Fv/F_0 of *Microcystis flos-aquae* increased at first and then decreased under different concentration/size of suspended particles. The relative electron transfer rate will increase or decrease. Over time, the algae cells can adjust themselves to restore the relative electron transfer rate to the normal level. The decrease of ETRmax and I_k showed that the utilization of light energy was not affected, but the tolerance to strong light was decreased.

6. Declarations

Ethics approval: The authors confirm that this article is original research and has not been published or presented previously in any journal or conference in any language (in whole or in part)

Consent to participate: Not applicable in this section.

Consent for publication: Not applicable in this section.

Competing interests: The authors declare no competing interests.

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Author contribution:

The participation of Yingting Nan, Hui Xing, Sijia Chen Bo Hu and Jie Liu includes the operation of experiments, and the participation of Yiting Nan and Peiyong Guo includes analyzing the results, and writing the article. All authors read and approved the final manuscript.

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Data availability: All necessary data are present in this paper. In case where they request more data details, it can be sent.

7. References

1. Alahverdi M, Savabieasfahani M (2012): Seaweed and Chlorophyll as Biomarkers of Metals in the Persian Gulf, Iran. Bulletin of Environmental Contamination and Toxicology 89 (3), 501-506
2. Gill SS, Tuteja N (2010): Reactive oxygen species and antioxidant machinery in abiotic stress tolerance in crop plants. Plant Physiology and Biochemistry 48 (12), 909-930
3. He Q, Qiu Y, Liu H, Sun X, Kang L, Cao L, Li H, Ai H (2017): New insights into the impacts of suspended particulate matter on phytoplankton density in a tributary of the Three Gorges Reservoir, China. Sci Rep 7 (1), 13518
4. He Y, Qin N, He W, Xu FL (2021): The impacts of algae biological pump effect on the occurrence, source apportionment and toxicity of SPM-bound PAHs in lake environment. Sci. Total Environ. 753 (2-4), 141980
5. Hong Y, Hong-Ying H, Li F-M (2008): Growth and physiological responses of freshwater green alga Selenastrum capricornutum to allelochemical ethyl 2-methyl acetoacetate (EMA) under different initial algal densities. Pesticide Biochemistry and Physiology 90 (3), 203-212
6. Houliez E, Lefebvre S, Lizon F, Schmitt FG (2017): Rapid light curves (RLC) or non-sequential steady-state light curves (N-SSLC): which fluorescence-based light response curve methodology robustly characterizes phytoplankton photosynthetic activity and acclimation status? Mar. Biol. 164 (8), 175
7. Huang M, Wong S, Weng J (2021): Rapid Light-Response Curve of Chlorophyll Fluorescence in Terrestrial Plants: Relationship to CO2 Exchange among Five Woody and Four Fern Species Adapted to Different Light and Water Regimes. Plants-Basel 10 (3), 445

8. Lu Y, Allen HE (2006): A predictive model for copper partitioning to suspended particulate matter in river waters. Environmental Pollution 143 (1), 60-72
9. Lushchak VI (2011): Adaptive response to oxidative stress: Bacteria, fungi, plants and animals. Comparative Biochemistry and Physiology C-Toxicology & Pharmacology 153 (2), 175-190
10. Metzler DM, Erdem A, Huang CP (2018): Influence of Algae Age and Population on the Response to TiO2 Nanoparticles. International Journal of Environmental Research and Public Health 15 (4), 585
11. Nie X-P, Liu B-Y, Yu H-J, Liu W-Q, Yang Y-F (2013): Toxic effects of erythromycin, ciprofloxacin and sulfamethoxazole exposure to the antioxidant system in Pseudokirchneriella subcapitata. Environmental Pollution 172, 23-32
12. Patil S, Pandit R, Lali A (2017): Photosynthetic acclimation of Chlorella saccharophila to heat stress. Phycological Research 65 (2), 160-165
13. Serodio J, Ezequiel J, Frommlet J, Laviale M, Lavaud J (2013): A Method for the Rapid Generation of Nonsequential Light-Response Curves of Chlorophyll Fluorescence. Plant Physiology 163 (3), 1089-1102
14. Swift TJ, Perez-Losada J, Schladow SG, Reuter JE, Jassby AD, Goldman CR (2006): Water clarity modeling in Lake Tahoe: Linking suspended matter characteristics to Secchi depth. Aquatic Sciences 68 (1), 1-15
15. Wang G, Chen K, Chen L, Hu C, Zhang D, Liu Y (2008): The involvement of the antioxidant system in protection of desert cyanobacterium Nostoc sp against UV-B radiation and the effects of exogenous antioxidants. Ecotoxicology and Environmental Safety 69 (1), 150-157
16. Wang M, Zhang Y, Guo P (2017): Effect of florfenicol and thiamphenicol exposure on the photosynthesis and antioxidant system of Microcystis flos-aquae. Aquatic Toxicology 186, 67-76
17. Wang Z, Nie X, Yue W (2011): Toxicological effects of cypermethrin to marine phytoplankton in a co-culture system under laboratory conditions. Ecotoxicology 20 (6), 1258-1267
18. Wrzaczek M, Brosche M, Kangasjarvi J (2013): ROS signaling loops - production, perception, regulation. Current Opinion in Plant Biology 16 (5), 575-582
19. Wu Y, Guo P, Zhang X, Zhang Y, Xie S, Den J (2019): Effect of microplastics exposure on the photosynthesis system of freshwater algae. Journal of Hazardous Materials 374, 219-227

Figures

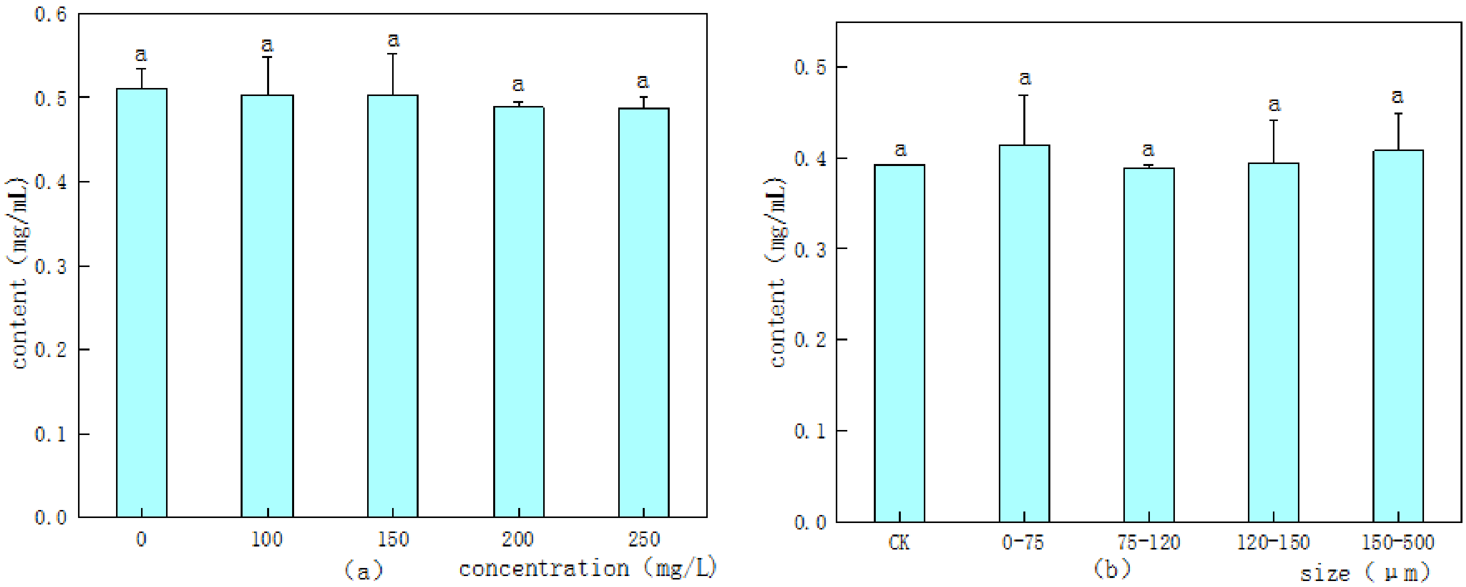

Figure 1

Effects of different concentrations or sizes suspended particulate matters on soluble protein of Microcystis flos-aquae

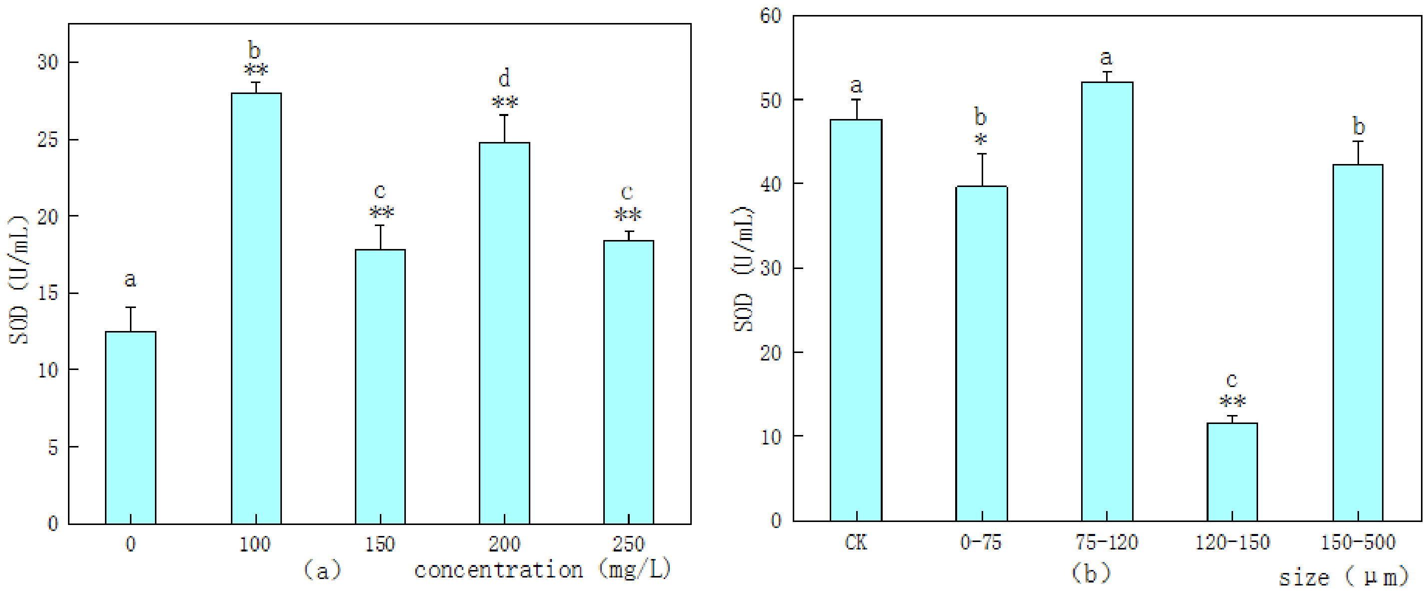

Figure 2

Effects of different concentrations or sizes suspended particulate matters on SOD activity of Microcystis flos-aquae

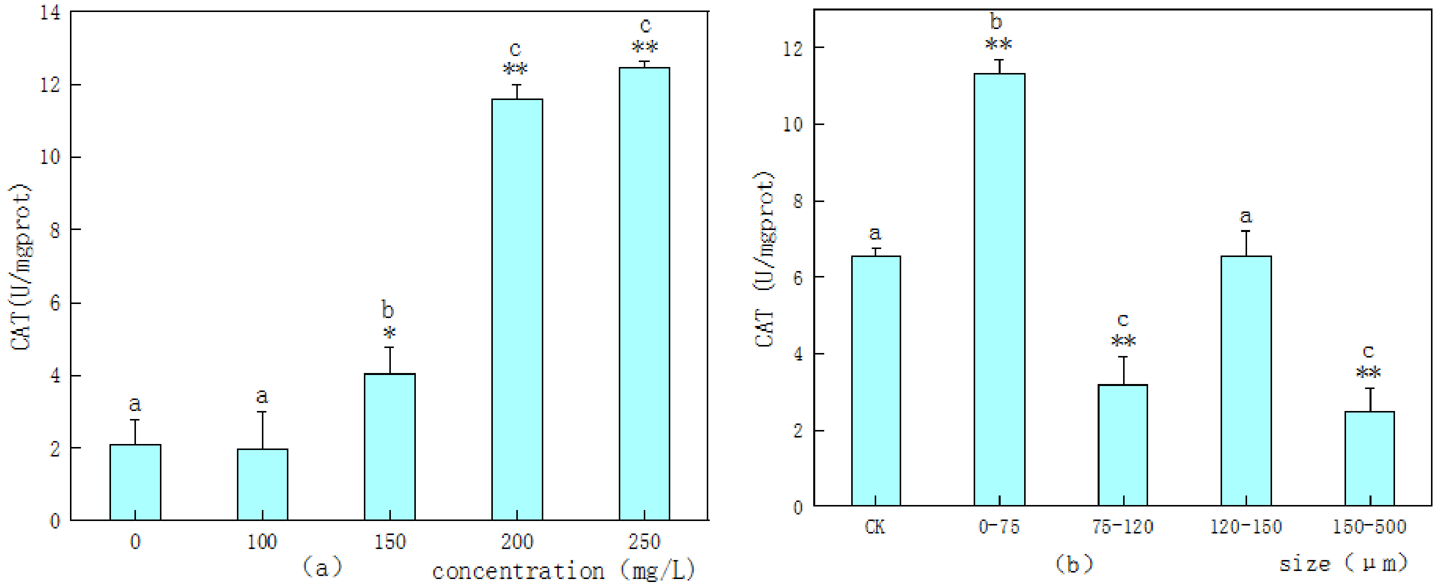

Figure 3

Effects of different concentrations or sizes suspended particulate matters on CAT activity of Microcystis flos-aquae

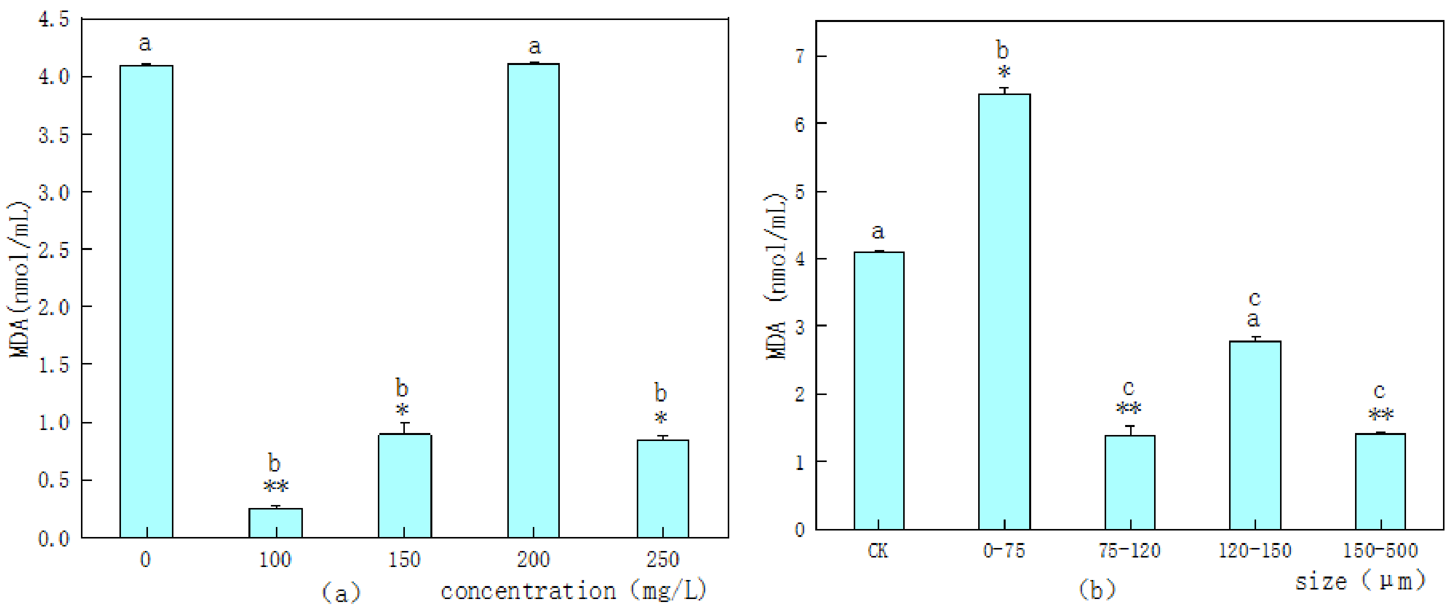

Figure 4

Effects of different concentrations or sizes suspended particulate matters on the MDA content of Microcystis flos-aquae

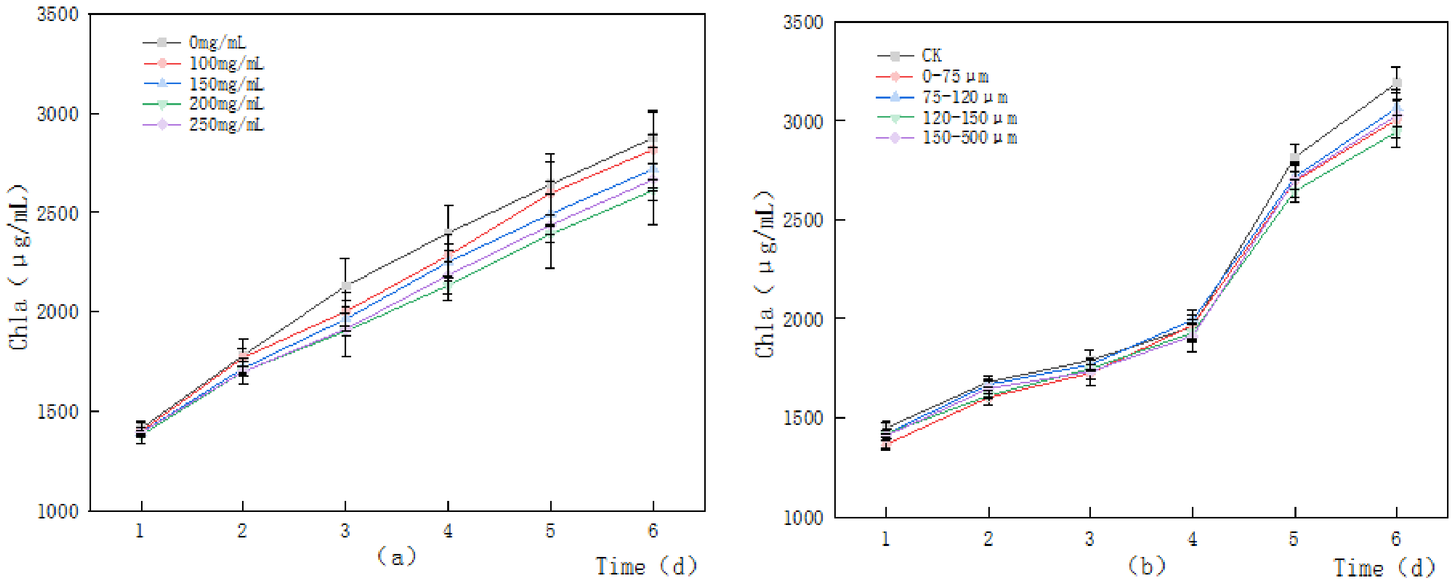

Figure 5

Effects of different concentrations or sizes suspended particulate matters on chlophyll a content of Microcystis flos-aquae

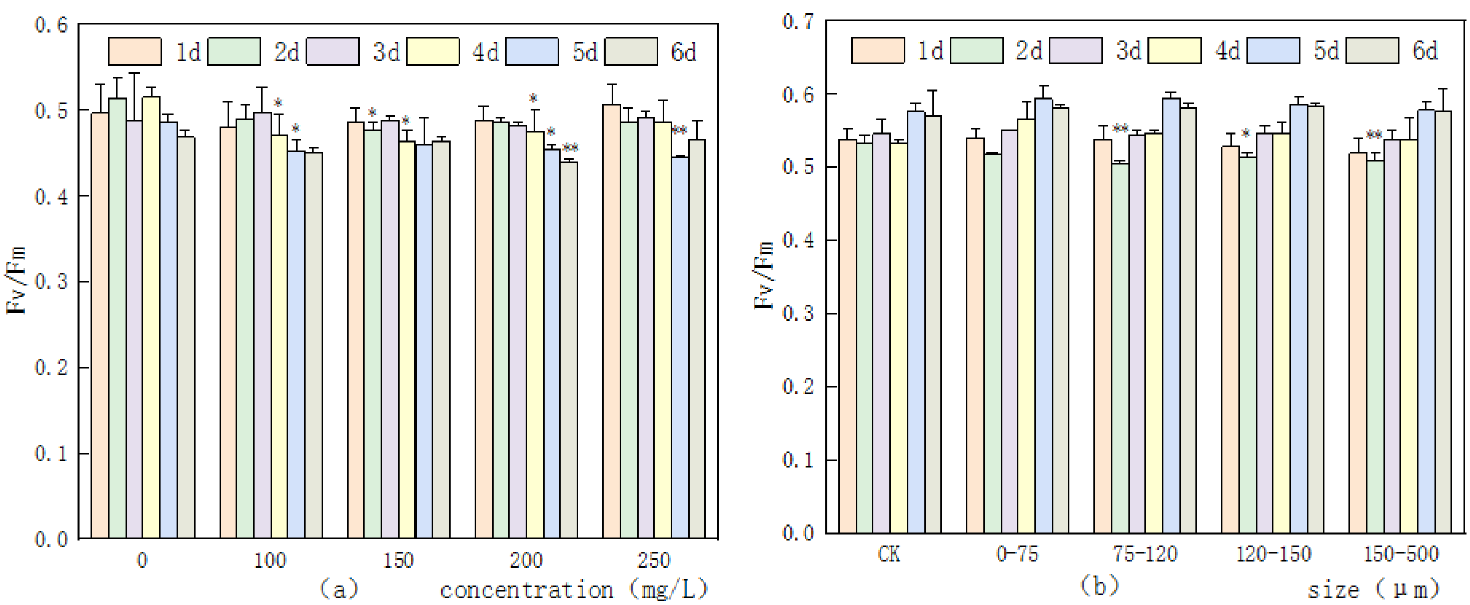

Figure 6

Effects of different concentrations or sizes suspended particulate matters on maximum quantum yield of Microcystis flos-aquae

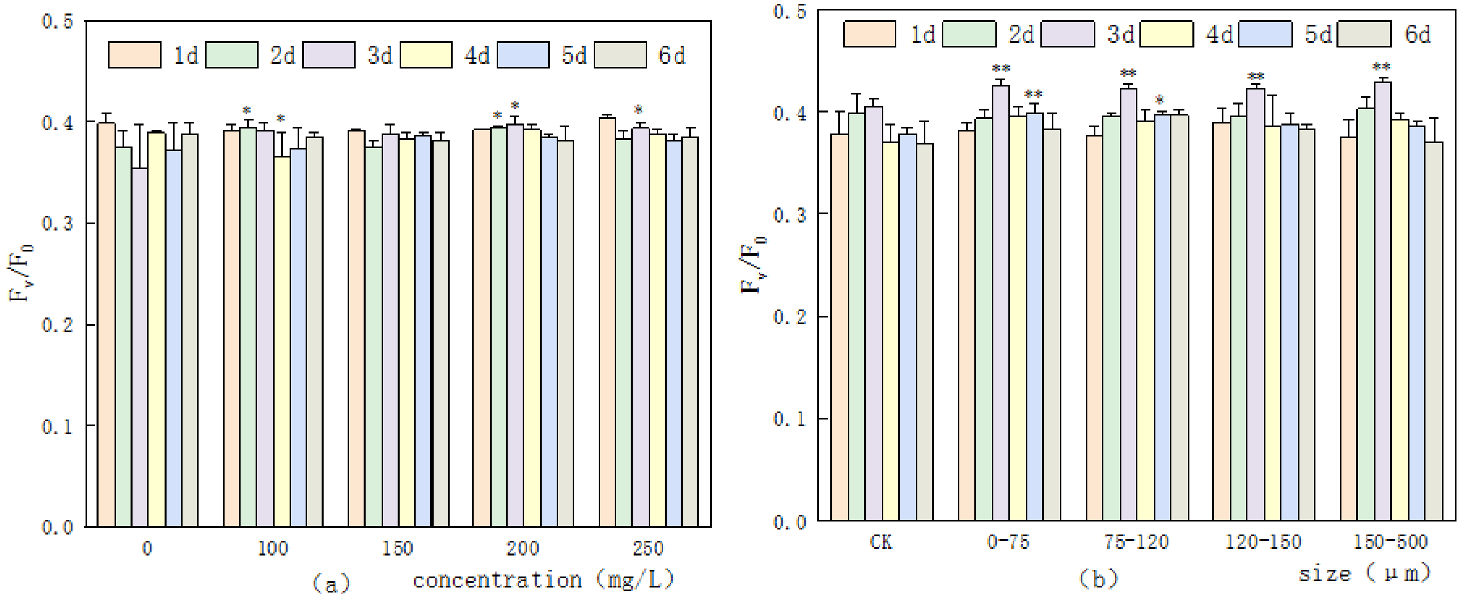

Figure 7

Effects of different concentrations or sizes suspended particulate matters on potential activity of Microcystis flos-aquae

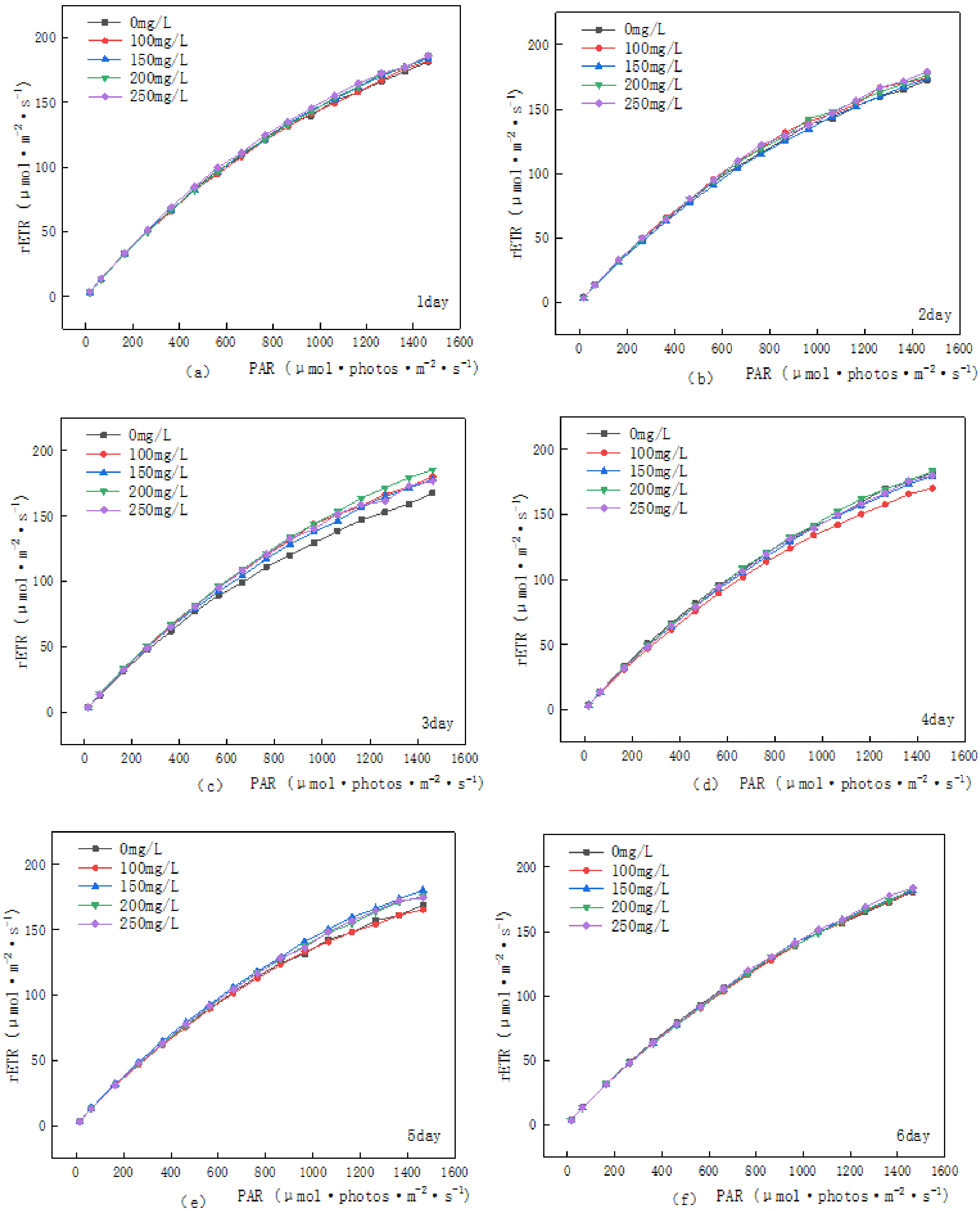

Figure 8

Effects of different concentrations suspended particulate matters on the Rapid light curves (RLCs) of Microcystis flos-aquae

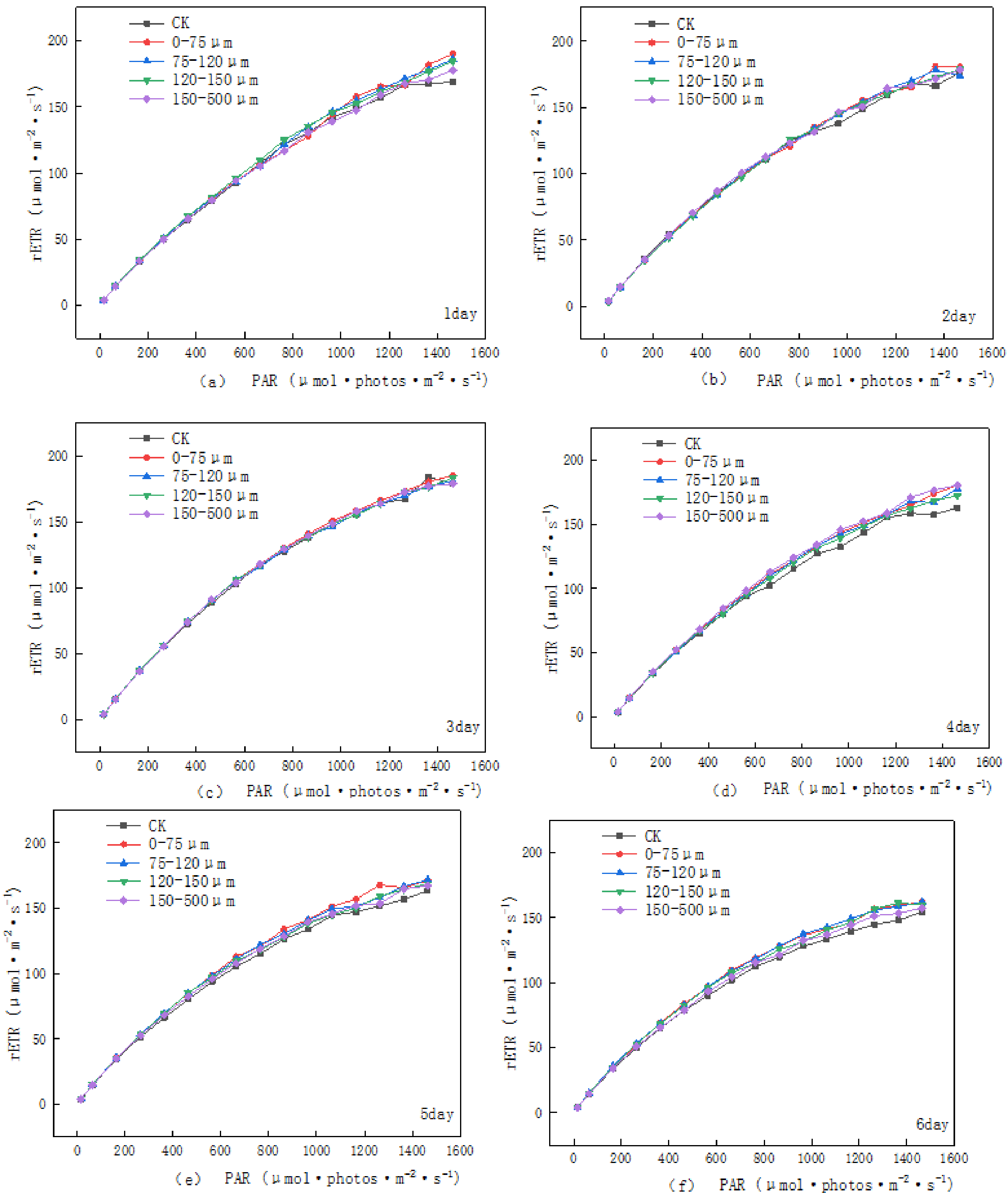

Figure 9

Effects of different sizes suspended particulate matters on the Rapid light curves (RLCs) of Microcystis flos-aquae

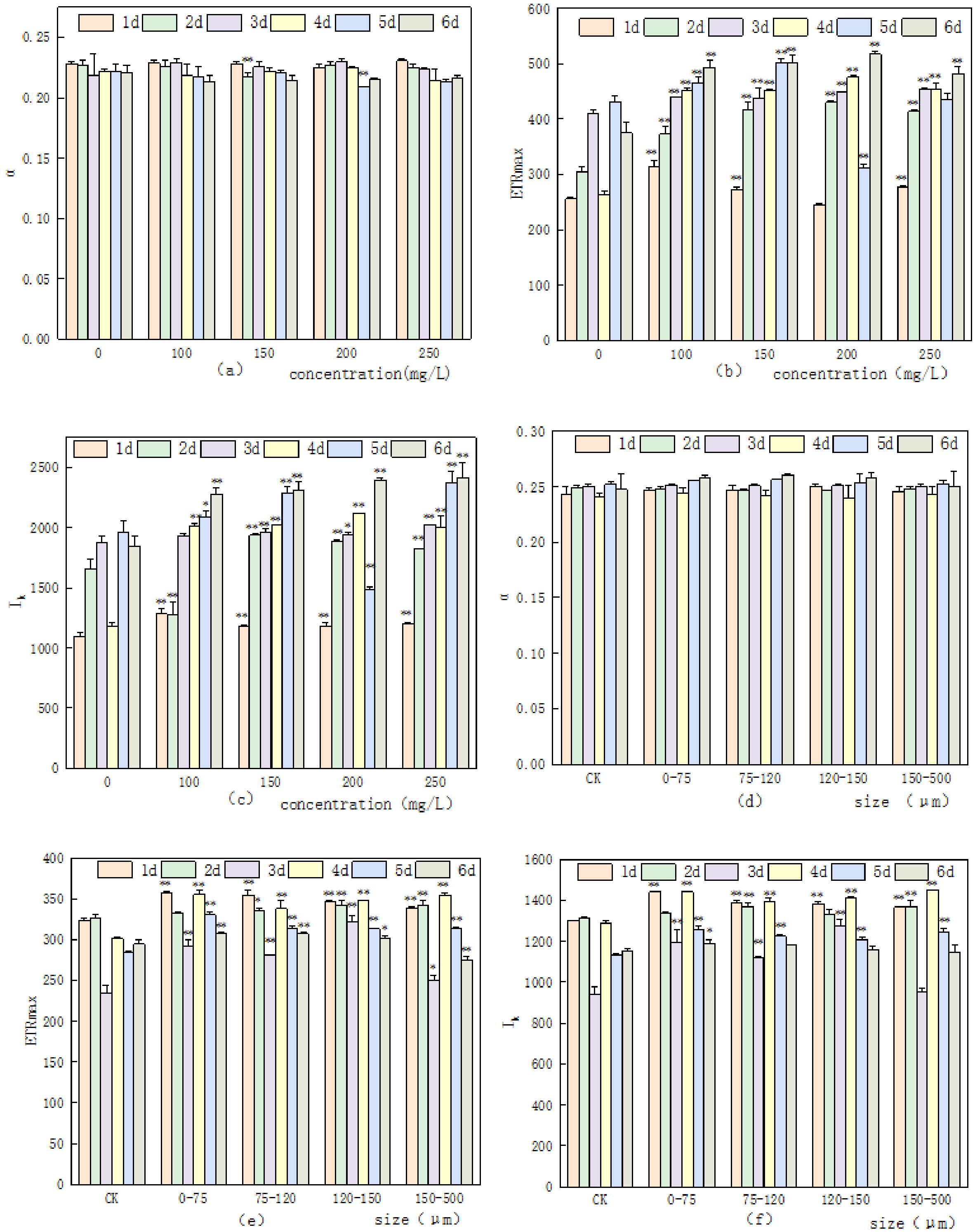

Figure 10

Effects of different concentrations or sizes suspended particulate matters on Microcystis flos-aquae RLCs parameters such as alpha ETRmax Ik