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DETECTION OF CYANOBACTERIAL TOXIN MICROCYSTINS IN TREATED DRINKING WATER: A PRELIMINARY STUDY IN SELANGOR, MALAYSIA

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ABSTRACT

This study aimed to quantify microcystins concentration in treated drinking water in Selangor, Malaysia. In June and July 2020, one hundred and twenty tap water samples were collected from eight districts. Samples were treated with sodium thiosulfate to neutralise chlorine before microcystins analysis using enzyme-linked immunosorbent assay (ELISA). The analyses revealed that microcystins were present at low concentrations in 22 percent (N=26) of the total collected samples. Microcystins were detected in 27 percent to 53 percent of samples collected from Hulu Langat, Sepang, Gombak, Petaling, and Kuala Langat districts. On average, microcystins concentrations were below the maximum acceptable limit of 1.0 ppb, and ranged from 0.10 to 0.68 ppb across the positive tested samples. The highest mean microcystins concentration of 0.368 ± 0.20 was detected in the Hulu Langat district. Overall, microcystins were detected from districts with their raw water sources derived from dams and retention ponds. These results suggest the presence of cyanobacterial toxin microcystins in treated drinking water supplies. Despite being detected at low prevalence and concentrations, microcystins in drinking water supplies could pose a potential health risk as toxin production at the cellular level can be highly variable on a temporal basis due to various environmental factors.

KEY WORDS : Microcystins, Drinking water, Dams, Cyanobacterial toxins, Health risk

INTRODUCTION

Cyanobacteria are photosynthetic prokaryotic organisms that are present as a natural part of phytoplankton assemblages in freshwater ecosystem. Cyanobacteria are also known as blue-green algae and are mainly composed of four main groups, including Chroococcales, Oscillatoriales, Nostocales, and Stigonematales (Bellinger *et al.*, 2015). Cyanobacteria may grow excessively under increased eutrophication and water temperatures (Deng *et al.*, 2019; Kitan *et al.*, 2020; Aeriyanie *et al.*, 2021a). It is well documented that cyanobacteria usually form thick and greenish surface bloom in freshwater ecosystems receiving high phosphorus and nitrogen inputs (Deng *et al.*, 2019; Aeriyanie *et al.*, 2021b). It is also widely reported that annual or even permanent cyanobacteria blooms are becoming common in lakes and dams used for drinking water supply (Zakaria A. Mohamed *et al.*, 2016; Sinden *et al.*, 2016; Piontek *et al.*, 2017; Recknagel *et al.*, 2017; Kim *et al.*, 2020).

High cyanobacterial biomass in water supply resources can cause unpleasant taste and odour to the drinking water (Watson *et al.*, 2016). The taste and odour are due to the release of cyanobacterial secondary metabolites such as 2-Methylisoborneol (Perkins *et al.*, 2019). Nevertheless, the major concern on the effects of cyanobacteria blooms is mainly on cyanobacterial toxins contamination (Golshan *et al.*, 2020). *Cyanobacteria genera*, including *Microcystis, Anabaena, Anabaenopsis,* and *Planktothrix*, can produce a range of toxins, which can have lethal and sub-lethal effects in both humans and animals. Commonly detected cyanobacterial toxins include hepatotoxins (e.g. microcystins), neurotoxins (e.g.

anatoxins), and skin irritants. Microcystins are widely found in surface water, and the primary route of human exposure is through drinking water consumption (WHO, 1998; Sukenik *et al.*, 2015; Reichwaldt *et al.*, 2016).

Microcystins can cause developmental effects and carcinogenicity on humans (Chorus *et al.*, 1999). To date, there is evidence which linked chronic consumption of microcystins-contaminated drinking water with the occurrence of liver and colorectal cancer (Hernández *et al.*, 2009; Greer *et al.*, 2018). Concerning public health protection, the World Health Organization (WHO) had set a provisional guideline value of 1.0 ppb microcystins in the treated drinking water (WHO, 1998). In Malaysia, the maximum acceptable limit for microcystins is also set as 1.0 ppb. However, microcystin is not listed as a parameter that needs to be monitored regularly in raw and treated drinking water (Department of Standards Malaysia, 2010).

To date, many studies have reported on microcystins contamination in domestic treated water supplies (Piontek *et al.*, 2012; Z. A. Mohamed *et al.*, 2015; Mokoena, 2016; Beversdorf *et al.*, 2018; Oliveira *et al.*, 2019; Tamele *et al.*, 2020). In the published studies, microcystins concentrations in drinking water supplies could present at elevated concentrations and higher than the WHO's established safe limit. In fact, various environmental factors can induce changes in microcystins production by cyanobacterial biomass. Therefore, the possible occurrences of microcystins in our drinking water supplies may have been neglected and pose a significant health risk. Thus, this study aims to determine the prevalence of cyanobacterial toxin microcystins and its concentrations in treated drinking water derived from selected raw water sources in Selangor state, Malaysia.

MATERIALS AND METHODS

Samplings were carried out between June and July 2020 at eight districts of Selangor state, Malaysia (Fig.1).

The sampled districts include Petaling, Sepang, Hulu Langat, Kuala Langat, Sabak Bernam, Kuala Selangor, Hulu Selangor, and Gombak. The respective water authority was notified before the commencement of water sampling. Throughout the sampling period, 120 samples were collected from water taps which are accessible for public use. For each sample, duplicated 30ml water was collected using acid-washed glass vials. Upon collection, samples were immediately treated with sodium thiosulfate at a final concentration of 100 mg/l to neutralise chlorine. Samples were stored at -20°C before microcystin analysis. Microcystins were quantified using Microcystins (ADDA)-DM ELISA kit (Abraxis, Product No. 522015). The samples' absorbances were measured at 450nm using a Spectramax microplate reader. The detection limit for this assay was set as 0.10 ppb following the kit's manufacturer. Microcystins concentrations were quantified based on five points linear standard

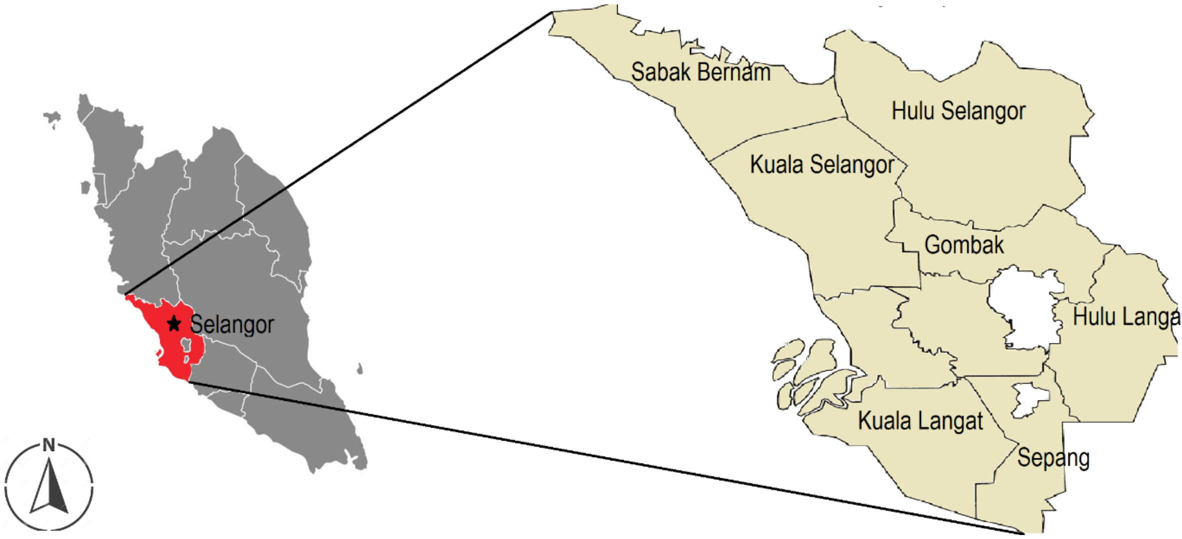


Fig. 1. Locations of eight districts in Selangor state, Malaysia

Fig. 2. Detected microcystins concentrations compared to the WHO guideline limit.

curve obtained from each assay. Data were analysed in Statistical Package for the Social Sciences (SPSS) and presented as descriptive statistics.

RESULTS AND DISCUSSION

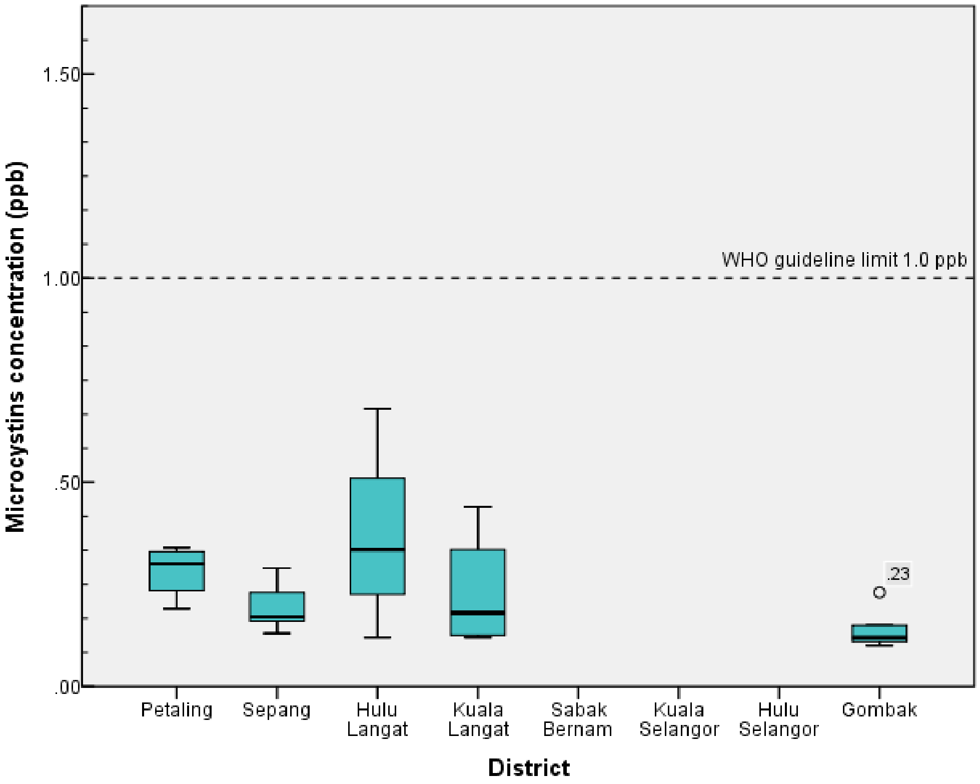
The microcystin enzyme-linked immunosorbent assays conducted on 120 tap water samples showed the presence of hepatotoxic microcystins in a small number of samples (Table 1). In total, microcystins were detected in 22 percent (N=26) of the tested samples. When analysed by the district, microcystins were detected in 53% of samples collected from Hulu Langat district. In Sepang and Gombak district, microcystins were detected in 33% of the samples. Meanwhile, microcystins were only detected in 27% of the samples collected from Petaling and Kuala Langat districts. Other than that, microcystins were not detected in any samples collected from Sabak Bernam, Kuala Selangor, and Hulu Selangor. Earlier studies have reported a wide range of microcystins prevalence in the treated water supplies. As published in WHO (2003), microcystins can be detected in up to 68% of the treated water. Lower prevalences of 17% and 30% microcystins detection in treated water were also reported in earlier studies (Haddix *et al.*, 2007; Szlag *et al.*, 2015; Addico *et al.*, 2017). In contrast, Szlag et al. (Szlag *et al.*, 2015) had reported that microcystins were not detectable in treated water despite the confirmed presence in the raw water. These scenarios can be possibly explained by UV and chemical compound potential to decompose extracellular microcystins during the water treatment process (Zhang *et al.*, 2017). Thus, microcystins concentrations in treated drinking water can be highly variable due to variation in exposure times to these factors.

In this study, microcystins were present at low concentrations in the positively tested samples (Table 1). On average, microcystins were detected at 0.257 ± 0.15 ppb across the tested samples. The highest mean microcystins concentration of 0.368 ± 0.20 ppb was detected in the Hulu Langat district. Lower mean microcystins concentrations of 0.283 ± 0.07 ppb and 0.230 ± 0.15 ppb were detected in Petaling and Kuala Langat districts, respectively. Meanwhile, microcystins concentrations of 0.196 ± 0.06 ppb and 0.142 ± 0.05 ppb were detected in Sepang and Gombak districts, respectively. The highest concentration of 0.68 ppb was detected in the Hulu Langat district. These concentrations were well below the recommended safe limit for drinking water of 1.0 ppb, as recommended by World Health Organization (WHO, 1998) (Fig.2).

Many studies have also reported microcystins at concentrations below 1.0 ppb in their treated drinking water samples (Ibrahim *et al.*, 2015; Addico *et al.*, 2017; Oliveira *et al.*, 2019). In contrast, microcystins contamination in treated water supplies could be in many folds higher than the WHO's established safe limit. Several examples of elevated microcystins concentrations were reported as 2.3 ppb (Mokoena, 2016), 3.6 ppb (Mohamed *et al.*, 2015), 7.8 ppb (Tamele *et al.*, 2020). Thus, it appears that microcystins may be persistent even after the conventional drinking water treatment processes. Chlorine is the most common oxidation agent applied in conventional drinking water treatment for disinfection purposes. Chlorination has been proven as the least effective for microcystins degradation (Sharma *et al.*, 2012). Meanwhile, effective microcystins removal can be achieved through advanced treatment processes such as nanofiltration and activated carbon (Ibrahim *et al.*, 2015). However, these advanced treatment processes are not applied in the local drinking water treatment process.

Table 1. Microcystins detection and concentrations in the tested samples

District	Number of samples	% samples with microcystins ≥ 0.1ppb	Microcystins concentration	
			Mean ± SD	Min -Max
Petaling	15	27	0.283±0.07	0.19-0.34
Sepang	15	33	0.196± 0.06	0.13-0.29
Hulu Langat	15	53	0.368±0.20	0.12-0.68
Kuala Langat	15	27	0.230±0.15	0.12-0.44
Sabak Bernam	15	0	-	
Kuala Selangor	15	0	-	
Hulu Selangor	15	0	-	
Gombak	15	33	0.142±0.05	0.10-0.23
Total	120	22	0.257±0.15	0.10-0.68



Water supplies in Selangor are mainly derived from dams and rivers (Jabatan Perancangan Bandar dan Desa Negeri Selangor, 2018). Raw water sources are commonly obtained from rivers which their flows are stabilised and regulated by a dam located upstream. In general, water supply sources in the Hulu Langat district are derived from Langat Dam and Semenyih Dam. Meanwhile, water supply sources in Kuala Langat, Petaling, and Sepang districts are originated from Semenyih Dam. Gombak district receives water sources from Batu Dam. Other than that, Kuala Selangor and Hulu Selangor districts obtain water sources from Selangor River Dam and Tinggi River Dam, with some areas also receiving water sources from rivers. Water sources in the Sabak Bernam district are mainly derived from the Bernam River. Other than dams and rivers, retention ponds also serve as alternative raw water sources for Petaling, Sepang, Hulu Langat, Kuala Langat, Kuala Selangor, and Hulu Selangor districts (Jabatan Perancangan Bandar dan Desa Negeri Selangor, 2018). In this study, microcystins are detected and quantified in areas receiving water sources from Langat Dam, Semenyih Dam, Batu Dam, and retention ponds. These findings are aligned with many earlier studies, which suggest that cyanobacterial toxins are commonly detected in water dams and reservoirs (Z. A. Mohamed *et al.*, 2015; Zakaria A. Mohamed *et al.*, 2016; Recknagel *et al.*, 2017; Golshan *et al.*, 2020; Kim *et al.*, 2020). In Malaysia, very few studies have focused only on cyanobacteria in the water supply reservoirs. For example, high cyanobacteria biomass with chlorophyll-*a* up to 50 ìg/L was reported in the Semberong Barat Dam, Johor state (Omar, 2016). On top of that, cyanobacteria were also detected in Air Itam Dam, Penang state (Rohaslinda, 2017). Even so, microcystins concentrations in the local water supply reservoir and its treated water distribution systems have never been reported in earlier studies. This issue has not been adequately addressed at this stage despite many lakes and reservoirs being eutrophic and prone to high cyanobacteria abundance. Therefore, this study is the first to report on microcystins presence in the local treated water supply, thus providing new research direction to highlight the significance of this issue.

CONCLUSION

This study suggests the need for a regular assessment of microcystins concentrations in the treated water supply. Our results revealed the presence of cyanobacterial toxin microcystins in the treated drinking water supplies in five out of eight sampled districts. Microcystins were detected at concentrations below the recommended safe limit of 1.0 ppb in the positively-tested samples. Even at low concentrations, these findings suggest that hepatotoxic microcystins could present in our drinking water. Therefore, microcystins could pose a public health risk as the concentrations are highly dynamic on temporal and spatial scales. Therefore, a more comprehensive study is needed to investigate the dynamics of microcystins at several points throughout the public water supply system.

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Conflict of Interest

The authors declare no conflict of interest.

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