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CURRENT EVIDENCE

Microcystin as a biogeochemical cycle: Pools, fluxes, and fates of the cyanotoxin in inland waters

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Scientific Significance Statement

There is a pressing need to understand the dynamics of microcystin, a toxin produced by some cyanobacteria, in the environment. Despite substantial advancements in our understanding of individual pools of microcystin, we lack a synthesized understanding of the sources, sinks, and movement of cyanotoxins within aquatic ecosystems. Using a literature synthesis approach, we developed a conceptual biogeochemical cycle of microcystin in lakes. We identified and synthesized the magnitude of four major pools of microcystin in lakes and reservoirs and nine major fluxes, including into the terrestrial environment (another major pool). Through this literature synthesis approach, we also identified understudied pools and fluxes. Adopting the framework of a "microcystin cycle" can provide new insights for the management and mitigation of microcystin exposure risks.

Abstract

Microcystin poses a serious threat to aquatic ecosystems and human health. There is a pressing need to understand the production, movement, and storage of microcystin in lakes. We constructed a conceptual biogeochemical model for microcystin through a comprehensive literature synthesis, identifying four major pools and nine major fluxes in lakes that also connect to the terrestrial environment. This conceptual model can be used as the framework for developing ecosystem mass balances of microcystin. We propose that the concentration of microcystin in the water column is the balance between the import, sediment translocation, production and degradation, uptake, burial, and export. However, substantial unknowns remain pertaining to the magnitude and movement of microcystin. Future investigations should focus on sediment fluxes, drivers of biodegradation, and seasonal dynamics. Adopting the framework of a "microcystin cycle" improves our understanding of processes driving toxin prevalence and helps to prioritize strategies for minimizing exposure risks.

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Additional Supporting Information may be found in the online version of this article.

The widespread eutrophication of inland waters combined with a changing climate is modifying the magnitude and severity of cyanobacteria blooms in some, but not all, waterbodies (Ho et al. 2019; Wilkinson et al. 2022). Cyanobacteria blooms can pose a serious threat to aquatic ecosystems and public health, particularly through the production of toxins that have the capacity to disrupt ecosystem services. Cyanotoxins create unsafe conditions for recreational water use and impede provisioning services such as fisheries, irrigation, and drinking water supplies (Carmichael and Boyer 2016). While there are numerous cyanotoxins, microcystin is among the most prevalent in inland waters (Rastogi et al. 2014). Microcystins are a group of monocyclic heptapeptides produced by numerous genera of cyanobacteria in both marine and freshwater ecosystems. Given the ubiquity of this toxin, persistence in the environment, and the potential for severe harm to humans and wildlife, there is a pressing need to understand the dynamics of when, where, and how microcystin is produced, transformed, moves, and accumulates.

As microcystin produced in the water column is a key reservoir and pathway for human exposure, a major research focus has been documenting the incidence and magnitude of microcystin concentrations in the water column (Loftin et al. 2016) and the environmental conditions that lead to microcystin production (Orihel et al. 2012; Harris et al. 2014). There has also been substantial effort to identify the organisms and processes that metabolize microcystin into less harmful molecules (Dziga et al. 2013; Schmidt et al. 2014; Massey and Yang 2020). In addition, there has been effort to understand the accumulation, transformation, and movement of microcystin in the aquatic food web (Kozlowsky-Suzuki et al. 2012; Flores et al. 2018), sediments (Zastepa et al. 2015; Wood et al. 2020), and the terrestrial environment (Ibelings and Chorus 2007). However, despite substantial advancements in our understanding of these individual pools of microcystin, we lack a synthesized understanding of the sources, sinks, and movement of cyanotoxins within aquatic ecosystems.

Our objective was to develop a biogeochemical model for microcystin from an ecosystem perspective that synthesizes production, movement, and storage in lakes. While our focus here is on lakes, the conceptual model we propose is adaptable to other aquatic ecosystems, including both marine and freshwater environments. Conceptual models of biogeochemical cycles provide a framework for examining the transport and transformation of molecules within and among ecosystems, including the interactions between abiotic and biotic components of the ecosystem. In addition to the more common elemental cycles, biogeochemical frameworks have recently been used to study contaminants such as plastic pollution (Hoellein and Rochman 2021), revealing important pathways for future research. We used this conceptual model to synthesize the current knowledge of the magnitudes of microcystin pools and fluxes in lakes, revealing gaps in our understanding of microcystin dynamics. By taking a comprehensive literature-review approach, we have been able to identify which pools and fluxes are well studied in lakes and which dynamics have received less attention, despite being potentially important pathways for human exposure. In addition, this conceptual framework can provide new insights for the management of microcystin exposure risks to humans and wildlife.

Constructing the microcystin cycle

To construct a comprehensive cycle of microcystin for inland waterbodies, specifically focusing on lakes, we reviewed and synthesized the current information on microcystin pools and fluxes in the literature. We performed a literature search in Web of Science using the terms "microcystin*" and "lake*", which returned 1781 articles. We supplemented this search with additional results by searching for "microcystin*" with "sediment*," "macrophyte*," "degradation*," and "aerosol*." Each article's abstract was reviewed to determine if the study contained information pertinent to our synthesis goals and had measurements from an inland waterbody. For studies that were deemed potentially pertinent based on the abstract, the main text was reviewed and if a pool or flux was measured, the estimate of the magnitude was extracted along with details about the ecosystem and methods (*see* Supporting Information Tables). In most studies, the magnitude was reported as a range of measured concentrations. We then synthesized this information to estimate the range of microcystin concentrations, the fluxes into and out of each pool, compare the magnitude and rates to biomass turnover times, and identify any gaps in our understanding of the processes that control microcystin dynamics within the pool. In total, we synthesized the quantitative results from 160 studies (*see* Supporting Information). While microcystin production and cycling also occurs in marine environments, we chose to limit the literature synthesis to lakes for this study (with some studies also reporting results for reservoirs). However, the conceptual microcystin cycle (pools and fluxes) that we constructed from this review is generally applicable across the aquatic continuum.

From our literature review, we identified four major pools of microcystin within lakes: *A. water column*, *B. sediment*, *C. macrophytes*, and *D. aquatic consumers* (letters correspond to major pools in Figs. 1, S1). Each of these major pools can be further divided into subpools based on the form (e.g., intercellular, extracellular in the water column and sediments) or trophic guild (e.g., zooplankton in aquatic consumers) that contribute to the dynamics in their major pools. These major pools are connected to each other and the *E. terrestrial environment* (another major pool with subpools) through nine major fluxes (numbered 1–9 in Fig. 1). These fluxes can be broadly categorized as *cellular* fluxes including lysis and

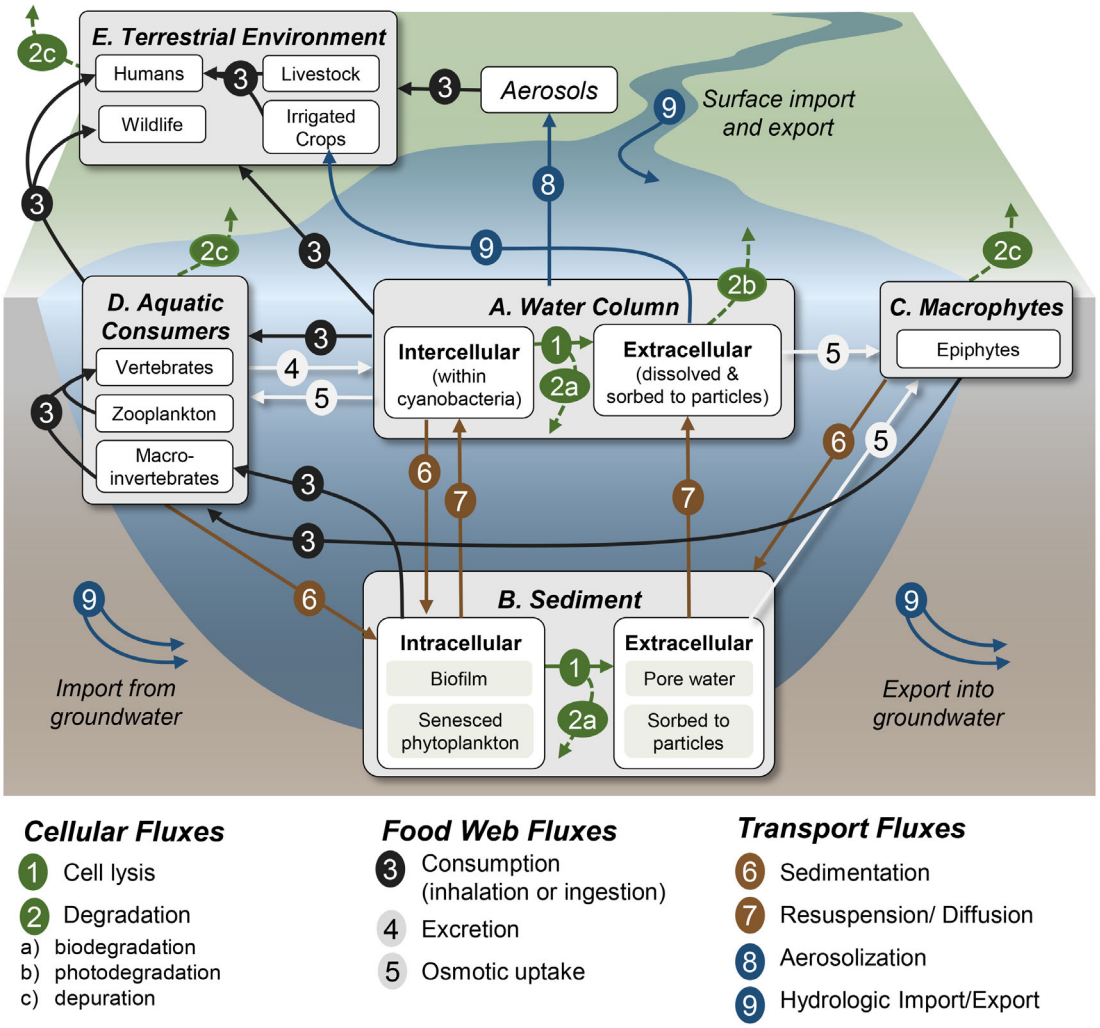

Fig. 1. A conceptual model of the microcystin cycle in lentic inland waters. The major pools (**A–E**) of microcystin and subpools (white boxes within major pools) are labeled in the diagram in light gray boxes. The major fluxes among these pools are denoted with the color-coded arrows. The numbers on the arrows correspond to the key of fluxes below the figure. The fluxes (arrows) are between the major pools and inclusive of all subpools unless arrows specifically connect two subpools (e.g., the diffusion flux [#7] between the extracellular sediment subpool and the extracellular water column subpool).

several forms of degradation, *food web* fluxes including consumption (direct ingestion or inhalation), excretion, and uptake (osmotic equilibration with biotic tissues), and *transport* fluxes including sedimentation, resuspension, aerosolization, and hydrologic import and export through surface and groundwater movement.

Based on this literature synthesis, we propose that concentrations of microcystin in the water column (the most frequently measured pool) are the balance between the import, translocation from the sediments, internal production of microcystin and the degradation, uptake, burial, and export of microcystin. In other words, water column concentrations are not reflective of intercellular microcystin production alone. Below, we describe each major pool including the subpools and fluxes that connect them and the environmental conditions that drive accumulation or loss from each pool.

A. Water column pool

Subpools in the water column

Microcystin is synthesized within the vegetative cells of cyanobacteria, forming the intercellular pool of microcystin. When microcystin-producing cyanobacteria are blooming (experiencing exponential population growth), the pool of intercellular microcystin in the water column can increase if toxigenic strains dominate the assemblage. When cells are lysed or damaged, intercellular microcystin is released into the extracellular microcystin pool. In its extracellular form, microcystin can adsorb to particles and organic matter or be subject to further degradation and loss from the ecosystem due to

ultraviolet radiation or bacterial metabolism (Munusamy et al. 2012; Massey and Yang 2020). High microcystin concentrations in lakes and reservoirs, reported as the intercellular, extracellular, or combined total concentrations, are associated with eutrophic conditions and low N : P ratios in the surface waters which favor cyanobacterial dominance (Orihel et al. 2012; Harris et al. 2014). In addition, warmer water temperatures and greater water column stability are conditions that favor cyanobacterial blooms leading to higher microcystin concentrations in the water column (Mantzouki et al. 2018). However, given the dynamic nature of blooms, the size of the microcystin pool in the water column is also dynamic.

Assessing the likelihood that a measurable pool of microcystin is present in the water column is challenging given the dynamic nature of cyanobacteria blooms and other fluxes (Fig. 1). Large, randomized surveys can provide a snapshot of microcystin pools among hundreds, or even thousands of lakes (Loftin et al. 2016), whereas longitudinal studies on a smaller number of waterbodies are more likely to capture brief episodes of toxin production. To quantify the incidence of a measurable microcystin pool in the water column of lakes and reservoirs, we compiled studies that reported surveying at least five waterbodies for microcystin concentrations. Surveys reported either intercellular, dissolved, or both concentrations combined for the water column. We used the information reported in these papers to calculate the percentage of waterbodies with detectable microcystin pools in the water column for each survey. In total, we reviewed 67 studies that reported on 69 surveys (Table S1). We did not discriminate among survey designs (e.g., statistically randomized, longitudinal, opportunistic); however, if microcystin was detected during any point in a repeated sampling design, the waterbody was considered to have detectable microcystin concentrations. We also categorized the frequency of sampling as reported in 62 of the survey as repeated (> 2 sampling events in each waterbody) or snapshot (1–2 sampling events only in a waterbody). Ten of the studies did not provide enough information to determine which waterbodies had detectable microcystin, only the fraction of samples that had measurable concentrations. For these 10 studies we calculated the percent of samples with detectable concentrations. The collated data is available from Wilkinson and Shingai (2022).

Among all the surveys, the presence of a microcystin pool in the water column ranged from 7.3% to 100% of waterbodies or samples, with a median of 78% and mode of 100% (Fig. 2A). This tallying exercise illustrates the ubiquity of microcystin in the water column of lakes and reservoirs. There was a significant negative correlation between the number of waterbodies or samples in a survey and the detection rate of microcystin (Fig. 2B; F value = 25.5, $p < 0.001$, $R^2 = 0.28$). We hypothesized that this relationship was likely the result of survey design: surveys of many lakes are more likely to be spatially randomized with a single sampling event whereas surveys with a smaller number of lakes are more likely to be longitudinal with repeated sampling events on the same waterbodies. Based on the sampling designs reported, studies with repeated sampling designs had a significantly higher percent detection of microcystin in the water column than studies with a "snapshot" design (Fig. 2C; one-way ANOVA $F_{1,60} = 5.365$, $p = 0.024$). In addition, snapshot surveys had a significantly higher number of waterbodies sampled compared to repeat sampling surveys (one-way ANOVA, $F_{1,60} = 6.03$, $p = 0.017$, number of waterbodies in each survey log-transformed). These findings support the hypothesis that the likelihood of microcystin being present in a waterbody sampled at one single point in time is lower than the likelihood of microcystin being present at any point over time in a waterbody sampled repeatedly.

Water column fluxes and fates

The pool of microcystin in the water column has many potential fates (Fig. 1). Two of the major fates for both intercellular and extracellular microcystin are degradation and transport into and out of the ecosystem through surface and groundwater flows, withdrawals for human use, and aerosolization. In addition, microcystin can be lost or gained from the water column pool with connections to the sediment, macrophyte, and aquatic consumer pools, detailed in sections below.

Degradation fluxes

Microcystin is removed from aquatic ecosystems through both photo- and biodegradation. Photodegradation rates are highest at ultraviolet wavelengths (Thirumavalavan et al. 2012) and lead to the rapid and efficient loss of extracellular microcystin from surface waters (Wörmer et al. 2010) (Table S2). This may be a particularly important mechanism in large shallow lakes with a high ratio of surface area to volume. The presence of humic substances may shield microcystin from photodegradation or act as a photosensitizer increasing degradation (Welker and Steinberg 2000). Biodegradation, performed by bacteria and fungi using hydrolytic enzymes to cleave the cyclic structure is another process that leads to substantial loss of microcystin from aquatic ecosystems (Dziga et al. 2013; Schmidt et al. 2014). Microbes that degrade cyanotoxins reside in both the water column and sediments and can even coexist with cyanobacteria cells themselves (Dziga et al. 2013). Among ecosystems, the abundance of microcystin-degrading microbes is tightly coupled to microcystin availability, highlighting the important relationship between these two bacterial communities (Lezcano et al. 2018).

Based on rates reported in the literature, the average half-life of microcystin in the environment ranges from 0.5 to 22 d (Table S2). Many studies report a lag phase between the introduction of microcystin in the environment and peak

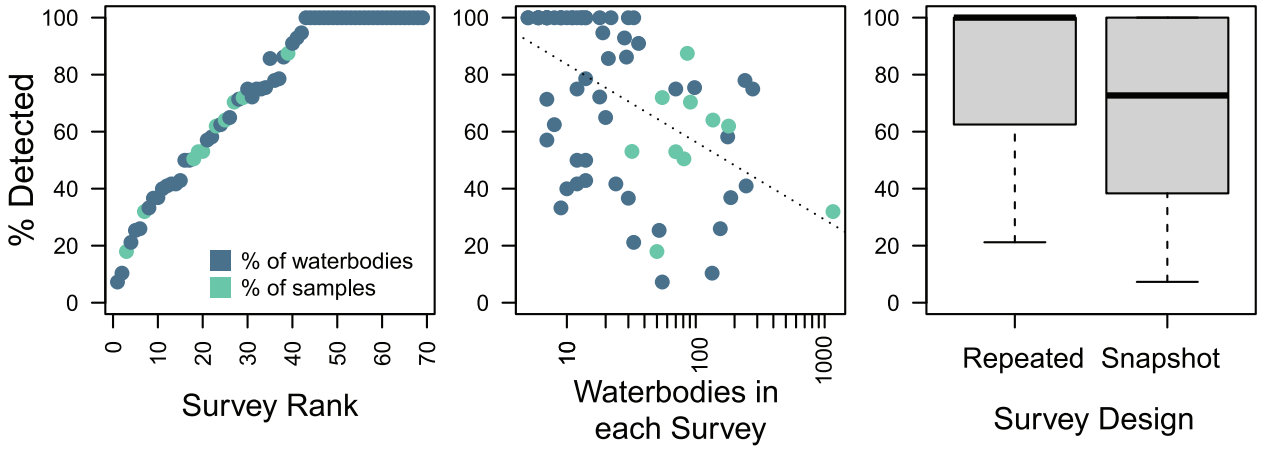

Fig. 2. The percent of waterbodies (blue) or water samples (teal) with (**A**) detectable microcystin from 69 surveys ranked from lowest to highest percent detected among surveys, and (**B**) the relationship between the number of waterbodies in each survey and the detection rate of microcystin in the water column (% detected = 111.06 − 27.4 × waterbodies in survey, $p < 0.001$, $R^2 = 0.27$), and (**C**) boxplots of the detection rate for repeated sampling surveys and snapshot (1–2 sampling events maximum) survey designs. There is a significantly higher detection rate among the population of repeated sampling surveys compared to the population of snapshot surveys reviewed in this study.

degradation rates (Lezcano et al. 2018). Variation in the conditions that favor higher rates of biodegradation such as warm temperatures, high pH, nutrient availability, and an oxic environment also contribute to the variation in rates among ecosystems and over time (Chen et al. 2010; Dziga et al. 2019). However, much of the information on microcystin biodegradation rates comes from studies performed in a water treatment setting. While advances have been made in isolating and identifying microcystin-degrading bacteria in waterbodies, additional data are needed to understand the seasonal dynamics and rates of biodegradation in aquatic ecosystems to adequately model the magnitude of this important flux at an ecosystem scale.

Transport fluxes

In addition to endogenous production of microcystin, toxins produced outside of the ecosystem can be imported from upstream and exported from the ecosystem through hydrologic flows and human transport of water. The relative importance of surface hydrologic connections on microcystin import and export fluxes is likely higher in river networks with reservoirs (Graham et al. 2012; Ge et al. 2021) and marine coastal habitats connected to inland waters (Miller et al. 2010; Umehara et al. 2019). Microcystin can also be exported from a waterbody into the surrounding groundwater (Yang et al. 2016; Zhang et al. 2021) and be produced by cyanobacteria active in the vadose zone; however, it is unclear how sediment sorption dynamics might influence this flux (*see* below). Further research is needed to quantify the magnitude and seasonality of hydrologically driven import and export fluxes of microcystin from waterbodies. Human water export for drinking, irrigation, and transport (e.g., ballast water) can also alter the size of the water column microcystin pool.

Besides hydrologic and human transport, microcystin also leaves waterbodies and enters the atmosphere through the formation of spray aerosols. Wave action, mainly driven by wind, entrains air into the water resulting in the formation of bubbles that eject cyanobacteria cells and extracellular microcystin into the atmosphere upon bursting (Plaas and Paerl 2021). There is evidence that droplets are enriched in hydrophobic congeners of microcystin relative to the bulk concentration in the water (Olson et al. 2020). These droplets, commonly formed by wave action along the shoreline, can be inhaled by terrestrial organisms, including humans. The concentration of microcystin in spray aerosols from lakes ranges from 0.0018 to 50 ng m^{-3} (Table S3), based on the few measurements reported in the literature. Ultraviolet radiation and ozone can quickly degrade microcystin contained in aerosols (Jang et al. 2020). The residence time of microcystin-laden aerosols in the atmosphere, the distance traveled by aerosols, and the dynamic nature of cyanobacteria bloom and aerosol formation all influence the magnitude of this flux yet are largely unresolved for freshwater ecosystems.

B. Sediment pool

Subpools in sediment

The bulk sediment pool of microcystin varies by orders of magnitude, from undetectable to 3 $\mu g\ g^{-1}$ dry weight (d.w.) among lakes and over time (Fig. 3; Table S4). The microcystin in the bulk sediment pool can be divided into microcystin bound in cells—either in biofilms, senesced, or dormant cells and colonies—dissolved in the pore water, and sorbed to sediment particles. The formation and persistence of microcystin-producing biofilms varies, but light-rich, shallow waters favor the development of

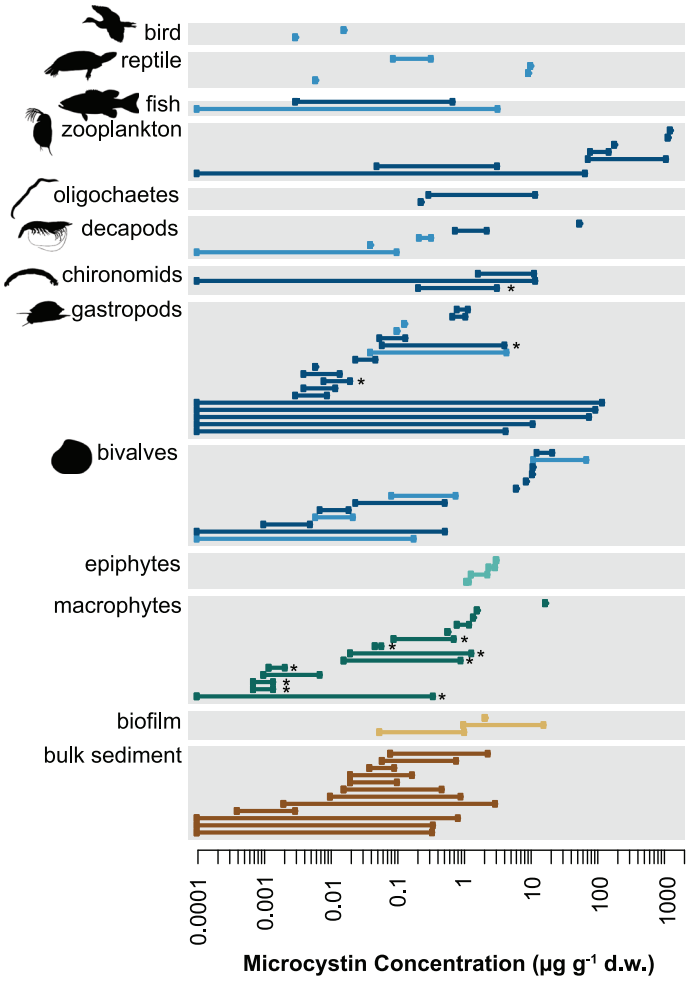

Fig. 3. The concentration of microcystin in various pools in comparable units (μg g^{-1} dry weight, d.w.; note the asterisk indicating the few measurements in μg g^{-1} wet weight, w.w.). each line or point is a single study of concentration, with the line spanning the range of values reported in the study. For animals, light blue lines are microcystin concentrations in muscle tissue (common tissue for human consumption) and dark blue lines are concentrations in the whole body (consumption-based exposure through predation). Concentrations in other tissues (e.g., liver, hepatopancreas) are listed in Tables S6, S7.

cyanobacterial mats. The intercellular concentration of microcystin in biofilms ranges from 0.06 to 16 μg g^{-1} d.w. (Fig. 3; Table S4).

In the dissolved form, microcystin can be found in the pore water between sediment particles. Microcystin can also adsorb to sediment particles, although there is a large range in maximum sorption capacity from 0.004 to 11.9 μg g^{-1} d.w. (Table S4) with some of the variation in sorption attributable to variation in congeners (Maghsoudi et al. 2015) and pH (de Maagd et al. 1999). In general, fine particles such as clay and sediments with high organic matter content have higher sorption capacity for microcystin (Munusamy et al. 2012).

Sediment fluxes and fates

As evidenced by the numerous subpools of microcystin in the sediments and the fluxes into, among, and out of these subpools (Fig. 1), the sediments are an important component of the microcystin cycle. However, there have been few ecosystem-level investigations of sediment microcystin fluxes (Song et al. 2015), limiting our understanding of the role of this pool in ecosystem dynamics and human exposure risk, overall. One of the main fates of microcystin in the sediments is biodegradation. Biodegradation rates in the sediments are generally higher than the water column, with rates as high as 35 times faster in the sediments of some eutrophic ecosystems compared to the water column (Li et al. 2016).

The sedimentation of microcystin-containing cells and colonies contributes to the biofilm pool. Through resuspension and migration, approximately 0.8–3% of colonies reinvade the water column (Feng et al. 2019), moving microcystin from the sediment pool into the intercellular water column pool. The rate of microcystin resuspension and residence time in the water column is not well quantified but could be a cryptic pathway of human exposure when water column production is otherwise low. Intracellular microcystin in the sediment pool is susceptible to movement into the extracellular pool through cell lysis or consumption and subsequent excretion by aquatic organisms (*see* "D. Aquatic Consumer Pool" section below). This dissolved pool in the sediments is subject to either diffusion back into the overlying water column, adsorption to sediment particles, or degradation by bacteria. In a rare comparison of rates within an ecosystem, Zastepa et al. (2017) found that the rate of microcystin diffusion from the sediments, at 1.38 ± 0.04 μg m^{-2} d^{-1}, was substantially higher than the burial rate, 0.13 ± 0.18 μg m^{-2} d^{-1}, in Lake of the Woods (North America), indicating that the sediments were a potential source of microcystin to the water column (Table S4).

C. Macrophyte pool

Subpools in macrophytes

Macrophytes accumulate extracellular, dissolved microcystin into their roots, stems, leaves, flowers, seeds, and bulbs (Romero-Oliva et al. 2014) with concentrations up to 16.9 μg g^{-1} d.w. in some instances (Fig. 3; Table S5). The allocation of microcystin among tissues within aquatic plants varies by species; however, the highest concentrations of microcystin are typically found in the roots and likely taken up from the sediment pool (Song et al. 2009). In addition to microcystin found within their tissues, macrophytes also provide the structural support for epiphytic cyanobacteria growth. While there is limited information regarding microcystin production by epiphytic cyanobacteria, reported concentrations vary from 1.16 to 3.12 μg g^{-1} d.w. of epiphyte biomass (Fig. 3; Table S5).

Macrophyte fluxes and fates

The rate of microcystin uptake by macrophytes spans orders of magnitude (1.9–544 $\mu g\ L^{-1}\ d^{-1}$; Table S5), with much of the variability attributable to time since exposure, variation among species, and variation in uptake rates of microcystin congeners (Romero-Oliva et al. 2015). While the capacity for macrophytes to accumulate microcystin make them a potentially large sink of cyanotoxin in the environment, there is evidence that microcystin exposure can inhibit macrophyte growth by inducing physiological stress (Ujvárosi et al. 2019), potentially altering the strength of this sink. Once incorporated into the tissues of macrophytes, microcystin is removed from this pool through biotransformation and degradation (Table S5) (Pflugmacher 2004; Romero-Oliva et al. 2015), consumption of macrophyte tissues by organisms, release during plant decomposition, or incorporation into the sediment pool upon senescence.

D. Aquatic consumer pool

Subpools of aquatic consumers

Aquatic consumer subpools include zooplankton, macroinvertebrates including shellfish and emergent insects, and vertebrates including fish that span trophic levels (Fig. 3). Microcystin is incorporated into the tissues of aquatic organisms, particularly primary consumers (Papadimitriou et al. 2012), either through direct consumption of intercellular toxins, osmotic uptake of extracellular toxins, or consumption of lower trophic levels that have microcystin in their tissues. While direct toxicity to aquatic consumers is not usually widespread at lower microcystin concentrations, sublethal effects such as disruption of reproductive development (Zhang et al. 2019), increased sensitivity of juveniles (Gérard et al. 2005), and genotoxicity (Juhel et al. 2007) can all have population-level effects that influence ecosystem processes (Gérard et al. 2009).

Zooplankton are a key link in the aquatic food web between intercellular microcystin in the water column and higher trophic levels (Rohrlack et al. 1999). Primary consumers such as zooplankton mainly accumulate microcystin through direct consumption of cyanobacteria cells. Cladocera such as *Daphnia* graze on phytoplankton in the water column and ingest intracellular microcystin through filter feeding. Whole-body concentrations of microcystin are up to an order of magnitude higher in *Daphnia* compared to other aquatic invertebrates (Fig. 3; Table S6) and can have adverse and sometimes lethal consequences for *Daphnia* (Rohrlack et al. 1999). Alternatively, microcystin exposure is hypothesized to provide medicinal protection against some parasites *Daphnia* (Sánchez et al. 2019).

In macroinvertebrates microcystin is introduced to tissues through consumption of cells and osmotic uptake of extracellular toxin through trans-tegument diffusion, oral water uptake, and gill or pulmonary breathing. While microcystin is detectable within whole-body tissues (Fig. 3), the highest concentrations in macroinvertebrates are usually found in the hepatopancreas and intestines (Table S6). Species that feed by ingesting sediment accumulate larger amounts of extracellular microcystin that is sorbed to sediment particles (Lance et al. 2010). Nonselective feeders may have higher susceptibility to microcystin accumulation; however, some macroinvertebrates have developed means for expelling, instead of ingesting, toxins (Juhel et al. 2006). Environmental and dietary exposure over longer periods of time can increase microcystin accumulation, however juveniles can have lower rates of accumulation, in part due to their less developed immune systems (Gérard et al. 2005).

Fish accumulate microcystin through epithelial uptake, direct consumption of phytoplankton, and bioaccumulation from prey (Zhang et al. 2009*a*; Flores et al. 2018). While studies of microcystin accumulation in fish most commonly evaluate concentrations in liver and muscle tissues (Fig. 3), a recent meta-analysis of fish tissues revealed that the toxin is also found in the blood, heart, reproductive organs, gut, gills, and skin of fishes (Flores et al. 2018) (Table S7). The highest reported concentration of microcystin contained within fish tissues was 375.3 $\mu g\ g^{-1}$ d.w. in the liver of planktivorous smelt (Flores et al. 2018). Microcystin accumulation in fish varies by species and location but is positively correlated with microcystin concentrations in the surrounding water column (Poste et al. 2011; Flores et al. 2018). Feeding strategy also influences microcystin accumulation with higher concentrations in omnivorous fish compared to planktivorous and piscivorous fishes.

Aquatic consumer fluxes and fates

Microcystin bioaccumulation is consistently observed in zooplankton, planktivorous fishes, and bivalves (Kozlowsky-Suzuki et al. 2012; Gibble et al. 2016). The rate of microcystin accumulation in aquatic consumers depends on animal behavior and environmental factors. For example, in zooplankton accumulation rates vary depending on environmental effects on filter feeding rates (e.g., temperature) as well as population-level adaptations to cyanotoxins such as avoidance of ingesting intercellular microcystin following exposure to the toxin (Tillmanns et al. 2011; Wojtal-Frankiewicz et al. 2013). Population-level effects of microcystin accumulation on fitness are also possible if the toxin accumulates in the reproductive tissues with the potential to be passed on to future (Zhang et al. 2007).

Predation is another flux of microcystin between aquatic consumers that leads to bioaccumulation, however, there is limited evidence that microcystin biomagnifies in the food chain (Kozlowsky-Suzuki et al. 2012; Papadimitriou et al. 2012). In some food chains there is evidence of microcystin biodilution (decreasing concentration with increasing trophic level), but this does not appear to be common. The potential flux of microcystin from prey to predator is dependent on the tissues consumed. Toxins are often concentrated in digestive organs such as the stomach, intestines, liver, or

hepatopancreas (*see* Tables S6, S7). As such, consumption of the whole organism will likely result in higher microcystin exposure than consumption of muscle tissue alone (e.g., humans consuming fish fillets).

Once consumed or absorbed and incorporated into organismal tissues, microcystin is excreted, egested, or undergoes biotransformation (i.e., depuration), resulting in detoxification (Schmidt et al. 2014). Biotransformation resulting in detoxification has been documented in *Daphnia* tissues (Wojtal-Frankiewicz et al. 2013), in the digestive glands of macroinvertebrates (Schmidt et al. 2014), and in fish (Flores et al. 2018). Excretion is another flux from aquatic consumers to other pools including the water column and sediment. While microcystin excretion has been documented in fish and other organisms, the sedimentation of fecal pellets and concentration of toxins is not well quantified.

E. Terrestrial environment

Subpools in the terrestrial environment

Microcystin has been found in the tissues of many terrestrial animals, with the highest concentrations in aquatic-associated wildlife including waterfowl, turtles, and reptiles (Fig. 3; Table S7). Microcystin can also accumulate in crops via contaminated irrigation water. An assay experiment to investigate bioaccumulation of microcystin in lettuce (*Lactuca sativa* L.) revealed that toxin the accumulated in the foliar tissues of the plants regardless of the concentration in the irrigation water (Romero-Oliva et al. 2014). There is also evidence that microcystin can accumulate in soils (Zhang et al. 2021). In livestock, microcystin can accumulate through watering from a microcystin-contaminated source and potentially be passed to other terrestrial consumers.

Terrestrial fluxes and fates

Microcystin found in terrestrial environments mainly originates from the water column and aquatic consumer pools. Originating from the water column, microcystin aerosol concentrations vary widely with bloom and wind conditions, but this is generally a diffuse flux. Microcystin-laden water withdrawn for drinking or irrigation purposes introduces the toxin to terrestrial consumers (i.e., wildlife, livestock, humans) and soils (Zhang et al. 2021). Animal movement between the aquatic and terrestrial environment (e.g., insect emergence and human recreation) as well as predation are another flux between these spheres (Moy et al. 2016). Of particular concern are the fluxes of microcystin that create cryptic pathways of human exposure.

Humans are exposed to microcystin through many pathways, including oral ingestion during recreation or from drinking water, consuming contaminated foods and supplements, dermal contact, and inhalation of aerosols (Carmichael and Boyer 2016). However, the predominant pathway of microcystin exposure to humans is ingestion of contaminated drinking water or ingestion during recreation (Giannuzzi et al. 2011). Communities that rely on untreated drinking water from lakes, reservoirs, and groundwater wells with microcystin concentrations that exceed recommended thresholds for ingestion are particularly vulnerable (Zhang et al. 2009*b*; Ruibal-Conti et al. 2019). In addition, when microcystin makes its way into domestic water supplies, hygienic activities such as bathing and hand washing become a pathway of exposure through respirable water particles (Benson et al. 2005).

Microcystin in animal tissues that humans consume is another cryptic pathway of exposure. In general, tissue concentrations in fish and shellfish are high when the surrounding water column concentrations are high (Ibelings and Chorus 2007; Poste et al. 2011; Flores et al. 2018). When microcystin concentrations are high in the water column, consumption of whole animals such as bivalves can result in 8–23.5 times the tolerable daily load for humans as defined by the World Health Organization (Chen and Xie 2005). Preparation method can also affect exposure risk, as boiling animal muscle tissue (e.g., fish fillets) has been shown to release microcystin otherwise bound to phosphate (Berry et al. 2011).

Comparing magnitudes of pools and fluxes

Quantitative comparison

Through the literature synthesis we identified studies of pools and fluxes from a diversity of inland waters employing a variety of measurement methods and reported units. While it is not feasible to construct a full quantitative cycle using the literature synthesis, we can further synthesize the reported values to compare the magnitude of pools and fluxes and identify poorly parameterized processes. To do this, we used a subset of studies that reported pools and fluxes in consistent and comparable units. For each of these studies, we extracted or calculated mean and standard deviations for pool and flux values reported. For studies without a reported variance, we estimated the standard deviation using the linear relationship between mean and standard deviation (SD) for all pools and fluxes ($SD = -1.35 \times 0.99 \times \text{mean}$; $F_{1,61} = 327.9$, $p < 0.001$, $R^2 = 0.84$). The population of means and standard deviations collected from the literature for a given pool or flux were used in a random effects model with a restricted maximum likelihood estimator method to estimate the mean pool or flux magnitude and 95% confidence intervals (CI) (Fig. 4). This analysis was performed using the *metafor* package in R version 4.1.3 (Viechtbauer 2010).

In general, there were few studies with comparable units for estimating the magnitude of pools and fluxes and high variance within and among studies (Fig. 4). Only 6 of the 14 mean estimates had CIs that did not include zero. Despite this high degree of uncertainty and variation in measurement methods, some patterns did emerge that can be cautiously interpreted. The mean concentration of microcystin in sediments (0.13 $\mu g\ g^{-1}$ d.w., [−0.01, 0.27 CI]) was an order of magnitude lower than the mean sediment sorption capacity

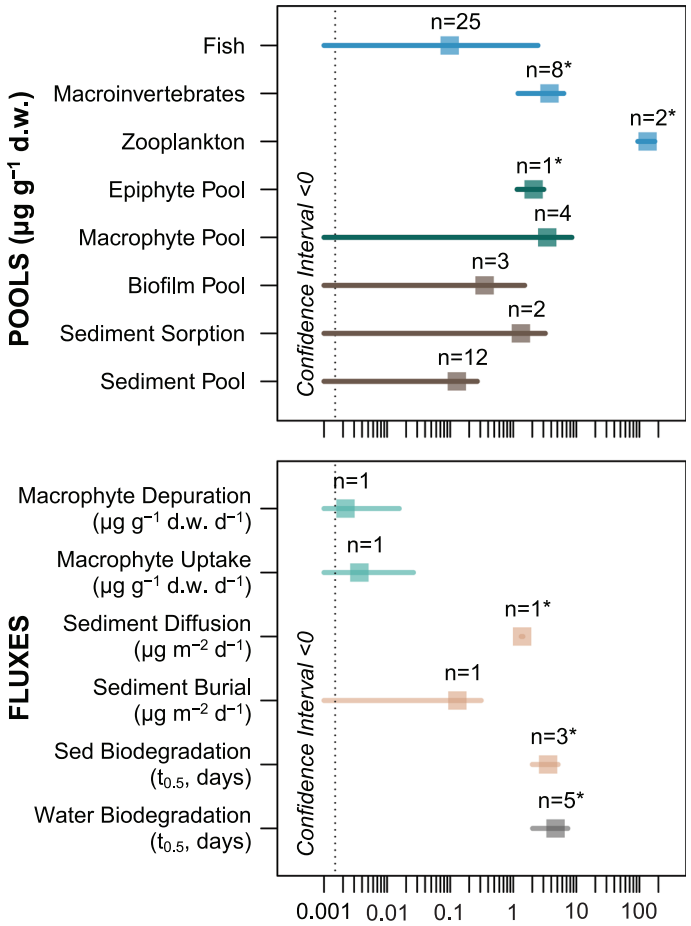

Fig. 4. The estimate of the mean magnitude (square) and 95% confidence interval (line) of various pools (top panel) and fluxes (bottom panel) in a random effects model using a subset of studies from the literature with comparable units of measurement. A log scale was used for visualization, and therefore confidence intervals that included zero were set to 0.001 for plotting and the confidence interval crosses the vertical dashed line denoting zero. Mean estimates with a 95% confidence interval not including zero have an asterisk next to the bar. The number of studies (*n*) contributing to each estimate is displayed above each estimate.

(1.3 μg g^{-1} d.w., [−0.57, 3.27 CI]), potentially indicating that fluxes resulting in loss of microcystin from the sediment pool (e.g., burial, diffusion) can actively reduce concentrations below sorption capacity. However, the estimates for both pools had CIs that included zero. The mean half-life for sediment biodegradation (3.57 d [2.01, 5.13 CI]) was similar to the half-life in the water column (4.67 d [2.028, 7.32 CI]) in this among-study comparison. The mean concentrations in macrophyte tissue (3.45 μg g^{-1} d.w., [−1.58, 8.49 CI]), the epiphyte pool (2.10 μg g^{-1} d.w., [1.15, 3.05 CI]), and the sediment biofilm pool (0.35 μg g^{-1} d.w., [−0.81, 1.51 CI]) indicate the magnitude of the primary producer reservoir of microcystin not found in the water column pool. For aquatic consumers, the mean zooplankton concentration was the highest of any pool (134.7 μg g^{-1} d.w., [94.5, 175.1 CI]), followed by macroinvertebrates (values only available for oligochaetes, bivalves, a gastropod, and chironomid; 3.75 μg g^{-1} d.w., [1.19, 6.32 CI]). The estimate for whole fish was taken from another meta-analysis that was based on 25 measurements (Flores et al. 2018).

Consideration of temporal dynamics

When cyanobacteria bloom—by definition, an episodic event—the pool of microcystin in the water column and/or biofilm can increase during this period. Given the detrimental effects of microcystin to human health, a great deal of research effort has focused on understanding the temporal dynamics of blooms and toxin production (Rastogi et al. 2015), even during periods of ice cover (Wejnerowski et al. 2018). The seasonality of blooms and toxin production likely also produces strong temporal dynamics in the other pools (e.g., animal and macrophyte tissues) and fluxes (e.g., sedimentation, biodegradation) of microcystin, particularly when coupled with variable turnover times in aquatic ecosystems.

Turnover times span many orders of magnitude, from a few minutes for a limiting reactant like microcystin for a biodegrading bacterium to months for animal tissues to years for sediment pools. This mismatch in turnover times allows legacies of past events to shape current ecosystem dynamics (Carpenter and Turner 2000). For organisms, tissue turnover time, microcystin accumulation rates, and toxin metabolism (Schmidt et al. 2014) combine with the availability of microcystin from other pools to dictate storage and persistence of the toxin in the aquatic food web. Organisms in lakes where blooms only occur seasonally may pose less of a threat to humans consuming them depending on the time since the bloom and opportunity for depuration and tissue turnover. On the other hand, the long tissue turnover times of some organisms such as bivalves and fish muscle (Vander Zanden et al. 2015) may result in a "hidden" pathway of human exposure when consumed weeks after a toxic bloom has subsided. Similarly, bloom seasonality in combination with plant phenology and tissue turnover may also affect microcystin accumulation in crops that are irrigated with microcystin-laden water supplies (Romero-Oliva et al. 2014).

Future directions and research needs

Constructing a comprehensive biogeochemical microcystin cycle revealed gaps in our understanding of ecosystem-scale microcystin dynamics. Despite the pools and fluxes identified from the literature synthesis, many unknowns remain pertaining to specific mechanisms and environmental factors that favor the movement and accumulation of microcystin within and among the pools. Future investigations of microcystin movement and accumulation in aquatic environments should focus particularly on fluxes to and from the sediment,

environmental drivers of biodegradation, and the seasonal dynamics of the aquatic food web pool.

Sediments are an active pool of microcystin in freshwater environments (Fig. 4; Zastepa et al. 2015). However, the current handful of ecosystem-level investigations of sediment microcystin fluxes limits our understanding of the magnitude and role sediments play in microcystin dynamics overall. For example, there is evidence indicating microcystin resuspension from sediments is a potential source of microcystin into the water column (Maghsoudi et al. 2015), but it is unclear when, where, and how much this flux contributes to the water column pool. Incorporating sediment–water exchange of microcystin into models and applying these to ecosystem-level investigations would generate valuable insight into the role of sediments in microcystin movement and accumulation.

Similarly, there is currently little information on the seasonal dynamics and environmental drivers of microcystin biodegradation in both the water column and sediments. While advances have been made in isolating and identifying microcystin-degrading bacteria, additional data are needed to understand the seasonal dynamics and mechanisms that control rates of biodegradation at an ecosystem scale. Accurately quantifying this important loss term in the microcystin cycle will require scaling bottle experiments from a controlled laboratory setting to the whole ecosystem scale which is heterogenous in both space and time.

While there are observational studies of microcystin dynamics in consumers in natural environments, much of the information we have regarding microcystin accumulation in aquatic organisms comes from toxicology-type studies with exposure treatments performed in a laboratory setting. Additional study of the duration and magnitude of microcystin accumulation in organism tissues seasonally and long-term would provide valuable insight into the dynamics of the aquatic consumer pool following bloom events. Another avenue of microcystin movement that remains poorly understood are the predominant uptake pathways in aquatic organisms and the role of environmental regulation of these rates. Similarly, the rates of microcystin excretion and egestion are poorly quantified. Further investigations of microcystin fluxes across aquatic–terrestrial interfaces and along the aquatic continuum (i.e., from inland to coastal ecosystems) are also needed to better capture the exogenous inputs available for consumer uptake. For example, limited information exists on microcystin flux via emergent aquatic insects from the aquatic to terrestrial environment (Moy et al. 2016). Similarly, the hydrologic fluxes of microcystin downstream into the marine environment poses a threat to wildlife (Miller et al. 2010) and humans consuming contaminated shellfish (Gibble et al. 2016).

Finally, the development of full microcystin budgets for watersheds requires investigators across various fields to report values in a "common currency" that can be incorporated into ecosystem models. For example, measurements of pools need to be in mass per unit area or volume and fluxes in mass per unit area or volume per unit time. While we were able to compare some microcystin pools across the literature ($\mu g\ g^{-1}$ d.w.; Figs. 3, 4), measurements reported in this common currency were lacking for some important pools, preventing us from developing a full, quantitative budget.

Conclusions

The conceptual biogeochemical model for microcystin that we constructed identified the major pools and fluxes of this toxin for inland waterbodies. From the quantitative synthesis we presented (Fig. 4) it is evident that microcystin is present and moves through many components of the ecosystem besides the water column. Given the many fluxes into and out of the water column (e.g., import from upstream, sediment diffusion), the visual presence of bloom is not necessarily indicative of exposure risk for humans. This conceptual model can be used as the framework for developing ecosystem mass balances of microcystin to quantify the transport and transformation of this toxin both in freshwater and marine ecosystems. Adopting the framework of a "microcystin cycle" will not only improve our understanding of processes driving toxin prevalence but will also help to prioritize effective strategies for the management of microcystin exposure risks to humans and wildlife.

References

Benson, J. M., J. A. Hutt, K. Rein, S. E. Boggs, E. B. Barr, and L. E. Fleming. 2005. The toxicity of microcystin LR in mice following 7 days of inhalation exposure. Toxicon **45**: 691–698. doi:10.1016/j.toxicon.2005.01.004

Berry, J. P., E. Lee, K. Walton, A. E. Wilson, and F. Bernal-Brooks. 2011. Bioaccumulation of microcystins by fish associated with a persistent cyanobacterial bloom in Lago de Patzcuaro (Michoacan, Mexico). Environ. Toxicol. Chem. **30**: 1621–1628. doi:10.1002/etc.548

Carmichael, W. W., and G. L. Boyer. 2016. Health impacts from cyanobacteria harmful algae blooms: Implications for the North American Great Lakes. Harmful Algae **54**: 194–212. doi:10.1016/j.hal.2016.02.002

Carpenter, S. R., and M. G. Turner. 2000. Hares and tortoises: Interactions of fast and slow Variablesin ecosystems. Ecosystems **3**: 495–497. doi:10.1007/s100210000043

Chen, J., and P. Xie. 2005. Seasonal dynamics of the hepatotoxic microcystins in various organs of four freshwater bivalves from the large eutrophic lake Taihu of subtropical China and the risk to human consumption. Environ. Toxicol. **20**: 572–584. doi:10.1002/tox.20146

Chen, X., X. Yang, L. Yang, B. Xiao, X. Wu, J. Wang, and H. Wan. 2010. An effective pathway for the removal of

microcystin LR via anoxic biodegradation in lake sediments. Water Res. **44**: 1884–1892. doi:10.1016/j.watres.2009.11.025

de Maagd, P. G.-J., A. J. Hendriks, W. Seinen, and D. T. H. M. Sijm. 1999. pH-dependent hydrophobicity of the cyanobacteria toxin microcystin-LR. Water Res. **33**: 677–680. doi:10.1016/S0043-1354(98)00258-9

Dziga, D., M. Wasylewski, B. Wladyka, S. Nybom, and J. Meriluoto. 2013. Microbial degradation of *Microcystins*. Chem. Res. Toxicol. **26**: 841–852. doi:10.1021/tx4000045

Dziga, D., M. Kokociński, J. Barylski, G. Nowicki, A. Maksylewicz, A. Antosiak, A. K. Banaś, and W. Strzałka. 2019. Correlation between specific groups of heterotrophic bacteria and microcystin biodegradation in freshwater bodies of Central Europe. FEMS Microbiol. Ecol. **95**: fiz162. doi:10.1093/femsec/fiz162

Feng, B., C. Wang, X. Wu, C. Tian, Y. Tian, and B. Xiao. 2019. Involvement of microcystins, colony size and photosynthetic activity in the benthic recruitment of *Microcystis*. J. Appl. Phycol. **31**: 223–233. doi:10.1007/s10811-018-1508-0

Flores, N. M., T. R. Miller, and J. D. Stockwell. 2018. A global analysis of the relationship between concentrations of Microcystins in water and fish. Front. Mar. Sci. **5**: 30. doi:10.3389/fmars.2018.00030

Ge, S., X. Qiao, X. Zhao, X. Li, and Y. Liu. 2021. Microcystin in source water: Pollution characteristics and human health risk assessment. RSC Adv. **11**: 6415–6422. doi:10.1039/D0RA08983D

Gérard, C., L. Brient, and B. Le Rouzic. 2005. Variation in the response of juvenile and adult gastropods (*Lymnaea stagnalis*) to cyanobacterial toxin (microcystin-LR). Environ. Toxicol. **20**: 592–596. doi:10.1002/tox.20147

Gérard, C., V. Poullain, E. Lance, A. Acou, L. Brient, and A. Carpentier. 2009. Influence of toxic cyanobacteria on community structure and microcystin accumulation of freshwater molluscs. Environ. Pollut. **157**: 609–617. doi:10.1016/j.envpol.2008.08.017

Giannuzzi, L., D. Sedan, R. Echenique, and D. Andrinolo. 2011. An acute case of intoxication with cyanobacteria and cyanotoxins in recreational water in Salto Grande Dam, Argentina. Mar. Drugs **9**: 2164–2175. doi:10.3390/md9112164

Gibble, C. M., M. B. Peacock, and R. M. Kudela. 2016. Evidence of freshwater algal toxins in marine shellfish: Implications for human and aquatic health. Harmful Algae **59**: 59–66. doi:10.1016/j.hal.2016.09.007

Graham, J. L., A. C. Ziegler, B. L. Loving, and K. A. Loftin. 2012. Fate and transport of cyanobacteria and associated toxins and taste-and-odor compounds from upstream reservoir releases in the Kansas River, Kansas, September and October 2011. Scientific Investigations Report 2012-5129. doi:10.3133/sir20125129

Harris, T. D., F. M. Wilhelm, J. L. Graham, and K. A. Loftin. 2014. Experimental manipulation of TN:TP ratios suppress cyanobacterial biovolume and microcystin concentration in large-scale *in situ* mesocosms. Lake Reserv. Manag. **30**: 72–83. doi:10.1080/10402381.2013.876131

Ho, J. C., A. M. Michalak, and N. Pahlevan. 2019. Widespread global increase in intense lake phytoplankton blooms since the 1980s. Nature **574**: 667–670. doi:10.1038/s41586-019-1648-7

Hoellein, T. J., and C. M. Rochman. 2021. The "plastic cycle": A watershed-scale model of plastic pools and fluxes. Front. Ecol. Environ. **19**: 176–183. doi:10.1002/fee.2294

Ibelings, B. W., and I. Chorus. 2007. Accumulation of cyanobacterial toxins in freshwater "seafood" and its consequences for public health: A review. Environ. Pollut. **150**: 177–192. doi:10.1016/j.envpol.2007.04.012

Jang, M., D. E. Berthold, Z. Yu, C. Silva-Sanchez, H. D. Laughinghouse IV, N. D. Denslow, and S. Han. 2020. Atmospheric progression of microcystin-LR from cyanobacterial aerosols. Environ. Sci. Technol. Lett. **7**: 740–745. doi:10.1021/acs.estlett.0c00464

Juhel, G., J. Davenport, J. O'Halloran, S. Culloty, R. Ramsay, K. James, A. Furey, and O. Allis. 2006. Pseudodiarrhoea in zebra mussels *Dreissena polymorpha* (Pallas)exposed to microcystins. J. Exp. Biol. **209**: 810–816. doi:10.1242/jeb.02081

Juhel, G., and others. 2007. In vivo exposure to microcystins induces DNA damage in the haemocytes of the zebra mussel, *Dreissena polymorpha*, as measured with the comet assay. Environ. Mol. Mutagen. **48**: 22–29. doi:10.1002/em.20271

Kozlowsky-Suzuki, B., A. E. Wilson, and A. da Silva Ferrão-Filho. 2012. Biomagnification or biodilution of microcystins in aquatic foodwebs? Meta-analyses of laboratory and field studies. Harmful Algae **18**: 47–55. doi:10.1016/j.hal.2012.04.002

Lance, E., L. Brient, A. Carpentier, A. Acou, L. Marion, M. Bormans, and C. Gérard. 2010. Impact of toxic cyanobacteria on gastropods and microcystin accumulation in a eutrophic lake (Grand-Lieu, France) with special reference to Physa (=Physella) acuta. Sci. Total Environ. **408**: 3560–3568. doi:10.1016/j.scitotenv.2010.04.050

Lezcano, M. Á., A. Quesada, and R. El-Shehawy. 2018. Seasonal dynamics of microcystin-degrading bacteria and toxic cyanobacterial blooms: Interaction and influence of abiotic factors. Harmful Algae **71**: 19–28. doi:10.1016/j.hal.2017.11.002

Li, J., J. Li, G. Shi, Z. Mei, R. Wang, and D. Li. 2016. Discerning biodegradation and adsorption of microcystin-LR in a shallow semi-enclosed bay and bacterial community shifts in response to associated process. Ecotoxicol. Environ. Saf. **132**: 123–131. doi:10.1016/j.ecoenv.2016.05.033

Loftin, K. A., J. L. Graham, E. D. Hilborn, S. C. Lehmann, M. T. Meyer, J. E. Dietze, and C. B. Griffith. 2016. Cyanotoxins in inland lakes of the United States: Occurrence and potential recreational health risks in the EPA National Lakes Assessment 2007. Harmful Algae **56**: 77–90. doi:10.1016/j.hal.2016.04.001

Maghsoudi, E., M. Prévost, S. Vo Duy, S. Sauvé, and S. Dorner. 2015. Adsorption characteristics of multiple microcystins

and cylindrospermopsin on sediment: Implications for toxin monitoring and drinking water treatment. Toxicon **103**: 48–54. doi:10.1016/j.toxicon.2015.06.007

Mantzouki, E., and others. 2018. Temperature effects explain continental scale distribution of cyanobacterial toxins. Toxins **10**: 156. doi:10.3390/toxins10040156

Massey, I. Y., and F. Yang. 2020. A mini review on Microcystins and bacterial degradation. Toxins **12**: 268. doi:10.3390/toxins12040268

Miller, M. A., and others. 2010. Evidence for a novel marine harmful algal bloom: Cyanotoxin (microcystin) transfer from land to sea otters. PLoS ONE **5**: e12576. doi:10.1371/journal.pone.0012576

Moy, N. J., J. Dodson, S. J. Tassone, P. A. Bukaveckas, and L. P. Bulluck. 2016. Biotransport of algal toxins to riparian food webs. Environ. Sci. Technol. **50**: 10007–10014. doi:10.1021/acs.est.6b02760

Munusamy, T., Y.-L. Hu, and J.-F. Lee. 2012. Adsorption and photodegradation of microcystin-LR onto sediments collected from reservoirs and rivers in Taiwan: A laboratory study to investigate the fate, transfer, and degradation of microcystin-LR. Environ. Sci. Pollut. Res. **19**: 2390–2399. doi:10.1007/s11356-012-0751-1

Olson, N. E., M. E. Cooke, J. H. Shi, J. A. Birbeck, J. A. Westrick, and A. P. Ault. 2020. Harmful algal bloom toxins in aerosol generated from inland Lake water. Environ. Sci. Technol. **54**: 4769–4780. doi:10.1021/acs.est.9b07727

Orihel, D. M., and others. 2012. High microcystin concentrations occur only at low nitrogen-to-phosphorus ratios in nutrient-rich Canadian lakes. Can. J. Fish. Aquat. Sci. **69**: 1457–1462. doi:10.1139/f2012-088

Papadimitriou, T., I. Kagalou, C. Stalikas, G. Pilidis, and I. D. Leonardos. 2012. Assessment of microcystin distribution and biomagnification in tissues of aquatic food web compartments from a shallow lake and evaluation of potential risks to public health. Ecotoxicology **21**: 1155–1166. doi:10.1007/s10646-012-0870-y

Pflugmacher, S. 2004. Promotion of oxidative stress in the aquatic macrophyte *Ceratophyllum demersum* during biotransformation of the cyanobacterial toxin microcystin-LR. Aquat. Toxicol. **70**: 169–178. doi:10.1016/j.aquatox.2004.06.010

Plaas, H. E., and H. W. Paerl. 2021. Toxic cyanobacteria: A growing threat to water and air quality. Environ. Sci. Technol. **55**: 44–64. doi:10.1021/acs.est.0c06653

Poste, A. E., R. E. Hecky, and S. J. Guildford. 2011. Evaluating microcystin exposure risk through fish consumption. Environ. Sci. Technol. **45**: 5806–5811. doi:10.1021/es200285c

Rastogi, R. P., R. P. Sinha, and A. Incharoensakdi. 2014. The cyanotoxin-microcystins: Current overview. Rev. Environ. Sci. Biotechnol. **13**: 215–249. doi:10.1007/s11157-014-9334-6

Rastogi, R. P., D. Madamwar, and A. Incharoensakdi. 2015. Bloom dynamics of cyanobacteria and their toxins: Environmental health impacts and mitigation strategies. Front. Microbiol. **6**: 1254. doi:10.3389/fmicb.2015.01254

Rohrlack, T., E. Dittmann, M. Henning, T. Börner, and J.-G. Kohl. 1999. Role of Microcystins in poisoning and food ingestion inhibition of *Daphnia galeata* caused by the cyanobacterium *Microcystis aeruginosa*. Appl. Environ. Microbiol. **65**: 737–739. doi:10.1128/AEM.65.2.737-739.1999

Romero-Oliva, C. S., V. Contardo-Jara, T. Block, and S. Pflugmacher. 2014. Accumulation of microcystin congeners in different aquatic plants and crops—A case study from Lake Amatitlán, Guatemala. Ecotoxicol. Environ. Saf. **102**: 121–128. doi:10.1016/j.ecoenv.2014.01.031

Romero-Oliva, C. S., V. Contardo-Jara, and S. Pflugmacher. 2015. Time dependent uptake, bioaccumulation and biotransformation of cell free crude extract microcystins from Lake Amatitlán, Guatemala by *Ceratophyllum demersum*, *Egeria densa* and *Hydrilla verticillata*. Toxicon **105**: 62–73. doi:10.1016/j.toxicon.2015.08.017

Ruibal-Conti, A. L., M. A. Ruiz, M. I. Rodriguez, D. Lerda, and M. D. Romero. 2019. Assessment of specific antibodies as biological indicators of human chronic exposure to microcystins. Ecotoxicol. Environ. Saf. **175**: 236–242. doi:10.1016/j.ecoenv.2019.03.071

Sánchez, K. F., N. Huntley, M. A. Duffy, and M. D. Hunter. 2019. Toxins or medicines? Phytoplankton diets mediate host and parasite fitness in a freshwater system. Proc. R. Soc. B **286**: 20182231. doi:10.1098/rspb.2018.2231

Schmidt, J., S. Wilhelm, and G. Boyer. 2014. The fate of Microcystins in the environment and challenges for monitoring. Toxins **6**: 3354–3387. doi:10.3390/toxins6123354

Song, H.-L., X.-N. Li, X.-W. Lu, and Y. Inamori. 2009. Investigation of microcystin removal from eutrophic surface water by aquatic vegetable bed. Ecol. Eng. **35**: 1589–1598. doi:10.1016/j.ecoleng.2008.04.005

Song, H., L. Coggins, E. Reichwaldt, and A. Ghadouani. 2015. The importance of Lake sediments as a pathway for microcystin dynamics in shallow Eutrophic Lakes. Toxins **7**: 900–918. doi:10.3390/toxins7030900

Thirumavalavan, M., Y.-L. Hu, and J.-F. Lee. 2012. Effects of humic acid and suspended soils on adsorption and photodegradation of microcystin-LR onto samples from Taiwan reservoirs and rivers. J. Hazard. Mater. **217–218**: 323–329. doi:10.1016/j.jhazmat.2012.03.031

Tillmanns, A. R., S. K. Burton, and F. R. Pick. 2011. *Daphnia* preexposed to toxic *Microcystis* exhibit feeding selectivity. Int. Rev. Hydrobiol. **96**: 20–28. doi:10.1002/iroh.201011298

Ujvárosi, A. Z., M. Riba, T. Garda, G. Gyémánt, G. Vereb, M. M.-Hamvas, G. Vasas, and C. Máthé. 2019. Attack of *Microcystis aeruginosa* bloom on a *Ceratophyllum submersum field*: Ecotoxicological measurements in real environment with real microcystin exposure. Sci. Total Environ. **662**: 735–745. doi:10.1016/j.scitotenv.2019.01.226

Umehara, A., T. Komorita, T. Takahashi, and H. Tsutsumi. 2019. Estimation of production and sedimentation of cyanobacterial toxins (microcystin) based on nutrient

budgets in the reservoir of Isahaya Bay, Japan. Ecotoxicol. Environ. Saf. **183**: 109477. doi:10.1016/j.ecoenv.2019.109477

Vander Zanden, M. J., M. K. Clayton, E. K. Moody, C. T. Solomon, and B. C. Weidel. 2015. Stable isotope turnover and half-life in animal tissues: A literature synthesis. PLoS ONE **10**: e0116182. doi:10.1371/journal.pone.0116182

Viechtbauer, W. 2010. Conducting meta-analyses in *R* with the metafor package. J. Stat. Soft. **36**: 1–48. doi:10.18637/jss.v036.i03

Wejnerowski, Ł., P. Rzymski, M. Kokociński, and J. Meriluoto. 2018. The structure and toxicity of winter cyanobacterial bloom in a eutrophic lake of the temperate zone. Ecotoxicology **27**: 752–760. doi:10.1007/s10646-018-1957-x

Welker, M., and C. Steinberg. 2000. Rates of humic substance photosensitized degradation of microcystin-LR in natural waters. Environ. Sci. Technol. **34**: 3415–3419. doi:10.1021/es991274t

Wilkinson, G. M. and Q. Shingai. 2022. Meta-analysis data for microcystin as a biogeochemical cycle: Pools, fluxes, and fates of the cyanotoxin in inland waters. ver 1. Environmental Data Initiative. doi:10.6073/pasta/0650f1cba18af503915e649f46e427e3

Wilkinson, G. M., J. A. Walter, C. D. Buelo, and M. L. Pace. 2022. No evidence of widespread algal bloom intensification in hundreds of lakes. Front. Ecol. Environ. **20**: 16–21. doi:10.1002/fee.2421

Wojtal-Frankiewicz, A., J. Bernasińska, T. Jurczak, K. Gwoździński, P. Frankiewicz, and M. Wielanek. 2013. Microcystin assimilation and detoxification by *Daphnia* spp. in two ecosystems of different cyanotoxin concentrations. J. Limnol. **72**: 13. doi:10.4081/jlimnol.2013.e13

Wood, S. A., and others. 2020. Toxic benthic freshwater cyanobacterial proliferations: Challenges and solutions for enhancing knowledge and improving monitoring and mitigation. Freshw. Biol. **65**: 1824–1842. doi:10.1111/fwb.13532

Wörmer, L., M. Huerta-Fontela, S. Cirés, D. Carrasco, and A. Quesada. 2010. Natural photodegradation of the cyanobacterial toxins microcystin and cylindrospermopsin. Environ. Sci. Technol. **44**: 3002–3007. doi:10.1021/es9036012

Yang, Z., F. Kong, and M. Zhang. 2016. Groundwater contamination by microcystin from toxic cyanobacteria blooms in Lake Chaohu, China. Environ. Monit. Assess. **188**: 280. doi:10.1007/s10661-016-5289-0

Zastepa, A., F. R. Pick, J. M. Blais, and A. Saleem. 2015. Analysis of intracellular and extracellular microcystin variants in sediments and pore waters by accelerated solvent extraction and high performance liquid chromatography-tandem mass spectrometry. Anal. Chim. Acta **872**: 26–34. doi:10.1016/j.aca.2015.02.056

Zastepa, A., F. R. Pick, and J. M. Blais. 2017. Distribution and flux of microcystin congeners in lake sediments. Lake Reserv. Manag. **33**: 444–451. doi:10.1080/10402381.2017.1362491

Zhang, D., P. Xie, Y. Liu, J. Chen, and G. Liang. 2007. Bioaccumulation of the hepatotoxic microcystins in various organs of a freshwater snail from a subtropical Chinese Lake, Lake Taihu, with dense toxic Microcystis blooms. Environ. Toxicol. Chem. **26**: 171–176. doi:10.1897/06-222R.1

Zhang, D., P. Xie, Y. Liu, and T. Qiu. 2009*a*. Transfer, distribution and bioaccumulation of microcystins in the aquatic food web in Lake Taihu, China, with potential risks to human health. Sci. Total Environ. **407**: 2191–2199. doi:10.1016/j.scitotenv.2008.12.039

Zhang, H., J. Zhang, and Y. Zhu. 2009*b*. Identification of microcystins in waters used for daily life by people who live on tai Lake during a serious cyanobacteria dominated bloom with risk analysis to human health. Environ. Toxicol. **24**: 82–86. doi:10.1002/tox.20381

Zhang, Y., H. Zhuang, H. Yang, W. Xue, L. Wang, and W. Wei. 2019. Microcystin-LR disturbs testicular development of giant freshwater prawn *Macrobrachium rosenbergii*. Chemosphere **222**: 584–592. doi:10.1016/j.chemosphere.2019.01.146

Zhang, Y., B. R. Husk, S. V. Duy, Q. T. Dinh, J. S. Sanchez, S. Sauvé, and J. K. Whalen. 2021. Quantitative screening for cyanotoxins in soil and groundwater of agricultural watersheds in Quebec, Canada. Chemosphere **274**: 129781. doi:10.1016/j.chemosphere.2021.129781

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