

Performance of a granular activated carbon biologically active filter (GAC-BAF) for removing microcystin-LR (MC-LR) from eutrophic water

Wei Xu, Meijun Kong, Jie Song and Zhen Zhang

ABSTRACT

Microcystin-LR (MC-LR) removals linked with empty bed contact time (EBCT), temperature and backwash strategies are important for biologically active filter (BAF) application but remain not entirely clear. In addition, there is still a lack of understanding about the bio-regeneration of MC-LR adsorption capacity in a BAF. This study examined MC-LR removals by a granular activated carbon-biologically active filter (GAC-BAF) as a function of EBCT, temperature, and backwash strategies. Results demonstrated that the optimum EBCT (1.2 h for this study) likely depended upon an optimum superficial velocity as well as the GAC column height. A superficial velocity not greater than 0.5 m/h was deemed appropriate according to this research. The MC-LR removals achieved at the optimum EBCT reached 54.8%–72.0%. A higher temperature resulted in a greater MC-LR removal; adsorption might contribute to effective MC-LR removal by the GAC-BAF at a lower temperature. A strategy (Air + air-water + water) was suggested as an optimal backwash strategy for GAC-BAFs in terms of MC-LR removal. A comparison of the effectiveness of the experimental GAC-BAF in continuous and discontinuous operations on MC-LR removals was conducted, from which indirect evidence for bio-regeneration of MC-LR adsorption capacity of the GAC was obtained.

Key words | biologically active filters, drinking water, eutrophication, GAC, microcystin-LR

Wei Xu (corresponding author)
Meijun Kong
Jie Song
Zhen Zhang
School of Resources & Environment,
Anhui Agricultural University,
Hefei, 230036,
China
E-mail: xuwei981@sina.com;
xuwei981@ahau.edu.cn

HIGHLIGHTS

- GAC-BAF achieved MC-LR removals of 54.8% ~ 72.0%.
- Optimal EBCT and backwash strategy for MC-LR removal by GAC-BAFs were investigated.
- MC-LR removals by a continuous flow GAC-BAF were linked with temperature.
- Indirect evidence for bioregeneration of MC-LR adsorption capacity of GAC was found.

INTRODUCTION

Microcystin-LR (MC-LR), which is one of the most common toxins produced by cyanobacteria, has raised worldwide concern for its potential to severely compromise human health. The World Health Organization (WHO) has recommended a guideline value of 1 µg/L for MC-LR in drinking water (Park *et al.* 2017). Dissolved MC-LR is not readily removed by conventional water treatments (Huang *et al.* 2007; Chang *et al.* 2015; Pestana *et al.* 2015; Park *et al.*

2017; Jeong *et al.* 2017). Therefore, it has become of urgent practical significance to study advanced water treatment processes to improve MC-LR removal from drinking water. Biologically active filters (BAFs) have been recognized as an effective process in removing MC-LR (Ho *et al.* 2006, 2007, 2012; Li *et al.* 2012, 2015; Zhu *et al.* 2016). Also, its low cost and energy requirements make it extremely promising for drinking water treatment (Benner *et al.* 2013). Moreover,

no cytotoxic by-products have been found from the MC-LR biodegradation processes (Ho *et al.* 2007). Among all types of BAFs, GAC-BAFs have proved more effective for water purification in drinking water treatment generally involving both biodegradation and adsorptive processes (Seredyńska-Sobecka *et al.* 2006; Aktaş & Çeçen 2007; Abromaitis *et al.* 2016; Lompe *et al.* 2016).

Empty bed contact time (EBCT), temperature and backwash strategies are essential influencing factors for the effectiveness of BAFs (Liu *et al.* 2016; Zhang *et al.* 2017). Nevertheless, to date, only a few studies have focused on the effect of those factors on MC-LR degradation by BAFs. To the best of our knowledge, few studies have linked the MC-LR removal of BAFs with EBCT. Only Ho *et al.* (2006) have investigated the performance of biologically active sand filters on microcystins' (including MC-LR) removal at different EBCTs (7.5 min, 10 min, 15 min, 30 min). Results showed that high microcystin removal was achieved at each EBCT. Furthermore, this research found that the lowest EBCT of approximately 4 min still yielded no toxin breakthrough. One limitation of this research was that it employed sand with negligible absorptivity as media and therefore the results were unlikely to reflect the influence of EBCT on comprehensive MC-LR removal by both biodegradation and adsorption that may co-exist in the BAFs packed with media with adsorption capacity. Regarding temperature, Wang *et al.* (2007) assessed MC-LR biodegradation at 22 °C, 25 °C, and 33 °C using three reactors inoculated with the bacteria from an experimental bio-filter. The results showed that bio-degradation of MC-LR was correlated to temperature, with a higher rate of degradation observed at 25 °C and 33 °C. Li *et al.* (2015) investigated the seasonal variation in MC-LR biodegradability of biofilms on a practical biological treatment facility (BTF). They revealed that MC-LR biodegradability was correlated to temperature, MC-LR, and chlorophyll-a concentration. Peaks of MC-LR degrader abundance and MC-LR biodegradability co-occurred following observed peaks of water temperature (29.7 °C) and MC-LR concentration. The two studies (Li *et al.* 2015; Wang *et al.* 2007) provided extremely useful information about the response of MC-LR biodegradation to temperature; nevertheless, there were still limitations. Both studies were conducted with batch reactors, which are not expected to include the

specific MC-LR mass transfer mechanism co-existing with biodegradation in a continuous flow bio-filter. Moreover, few studies have evaluated MC-LR removal by BAFs as a function of temperature based on a BAF system including both biodegradation and adsorption. As for backwash strategies, though it can be an important influencing factor for BAFs, the effect of this factor on BAFs in MC-LR reduction has scarcely been reported in the literature.

Another issue addressed by the current study is that there is a lack of understanding regarding bio-regeneration of the MC-LR adsorption capacity of GAC. Several researchers have found that the bio-regeneration of MC-LR adsorption capacity of activated carbon might occur in the BAF (Aktaş & Çeçen 2007). However, direct evidence for that effect is difficult to obtain.

The main objectives of this study are to: (1) examine MC-LR removal by a GAC-BAF as a function of three critical factors – EBCT, temperature, and backwash strategy – to provide more information for determining optimal operational parameters for the application of BAFs in treating MC-LR; (2) discover if bio-regeneration of MC-LR adsorption capacity of the GAC could occur by comparing the performance of an experimental GAC-BAF in continuous and discontinuous operations on MC-LR removals.

MATERIALS AND METHODS

Experimental filters' design and operation

A bench-scale GAC-BAF made of acrylic sheet was employed to assess the effect of EBCT, temperature, and backwash strategies, as well as to investigate the bio-regeneration of MC-LR adsorption capacity of GAC (Figure 1). It was packed with granular activated carbon (GAC) (iodine number: 800–1,000 mg/g; diameter of 1.25–2.5 mm; bulk density of 0.32 g/cm³; Pingdingshan Xinzhiyuan Activated Carbon Co., Ltd, China) and sand (diameter of 1.2–2.0 mm; bulk density of 1.51 g/cm³). The total media depth was 1.0 m, with a depth ratio of GAC to sand of 3:2. The function of sand has been explained by our previous study (Xu *et al.* 2017); negligible MC-LR removals through the sand layer were observed in the current study (data are not shown in this paper).

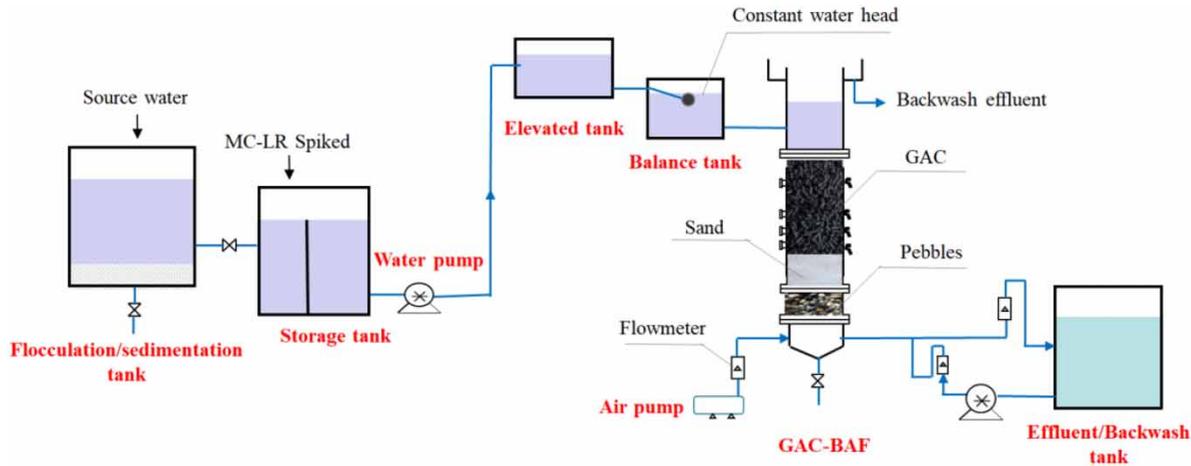


Figure 1 | Schematic diagram of the experimental system.

The filter column was down-flow and had an internal diameter of 40 mm and overall height of 2.0 m. Support was provided by 0.4 m of pebbles at the bottom of the filter. Four ports for media sampling were located at column depths of 5, 30, 45, and 60 cm.

As shown in Figure 1, source water, which was collected from a polluted lake located on the campus of Anhui Agricultural University, was pretreated with flocculation and sedimentation (FS) before flowing into the storage tank. To simulate eutrophic water, a specific volume of frozen MC-LR stock solution was thawed and subsequently spiked into the storage tank at an MC-LR concentration in the desired range (10–50 µg/L). Water from the storage tank (influent water of the experiment GAC column) was then sent into the filter column through a pump, an elevated tank, and a balance tank. A regular backwash was conducted every 3–4 days according to the methods of our previous study (Xu et al. 2017). Strategy 3 described in the section ‘Assessment of the effect of EBCT, temperature, and backwash strategies on MC-LR removals by the GAC-BAF’ (Table 1) was used in the experiments except in the test of impact of backwash strategies on MC-LR removals. The water for the backwash was from the backwash tank, with air for the backwash supplied by an air pump. All the water and air flow rates were adjusted using flow meters. A heating device, which was controlled by a temperature sensor, was used to increase the temperature of the water inside the filter column, when needed.

Table 1 | Operational conditions for the tests of influencing factors (EBCT, temperature, and backwash strategies)

Stages	EBCT (h)	Temperature (°C)	Backwash strategies	Running time
A	0.125 0.25 0.6 0.8 1.2 1.6 2 2.4	23–25	Strategy 3	7 days for each EBCT
B	1.2	< 10 15 25 30	Strategy 3	10 days for each temperature
C	1.2	19–21	Strategy 1 Strategy 2 Strategy 3	14 days for each backwash strategy (backwash for two times)
Strategy 1 (Water only) : ①Water backwash at 15 L/m ² ·s for 10 min with an expansion of 25%				
Strategy 2 (Air + water): ①Air backwash at 6 L/m ² ·s for 6 min with an expansion of 2% ②Water backwash at 8 L/m ² ·s for 10 min with an expansion of 15%				
Strategy 3 (Air + air-water + water): ①Air backwash at 9 L/m ² ·s for 3 min with and expansion of 5% ②Simultaneous air backwash 8 L/m ² ·s and water backwash at 6 L/m ² ·s for 5 min twice with a 1 min interval with an expansion (%) :10% ③Water backwash at 6 L/m ² ·s for 3 min with an expansion(%) :11%				

Conditions for FS treatment for source water were: flocculation for 25 min and sedimentation for 1.5 h, with polyaluminum chloride (30 mg/L) and polyacrylamide (0.2 mg/L) used as the flocculant. The MC-LR extraction was performed by ultrasonication (1,200 w output) of the cyanobacteria slurry (5 L for one time) collected from Chaohu Lake in China. Cell debris was removed by centrifugation at 10,000 r/min and filtration through a 0.45 µm membrane. The filtrate was stored frozen as MC-LR stock solution.

The pH of the influent water of the experiment GAC column was 6.5–7.8; concentrations of COD_{Mn}, ammonium, nitrite and MC-LR in the influent water were 5.32–13.2 mg/L, 0.96–2.65 mg/L, 0.14–3.12 mg/L, and 14.3–49.0 µg/L, respectively; influent dissolved oxygen of the filter columns during the experiments was 5.2–6.1 mg/L.

Biofilm cultivation

The biofilm immobilized on the surface of the media in the filter column was cultivated under continuous-flow operation of the column with the indigenous microorganisms in influent water as inocula. Superficial velocity, which equals flow rate divided by cross-sectional area of a BAF, is an important parameter for BAF. A low superficial velocity (0.2 m/h) was employed for the filter to decrease the shearing force of the water on the biofilm. At this stage, the water temperature was within 18–25 °C.

Assessment of the effect of EBCT, temperature, and backwash strategies on MC-LR removals by the GAC-BAF

To assess the effect of EBCT, temperature, and backwash strategies on MC-LR removals by a GAC-BAF, the experimental GAC-BAF was operated under the conditions listed in Table 1 after the biomass in it reached a steady state. The data from Stages A, B, and C were used to examine the impact of EBCT, temperature, and backwash strategies on MC-LR removal, as a sole variable, respectively.

Exploring if bio-regeneration of MC-LR adsorption capacity of the GAC could occur in a GAC-BAF

Bio-regeneration of MC-LR adsorption capacity of GAC is defined in this study as biodegradation-driven release of the sites (in the GAC) for MC-LR adsorption.

During a GAC-BAF's shut-down period, there are neither additional MC-LR nor any other compound transported into the BAF (Figure S1) and, consequently, a significant concentration gradient of MC-LR (and/or some other compounds) between the GAC surface and biofilm was likely created due to the degradation of MC-LR (and/or some other compounds) in biofilm, driving the transfer of those compounds from the GAC surface to the biofilm, which would lead to a release of adsorption sites (bio-regeneration of adsorption capacity). Accordingly, an idle period is expected to be favorable for the bio-regeneration of MC-LR adsorption capacity of the GAC. As a result, the GAC-BAF likely had a heightened efficiency in MC-LR removals when restarted after its idle time. Hence, a GAC-BAF operated discontinuously is likely to achieve greater MC-LR removal compared to that in continuous operation, in the case that bio-regeneration of MC-LR adsorption capacity of GAC could occur.

Based on the above hypothesis, this study attempted to obtain indirect evidence for the possible occurrence of bio-regeneration of MC-LR adsorption capacity of the GAC by comparing the MC-LR removals of the experimental GAC-BAF in continuous and discontinuous operations. Each operation lasted for 25 days. The experiment was conducted at EBCT of 1.2 h and temperature of 25 °C (±1 °C).

Analytical methods

The MC-LR was quantified by high performance liquid chromatography (HPLC) (Eleuterio & Batista 2010). The samples were concentrated using C18 solid-phase extraction cartridges (we followed the procedure used by Nicholson et al. (1994)) before being analyzed by the HPLC. The biomass was represented by the phosphatide amount in terms of nanomoles of phosphorus (nmol P) (Liu et al. 2001). TTC dehydrogenase activity (TTC-DHA) was analyzed using the method found in Niu et al. (2013), and 3 g media coated by the biofilm was sampled for detection each time.

RESULTS AND DISCUSSION

MC-LR removal in the GAC-BAF at the startup phase

The startup phase was defined as the period from the start of the GAC-BAF to the time when the biomass (in the GAC-BAF) reached a steady state. The biomass required 32 days

to reach its steady state (Figure 2(a)). The biomass from top to bottom at steady state was 93.5 nmolP/g_{media} to 9.5 nmolP/g_{media}. The GAC-BAF had achieved a high efficiency in removing MC-LR on the first day when it started, with a MC-LR removal greater than 70% (Figure 2(b)). Since very small amounts of biomass existed in the GAC-BAF (Figure 2(a)) at this time, the good MC-LR removal

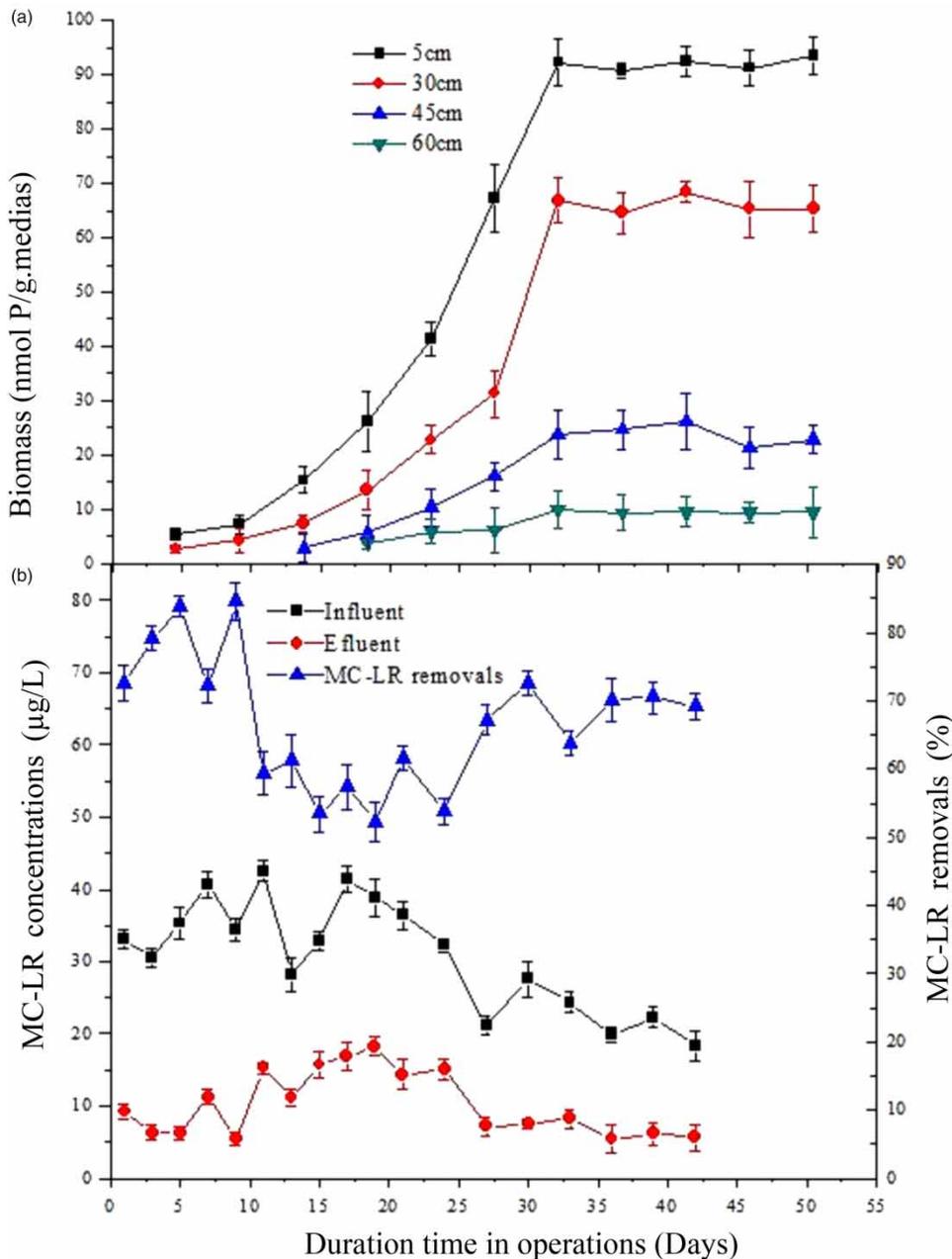


Figure 2 | Dynamic biomass of the biofilm in the filter column (nmol P/g) and MC-LR removals during the startup phase: (a) biomass of the biofilm in the filter column; (b) MC-LR removals.

effectiveness should be attributed to adsorption exerted by the GAC. A decrease in MC-LR removals was observed after 9 days. This result was interpreted as due to the deterioration of MC-LR adsorption, which might be caused by a reduction of the adsorption capacity and/or increased resistance to MC-LR transfer into the GAC by the biofilm. However, MC-LR removals began to pick up from day 27, possibly due to a substantial biodegradation that had existed from that day. Interestingly, the MC-LR removals tended to vary slightly right after 32 days, when the biomass reached steady state, further indicating that the recovery of MC-LR removals might be correlated with the improvement of biomass; biodegradation likely contributed to MC-LR removals in our experiments.

Effects of EBCT, temperature, and backwash strategies on MC-LR removals by a GAC-BAF

Figure 3(a) presents the average removals through the experimental GAC-BAF at each EBCT, from which we can see that EBCT significantly impacted the GAC-BAF on MC-LR removals. The MC-LR removals were improved from 16.8% to 64.5%, by increasing EBCT from 0.125 h to 2.4 h. However, EBCT could affect MC-LR removals slightly when greater than 1.2 h, probably because the MC-LR removal at that EBCT had been close to its limitation possibly determined by the maximum mass transfer rate corresponding to the influent MC-LR concentration. Accordingly, 1.2 h is suggested as an optimum EBCT under the operational conditions of this study. This value is far greater than the EBCT (7.5–30 min) adopted in the experiments conducted by Ho et al. (2006), where high efficiencies of microcystins' removal were achieved by use of a biologically active sand filter. Nevertheless, the corresponding superficial velocity of the optimum EBCT (Table S1) was within the range of the superficial velocity (0.3–1.2 m/h) employed by Ho et al. (2006). As EBCT equals column height divided by superficial velocity, the greater optimum EBCT suggested by this study was primarily attributable to the greater GAC column height (60 cm) compared to the media height (15 cm) employed by Ho et al. (2006). It could be further concluded that the optimum EBCT for a GAC-BAF is likely to be dependent upon an optimum superficial velocity as well as the depth of GAC column. More

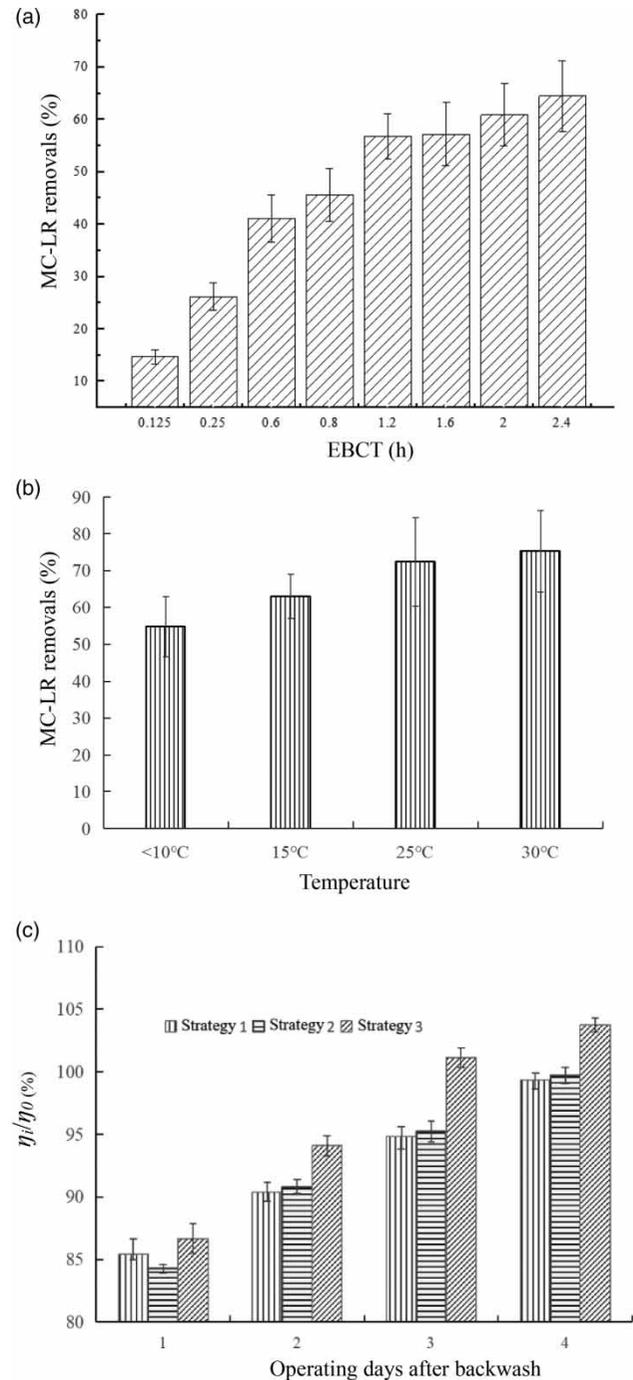


Figure 3 | (a) MC-LR removal rate as a function of the EBCT (influent MC-LR: 15.1–23.7 $\mu\text{g/L}$). (b) MC-LR removals through the GAC-BAF as a function of temperature (influent MC-LR: 14.3–21.5 $\mu\text{g/L}$). (c) Impact of backwash strategies on MC-LR removals (influent MC-LR: 15.6–23.9 $\mu\text{g/L}$).

studies are still needed to further test this conclusion. A superficial velocity not greater than 0.5 m/h was deemed appropriate for the experimental GAC-BAF according to

the results shown in Figure 3(a) and Table S1. However, high MC-LR removal (approximately 100%) was achieved at a superficial velocity of 1.2 m/h in the study of Ho *et al.* (2006). This difference in results may be due to the different packing and/or inocula employed by the two studies, as well as the possible difference of the compounds coexisting with MC-LR in water.

Average MC-LR removals at four temperatures (<10 °C, 15 °C, 25 °C, and 30 °C) are presented in Figure 3(b). Results show that increasing temperature could result in an improvement of MC-LR removal. The greatest MC-LR removal (72.0%) was achieved at 30 °C.

There are two possible mechanisms for MC-LR removal by a GAC-BAF, adsorption and biodegradation (Wang *et al.* 2007). Based on a thermodynamic analysis, Huang *et al.* (2007) concluded that the MC-LR adsorption on activated carbon is exothermic. Accordingly, a higher water temperature should not be favorable for MC-LR adsorption. Thus, the greater MC-LR removals at a higher water temperature might be due to the enhancement of MC-LR biodegradation with an increase in water temperature, from a possible stimulation of the activity of enzymes related to MC-LR biodegradation and/or acceleration of the proliferation of MC-LR degraders (Li *et al.* 2015). The performance of the GAC-BAF in MC-LR removal at low temperatures (<10 °C) was not as low as expected, as results showed an MC-LR removal of 54.8% at these temperatures (Figure 3(b)). Adsorption, which might still remain in the filter at the testing time, possibly played an important role in maintaining the effectiveness of the GAC-BAF in MC-LR removals at a lower temperature. This conjecture still needs to be further explored.

The impact of three prevalent backwash strategies (Table 1) on MC-LR removals through GAC-BAFs were calculated in terms of the ratio of η_i to η_0 (η_i/η_0), where η_i is the MC-LR removal i days after a backwash ($i = 1-4$); η_0 represents the MC-LR removal the day before a backwash. Average values of η_i/η_0 for each backwash strategy, which are presented by Figure 3(c), were estimated using data from two backwash cycles.

Backwash with each strategy could result in a decline of MC-LR removal, indicated by η_i/η_0 less than 100% at the first day after backwash (Figure 3(c)). Biomass loss caused by backwash was observed (Table S2) in this

study and is likely to be an important reason for the deterioration of MC-LR removals. An obvious increase in values of η_i/η_0 with the filtration time after backwash with each strategy is shown in Figure 3(c), implicating a recovery of MC-LR removal performance during the filtration period. MC-LR removals recovered at the greatest rate in the case that strategy 3 was employed and, therefore, strategy 3 seemed to be optimal for BAFs. Recovery rates of MC-LR removals after backwash were expected to be correlated to biomass loss and/or microbial activity increase (represented by TTC-DHA) resulting from the backwash in this study (Table S2). It is reasonable to hypothesize that TTC-DHA increase is favorable for MC-LR recovery, whereas biomass loss has an opposite effect. Strategy 3 achieved the greatest TTC-DHA increase ratio and a relatively low biomass just slightly greater than the lowest in the experiment (Table S2), which could explain why strategy 3 was more advantageous compared to the other two strategies.

Water shear to the biofilm is a possible mechanism for biomass loss during backwash in this experiment (Liu *et al.* 2016). Linking the biomass loss to water intensity employed in each strategy (Table S2 and Table 1), it is concluded that higher water backwash intensity seemed to create a greater biomass loss. Washing out aged and detached biofilm (hindering transfer of oxygen from aqueous phase to biofilm) from the bio-filter might contribute to TTC-DHA increase.

Indirect evidence for the possible occurrence of bio-regeneration of MC-LR adsorption capacity of the GAC

As stated in the section 'Analytical methods', this study explores whether a bio-regeneration of MC-LR adsorption capacity of GAC might occur in a BAF by comparing the performance of the experimental GAC-BAF on MC-LR removals in continuous and discontinuous operations.

As shown by Figure 4, an observable improvement in MC-LR reduction was observed when the experimental GAC-BAF was shifted from continuous operation to discontinuous operation, with the average MC-LR removal increasing from 59.3% to 67.5%. The current experiments were conducted after the biomass in the experimental filter reached steady state. Therefore, the difference between the MC-LR removals in the two different operations was not

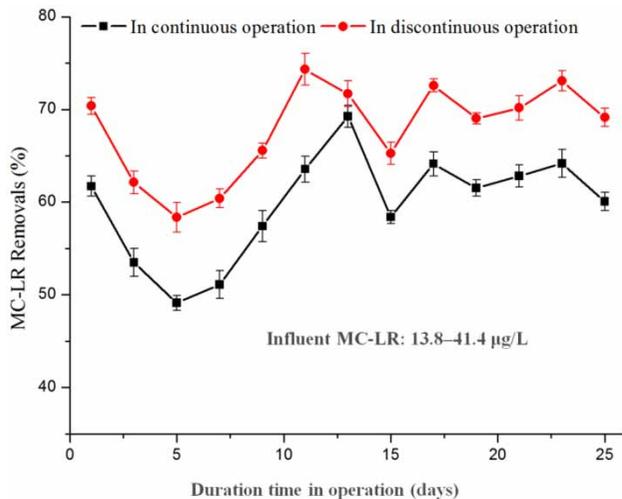


Figure 4 | MC-LR removals through the GAC-BAF in continuous and discontinuous operations. Influent MC-LR concentrations at a given duration time in two operations were approximately identical. The differences between the MC-LR removals at all points were significant with $P < 0.05$, except the data at the 13th day.

expected to be due to a variation of biomass. The only reasonable explanation for the observed results is that bio-regeneration of the MC-LR adsorption capacity of GAC might occur during the GAC-BAF's shut-down period, as illustrated in the section 'Exploring if bio-regeneration of MC-LR adsorption capacity of the GAC could occur in a GAC-BAF'. This result could be indirect evidence for the occurrence of bio-regeneration of MC-LR adsorption capacity of the GAC.

CONCLUSIONS

Conclusions were drawn as follows:

- (1) This study assessed the efficiency of a GAC-BAF in MC-LR removal as a function of EBCT, temperature, and backwash strategies. EBCT was shown to be a significant influencing parameter of GAC-BAF on MC-LR removal. The optimum EBCT was suggested as 1.2 h in this study, as influenced by an optimum superficial velocity of not more than 0.5 m/h, as well as the depth of GAC column (60 cm) employed here. MC-LR was removed effectively at all investigated temperatures, with removals of 54.8%–72.0% (at the optimum

EBCT). Increasing temperature could promote the elimination of MC-LR in GAC-BAF due to an improvement of MC-LR biodegradation under higher temperatures; adsorption might play an important role in maintaining the effectiveness of the GAC-BAF on MC-LR removal at a lower temperature. The strategy (Air + air-water + water) was found to be an optimal backwash strategy for GAC-BAF in terms of MC-LR removal.

- (2) The MC-LR removals of the GAC-BAF in continuous and discontinuous operations were compared. Results showed an observable improvement in MC-LR reduction when the GAC-BAF was shifted from continuous operation to discontinuous operation, providing indirect evidence for the possible occurrence of bio-regeneration of MC-LR adsorption capacity of the GAC.

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DATA AVAILABILITY STATEMENT

All relevant data are included in the paper or its Supplementary Information.

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