Chapter 36: Effective doses, guidelines & regulations

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Introduction

Cyanobacteria are an important ecological component of all freshwater, estuarine and marine ecosystems worldwide. They contribute significantly to ecosystem productivity – sometimes excessively. When this occurs they also form water 'blooms' which are now recognised as a serious water quality problem with our use of water both for drinking water supply, recreational amenity and for agricultural use. Blooms are a symptom of eutrophication and are evidence of the deterioration of our water resources as a result of effluent discharge, poor land and catchment management, and often also of poor water allocation practices in rivers. This is now becoming better understood and acknowledged both in the United States and worldwide (Burns 2005, Chorus and Bartram, 1999).

The conditions which favour the growth of cyanobacteria and lead to blooms are nutrient enrichment (largely phosphorus but also nitrogen), warm temperatures, and calm stable water conditions such as those occurring in thermally stratified lakes and slow-flowing rivers. The latter may be the result of hydrology altered by abstraction practices. These conditions are often caused by human actions and activities, but can also be associated with natural climatic cycles such as droughts.

There is an international consensus that there has been an increase in frequency and severity of harmful algal blooms in both the marine and freshwater environments. In the case of the fresh water environment, the occurrence of toxic cyanobacterial blooms (CyanoHABs) presents problems for treatment, management and regulation of the quality of drinking water supplies. In regard to cyanotoxins a number of countries however have developed regulations or guidelines for cyanotoxins and cyanobacteria in drinking water, and in some cases in water used for recreation and agriculture.

There are currently no federal regulations or guidelines in the US for protecting human health and ecosystem viability from cyanobacterial harmful algal blooms (CyanoHABs) that occur in fresh, estuary, and marine water environments. This paper will explore the regulations and guidelines that have been developed around the world, identify others that may be needed, identify research needed to support their development, and identify factors that would be needed in a model to predict the need for revised and/or additional regulations or guidelines concerning CHABs.

Regulations and Guidelines

A number of countries have developed regulations or guidelines for cyanotoxins and cyanobacteria in drinking water, and in some cases in water used for recreational activity and agriculture. The approaches taken and the degree of adoption of guidelines are summarised in a comprehensive recent international compilation by Chorus (2005a). The main focus internationally has been upon microcystin toxins, produced by *Microcystis aeruginosa* and *Planktothrix agardhii*. This is because microcystins are widely regarded as the most significant potential source of human injury from cyanobacteria on a world-wide scale. Many international guidelines have taken their lead from the World Health Organization's (WHO) provisional guideline of 1 $\mu g\ L^{-1}$ for microcystin-LR in drinking-water released in 1998 (WHO 2004). The WHO guideline value is stated as being 'provisional', - "because it covers only microcystin-LR, for reasons that the toxicology is limited and new data for toxicity of cyanobacterial toxins are being generated". A comprehensive list of guidelines from various countries for toxins in drinking water is given in Table 1, and for recreational water in Table 2.

Table 1. Status of guidelines or standards for cyanobacterial toxins in drinking water for various countries. Information is also included on countries that are considering guidelines. (Information derived from websites and Chorus, 2005; Codd et al., 2005).

Country	Guideline Value/Standard	Comments/Explanations
Argentina	Under review	(Codd et al., 2005)
Australia	1.3 μg L^{-1} Total Microcystins, expressed as toxicity equivalents of microcystin-LR	From the Australian Drinking Water Guidelines see text for explanation on the process of guideline derivation. (NHMRC/NRMMC, 2004). http://www.nhmrc.gov.au/publications/synopses/eh19syn.htm
Brazil	1.0 μg L^{-1} for microcystins 3.0 μg L^{-1} for saxitoxins (equivalents) 15 μg L^{-1} for cylindrospermopsin	Guideline values for microcystins, saxitoxins and cylindrospermopsin, along with biomass monitoring programs. Guideline value for microcystins adopted as mandatory. Guideline values for equivalents of saxitoxins and for cylindrospermopsin included as recommendations. Use of algicides prohibited and toxicity testing/toxin analysis is required when cell counts exceed 10,000 cells mL^{-1} or 1mm^3 L^{-1} biovolume. (Codd et al., 2005)

Country	Guideline Value/Standard	Comments/Explanations
Canada	1.5 μg L^{-1} cyanobacterial toxins as microcystin-LR MAC	Canada uses guidelines as the standard of water quality. The guidelines are expressed with the unit of Maximum Acceptable Concentration (MAC). These are derived from tolerable daily intake (TDI), which in turn is derived from a calculated no-observed adverse effect level (NOAEL) from data from human or animal studies. To derive a MAC from a TDI, adjustments are made for average body weight and drinking water consumption, as well as other considerations. In terms of health the guidelines ensure that the MACs are far below exposure levels at which adverse effects have been observed. For the case of cyanobacterial toxins the guideline is considered protective of human health against exposure to other microcystins (total microcystins) that may also be present. (Health Canada, 2002) http://www.hc-sc.gc.ca/ewh-semt/pubs/water-eau/doc_sup-appui/sum_guide-res_recom/index_e.html
Czech Republic	1 μg L^{-1} microcystin-LR	Value as national legislation, follows WHO provisional guideline value. (Codd et al., 2005)
China	1 μg L^{-1} microcystin-LR	WHO provisional guideline for microcystin-LR. (Codd et al., 2005)
France	1 μg L^{-1} microcystin-LR	Drinking water decree. (Codd et al., 2005)
Italy	0.84 μg L^{-1} total microcystins	WHO provisional guideline for microcystin-LR used as a reference by local authorities. (Codd et al., 2005)
Japan	1 μg L^{-1} microcystin-LR	WHO provisional guideline for microcystin-LR. (Codd et al., 2005)
Korea	1 μg L^{-1} microcystin-LR	WHO provisional guideline for microcystin-LR. Algal alarming system – based on cell numbers and chlorophyll a. (Codd et al., 2005)

Country	Guideline Value/Standard	Comments/Explanations
New Zealand	For cyanobacteria: <1 potentially toxic cyanobacterium present in 10 mL of sample. PMAV for cyanobacterial toxins: Anatoxin: 6.0 µg L^{-1} Anatoxin-a (S): 1.0 µg L^{-1} Cylindrospermopsin: 1.0 µg L^{-1} Homoanatoxin-a: 2.0 µg L^{-1} Microcystin-LR Toxicity Eq: 1.0 µg L^{-1} Nodularin: 1.0 µg L^{-1} Saxitoxins (as STX-eq) :3.0 µg L^{-1}	Maximum acceptable values (MAVs) are applied for micro-organisms or organic contaminants of health significance. Provisional MAV (PMAV) have been recommended for cyanobacterial toxins. MAVs are based on the WHO 'Guidelines for Drinking Water Quality'. They are the concentration of a determinand, which is not considered to cause any significant risk to the consumer over a lifetime of consumption of water. The method of derivation varies according to NZ conditions and the way in that the determinand presents a risk. However they are derived with the use of a TDI. The MAVs are standards in NZ. The Standards provide compliance criteria and compliance is routinely monitored. (Kouzminov, 2005). http://www.moh.govt.nz/moh.nsf/c7ad5e032528c34c4c2566690076db9b/70727db605b9f56a4c25696400802887?OpenDocument
Norway	1 µg L^{-1} microcystin-LR	Provisional WHO guideline for drinking water adopted. (Codd et al. 2005)
Poland	1 µg L^{-1} microcystin-LR	National legislation for guideline value in drinking water.
South Africa	0-0.8 µg L^{-1} for microcystin-LR	Guideline levels for microcystins in potable water as a "Target Water Quality Range". http://www.dwaf.gov.za/IWQS/wq_guide/
Spain	1 µg L^{-1} microcystins	National legislation, maximum permissible amount in drinking water. (Codd et al., 2005)
Thailand	No guideline currently	Awareness of the need for guidelines. (Codd et al. 2005)

Country	Guideline Value/Standard	Comments/Explanations
United States of America	No guideline currently.	Maximum Contaminant Levels (MCLs) are the highest level of a contaminant that is allowed in drinking water. They are enforceable standards. Cyanobacteria and their toxins are listed as microbiological contaminants on the contaminant candidate list (CCL). This means that they are currently recognised as unregulated contaminants, but are known to occur in public water systems and may require regulation under the Safe Drinking Water Act. Contaminants on the CCL are a priority for the US Environmental Protection Agency with the aim to set MCLs. http://www.epa.gov/safewater/mcl.html
Uruguay	Under review	(Codd et al. 2005)
World Health Organization	1 μg L^{-1} for microcystin-LR GV	World Health Organization (2004) http://www.who.int/water_sanitation_health/dwq/gdwq3/en/

Table 2. Status of guidelines or standards for cyanobacterial toxins in water used for recreation or bathing water for various countries.

Country	Guideline /Standard	Comments
Australia	Two Level Guideline Level 1: 10 μg L^{-1} total microcystins or >50,000 cells mL^{-1} toxic *M. aeruginosa* or biovolume equivalent of >4 mm^3 L^{-1} for the combined total of all cyanobacteria where a known toxin producer is dominant in the total biovolume. Level 2: either the total biovolume of all cyanobacterial material exceeds 10 mm^3 L^{-1} or scums are consistently present	The definitions of the levels are as follows: Level 1: Probability of adverse health effects due to known toxins Level 2: Probability of adverse health effects due to high levels of cyanobacterial material where known toxins are not present Closure is recommended at either level A 3-stage Alert Level Framework is used for situation assessment up to the guideline level. (NHMRC, 2006)
Germany	Three Level Guideline Level 1: <10 μg L^{-1} microcystins Level 2: >10 - <100 μg L^{-1} microcystins Level 3: >100 μg L^{-1} microcystins	The definitions and recommended actions for each of the levels are as follows: Level 1: Monitor cyanobacteria in a routine surveillance program (14-d) Level 2: Publish warnings, discourage bathing, and consider temporary closure. Level 3: Publish warnings, discourage bathing, temporary closure recommended. (Chorus, 2005a)

Country	Guideline /Standard	Comments
Netherlands	20 µg microcystin-LR L^{-1}	Note that this value is not a mandatory standard. Provinces use the guideline in a management framework as follows: MC-LR > 10 µg L^{-1}: issue warning MC-LR > 20 µg L^{-1}: issue warning and continue monitoring; if levels are persistently high close the bathing facility Presence of scums: at least a warning and continued monitoring (Ibelings, 2005)
World Health Organization	Three Level Guideline Level 1: 20,000 cyanobacterial cells mL^{-1} Level 2: 100,000 cyanobacterial cells mL^{-1} Level 3: Presence of scums	The definitions of the levels are as follows: Level 1: Relatively low probability of adverse health effects Level 2: Moderate probability of adverse health effects Level 3: High probability of adverse health effects The recommendation at Level 3 is for 'Immediate action to control scum contact' (Chorus & Bartram, 1999)
France	Three Level Guideline Level 1: 20,000 cyanobacterial cells mL^{-1} Level 2: >20,000 -<100,000 cyanobacterial cells mL^{-1} Level 3: Presence of scums	The actions the levels are as follows: Level 1: Monitoring intensified to fortnightly Level 2: Microcystins analysed. If > 25 µg MC-LR eq L^{-1}, swimming is prohibited Level 3: All activities are prohibited

The countries that have adopted the WHO provisional guideline for microcystin-LR for drinking water directly include the Czech Republic, France, Japan, Korea, New Zealand, Norway, Poland, Brazil and Spain.

Chorus (2005b) provides a summary of the application of the guideline in a range of European countries: "Polish regulations require a limit of 1 μg L^{-1} to be met for microcystin-LR in drinking-water. Czech legislation requires monitoring of tap water for microcystin-LR with a limit of 1 μg L^{-1}, and an update of the ordinance is expected in 2005 which will include alternatives to microcystin analysis such as quantification of cyanobacterial biomass in raw water or bioassays in conjunction with cell counts, requiring toxin analyses only if thresholds for cyanobacterial biomass are exceeded. The French Drinking-water Decree includes a maximum limit of 1 μg L^{-1} microcystin-LR, with analyses being required in the event of cyanobacterial proliferation in the raw water. The Spanish decree establishing the water quality criteria for human consumption includes a limit for "microcystin" (variants not specified) of 1 μg L^{-1}, to be reviewed at 5-year intervals, with sampling regimes specified in relation to size of population served.

In two countries, the provisional WHO Guideline value for microcystin-LR is important for implementation of regulations which do not explicitly address microcystins: In Germany, and very similarly in Finland, the national Drinking-water Ordinances stipulate that drinking-water should contain no substances in concentrations that may be harmful to human health, and the provisional WHO value for microcystin-LR provides an important definition of such concentrations. A prerequisite for this approach was that drinking-water suppliers using surface water run long-established phytoplankton monitoring programmes as basis for adapting treatment to raw water quality, usually have effective treatment in place, and are aware of the cyanotoxin hazard. In Italy also, no limit value has been implemented, but the national drinking-water decree considers algae as an accessory parameter to be monitored in case local authorities suspect a risk to human health, with the provisional WHO Guideline of 1μg L^{-1} micocystin-LR used as basis for this assessment.

In Hungary: the decree on drinking-water quality and the ordinance on monitoring include cyanobacteria among biological parameters to be monitored by microscopy, though no limit is given for cyanotoxins, only for the number of cyanobacterial cells. In Finland, starting in the late 1980's the waterworks have also been advised to monitor cyanobacteria microscopically, and if cyanobacterial cells occur in raw or treated water, to analyse toxins."

In addition, other countries (e.g. Australia and Canada) have decided to develop slight variants of the guideline based upon their local require-

ments. For example, Australia has developed a guideline for Total Microcystins of 1.3 μg L^{-1}, expressed as toxicity equivalents of microcystin-LR (NHMRC/NRMMC, 2004). This derivation has essentially used the same animal studies, the TDI and derivation convention as WHO which treats microcystins as a threshold (non-cancer) toxicant. The Australian Guidelines however is for total microcystins and the rationale for this is that blooms of *Microcystis aeruginosa,* which is the most common toxin-producing cyanobacterium in Australia, can contain a wide range of variants of microcystin in varying amounts. Experience indicates that the number of variants in an individual sample can range from a few to up to more than 20 in some cases. It is the cumulative toxicity of the microcystins in total that represents the potential hazard to human health from ingestion via drinking water. Therefore the unit recommended for the quantitative expression of this cumulative toxicity in the guideline is total microcystins expressed as toxicity equivalents of microcystin-LR. There are some issues for compliance monitoring in relation to this guideline, particularly in relation to the availability of analytical standards for microcystins and for the selection of appropriate analytical methods, and these are discussed in Nicholson and Burch (2001).

The WHO recognized that the recommendation of a guideline for a single microcystin congener (microcystin-LR) where more than 80 variants are known could make it problematic to express and interpret quantitative results from analysis or assays for these other toxins in relation to the guideline value. A discussion about the calculation and expression and limitations of the interpretation of microcystin concentrations other than microcystin-LR, in terms of "concentration equivalents (CE)" and "toxicity equivalents (TE)" is given by Falconer, et al. (1999).

In 1999, Canada set a *maximum accepted concentration* (MAC) for microcystin-LR in drinking-water of 1.5 μg L^{-1}, and research by Health Canada has been addressing the need to comprehensively include other microcystin variants in surveys and in monitoring (Health Canada, 2002). Brazilian Federal legislation is perhaps the most comprehensive and includes a mandatory standard of 1 μg L^{-1} for microcystins (variants not specified), and recommendations are given for saxitoxins (3 μg L^{-1}) and for cylindrospermopsin (15 μg L^{-1}) (Azevedo 2005).

Guidelines for cyanotoxins and/or cyanobacterial cell numbers for recreational waters are in place in a number of countries (Table 2). The World Health Organization considered that for recreational waters a single guideline value for cyanobacteria or cyanotoxins is not appropriate (Chorus and Bartram 1999; WHO 2003). Rather, a series of guideline values associated with incremental severity and probability of health effects were defined at

three levels based upon cyanobacterial cell densities and the presence of scums at the upper level (Table 2).

Other national guidelines for recreational water are also related to risk of adverse outcomes from ingestion of known toxins (Netherlands: 20 μg microcystin L^{-1}). On the other hand, the new Australian guideline has 2 levels defined by probability of adverse outcomes based upon microcystin ingestion (Level 1: 10 μg microcystin L^{-1}) and also from the probability of adverse health effects due to high levels of cyanobacterial material where known toxins are not present (Level 2: Total biovolume of all cyanobacterial material exceeds 10 mm^3 L^{-1} or scums) (Table 2).

Australia has also developed Livestock Drinking Water Guidelines for Cyanobacteria. These form part of the Australian and New Zealand Guidelines for Fresh and Marine Water Quality (ANZECC/ARMCANZ 2000). The guidelines for livestock are referred to as trigger values, which have the following definition and application: "Below the trigger value there should be little risk of adverse effects on animal health. Above the trigger value, investigations are recommended (e.g. of other factors such as age, condition, other dietary sources) to further evaluate the situation" (ANZECC/ARMCANZ 2000). The trigger values were developed using data on chronic and toxic effect levels on animals, taking into consideration animal weights, percentage intake from water, and safety factors for data not specific to the species. The summarised advice in the livestock drinking water guidelines is as follows: "Algal blooms should be treated as possibly toxic and the water source should be withdrawn from stock until the algae are identified and the level of toxin determined. An increasing risk to livestock health is likely when cell counts of *Microcystis* exceed 11,500 cells mL^{-1} and/or concentrations of microcystins exceed 2.3 μg L^{-1} expressed as microcystin-LR toxicity equivalents. There are insufficient data available to derive trigger values for other species of cyanobacteria" (ANZECC/ARMCANZ 2000). The guidelines provide individual derivations of trigger values for cattle, sheep, pigs, chickens and horses which take into account interspecies sensitivity.

Although there are no national guidelines for cyanotoxins in fish or shellfish in Australia, a 'health alert' level has been derived for toxins in fish, prawns and mussels in the state of Victoria, Australia (Van Buynder et al. 2001). The health alert level for microcystins and nodularin toxins in seafood is as follows: fish (250 μg kg^{-1}), prawns (1,100 μg kg^{-1}) and mussels (1,500 μg kg^{-1}). They were derived by determining a tolerable daily intake level for adults and modified to be protective for short-term exposure.

Role of Risk Assessment in Guideline Development

All of the international guidelines developed so far for microcystin in drinking-water have been based upon the World Health Organization's (WHO) provisional guideline of 1 μg L^{-1} for microcystin-LR released in 1998.

The derivation of this guideline is based upon data that there is reported human injury related to consumption of drinking water containing cyanobacteria, or from limited work with experimental animals. It was also recognised that at present the human evidence for microcystin tumor promotion is inadequate and animal evidence is limited. As a result the guideline is based upon the model of deriving a Tolerable Daily Intake (TDI) (i.e., Reference Dose; RfD) from an animal study No Observed Adverse Effects Level (NOAEL), with the application of appropriate safety or uncertainty factors. The resultant WHO guideline by definition is the concentration of a toxin that does not result in any significant risk to health of the consumer over a lifetime of consumption (WHO, 2004). Briefly the details of the calculation of the guideline are given in Chorus and Bartram (1999):

> "A 13-week mouse oral (by gavage) study with pure microcystin-LR is considered the most suitable for the derivation of a guideline value for microcystin-LR. In that study, a NOAEL of 40 μg/kg bw/day was determined, based on liver histopathology and serum enzyme level changes (Fawell et al., 1994). By applying a total uncertainty factor of 1000 (10 for intra-species variability, 10 for inter-species variability and 10 for lack of data on chronic toxicity and carcinogenicity), a provisional TDI of 0.04 μg/kg bw/day is determined for microcystin-LR….which….was used in deriving a provisional guideline value".

As indicated, where other countries have developed guidelines for microcystins that differ numerically from the WHO provisional guideline, this is due to the application of different scaling factors in the calculation. For example the same ingestion study in mice was used to calculate the Australian guideline of 1.3 μg L^{-1} total microcystin (Table 1) the WHO provisional guideline of 1 μg L^{-1} microcystin-LR. The guidelines differ due to the incorporation of a different average body weight for an adult (70 kg versus 60 kg), and to a difference with regard to the proportion of the daily intake of microcystin being attributed to the consumption of drinking water. The proportion for the Australian situation is regarded to be 0.9, which is higher than 0.8 selected by WHO. This is due to what was regarded as lower potential for exposure from other environmental sources, such as contaminated bathing water or via dietary supplements potentially containing microcystins. Similarly the Canadian guideline of 1.5 μg L^{-1}

(Table 1) results from incorporation of different scaling factors in the calculation for body-weight (70 kg versus 60 kg), and assumption of daily water consumption of 1.5 L as opposed to 2 L used by WHO.

In relation to recreational water guidelines for cyanotoxins and cyanobacteria, a number of countries have developed guidelines based either upon microcystin concentrations or equivalent cell densities of cyanobacteria. These guidelines essentially use a similar derivation process or are a translation from the drinking water guidelines for microcystin LR, while accounting for different exposures in recreational or bathing situations.

The World Health Organization was first to review the basis for guidelines for recreational water environments (WHO 2003). They recommended that in developing guidelines for cyanobacteria in freshwater the approach should consider:

1. The occurrence of cyanobacteria in general (in addition to known toxins) as part of the hazard: as it is not clear that all known toxic components have been identified and irritation symptoms reported may be due to these unknown substances.
2. The particular hazard due to the well known microcystin toxins.
3. The hazard associated with the characteristic or tendency for the occurrence of a heterogeneous distribution of many cyanobacterial populations in freshwater environments, which can result in the potential for scum formation.

The potential health effects were considered to be in two classes:

- chiefly irritative symptoms caused by unknown cyanobacterial substances
- the potentially more severe hazard of exposure to high concentrations of known cyanotoxins, particularly microcystins.

The result of this review was that they indicated that a single guideline value is not appropriate. Rather, a series of guideline values associated with incremental severity and probability of health effects were defined at three Levels (see Table 2). While these cell densities for the three Levels are not specifically indicated as being for toxic *Microcystis aeruginosa*, the Level 2 densities are cross-correlated to microcystin concentrations at 'the level of 20 μg microcystin Litre^{-1} which is equivalent to 20 times the WHO provisional guideline value concentration for microcystin-LR in drinking-water (WHO 1998) and would result in consumption of an amount close to the tolerable daily intake (TDI) for a 60 kg adult consuming 100 mL of water while swimming (rather than 2 litres of drinking-water)'.

A similar approach has been followed by the Netherlands, which is described by Ibelings (2005). This involved using the tolerable daily intake for microcystins (MC-LR < 0.04 μg per kg bodyweight), and "assuming that a swimmer ingests 100 mL of water and thereby calculating an exposure limit of 20 μg MC-LR L^{-1} of bathing water (Table 2). While the derivation assumes that bathing occurs 365 days per year – it is recognised that a more likely exposure would be less than 35 days (Ibelings 2005).

The recent Australian approach for guideline derivation (NHMRC, 2006) is somewhat different from the WHO and recommends a two-level guideline based upon:

1. Level 1: The probability of adverse health effects from ingestion of known toxins, in this case based upon the toxicity of microcystins.
2. Level 2: The probability of increased likelihood of non-specific adverse health outcomes, principally respiratory, irritation and allergy symptoms, from exposure to very high cell densities of cyanobacterial material irrespective of the presence of toxicity or known toxins.

The Level 1 guideline uses animal toxicity data for microcystin toxins and conventional toxicological calculations to derive a guideline for short-term (14-day) exposure to microcystins via ingestion for both children and adults based upon typical bodyweights (Table 2). The derivation of the guideline selects the LOAEL from the 44-day pig study (Kuiper-Goodman et al, 1999) as the most suitable data to derive a shorter-term exposure LOAEL (i.e., 14 days) that is representative of a period of repeated daily exposure for an uninterrupted period of up to 2 weeks. This is regarded as a likely, but albeit rather intense, continuous exposure for swimming and aquatic recreation in a summer season (e.g., a holiday period exposure).

The second guideline level (Level 2) is recommended for circumstances where high cell densities or scums of 'non-toxic' cyanobacteria are present, i.e. where the population has been tested and shown not to contain known toxins (microcystin, nodularin, cylindrospermopsin or saxitoxins). In this case where the microcystin-related biovolume guideline is exceeded, and there are no microcystins or other toxins present, it is felt appropriate to issue warning where either the total biovolume of all cyanobacterial material exceeds 10 mm^3 L^{-1} or scums are consistently present, i.e. are seen at some time each day at the recreational bathing site. This guideline level is recommended based upon the work of Stewart (2004), where it was shown that there was an increase in likelihood of symptom reporting in bathers above this approximate biovolume. The potential

symptoms reported above this level of cyanobacteria were primarily mild respiratory symptoms Stewart (2004).

Need for Additional Guidelines

As discussed, it is clear that considerable and growing international attention has been given to development of guidelines for cyanotoxins in drinking water and to a lesser extent for cyanobacterial numbers and toxins in recreational waters. Internationally there has been a "pronounced demand" for the WHO to develop a guideline for cylindrospermopsin (Chorus, 2005a), to provide authoritative guidance for countries to develop a risk-based approach to health hazards from cyanotoxins. The WHO guidelines provide a scientifically-substantiated target with a transparent explanation of the health-based considerations that lead to derivation of the value.

In relation the need for development or adoption of regulations for the US, an important part of the process is to carry out further surveys as part of hazard and exposure assessments for the various toxin types that have been found to occur. The information base for these assessments is currently being assembled from a range of studies in the US. In the context of Hazard Identification, a recent review of cyanotoxin occurrence in the US by Burns (2005) indicated that cyanobacteria are common in surface waters throughout the USA and that toxins have been detected in water bodies in the states of Florida, Indiana, Nebraska, Missouri, New York and Wisconsin, and in the Great Lakes. In addition a survey over 1996-1998, twenty-four public water systems in the USA and Canada were surveyed for microcystins (AwwaRF 2001). The results indicated that 80% of the 677 samples tested were positive for microcystin. A range of other investigations and surveys (e.g. SJRWMD 2001) have detected microcystin, cylindrospermopsin and anatoxin-a in finished drinking water, whereas saxitoxins have been found in freshwater cyanobacteria (Burns 2005). The surveys of toxin occurrence from New York State and the lower Great Lakes System reported by Boyer (see this volume) also support this incidence trend, where microcystin were by far the most common toxin (38% of samples tested), followed by anatoxin (8%), with a relatively low incidence of cylindrospermopsin (2%).

Certainly with regard to the need for guidelines, the incidence of microcystins, cylindrospermopsin and anatoxin in public water supply would suggest these are priority compounds for consideration for regulation in the US.

In relation to the use of water for other than drinking, some countries have developed guidelines for livestock-drinking protection (e.g., Australia: "trigger levels": for microcystins only) and local regulations for seafood harvesting (Victoria, Australia) (Burch and House, 2005). It is considered however, that there is currently insufficient information to derive sound guidelines for the broader range of toxins that could be present for the use of water contaminated by cyanobacteria or toxins for irrigated agricultural production, fisheries and aquaculture. The potential for algal toxin, odour or tainting residues in certain produce or commodities is an issue for agricultural activities which use contaminated water for irrigation or processing. Aquaculture and fish harvesting are particularly susceptible to residues, and this industry needs to be aware of algal blooms and their potential importance. The knowledge gaps in this area pose a business risk for these industries. The probability of this contamination may be quite low, and any possible public health risk may be small to negligible, however the damage to reputation is potentially more significant. This is because of the negative emotive image of produce contaminated with toxins and the absence of any guidance on acceptable background levels of residues or MCLs for these compounds in food (with the exception of PSPs in shellfish from a marine situation).

The other important area that requires further research is the issue of toxins and ecosystem protection. This is a developing research area, where more information on impact of toxins is required (See Ibelings this volume).

Research Needs

Additional research is required to support guideline development, including both short-term and whole-of–life animal studies with each of the known cyanotoxins. In view of the animal studies that indicate that microcystins may act as tumor promoters, and also some evidence of genotoxicity and carcinogenicity for cylindrospermopsin, it may be appropriate to carry out whole-of-life animal studies with both toxicity and carcinogenicity as end-points.

As part of the expert review by WHO in 1997, prior to the development of the guideline for microcystin-LR, it was revealed that there was insufficient information available for calculation of a TDI for most of the cyanotoxins including nodularin, cylindrospermopsin, anatoxin-a, homoanatoxin-a, anatoxin-a(S), and saxitoxins (Kuiper-Goodman et al. 1999). There has however been a recent sub-chronic, repeat animal oral-

toxicity assessment study for cylindrospermopsin which can be used to determine a NOAEL, which in turn has been used to calculate a TDI and propose a drinking water guideline for cylindrospermopsin (Humpage and Falconer 2003).

In relation to microcystins, it is known that there are a large number of congeners, and the toxico-dynamics and kinetics of these variants are not well understood. Further research is needed to consider the approach to take in formulating health advisories or regulations for toxin mixtures, i.e. multiple microcystins, or mixtures of toxin types.

Applicability of International Guidelines to the United States

The cyanotoxins must be subjected to the same complete risk assessment procedure as any other environmental contaminant prior to the development of regulations in the US. This risk assessment includes the four steps of

1. Hazard identification;
2. Dose-response assessment;
3. Exposure assessment; and
4. Risk characterization,

with risk characterization being the transitional step to risk management (ref - NCEA website). The suitability of existing regulations for use in the US needs to be considered in the context of the above assessment process.

Current efforts and investigations in the US are strongly focussed on collecting information and developing knowledge in the area of hazard identification. In addition proposals are well advanced for research on dose-response assessment. However, very little has been done in a coordinated way with regard to exposure assessment and risk characterization.

In the above context then, none of the current international guidelines for toxins in drinking water would appear to be immediately suitable for adoption as regulations in the US. The major limitations that need to be overcome include: the capacity to deal with multiple toxin congeners, the absence of robust analytical methods for compliance monitoring, and the absence of certified toxin standards to support analyses.

The limitation of the WHO guideline for Microcystin-LR in relation to its use as a regulation is that it does not deal with congeners other than microcystin-LR. This is of course why WHO have recommended it as ‘provisional’. Consideration needs to be given to the issue of mixtures and the

incidence of the most common microcystin congeners in the US for regulation development.

The second issue for both the US and indeed international context is the requirement for appropriate and robust compliance monitoring and analytical protocols to accompany to any regulations. Although there is considerable analytical capability for cyanotoxins within the research community, it cannot be said that there are that rapid and economical screening or quantitative analytical methods that are readily available to the water industry. Organisations such as AwwaRF and the EPA are supporting research to develop validated methods. In this context there are also no validated analytical standards available commercially for the major toxins, i.e. microcystins, cylindrospermopsin and Anatoxin-a, although this may change in the near future.

The current WHO provisional guideline for microcystin, or the other national guideline variants that are based upon it, (e.g., Canadian, Australian) may be appropriate to adopt as a health advisory. The advantage of having a health advisory as a target is that it will assist in the development of a database on occurrence of cyanotoxins and contribute complementary information on exposure assessment, where local or state-based monitoring programs become established.

The bathing and recreational water guidelines developed in other countries could also be translated for the protection of public health in recreational situation in the US, by whatever state mechanisms are appropriate. The WHO guidelines that are widely used in Europe have provided a useful target for local authorities to manage hazards from cyanobacterial blooms in recreational water situations. The recent Australian guidelines for cyanobacteria in bathing waters (NHMRC 2006) are the most recent review of recreational water hazards from toxic cyanobacteria. These guidelines are evidence–based and deal with exposure to both toxic and non-toxic cyanobacteria, and also provide recommendations on sampling strategies and development of staged monitoring programs.

Predictive Models

The occurrence of CHABs and their toxins are stressors that are multifacetted in impact. They impact upon humans, ecosystems and biota, and natural resources in interdependent ways. Regulations and guidelines need to consider the impact upon human health via various exposure pathways (drinking water, food, recreational exposure). Similarly the integrated economic, environmental and social impact of CHABs should somehow be re-

flected in the formulation of ecosystem protection guidelines for CHABs. In this context the integrated human health and ecological risk assessment models proposed by Orme-Zavaleta and Munns (2007) offer potential to characterise the human-environment risk assessment for a stressors such as CHABs and toxins. The models offer ways to consider linkages of stressors and impacts in complex systems, and may provide a useful framework to apply to CHABs.

Conclusion and Summary

A number of countries have developed regulations or guidelines for cyanotoxins and cyanobacteria in drinking water, and in some cases in water used for recreational activity and agriculture. The main focus internationally has been upon microcystin toxins, produced predominantly by *Microcystis aeruginosa*. This is because microcystins are widely regarded as the most significant potential source of human injury from cyanobacteria on a world-wide scale. Many international guidelines have taken their lead from the World Health Organization's (WHO) provisional guideline of 1 $\mu g\ L^{-1}$ for microcystin-LR in drinking-water released in 1998 (WHO 2004).

The WHO guideline value is stated as being 'provisional', because it covers only microcystin-LR, for reasons that the toxicology is limited and new data for toxicity of cyanobacterial toxins are being generated. The derivation of this guideline is based upon data that there is reported human injury related to consumption of drinking water containing cyanobacteria, or from limited work with experimental animals. It was also recognised that at present the human evidence for microcystin tumor promotion is inadequate and animal evidence is limited. As a result the guideline is based upon the model of deriving a Tolerable Daily intake (TDI) from an animal study No Observed Adverse Effects Level (NOAEL), with the application of appropriate safety or uncertainty factors. The resultant WHO guideline by definition is the concentration of a toxin that does not result in any significant risk to health of the consumer over a lifetime of consumption.

Following the release of this WHO provisional guideline many countries have either adopted it directly (e.g., Czech Republic, France, Japan, Korea, New Zealand, Norway, Poland, Brazil and Spain), or have adopted the same animal studies, TDI and derivation convention to arrive at slight variants based upon local requirements (e.g., Australia, Canada). Brazil currently has the most comprehensive federal legislation which includes a mandatory standard of 1 $\mu g\ L^{-1}$ for microcystins, and also recommendations for saxitoxins (3 $\mu g\ L^{-1}$) and for cylindrospermopsin (15 $\mu g\ L^{-1}$).

Although guidelines for cyanotoxins and cyanobacterial cell numbers for recreational waters are in place in a number of countries, it is considered that there is currently insufficient information to derive sound guidelines for the use of water contaminated by cyanobacteria or toxins for agricultural production, fisheries and ecosystem protection.

In relation to the need for specific regulations for toxins for the US, the surveys that have been carried out to date would indicate that the priority compounds for regulation, based upon their incidence and distribution, are microcystins, cylindrospermopsin and Anatoxin-a.

Additional research is required to support guideline development, including whole-of-life animal studies with each of the known cyanotoxins. In view of the animal studies that indicate that microcystins may act as tumor promoters, and also some evidence of genotoxicity and carcinogenicity for cylindrospermopsin, it may be appropriate to carry out whole-of-life animal studies with both toxicity and carcinogenicity as end-points. In relation to microcystins, it is known that there a large number of congeners, and the toxico-dynamics and kinetics of these variants are not well understood. Further research is needed to consider the approach to take in formulating health advisories or regulations for toxin mixtures, i.e. multiple microcystins, or mixtures of toxin types.

An important requirement for regulation is the availability of robust monitoring and analytical protocols for toxins. Currently rapid and economical screening or quantitative analytical methods are not available to the water industry or natural resource managers, and this is a priority before the release of guidelines and regulations.

There is insufficient information available in a range of the categories usually required to satisfy comprehensive risk assessment process for the major toxins to currently adopt any of the international guidelines as regulations in the US. The major limitations that need to be overcome include: the capacity to deal with multiple toxin congeners, the absence of robust analytical methods for compliance monitoring, and the absence of certified toxin standards to support analyses.

However, the current WHO provisional guideline for microcystin-LR, or the other national guideline variants that are based upon it, (e.g., Canadian, Australian) may be appropriate to adopt as a health advisory in the short-term, while regulations are developed. The bathing and recreational water guidelines developed in other countries could also be translated for use as recreational water guidelines situation in the US.

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