Available online at www.sciencedirect.com

ScienceDirect

www.journals.elsevier.com/journal-of-environmental-sciences

JOURNAL OF ENVIRONMENTAL SCIENCES
www.jesc.ac.cn

Influence of zeta potential on the flocculation of cyanobacteria cells using chitosan modified soil

*Liang Li, Honggang Zhang, Gang Pan**

Research Center for Eco-Environmental Sciences, Chinese Academy of Sciences, Beijing 100085, China. E-mail: ironlee@sina.com

ARTICLE INFO

Article history:
Received 20 March 2014
Revised 17 April 2014
Accepted 22 April 2014
Available online 15 November 2014

Keywords:
Cyanobacteria blooms
Flocculation
Chitosan
Modified soil
Zeta potential

ABSTRACT

Using chitosan modified soil to flocculate and sediment algal cells has been considered as a promising strategy to combat cyanobacteria blooms in natural waters. However, the flocculation efficiency often varies with algal cells with different zeta potential (ZP) attributed to different growth phases or water conditions. This article investigated the relationship between ZP of *Microcystis aeruginosa* and its influence to the flocculation efficiency using chitosan modified soil. Results suggested that the optimal removal efficiency was obtained when the ZP was between −20.7 and −6.7 mV with a removal efficiency of more than 80% in 30 min and large floc size of >350 μm. When the algal cells were more negatively charged than −20.7 mV, the effect of chitosan modified soil was depressed (<60%) due to the insufficient charge density of chitosan to neutralize and destabilize the algal suspension. When the algal cells were less negative than −6.7 mV or even positively charged, a small floc size (<120 μm) was formed, which may be difficult to sink under natural water conditions. Therefore, manipulation of ZP provided a viable tool to improve the flocculation efficiency of chitosan modified soil and an important guidance for practical engineering of cyanobacteria bloom control.

Introduction

Excess qualities of nutrients have been discharged into fresh waters, inducing a global environmental epidemic of cyanobacteria blooms (Paerl and Huisman, 2008). Such blooms pose serious threats to aquatic life, fish industry, local tourism, and water quality in lakes, rivers and reservoirs (Beaulieu et al., 2005). They also threaten drinking water safety, such as the drinking water crisis in Wuxi City, China in 2007 (Guo, 2007).

Over the past several decades, many efforts have been done to combat the cyanobacteria blooms (HABs). Among the technologies of mechanical, biological, chemical, genetic and environmental control (Anderson, 2009), significant attention has been focused on the use of clay to flocculate and settle the cyanobacteria cells in natural waters (Anderson, 1997; Sengco et al., 2001; Yu et al., 1994). However, the efficiency of clay alone was low and high loads of clay (0.25–2.5 g/L) (Pan et al., 2006; Sengco et al., 2001; Sun et al., 2004) often lead to various ecological concerns (Lee et al., 2008). Pan et al. (2006) found that local soil/sand collected from lake shore after modified by chitosan could be turned into effective flocculants to remove cyanobacteria blooms and improve water quality, which greatly reduced the dosage to 11 mg/L and hence minimized the costs and the use of exogenous materials to the aquatic environments. Chitosan, a commercially available product of edible food additives, is derived from the alkaline deacetylation of crustacean chitin and known to be a biodegradable and non-toxic natural polymer. A field application of chitosan modified soil in Lake Taihu and the study of ecological

* *Corresponding author*. E-mail: gpan@rcees.ac.cn (Gang Pan).

http://dx.doi.org/10.1016/j.jes.2014.04.017

response in time scale of months to year proved its efficiency and ecological safety, 0.1 km^2 of the HAB layer disappeared in 10 hr after the dispersion of the chitosan modified soil and the submerged vegetation was successfully restored after 4 months due to the improved water quality (Pan et al., 2011b).

The key mechanism of chitosan modified soil/sand to remove cyanobacteria blooms was that the chitosan with long polymer chain and positively charged groups ($-NH_3^+$) captured and linked the negatively charged algal cells and other particles, the soils then provided the mass or ballast to carry the flocs to the water sediment (Zou et al., 2006). Therefore, the surface charge of algal cells was critically important for the flocculation process. However, the zeta potential (ZP), which gives a measurement of the apparent surface charge of algal cells, often changed because of different growth phases (Henderson et al., 2008a) or water conditions (Zou et al., 2005), which caused the flocculation efficiency of chitosan modified soil variable. For example, when *Microcystis aeruginosa* (M.A.), the main species forming cyanobacteria blooms in Lake Taihu was firstly harvested from the culture medium by centrifugation and then re-dispersed into 0.5% NaCl solution, Zou et al. (2006) reported that 80% of the algal cells were removed by 1 mg/L chitosan modified 10 mg/L soil in 30 min. However, if directly flocculated in the culture medium, maximally 60% were achieved in 4 hr with the same dosage (Li and Pan, 2013). Further studies proved that after the pretreatment, the magnitude of ZP was significantly reduced from −67.9 to −30 mV, which greatly increased the flocculation potential of M.A. cells and hence achieved higher removal efficiency (Li and Pan, 2013).

Reducing the magnitude of negative ZP means charge neutralization and destabilization, which established the polymer flocculation mechanism (Hjorth and Jorgensen, 2012). Although the ZP as an influencing factor affecting the flocculation ability of chitosan has been proposed (Renault et al., 2009), little progress has been done to quantify the effects and study the mechanism on how it affected the flocculation efficiency. The use of ZP for monitoring and controlling the coagulation of algal cells using aluminium sulfate has been well researched and found to be of great benefit (Henderson et al., 2008b), it was reported that the optimum removal was measured when the ZP of algal cells was controlled between −8 and +2 mV. However, the main mechanism of chitosan to remove particles in water was the long polymer chain with netting and bridging function (Huang and Chen, 1996; Zou et al., 2006), which was significantly different from the aluminium sulfate functioned mainly as a charge neutralizer, the results and mechanism on how the ZP affect the removal efficiency of chitosan may be also different. Therefore, if the flocculation behavior of chitosan modified soil to cyanobacteria cells with different ZP can be cleared, it will give an insight for understanding the flocculation mechanism and provide a useful guidance for practical engineering of cyanobacteria bloom control.

Here, the M.A. cells, main species forming cyanobacteria blooms was selected in different growth phases and adjusted to possess different ZP by a positively charged protein, *Moringa oleifera* seed extract (MO). The relationship between ZP of algal cells and flocculation ability of chitosan was studied. The main objective of this research was to find how the ZP of particles affects the flocculation behavior of chitosan, including removal efficiency, sedimentation kinetics, floc structure and floc size growth. According to these results, an optimized ZP range for algal flocculation using chitosan modified soil was proposed.

1. Materials and methods

1.1. Algae culture

The M.A. cells were obtained from Freshwater Algae Culture Collection at the Institute of Hydrobiology, Chinese Academy of Sciences in Hubei province, China. The culture medium, BG11, was adjusted to pH =8.0 by adding either 0.1 mol/L HCl solution or 0.1 mol/L NaOH solution before autoclaving. The sterilized 500 mL glass flasks containing 300 mL aqueous M.A. medium were maintained at 25 ± 1°C under a cool white fluorescent light of 2000–3000 lx on a 12 hr light and 12 hr darkness regimen in the illuminating incubator (LRH-250-G, Guangdong Medical Apparatus Co. Ltd., Guangdong, China).

1.2. MO, chitosan, soil and modification process

MO were cationic proteins with a molecular mass of 6.5–13 kDa and isoelectric points in the range of pH 9.6–11 (Ghebremichael et al., 2005). It was chosen as the ZP adjuster for M.A. cells since as reported, it can significantly reduce ZP of particles (Ndabigengesere et al., 1995). MO seeds were purchased from Shaoguan city (South China) in dry form, having already been removed from the pod. The healthy seeds (about 1.0 cm) were selected and deshelled. The kernels were grounded in a coffee grinder to become particles of about 300 μm, stored at room temperature in an airtight container and used for one month (Katayon et al., 2006). To extract the active proteins, 5 g of the seed powder was suspended in 100 mL of 1.0 mol/L NaCl solution and the suspension was stirred using a magnetic stirrer for 30 min (Okuda et al., 2001). The solution was then filtered through a glass microfiber filter of 0.45 μm pore size (Whatman GF/C, UK) and the filtrate was used as the ZP adjuster.

Chitosan was purchased from Qingdao Yunzhou Bioengineering Co. Ltd., Shandong, China. The chitosan flakes were dissolved by adding 500 mg chitosan to 100 mL of 0.5% HAc and stirred until all the chitosan was dissolved. This solution was then diluted with deionized water to obtain a final concentration of 1 g/L before use. The MO and chitosan were prepared freshly for each experiment.

The soil was collected from lakeshore of Meiliang Bay, Lake Taihu, washed with deionized water, dried at 100°C for 10 hr, and then grounded and sieved through 180 mesh (<90 μm).

To modify the soil, a certain volume of chitosan solution (1 mg/L) was added to a clay suspension (10 mg/L). The mixture was well stirred and then ready for use in the flocculation experiment. As the surface properties have been changed after Al_{13}-modification reported by Zhao et al. (2012), the netting and bridging modifications using chitosan also changed the physico-chemical characteristics of soil and hence affected the flocculation behavior, more detailed information can be obtained from the previous publications (*i.e.*, Pan et al., 2006 and Zou et al., 2006).