PERSPECTIVE

Title: **Low levels of contaminants stimulate harmful algal organisms and enrich their toxins**

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Abstract: A widespread increase in intense phytoplankton blooms has been noted in lakes worldwide since the 1980s, with the summertime peak intensity amplifying in most lakes. Such blooms cause annual economic losses of multi-billion USD and present a major challenge, affecting eleven out of the seventeen Sustainable Development Goals. Here, we evaluate recent scientific evidence for hormetic effects of emerging contaminants and regulated pollutants on *Microcystis* sp., the most notorious cyanobacteria forming harmful algal blooms and releasing phycotoxins in eutrophic freshwater systems. This new evidence leads to the conclusion that pollution is linked to algal bloom intensification. Concentrations of contaminants that are considerably smaller than the threshold for toxicity enhance the formation of harmful colonies, increase the production of phycotoxins and their release into

the environment, and lower the efficacy of algaecides to control algal blooms. The low-dose enhancement of microcystins is attributed to the up-regulation of a protein controlling microcystin release (McyH) and various microcystin synthetases in tandem with the global nitrogen regulator Ycf28, nonribosomal peptide synthetases, and several ATP-binding cassette transport proteins. Given that colony formation and phycotoxin production and release are enhanced by contaminant concentrations smaller than the toxicological threshold and widely occurring in the environment, the effect of contaminants on harmful algal blooms is more prevalent than previously thought. Climate change and nutrient enrichment, known mechanisms underpinning algal blooms, are thus joined by low-level pollutants as another causal mechanism.

Keywords: biphasic response; cyanobacteria; dose-response relationship; emerging contaminants; harmful microalgal bloom; hormetic effect

Introduction

Aquatic harmful algal blooms (HABs) are considered a climate change co-stressor in freshwater and marine ecosystems [1] as well as a major environmental issue that can severely affect aquatic ecosystems, human health, and economy [2] (see also https://www.epa.gov/nutrientpollution/harmful-algal-blooms). HABs can result in acute human illness, primarily due to phycotoxins ingested through contaminated seafood, direct skin contact, or inhalation [3–5]. Blooms of harmful cyanobacteria also enter water supply systems in all major continents but Antarctica [5–7]. The toxic effects of HABs are often similar to infectious diseases, such as norovirus, thus making their diagnosis difficult or impossible [4]. Health risks due to toxic HABs are linked with socioeconomic impacts, especially for human sub-populations whose wellbeing depends on aquacultures and shellfish cultivation [4]. Lethal harmful blooms might also be responsible for still mysterious species extinctions, e.g. dinosaurs [8]. They may even delay ecosystem recovery following extinction events, as was the case after the end-Permian extinction [9].

Algal blooms are linked with economic losses exceeding US$4 billion per year in the US alone [10]. The dimension of the issue of HABs is gigantic considering that it affects 11 of the 17 UN Sustainable Development Goals set forth to be achieved by 2030, namely no poverty, zero hunger, good health and wellbeing, clean water and sanitation, affordable and clean energy, decent work and economic growth, sustainable cities and communities, sustainable consumption and production, life below water, life on land, and partnerships for the goals (https://www.unep.org/news-and-stories/press-release/tackling-harmful-algal-blooms) [10]. Hence, control and management of HABs [11] is of utmost importance for human welfare and wellbeing, ecosystems health, and biosphere sustainability.

A widespread increase in intense freshwater blooms of phytoplankton has been noted worldwide since the 1980s, with the summertime peak intensity increasing in most lakes [10, 12, 13]. Importantly, lake algal blooms exhibited a pronounced increase in the 2010s (except in

Oceania) [14]. Climate change affects HABs in various ways, and climate simulations suggest species-specific changes in the abundance of harmful algae in the next decades [4, 10, 15, 16]. However, the reasons of the global increase in intense blooms since the 1980s remain unclear [17]. The phenomenon cannot be fully explained only by previously hypothesized environmental drivers, such as fertilizer use, precipitation, and temperature, because of the absence of consistent temporal matching [10, 12]. An analysis of about 9,500 events of HABs also suggests that the intensification of HABs may be linked to increased aquaculture industry and marine exploitation [17]. Moreover, anthropogenic factors (e.g. fertilizer, gross domestic product, and population) may be stronger drivers of global algal bloom intensification than climatic factors (e.g. temperature, wind speed, pressure, and rainfall) [12]. Stronger intensification of lake algal blooms occurs in Asia, South America and Africa than in other regions, and is linked to the persistent reliance of developing countries on agricultural fertilizers [12, 14]. However, these regions are also often hot spots of contamination, including pharmaceutical pollution [18–20], a factor hitherto unaccounted for in the evaluations of potential HABs driving mechanisms.

Hormesis is a biphasic dose response that is increasingly revealed in a vast array of plants and other aquatic and terrestrial organisms exposed to a plethora of contaminants applied individually or in mixtures [21–28]. That is, the responses to contaminants are opposite between low sub-toxic doses and high toxic doses, with low doses commonly inducing positive effects on individual organisms (Fig. 1). Such low doses are considerably below what was previously thought to be a toxicological threshold below which no effects occur, and are now widely shown to enhance the defense capacity of organisms, protect them against harmful drugs and other stresses, and promote the development of resistance [21–28]. Such effects of low doses of contaminants would be profoundly important for the control and management of HABs [11], e.g. due to higher doses of algaecides that would be needed to counteract low-dose contaminant

enhancement and prevent stimulation by low-dose algaecides. However, low-dose effects are not accounted for in current HAB control strategies.

In this article we review evidence pertaining to the potential of contaminants to induce hormesis in HABs-forming organisms. We discuss how this could affect HABs and control and management programs, thus offering a novel perspective to address the HAB problem (Fig. 2). We focus on *Microcystis* sp., which produce peptide hepatotoxins and neurotoxins, and which are the most notorious HAB-forming cyanobacteria in eutrophic fresh water systems [5, 10, 29]. *Microcystis* sp. also dominated about one third of 76 lakes studied worldwide and are commonly linked to exacerbation of bloom conditions [10]. While changes in harmful algae abundance and HAB intensification are largely linked to climate change and eutrophication [4, 14–16], this review suggests the possibility that pollution contributes to the intensification of HAB problem, enhancing algal colony formation and boosting synthesis and release of harmful toxins, even at pollutant concentrations hundreds-fold smaller than those considered toxic.

Occurrence of hormesis in harmful algae

This review revealed numerous studies showing that various chemicals often induce hormesis in HAB-forming and toxin-producing cyanobacteria (blue-green algae) [30], in particular in different strains of *Microcystis aeruginosa* [31–59] and in *M. wesenbergii* [60, 61]. However, hormetic responses were also revealed in the neurotoxin-producing *Anabaena flos-aquae* [62] and the bloom-forming haptophyte *Prymnesium parvum*, which produces the phycotoxin prymnesi [63]. Such hormetic responses were further identified in *Synechocystis* sp. [51], which may also contribute to blooms formation [64]. These data indicate that contaminants widely induce hormesis in some of the most abundant bloom-forming and toxin-producing cyanobacteria (Fig. 1).

Hormetic responses of HAB-forming and toxin-producing cyanobacteria are induced by many chemicals, such as arsenate [44, 45], environmental estrogens [65], chlorinated

organophosphorus flame retardants (Cl-OPFRs) [52], halogenated organic compounds [50], heavy metals [37], hydrogen peroxide (algaecide) [30], and the principal compound of yellow dye luteolin [32] and other allelopathic chemicals/algaecides [42, 58, 59, 62]. They were also induced by various antibiotics [31, 34, 36, 39, 40, 43, 46–48, 51, 53, 54, 66, 67], nanomaterials [56], polycyclic aromatic hydrocarbons [49], rare earths [33, 35], and several pesticides and disinfectants [34, 38, 41, 57, 60, 63]. Hence, hormetic responses represent a universal phenomenon across chemically diverse contaminants. The studies providing such evidence commonly apply environmentally-relevant concentrations, demonstrating that realistic concentrations widely enhance harmful algae and the production of microcystins. The occurrence of contaminants (e.g. hydrophobic organic compounds) in the surface layers of some lakes with frequent *Microcystis* blooms in the last decades provides support for this hypothesis [19]. Furthermore, concentrations of such contaminants (polycyclic aromatic hydrocarbons) are positively associated with phytoplankton biomass in lakes, with biomagnification of these contaminants during phytoplankton blooms [20].

The hormesis-inducing contaminants include regulated pollutants (e.g. heavy metals) but also many unregulated global contaminants of emerging concern (not subject to regulation limiting their concentrations in the environment), such as various agrochemicals, nanomaterials, and pharmaceuticals [31–48, 50, 51, 53, 54, 56–60, 62, 63, 65–67]. This is of profound importance since sub-NOAEL stimulation of such HABs-forming and toxin-producing organisms cannot be captured by the traditional dose-response models. Importantly, multi-component mixtures of same or different (e.g. herbicide-antibiotics) types of contaminants widely induce hormesis in these cyanobacteria, which can persist in the presence of other co-stressors too [31, 34, 66, 67, 35, 43, 48–51, 53, 54]. The presence of multiple contaminants can change the concentrations at which low-dose stimulation occurs. For example, cell density of *M. aeruginosa* significantly increased by the singular antibiotics ciprofloxacin and sulfamethoxazole

and their combination at 0.05-0.2, 0.1-0.2, and 0.02-0.1 μg L^{-1}, respectively, after 8 days of exposure [48]. In another example, one-week-long singular exposures of *M. aeruginosa* to the polycyclic aromatic hydrocarbons phenanthrene and benzo[α]pyrene and their mixture revealed growth hormesis at ≤1000 μg L^{-1} phenanthrene and at ≤279 μg L^{-1} mixture, whereas singular benzo[α]pyrene significantly inhibited growth at all tested concentrations (≥200 μg L^{-1}) [49]. Concurrent contaminants may even produce additive or synergistic effects on the low-dose hormetic stimulation of microcystins and growth, indicating magnified hazard and risks [34, 48, 50]. Thus, the issue of contaminant-induced hormesis in such cyanobacteria becomes more pressing because mixture effects are commonly neglected in ecological risk assessments.

Biological mechanisms of hormesis: driving harmful algal blooms

Hormetic responses of HAB-forming and toxin-producing cyanobacteria appear in electron transport rate, fluorescence intensity, and photochemical quantum yield of PSII [31, 33, 54, 56, 57, 60, 61, 66, 67, 34–36, 41, 43, 48, 51, 53]. Chlorophylls and less frequently carotenoids also respond in a hormetic fashion [33, 35, 38, 41, 43, 45–48, 52, 53, 61, 62, 65, 67]. For example, tris(1,3-dichloro-2-propyl) phosphate (TDCPP), a Cl-OPFR, significantly increased chlorophyll *a* and carotenoid concentrations in *M. aeruginosa* by ≈27-32 % at 0.1 and 1 mg L^{-1}, whereas the response returned to levels similar to the control or below [52]. Increasing concentrations of reactive oxygen species (ROS) and decreasing ratio of high-potential to low-potential form of cytochrome b_{559} also occur, suggesting thermodynamic transformation of cytochrome b_{559}, whose states are modulated by nitric oxide, to yield mild ROS and enhance stimulation [31, 50, 52, 56–58]. ROS such as hydrogen peroxide are key molecules and essential in cell signalling [68]. This mild increase in ROS contributes to overcompensation response of photosynthesis, while cytochrome P450 is also an important component of the stress response and contaminant biodegradation [31, 48]. Low doses of contaminants can also activate clathrin-mediated

endocytosis to facilitate a swift absorption of macro- (C, N, P) and micro-nutrients (Ca, K, Mg) to enhance chlorophylls, photosynthesis, growth, and microcystins production [33, 35].

As a result of the physiological hormetic controls (Fig. 3), cell density and proliferation, growth rate, as well as biomass exhibit hormetic responses to contaminants [30, 31, 42, 43, 46–53, 33, 54, 56–59, 66, 67, 34–37, 39–41]. These hormetic responses appear generally across different stresses, highlighting that low, sub-NOAEL doses of contaminants can act in promoting the growth of the population of harmful algae, forming more robust, dense, and competitive colonies. The broad hormesis literature, including algae, demonstrates that the low-dose enhancement is restricted by the limits of biological plasticity [21, 69–73]. Thus, the stimulation is modest in amplitude, typically 30-60%, and rarely exceeding 100%, independently from the organisms, biological mechanisms, and stressors [21, 69–73]. Similarly modest, but significant, are also the responses of chlorophylls, photosynthesis, and harmful algal growth/densities or proliferation to low doses of contaminants [31–47, 49–54, 56–63, 65–67, 74]. For instance, the stimulation of *M. aeruginosa* growth (cell density) by antibiotics was commonly less than 60% and as a rule smaller than 100% [31, 34, 36, 43, 46–48, 53, 54, 66, 67]. These suggest a lower effect amplitude of low, sub-NOAEL doses of contaminants to enhance HABs compared to fertilization effect of N and P, which are essential nutrients providing substance for physiological functioning and growth. However, regarding effect amplitude range, the contaminants enhancement is similar to that of atmospheric partial pressure of CO_2 (pCO_2) and warming effect on marine harmful algae, mostly consisted of dinoflagellates [75], suggesting that contaminants enhancement is equally important. Elevated pCO_2 overall increases HABs growth rate by 20%, but the effect varies among species and strains, often being null or negative [75]. The growth response of harmful algae to warming (+3-5 ^{o}C) is also highly variable and inconsistent across species, strains, and latitude, including not only positive effect but often null or negative effect [75]. Hence, regarding effect direction, it emerges that sub-NOAEL doses of contaminants have

a similar potential to enhance harmful algal growth with the two major global change factors, pCO_2 and temperature. Similar to the inconsistency and variability of N and P inputs, pCO_2 and temperature effects [76], the contaminant effects, and thus the NOAELs, the sub-NOAEL concentrations stimulating growth, toxicities, and algaecide resistance, would vary spatiotemporally and with chemical mode of action. However, except antibiotics, the number of studies is limited for each of the many contaminants reported to induce hormesis, not permitting a scientifically sound comparison among contaminants at this point of time. It is also possible but not irrefutable that sub-NOAEL doses of contaminants may induce a more consistent and universal enhancement of harmful algal growth than the two global change factors, pCO_2 and temperature, a hypothesis that remains to be tested.

Recent studies now shed light on the molecular mechanisms explaining the hormetic responses of these cyanobacteria to antibiotics, widely applied as multi-component mixtures (Fig. 3). The enhanced cell density or proliferation, growth rate, and photochemical quantum yield of PSII by different multi-component mixtures of antibiotics alone or with herbicides in *M. aeruginosa* is linked with increased energy generation by excitation of carbon metabolism and photosynthesis, as indicated by several transcriptomic/proteomic alterations, as well as promoted replication of DNA [31, 34, 43, 48, 54, 66, 67]. There are many genes involved in these hormetic responses, and numerous proteins are up- or down-regulated to modify ATP, biosynthesis, carbohydrate metabolism, carbon fixation/utilization, cell division, cell redox homeostasis, chlorophyll synthesis, circadian rhythms, pentose phosphate, photosynthesis, gene/protein transcription and expression, oxidation-reduction, quorum sensing, ribosome, translation, and DNA and its repair [31, 34, 43, 46–48, 51, 53, 66, 67]. These genetic changes also modulate transport proteins, ion homeostasis, cell division inhibitors, N compound metabolism, P metabolism, and stress response [31, 34, 43, 46–48, 51, 53, 66, 67].

This review revealed lack of studies reporting underlying molecular mechanisms of hormesis induced by contaminant types other than antibiotics. Nevertheless, a limited number of studies suggest similar molecular mechanisms for different contaminants. Specifically, low-dose graphene oxide nanomaterials improved photosynthetic performance of *M. aeruginosa* and enhanced the production of microcystins while increasing the abundance of *sul1*, *sul2*, *tetW*, and *tetM* in wastewater and the gene copy numbers of *mcyA-J* [56]. The hormetic stimulation of *M. aeruginosa* by sub-NOAEL doses of Cl-OPFRs also occurred in tandem with upregulation of the type I NADH dehydrogenase (NDH-1) complex (*ndhD1*, *ndhG*, *ndhH*, *ndhI*, *ndhJ*, *ndhL*, *ndhM*) and its mediated cyclic electron transfer pathway [52]. More studies are needed to unravel the underlying molecular mechanisms of sub-NOAEL stimulation by various kinds of contaminants as well as to understand how the composition of complex chemical mixtures affects the underlying molecular mechanisms.

Microcystins

There are several toxins produced by HABs, which are a chemically diverse group of secondary metabolites, posing a threat of aquatic resources and human health [77–80]. Species of the genus *Microcystis* produce the hepatotoxins microcystins [5]. Such toxins can cause profound effects on wildlife. For example, long-term studies recently suggest that neurotoxins produced by the cyanobacterium *Aetokthonos hydrillicola* cause a neurological disease (vacuolar myelinopathy) and lead to mass eagle deaths [81]. Not only do cyanotoxins affect other toxigenic cyanobacteria [5], but microcystins are also found in drinking water, often at levels raising concerns for human health [7]. *Microcystis* blooms occur in at least 108 countries, in 73% of which microcystin is also detected [29]. Microcystins at concentrations found in the environment during blooms (e.g. <1 to 300 μg L^{-1}) dysregulate proteins, impair metabolism, modify DNA repair, inhibit photosynthesis, and negatively affect the growth and reproduction of various organisms [80, 82–85]. For example, concentrations of microcystins inhibiting photosynthesis of

aquatic plants ranged from as little as ≤ 1 μg L^{-1} to < 100 μg L^{-1} after short- (≤1 d) to long-term (>7 d) exposures [80]. They can even be lethal and reduce population density, albeit low doses of microcystins may initially produce positive effects, such as increased population density and longevity, before turning into adverse [80, 82–84]. For instance, the LC50 values (dose killing 50% of the population) for *Ceriodaphnia dubia* and *Daphnia magna* were 5.5 and 58.7 μg L^{-1} [82].

In recognition of the profound effects of microcystins on ingesting organisms, we evaluated how contaminants affect microcystins. We found that various contaminants induce hormesis, promoting microcystins synthesis and elevating intracellular microcystins concentrations [31–34, 43–48, 53–56, 65–67]. Microcystins are typically enhanced within 1-4 days and increase further and often remain elevated for nearly four weeks from exposure to protect against stress in early stages and enhance the survival odds of alive cells in the algal population [31, 32, 34–36, 43, 48, 53, 54, 56]. Even growth-inhibitory high doses of contaminants such as antibiotics and microplastics can enhance microcystin production and concentrations and release in the environment before suppressing it [36, 55, 59, 74]. Hence, low or high sub-lethal doses can also increase the release of microcystins in the environment, and total or extracellular microcystins decrease at lethal doses due to decreased cell density [31, 32, 36, 43, 44, 46–49, 54–56, 58, 65, 67, 86].

We estimated the average stimulation of the production ability or concentration of microcystins by low, sub-NOAEL doses of contaminants at 57.9 % of the control value (95% CI estimated at 53.7-62.1%, n = 203 dose responses) [30–36, 43–47, 53, 54, 56, 65–67]. This stimulation was induced by concentrations on average 280.9 μg L^{-1} (95% CI estimated at 146.9-414.8 μg L^{-1}, n = 203 dose responses) [30, 31, 46, 47, 53, 54, 56, 65–67, 32–36, 43–45]. Low doses enhancing production of microcystins are even 100-20,000 times smaller than the lowest toxic concentrations inhibiting algal growth and production of microcystins [34, 36, 44, 45, 56]. Among the 203 dose responses analyzed, 93.6% were induced by concentrations ≤100 μg L^{-1},

while the majority (67.5%) of the dose responses were induced by concentrations ≤10 μg L^{-1}. As many as 60.6% of the dose responses were induced by concentrations as little as ≤0.6 μg L^{-1}. Such concentrations occur widely in the environment. The majority of dose-response entries concerned antibiotics (65.0%), followed by rare earth elements (22.7%), glyphosate (4.9%), forms of arsenic (3.9%), and other stressors (hydrogen peroxide, luteolin, nanomaterials, nonylphenol; 3.4%). The concentrations of antibiotics that enhanced microcystins ranged from 0.1 to 2000 μg L^{-1} (average: 71.1 μg L^{-1}), and 87.1% of these antibiotic stimulations were induced by concentrations ≤0.6 μg L^{-1}. The only concentration used for rare earth elements to produce significant stimulation was 30 μg L^{-1}. For glyphosate, the concentrations significantly enhancing microcystins ranged between 0.5 and 5 μg L^{-1}, a 10-fold range. However, a 10,000-fold concentration range was used for arsenic forms (0.01-100 μg L^{-1}). For the limited number of entries concerning hydrogen peroxide, luteolin, nanomaterials, and nonylphenol, the stimulatory concentrations were 200, 6,500, 10, and 200 μg L^{-1}, respectively. These results indicate that highly variable concentrations of contaminants can enhance microcystins. Recalculation of the doses after excluding the 13 dose-response entries with inducing concentrations ≥100 μg L^{-1}, the average concentration was 9.9 μg L^{-1} (95% CI estimated at 8.6-11.1 μg L^{-1}, n = 190 dose responses) and the stimulation similar with the analysis including all dose responses (average=59.2%; 95% CI estimated at 54.7-63.7%, n = 190 dose responses).

Microcystins are also enhanced by high sub-lethal doses that are inhibitory at the level of individual organisms. We found this stimulation to be on average 6.5 (95% CI estimated at 5.4-7.5 times; n = 25 dose-response assays) and often about 10-22 fold higher than the control value. These are induced by very high or extreme concentrations of contaminants, for example concentrations that can be 10^6 higher than those occurring in the environment increased the early microcystin production 5.7 times [55]. Such stimulation is well beyond the known common range of hormetic low-dose stimulation and may indicate a failure to keep microcystin

318 production below the ranges of biological plasticity, indicating a forthcoming damage to the
319 organism. Increased release of microcystins in the environment due to very high, toxic doses can
320 be explained by such doses causing cell rupture and release of microcystins from the cells into
321 the extracellular space [49, 56]. Thus, we show here that sub-lethal doses of contaminants, both
322 low sub-NOAEL and high super-NOAEL, enhance microcystins.
323 The general increase of microcystins by low, sub-NOAEL (and often sub-lethal)
324 concentrations of contaminants suggests that contaminants can intensify not only HABs but also
325 phycotoxins. This effect of contaminant on microcystins may be more consistent and universal
326 than what is currently known for other HABs-forming organisms and toxins in response to
327 climate change factors and nutrients; a hypothesis requiring experimental validation. For
328 example, overall toxin content in marine harmful algae does not show a significant response to
329 elevated pCO_2 or warming across studies, while some toxins (e.g. brevetoxins and paralytic
330 shellfish poisoning) produced by specific species or strains (e.g. *Alexandrium* spp. and *Karenia*
331 *brevis*) even decrease [75]. Cellular microcystin and paralytic shellfish poisoning toxins also
332 show an overall non-significant response to N and are increased by P limitation by 88 and 100%
333 respectively, across studies and species [87]. Across taxa, N-rich phycotoxins decrease by 60%
334 under N limitation and increase by 71% under P limitation [87], suggesting a potential
335 antagonistic effect between N and P on phycotoxins. However, the response to N and P
336 limitation varies across strains, species, and genera [87]. Especially cyanobacteria, and some
337 species or strains, including *Microcystis* strains, exhibit no significant or opposite responses than
338 the majority of species or strains [87]. Hence, an emerging hypothesis is that the effects of
339 contaminants at low sub-NOAEL and sub-lethal concentrations on phycotoxins deserve equal
340 consideration.
341 The molecular mechanisms controlling the hormetic responses of microcystins to
342 contaminants are now revealed (Fig. 3). The enhanced synthesis and concentrations of

343 microcystins are due to the up-regulation of microcystin synthetases and McyH, the protein
344 regulating microcystin release, in tandem with nonribosomal peptide synthetases, the global
345 nitrogen regulator Ycf28 binding the mcyA/D initiation codon of the microcystin synthetase
346 gene cluster, and several ATP-binding cassette transport proteins [34, 43, 46–48, 53, 54, 56, 66,
347 67]. These molecular mechanisms now provide the opportunity to develop relevant chemicals
348 blocking the expression of these proteins and genes to inhibit the synthesis and release of
349 microcystins into the environment.

350 **Unanswered questions and the path forward**

351 Our research synthesis now suggests that emerging contaminants and regulated pollutants
352 can contribute to intensification of HABs and enrichment of phycotoxins. While the prediction of
353 the time and place of the occurrence of HABs is advancing, such hormetic effects are not
354 considered, presenting a challenge for the monitoring and early warning systems of (harmful)
355 algal blooms [88–90]. These hormetic effects also limit the effectiveness of climate change
356 simulations, indicating the need for their representation in climate-HABs models as well as
357 climate change impact predictions [88, 91]. The enhancement of harmful algal growth by
358 contaminants is of similar magnitude and importance with that of pCO_2 and temperature, thus
359 contaminants effects should be given at least the same weight as for pCO_2 and temperature in
360 climate-HABs models.

361 The hormetic enhancement of harmful algae by low, sub-NOAEL doses of contaminants
362 suggests potential changes in the disease burden epidemiology, with likely effects beyond areas
363 that are currently known to be at risk [3]. This becomes of even greater concern in the light of
364 unknown interactive effects between climate change (e.g. documented changes due to global
365 warming [2, 4]) and environmental pollution, which could lead to antagonistic, additive, or
366 synergistic effects at low doses.

Algal growth is restricted in nutrient-limited conditions [41], and hormetic responses (including microcystins) depend on nutrient conditions. For example, elevated or limited phosphate and nitrogen can accordingly enhance or limit the biodegradation of contaminants and the low-dose stimulation [40, 44, 46, 47, 61]. Hence, integrated management is needed within the framework of which nutrients will be optimized considering local levels of contamination, but this requires further research. Nitrogen loading also promotes the abundance of microcystins-synthesizing strains in particular (e.g. *Microcystis* and *Planktothrix*) [92], suggesting that HABs may be promoted by atmospheric N deposition [93]. These also suggest interactive effects between air pollutants and water contaminants, complicating the programs targeting the control of HABs. Such interactive effects require further studies, considering also that the ratio of concentrations between nitrogen and phosphorus may be more important in driving microcystins responses to contaminants than the concentration of each nutrient separately [94–96]. The level of phycotoxins depends on the cellular N:P ratio, and the importance of this becomes greater in the light of the global N-P imbalance [87, 97].

Global warming can also extend the growing season, impacting the life cycle of HABs [98]. Harmful blooms might have intensified in the last decades due to eutrophication, elevated CO_2 concentrations, and global warming, and further increases in atmospheric CO_2 and global warming may signify the problem of intensified HABs [11, 14, 98, 99] (but see also [17]). Nutrient limitation can also reduce the response of the growth of HABs to elevated CO_2 and climate warming [98], suggesting that nutrient management can offer double benefits, i.e. reducing the effects of both water contaminants and climate change. Since HABs may be more influenced by climate change in eutrophic and hypertrophic lakes, these systems may be more vulnerable in the presence of low levels of contaminants [98].

Contaminant-induced hormesis of HABs-forming algae impedes the efficacy of treatments against HABs, since hormetic stimulation of such algae has various positive effects, promoting

392 the recovery from and reducing the efficacy of anti-algae treatments such as H_2O_2, UVB-, UV-C,
393 and $CuSO_4$ and $KMnO_4$ algaecides [43, 53, 54, 66, 67]. These findings suggest that higher
394 amounts of algaecides would be needed to control HABs in contaminated environments.
395 Especially, algaecides also cause hormesis with significant enhancement of intra- and extra-
396 cellular microcystins at sub-NOAEL doses, as opposed to inhibitory effects of super-NOAEL
397 doses [30]. However, this would lead to further contamination with unknown implications for
398 non-target organisms and the ecosystem. In the race to discover novel classes of algaecides with
399 improved algaecidal properties [11, 100], hormetic effects should be considered in the effect
400 testing and selection to avoid undesirable effects that may be opposite to the desired ones.
401 Our study leads us to the novel conclusion that contaminants produce equally important
402 enhancement of phycotoxin with nutrient and climate change factors, indicating a potentially
403 higher risk of contaminants for ecological and human health and adding a new dimension to the
404 issue of HABs. The responses can be microcystin-specific, e.g. among microcystins LR, LW,
405 and YR [35, 45]. Hence, further studies are needed to identify which microcystins are affected
406 most by low, sub-NOAEL doses of contaminants and which pose the highest risk for toxicity to
407 humans and other organisms via the food chain or direct interaction in contaminated
408 environments. Effects of mixtures of biotoxins are under-investigated [4], and how low-dose
409 effects modify mixtures of biotoxins and their effects on organisms directly interacting with the
410 biotoxins or indirectly ingesting them via the food chain is currently unknown. Initially toxic
411 effects of contaminants can change into stimulation (and vice versa) over time, and 1-3 weeks
412 may be needed to reach the maximum stimulatory response [31, 36, 37, 39, 41, 45, 46, 48, 51,
413 52, 60–62, 65–67]. Even if the stimulation is short-term or transient in the absence of renewed or
414 continued exposure, this might translate to acute expansion of HABs that could further
415 contaminate environment and pose risks to humans and other creatures.

Low, sub-NOAEL concentrations of contaminants may also lead to changes in the species composition of algal assemblages [30, 51, 101]. They may even impair the chemosensory system of organisms depending on dissolved chemical cues for their survival by depleting inorganic carbon and highly elevating pH due to increased photosynthesis [102, 103]. For example, low, environmentally-relevant, stimulatory doses of a mixture of ciprofloxacin, sulfamethoxazole, and tetracycline antibiotics can enhance the competitiveness and increase the proportion of the 'harmful' *M. aeruginosa* in a mixed culture of four phytoplankton species [51]. Similarly, algaecidal hydrogen peroxide at 0.2 mg L^{-1} increased and at 0.5-1.5 mg L^{-1} decreased the relative abundance of *Anabeana*, *Microcystis*, and *Oscillatoria* within a community of cyanobacteria and the relative abundance of Cyanobacteria within the prokaryotic community [30]. Considering also the toxic or even lethal effect of phycotoxins on interacting organisms, potential threats of such low concentrations of emerging contaminants and regulated pollutants to biodiversity and ecosystem services should also be considered in the agendas for protecting biodiversity and ecosystems.

The contaminant-induced hormesis in HAB-forming organisms creates a new challenge for traditional risk assessment to include effects below the traditional toxicological threshold. It also suggests that standard approaches cannot capture these effects of low and widely occurring concentrations of contaminants, urging for scientifically flexible approaches to permit more holistic evaluations of ecological risks.

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References

1. Griffith AW, Gobler CJ. Harmful algal blooms: A climate change co-stressor in marine and freshwater ecosystems. *Harmful Algae* 2020; **91**: 101590.
2. Anderson DM, Fachon E, Pickart RS, Lin P, Fischer AD, Richlen ML, et al. Evidence for massive and recurrent toxic blooms of Alexandrium catenella in the Alaskan Arctic. *Proc Natl Acad Sci U S A* 2021; **118**: e2107387118.
3. Young N, Sharpe RA, Barciela R, Nichols G, Davidson K, Berdalet E, et al. Marine harmful algal blooms and human health: A systematic scoping review. *Harmful Algae* 2020; **98**: 101901.
4. Berdalet E, Fleming LE, Gowen R, Davidson K, Hess P, Backer LC, et al. Marine harmful algal blooms, human health and wellbeing: challenges and opportunities in the 21st century. *J Mar Biol Assoc United Kingdom* 2016; **96**: 61–91.
5. Carmichael WW, Boyer GL. Health impacts from cyanobacteria harmful algae blooms: Implications for the North American Great Lakes. *Harmful Algae* 2016; **54**: 194–212.
6. He X, Liu YL, Conklin A, Westrick J, Weavers LK, Dionysiou DD, et al. Toxic

cyanobacteria and drinking water: Impacts, detection, and treatment. *Harmful Algae* 2016; **54**: 174–193.

7. Bullerjahn GS, McKay RM, Davis TW, Baker DB, Boyer GL, D'Anglada L V., et al. Global solutions to regional problems: Collecting global expertise to address the problem of harmful cyanobacterial blooms. A Lake Erie case study. *Harmful Algae* 2016; **54**: 223–238.

8. Gramling C. Toxic algae may be culprit in mysterious dinosaur deaths. *Science* 2017; **357**: 857.

9. Mays C, McLoughlin S, Frank TD, Fielding CR, Slater SM, Vajda V. Lethal microbial blooms delayed freshwater ecosystem recovery following the end-Permian extinction. *Nat Commun* 2021; **12**: 5511.

10. Ho JC, Michalak AM, Pahlevan N. Widespread global increase in intense lake phytoplankton blooms since the 1980s. *Nature* 2019; **574**: 667–670.

11. Sha J, Xiong H, Li C, Lu Z, Zhang J, Zhong H, et al. Harmful algal blooms and their eco-environmental indication. *Chemosphere* 2021; **274**: 129912.

12. Fang C, Song K, Paerl HW, Jacinthe PA, Wen Z, Liu G, et al. Global divergent trends of algal blooms detected by satellite during 1982-2018. *Glob Chang Biol* 2022; **28**: 2327–2340.

13. Anderson DM, Fensin E, Gobler CJ, Hoeglund AE, Hubbard KA, Kulis DM, et al. Marine harmful algal blooms (HABs) in the United States: History, current status and future trends. *Harmful Algae* 2021; **102**: 101975.

14. Hou X, Feng L, Dai Y, Hu C, Gibson L, Tang J, et al. Global mapping reveals increase in lacustrine algal blooms over the past decade. *Nat Geosci* 2022; **15**: 130–134.

15. Boivin-Rioux A, Starr M, Chassé J, Scarratt M, Perrie W, Long Z, et al. Harmful algae and climate change on the Canadian East Coast: Exploring occurrence predictions of

Dinophysis acuminata, D. norvegica, and Pseudo-nitzschia seriata. *Harmful Algae* 2022; **112**: 102183.

16. Zhang J, Yang Q, Liu Q, Liu S, Zhu Y, Yao J, et al. The responses of harmful dinoflagellate Karenia mikimotoi to simulated ocean acidification at the transcriptional level. *Harmful Algae* 2022; **111**: 102167.

17. Hallegraeff GM, Anderson DM, Belin C, Dechraoui Bottein M-Y, Bresnan E, Chinain M, et al. Perceived global increase in algal blooms is attributable to intensified monitoring and emerging bloom impacts. *Commun Earth Environ* 2021; **2**: 1–10.

18. Wilkinson JL, Boxall ABA, Kolpin DW, Leung KMY, Lai RWS, Galbán-Malagón C, et al. Pharmaceutical pollution of the world's rivers. *Proc Natl Acad Sci* 2022; **119**: e2113947119.

19. Liu G, Zhang G, Jin Z, Li J. Sedimentary record of hydrophobic organic compounds in relation to regional economic development: A study of Taihu Lake, East China. *Environ Pollut* 2009; **157**: 2994–3000.

20. Tao Y, Yu J, Liu X, Xue B, Wang S. Factors affecting annual occurrence, bioaccumulation, and biomagnification of polycyclic aromatic hydrocarbons in plankton food webs of subtropical eutrophic lakes. *Water Res* 2018; **132**: 1–11.

21. Sun T, Ji C, Li F, Wu H. Hormetic dose responses induced by organic flame retardants in aquatic animals: Occurrence and quantification. *Sci Total Environ* 2022; **820**: 153295.

22. Erofeeva EA. Plant hormesis and Shelford's tolerance law curve. *J For Res* 2021; **32**: 1789–1802.

23. Erofeeva EA. Environmental hormesis of non-specific and specific adaptive mechanisms in plants. *Sci Total Environ* 2022; **804**: 150059.

24. Jalal A, Oliveira Junior JC de, Ribeiro JS, Fernandes GC, Mariano GG, Trindade VDR, et al. Hormesis in plants: Physiological and biochemical responses. *Ecotoxicol Environ Saf*

2021; **207**: 111225.

25. Duke SO. Glyphosate: Uses other than in glyphosate-resistant crops, mode of action, degradation in plants, and effects on non-target plants and agricultural microbes. *Rev Environ Contam Toxicol* 2021; **255**: 1–65.

26. Tang L, Zhou Y, Zhang Y, Sun H. The role of energy/substrate in microbial hormesis. *Curr Opin Toxicol* 2022; **29**: 10–18.

27. Sun T, Zhan J, Li F, Ji C, H. W. Effect of microplastics on aquatic biota: A hormetic perspective. *Environ Pollut* 2021; **285**: 117206.

28. Agathokleous E, Barceló D, Rinklebe J, Sonne C, Calabrese EJ, Koike T. Hormesis induced by silver iodide, hydrocarbons, microplastics, pesticides, and pharmaceuticals: Implications for agroforestry ecosystems health. *Sci Total Environ* 2022; **820**: 153116.

29. Harke MJ, Steffen MM, Gobler CJ, Otten TG, Wilhelm SW, Wood SA, et al. A review of the global ecology, genomics, and biogeography of the toxic cyanobacterium, Microcystis spp. *Harmful Algae* 2016; **54**: 4–20.

30. Jiang Y, Fang Y, Liu Y, Liu B, Zhang J. Community succession during the preventive control of cyanobacterial bloom by hydrogen peroxide in an aquatic microcosm. *Ecotoxicol Environ Saf* 2022; **237**: 113546.

31. Jiang Y, Liu Y, Zhang J. Mechanisms for the stimulatory effects of a five-component mixture of antibiotics in Microcystis aeruginosa at transcriptomic and proteomic levels. *J Hazard Mater* 2021; **406**: 124722.

32. Li J, Hu J, Cao L, Yuan Y. Growth, physiological responses and microcystin-production/-release dynamics of Microcystis aeruginosa exposed to various luteolin doses. *Ecotoxicol Environ Saf* 2020; **196**: 110540.

33. Liu Y, Yang Q, Zhu M, Wang L, Zhou Q, Yang Z, et al. Endocytosis in Microcystis aeruginosa accelerates the synthesis of microcystins in the presence of lanthanum(III).

Harmful Algae 2020; **93**: 101791.

34. Xu S, Liu Y, Zhang J, Gao B. Proteomic mechanisms for the combined stimulatory effects of glyphosate and antibiotic contaminants on Microcystis aeruginosa. *Chemosphere* 2021; **267**: 129244.

35. Yang Q, Liu Y, Wang L, Zhou Q, Cheng M, Zhou J, et al. Cerium exposure in Lake Taihu water aggravates microcystin pollution via enhancing endocytosis of Microcystis aeruginosa. *Environ Pollut* 2022; **292**: 118308.

36. Zhang M, Steinman AD, Xue Q, Zhao Y, Xu Y, Xie L. Effects of erythromycin and sulfamethoxazole on Microcystis aeruginosa: Cytotoxic endpoints, production and release of microcystin-LR. *J Hazard Mater* 2020; **399**: 123021.

37. Bi X, Yan R, Li F, Dai W, Jiao K, Zhou Q, et al. Sequestration and distribution characteristics of Cd(II) by Microcystis aeruginosa and its role in colony formation. *Biomed Res Int* 2016; **2016**: 9837598.

38. de Morais P, Stoichev T, Basto MCP, Ramos V, Vasconcelos VM, Vasconcelos MTSD. Cyanobacterium Microcystis aeruginosa response to pentachlorophenol and comparison with that of the microalga Chlorella vulgaris. *Water Res* 2014; **52**: 63–72.

39. Guo RX, Chen JQ. Phytoplankton toxicity of the antibiotic chlortetracycline and its UV light degradation products. *Chemosphere* 2012; **87**: 1254–1259.

40. Liu Y, Wang F, Chen X, Zhang J, Gao B. Cellular responses and biodegradation of amoxicillin in Microcystis aeruginosa at different nitrogen levels. *Ecotoxicol Environ Saf* 2015; **111**: 138–145.

41. Qiu H, Geng J, Ren H, Xia X, Wang X, Yu Y. Physiological and biochemical responses of Microcystis aeruginosa to glyphosate and its Roundup® formulation. *J Hazard Mater* 2013; **248**–**249**: 172–176.

42. Sun H, Zheng M, Song J, Huang S, Pan Y, Gong R, et al. Multiple-species hormetic

570 phenomena induced by indole: A case study on the toxicity of indole to bacteria, algae and
571 human cells. *Sci Total Environ* 2019; **657**: 46–55.
572 43. Jiang Y, Liu Y, Zhang J. Antibiotic contaminants reduced the treatment efficiency of UV-
573 C on Microcystis aeruginosa through hormesis. *Environ Pollut* 2020; **261**: 114193.
574 44. Gong Y, Chou HN, Tu CD, Liu X, Liu J, Song L. Effects of arsenate on the growth and
575 microcystin production of Microcystis aeruginosa isolated from Taiwan as influenced by
576 extracellular phosphate. *J Appl Phycol* 2008; **21**: 225–231.
577 45. Gong Y, Ao HY, Liu BB, Wen S, Wang Z, Hu DJ, et al. Effects of inorganic arsenic on
578 growth and microcystin production of a Microcystis strain isolated from an algal bloom in
579 Dianchi Lake, China. *Chinese Sci Bull* 2011; **56**: 2337–2342.
580 46. Liu Y, Chen X, Zhang J, Gao B. Hormesis effects of amoxicillin on growth and cellular
581 biosynthesis of Microcystis aeruginosa at different nitrogen levels. *Microb Ecol* 2014; **69**:
582 608–617.
583 47. Liu Y, Chen S, Chen X, Zhang J, Gao B. Interactions between Microcystis aeruginosa and
584 coexisting amoxicillin contaminant at different phosphorus levels. *J Hazard Mater* 2015;
585 **297**: 83–91.
586 48. Liu Y, Chen S, Zhang J, Li X, Gao B. Stimulation effects of ciprofloxacin and
587 sulphamethoxazole in Microcystis aeruginosa and isobaric tag for relative and absolute
588 quantitation-based screening of antibiotic targets. *Mol Ecol* 2017; **26**: 689–701.
589 49. Wan X, Guo Q, Li X, Wang G, Zhao Y. Synergistic toxicity to the toxigenic Microcystis
590 and enhanced microcystin release exposed to polycyclic aromatic hydrocarbon mixtures.
591 *Toxicon* 2022; **210**: 49–57.
592 50. Zhang Y, Gao Q, Liu S, Tang L, Li X-G, Sun H. Hormetic dose-response of halogenated
593 organic pollutants on Microcystis aeruginosa: Joint toxic action and mechanism. *Sci Total*
594 *Environ* 2022; **829**: 154581.

595 51. Xu S, Liu Y, Zhang J. Transcriptomic mechanisms for the promotion of cyanobacterial
596 growth against eukaryotic microalgae by a ternary antibiotic mixture. *Environ Sci Pollut*
597 *Res* 2022; **In Press**.
598 52. Zhang X, Ai S, Wei J, Yang X, Huang Y, Hu J, et al. Biphasic effects of typical
599 chlorinated organophosphorus flame retardants on Microcystis aeruginosa. *Ecotoxicol*
600 *Environ Saf* 2022; **241**: 113813.
601 53. Jiang Y, Liu Y, Zhang J. Antibiotics induced alterations in cell density, photosynthesis,
602 microcystin synthesis and proteomic expression of Microcystis aeruginosa during CuSO4
603 treatment. *Aquat Toxicol* 2020; **222**: 105473.
604 54. Liu Y, Cui M, Zhang J, Gao B. Impacts of antibiotic contaminants on Microcystis
605 aeruginosa during potassium permanganate treatment. *Harmful Algae* 2020; **92**: 101741.
606 55. Pan M, Lyu T, Zhan L, Matamoros V, Angelidaki I, Cooper M, et al. Mitigating antibiotic
607 pollution using cyanobacteria: Removal efficiency, pathways and metabolism. *Water Res*
608 2021; **190**: 116735.
609 56. Wu S, Ji X, Li X, Ye J, Xu W, Wang R, et al. Mutual impacts and interactions of
610 antibiotic resistance genes, microcystin synthetase genes, graphene oxide, and Microcystis
611 aeruginosa in synthetic wastewater. *Environ Sci Pollut Res* 2021; **29**: 3994–4007.
612 57. Zhang Y, Calabrese EJ, Zhang J, Gao D, Qin M, Lin Z. A trigger mechanism of herbicides
613 to phytoplankton blooms: From the standpoint of hormesis involving cytochrome b559,
614 reactive oxygen species and nitric oxide. *Water Res* 2020; **173**: 115584.
615 58. Zuo S, Yang H, Jiang X, Ma Y. Magnetic Fe3O4 nanoparticles enhance cyanobactericidal
616 effect of allelopathic p-hydroxybenzoic acid on Microcystis aeruginosa by enhancing
617 hydroxyl radical production. *Sci Total Environ* 2021; **770**: 145201.
618 59. Li J, Cao L, Guo Z, An G, Li B, Li J. Time- and dose-dependent allelopathic effects and
619 mechanisms of kaempferol on toxigenic Microcystis growth. *Ecotoxicol Environ Saf*

2021; **222**: 112508.

60. Sun K, Liu W, Liu L, Wang N, Duan S. Ecological risks assessment of organophosphorus pesticides on bloom of Microcystis wesenbergii. *Int Biodeterior Biodegradation* 2013; **77**: 98–105.

61. Sun KF, Xu XR, Duan SS, Wang YS, Cheng H, Zhang ZW, et al. Ecotoxicity of two organophosphate pesticides chlorpyrifos and dichlorvos on non-targeting cyanobacteria Microcystis wesenbergii. *Ecotoxicology* 2015; **24**: 1498–1507.

62. Zhao S, Pan W bin, Ma C. Stimulation and inhibition effects of algae-lytic products from Bacillus cereus strain L7 on Anabaena flos-aquae. *J Appl Phycol* 2011; **24**: 1015–1021.

63. Dabney BL, Patiño R. Low-dose stimulation of growth of the harmful alga, Prymnesium parvum, by glyphosate and glyphosate-based herbicides. *Harmful Algae* 2018; **80**: 130–139.

64. Maeda K, Okuda Y, Enomoto G, Watanabe S, Ikeuchi M. Biosynthesis of a sulfated exopolysaccharide, synechan, and bloom formation in the model cyanobacterium Synechocystis sp. strain PCC 6803. *Elife* 2021; **10**: e66538.

65. Wang J, Xie P, Guo N. Effects of nonylphenol on the growth and microcystin production of Microcystis strains. *Environ Res* 2007; **103**: 70–78.

66. Jiang Y, Liu Y, Zhang J, Gao B. Antibiotics promoted the recovery of Microcystis aeruginosa after UV-B radiation at cellular and proteomic levels. *Ecotoxicol Environ Saf* 2020; **190**: 110080.

67. Liu Y, Zhang J, Gao B. Proteomic mechanisms for the stimulatory effects of antibiotics on Microcystis aeruginosa during hydrogen peroxide treatment. *Chemosphere* 2020; **247**: 125837.

68. Soares C, Carvalho MEA, Azevedo RA, Fidalgo F. Plants facing oxidative challenges—A little help from the antioxidant networks. *Environ Exp Bot* 2019; **161**: 4–25.

645 69. Calabrese EJ, Agathokleous E, Kozumbo WJ, Stanek EJ, Leonard D. Estimating the range
646 of the maximum hormetic stimulatory response. *Environ Res* 2019; **170**: 337–343.
647 70. Agathokleous E, Kitao M, Calabrese EJ. Hormesis: Highly generalizable and beyond
648 laboratory. *Trends Plant Sci* 2020; **25**: 1076–1086.
649 71. Agathokleous E, Feng Z, Iavicoli I, Calabrese EJ. The two faces of nanomaterials: A
650 quantification of hormesis in algae and plants. *Environ Int* 2019; **131**: 105044.
651 72. Agathokleous E. The rise and fall of photosynthesis: Hormetic dose response in plants. *J*
652 *For Res* 2021; **32**: 789–803.
653 73. Cedergreen N, Streibig JC, Kudsk P, Mathiassen SK, Duke SO. The occurrence of
654 hormesis in plants and algae. *Dose Response* 2007; **5**: 150–62.
655 74. Wan Q, Li J, Chen Y. Comparative growth and cellular responses of toxigenic Microcystis
656 exposed to different types of microplastics at various doses. *Environ Pollut* 2021; **290**:
657 117950.
658 75. Brandenburg KM, Velthuis M, Van de Waal DB. Meta-analysis reveals enhanced growth
659 of marine harmful algae from temperate regions with warming and elevated CO2 levels.
660 *Glob Chang Biol* 2019; **25**: 2607–2618.
661 76. Tian D, Xie G, Tian J, Tseng KH, Shum CK, Lee J, et al. Spatiotemporal variability and
662 environmental factors of harmful algal blooms (HABs) over western Lake Erie. *PLoS One*
663 2017; **12**: e0179622.
664 77. Cusick KD, Widder EA. Bioluminescence and toxicity as driving factors in harmful algal
665 blooms: Ecological functions and genetic variability. *Harmful Algae* 2020; **98**: 101850.
666 78. Huang IS, Zimba P V. Cyanobacterial bioactive metabolites—A review of their chemistry
667 and biology. *Harmful Algae* 2019; **83**: 42–94.
668 79. Preece EP, Hardy FJ, Moore BC, Bryan M. A review of microcystin detections in
669 Estuarine and Marine waters: Environmental implications and human health risk. *Harmful*

Algae 2017; **61**: 31–45.

80. Zhang Y, Vo Duy S, Munoz G, Sauvé S. Phytotoxic effects of microcystins, anatoxin-a and cylindrospermopsin to aquatic plants: A meta-analysis. *Sci Total Environ* 2022; **810**: 152104.

81. Breinlinger S, Phillips TJ, Haram BN, Mareš J, Martínez Yerena JA, Hrouzek P, et al. Hunting the eagle killer: A cyanobacterial neurotoxin causes vacuolar myelinopathy. *Science (80-)* 2021; **371**.

82. Shahmohamadloo RS, Poirier DG, Ortiz Almirall X, Bhavsar SP, Sibley PK. Assessing the toxicity of cell-bound microcystins on freshwater pelagic and benthic invertebrates. *Ecotoxicol Environ Saf* 2020; **188**: 109945.

83. Shahmohamadloo RS, Ortiz Almirall X, Simmons DBD, Lumsden JS, Bhavsar SP, Watson-Leung T, et al. Cyanotoxins within and outside of Microcystis aeruginosa cause adverse effects in rainbow trout (Oncorhynchus mykiss). *Environ Sci Technol* 2021; **55**: 10422–10431.

84. De Senerpont Domis LN, Bartosiewicz M, Davis C, Cerbin S. The effect of small doses of toxic cyanobacterial food on the temperature response of Daphnia galeata: is bigger better? *Freshw Biol* 2013; **58**: 560–572.

85. He J, Chen J, Chen F, Chen L, Giesy JP, Guo Y, et al. Health Risks of Chronic Exposure to Small Doses of Microcystins: An Integrative Metabolomic and Biochemical Study of Human Serum. *Environ Sci Technol* 2022; **56**: 6548–6559.

86. Lin Y, Chen A, He Y, Qing C, Peng L, Luo S, et al. Responses of Microcystis aeruginosa (Cyanobacteria) to sanguinarine stress: morphological and physiological characteristics associated with competitive advantage. *Phycologia* 2019; **58**: 260–268.

87. Brandenburg K, Siebers L, Keuskamp J, Jephcott TG, van de Waal DB. Effects of nutrient limitation on the synthesis of N-rich phytoplankton toxins: A meta-analysis. *Toxins*

(Basel) 2020; **12**: 221.

88. Guan W, Bao M, Lou X, Zhou Z, Yin K. Monitoring, modeling and projection of harmful algal blooms in China. *Harmful Algae* 2022; **111**: 102164.

89. Durán-Vinet B, Araya-Castro K, Chao TC, Wood SA, Gallardo V, Godoy K, et al. Potential applications of CRISPR/Cas for next-generation biomonitoring of harmful algae blooms: A review. *Harmful Algae* 2021; **103**: 102027.

90. Tian R, Chen J, Sun X, Li D, Liu C, Weng H. Algae explosive growth mechanism enabling weather-like forecast of harmful algal blooms. *Sci Rep* 2018; **8**: 1–7.

91. Hennon GMM, Dyhrman ST. Progress and promise of omics for predicting the impacts of climate change on harmful algal blooms. *Harmful Algae* 2020; **91**: 101587.

92. Gobler CJ, Burkholder JAM, Davis TW, Harke MJ, Johengen T, Stow CA, et al. The dual role of nitrogen supply in controlling the growth and toxicity of cyanobacterial blooms. *Harmful Algae* 2016; **54**: 87–97.

93. de Vries W. Impacts of nitrogen emissions on ecosystems and human health: A mini review. *Curr Opin Environ Sci Heal* 2021; **21**: 100249.

94. Orihel DM, Bird DF, Brylinsky M, Chen H, Donald DB, Huang DY, et al. High microcystin concentrations occur only at low nitrogen-to-phosphorus ratios in nutrient-rich Canadian lakes. *Can J Fish Aquat Sci* 2012; **69**: 1457–1462.

95. Brandenburg KM, de Senerpont Domis LN, Wohlrab S, Krock B, John U, van Scheppingen Y, et al. Combined physical, chemical and biological factors shape Alexandrium ostenfeldii blooms in The Netherlands. *Harmful Algae* 2017; **63**: 146–153.

96. Anderson DM, Glibert PM, Burkholder JM. Harmful algal blooms and eutrophication: Nutrient sources, composition, and consequences. *Estuaries* 2002; **25**: 704–726.

97. Peñuelas J, Sardans J. The global nitrogen-phosphorus imbalance. *Science (80-)* 2022; **375**: 266–267.

98. Visser PM, Verspagen JMH, Sandrini G, Stal LJ, Matthijs HCP, Davis TW, et al. How rising CO2 and global warming may stimulate harmful cyanobacterial blooms. *Harmful Algae* 2016; **54**: 145–159.
99. Glibert PM, Seitzinger S, Heil CA, Burkholder JM, Parrow MW, Codispoti LA, et al. The role of eutrophication in the global proliferation of harmful algal blooms. *Oceanography* 2005; **18**: 198–209.
100. Wang Y, Liu Q, Wei Z, Liu N, Li Y, Li D, et al. Thiazole amides, a novel class of algaecides against freshwater harmful algae. *Sci Rep* 2018; **8**: 8555.
101. Park MH, Kim K, Hwang SJ. Differential effects of the allelochemical juglone on growth of harmful and non-target freshwater algae. *Appl Sci* 2020; **10**: 2873.
102. Chislock MF, Doster E, Zitomer RA, Wilson AE. Eutrophication: Causes, consequences, and controls in aquatic ecosystems. *Nat Educ Knowl* 2013; **4**: 10.
103. Turner AM, Chislock MF. Blinded by the stink: Nutrient enrichment impairs the perception of predation risk by freshwater snails. *Ecol Appl* 2010; **20**: 2089–2095.

Figure captions

Fig. 1. Response of *Microcystis aeruginosa* to low, environmentally occurring concentrations of antibiotics mixtures. At these trace concentrations included in the study, there were no negative effects on these studied endpoints; however, the concentration-response relationship suggest negative effects would be expected at antibiotics concentrations larger than 0.5 μg L^{-1}. The antibiotics were amoxicillin, ciprofloxacin, spiramycin, sulfamethoxazole, and tetracycline, and the exposure lasted 14 days under aseptic conditions in a constant-temperature illuminating incubator. F_v/F_m: maximum photochemical quantum yield of photosystem II (PSII). Microcystins refer to the total concentrations (μg mL^{-1}). The data are based on [31]. Data extraction and calculation are described in Supporting Materials.

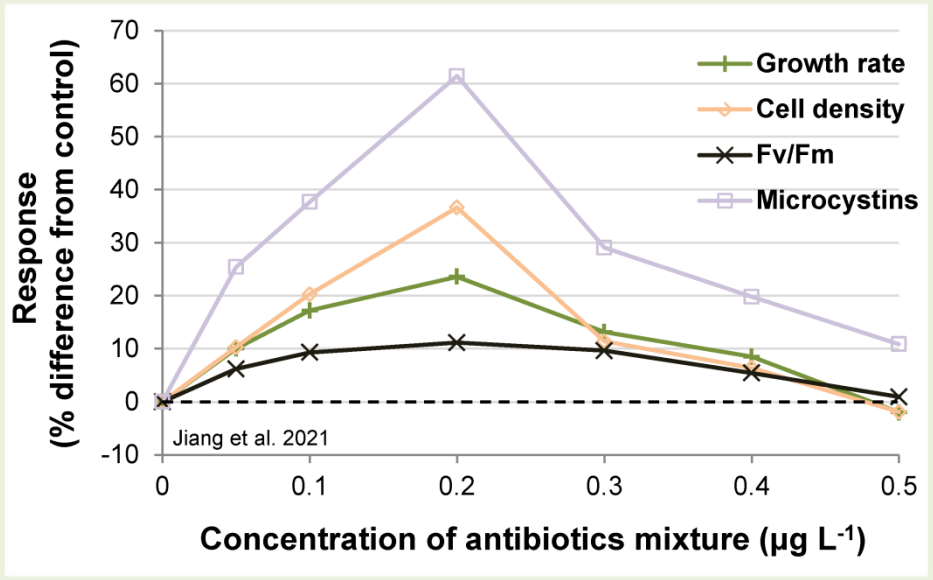

Fig. 2. Conceptual diagram of stimulation of cyanobacteria forming harmful algal blooms (HABs) by contaminants. The progressive anthropogenic impact on water bodies and the subsequent effects on algal ecology are of concern. Nutrient over-supply, leading to eutrophication and nutrient imbalance in water bodies, has long been known as a major factor driving algal ecology and thus HABs. However, recent studies now provide substantial evidence that trace chemicals in the waters exert significant influence on the ecology of major HABs-forming organisms, such as the notorious cyanobacterium *Microcystis aeruginosa*. Trace chemicals enhance algal growth, biomass, and proliferation and boost the synthesis of harmful phycotoxins and their release in the environment. Such effects of low concentrations of chemicals alter population dynamics and can change algal community structure, containing toxigenic strains and potentially composed of more toxigenic individuals with more abundant phycotoxins. The broad hormesis literature indicates a highly generalized stimulation amplitude across species, suggesting the degree of stimulation of HABs-forming species is not expected to differ from that of non-HABs-forming plankton species. However, it also suggests that resistant HABs-forming organisms are expected to have a broader range of stimulatory zone and undergo stimulation at concentrations not affecting the average population or inhibiting susceptible subpopulation groups. These would facilitate the dominance of resistant toxigenic individuals within HABs due to their stimulation and dominance over non-resistant, non-toxigenic individuals. These indicate the possibility that HABs with enriched toxigenic individuals may increase by increased concentrations of trace contaminants. Light gray color arrows indicate causal drivers of human origin, whereas dark gray color arrows indicate major changes in water quality that are associated with HABs. Black arrows indicate HABs-related effects (bold text) that are now attributed to trace chemicals in the water bodies.

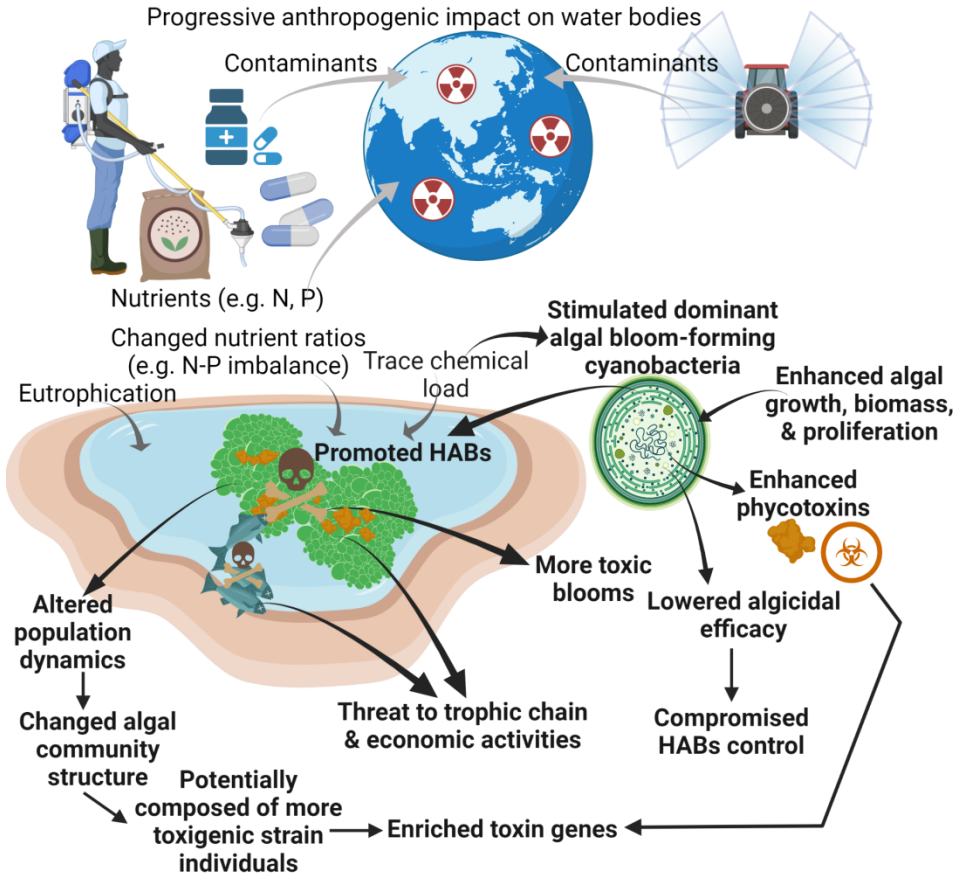

Fig. 3. Underlying mechanisms of *Microcystis aeruginosa* stimulation by low-level antibiotics contamination. These responses occur at doses of contaminants that are smaller than the no-observed-adverse-effect-level (NOAEL), i.e. toxicological threshold. The graphic illustrates major genes and proteins that are up- or down-regulated (oval boxes) and the underlying mechanisms they control (rectangle boxes). Further details about the molecular mechanisms can be found in the reviewed literature [31, 34, 66, 67, 43, 46–48, 51, 53, 54, 56].