ENVIRONMENTAL BIOTECHNOLOGY 10 (1) 2014, 26-43; doi:10.14799/ebms246

Neurotoxicity of cyanobacterial toxins

Maciej Florczyk, Alicja Łakomiak, Maciej Woźny, Paweł Brzuzan

Department of Environmental Biotechnology, Faculty of Environmental Sciences, University of Warmia and Mazury in Olsztyn, Poland

Corresponding author: Maciej Florczyk, Department of Environmental Biotechnology, Faculty of Environmental Sciences, University of Warmia and Mazury in Olsztyn, Słoneczna 45G, 10-709 Olsztyn, Poland; Phone: +48 89 523 4164; Fax: +48 89 523 4131; E-mail: maciej.florczyk@uwm.edu.pl

Keywords: blue-green algae, cyanotoxins, drugs, medicine, microcystin, neurotoxicity

Received in July 2014. Published in December 2014.

ABSTRACT

Eutrophication of marine- and fresh-waters can lead to excessive development of cyanobacterial blooms, which may contain strains that produce toxins. These toxins are secondary metabolites which can accumulate in the food chain and contaminate drinking water, thus posing a potential threat to the health of humans and aquatic organisms. These toxins include a variety of compounds with different mechanisms. This review focuses on the neurotoxicity of microcystin and other cyanotoxins. Although the hepatotoxic action of microcystins is commonly known, its neurotoxic effects have also been described, e.g. oxidative stress, cytoskeletal changes and changes in protein phosphatase activity. These effects have been partially explained by the discovery in the blood-brain barrier of the same membrane transporters involved in microcystins hepatotoxic mechanisms. Additionally, this paper reviews other cyanotoxins that are known or suspected to target cholinergic synapses and voltage-gated channels, including anatoxin-a, anatoxin-a(s), antillatoxins, cylindrospermopsin, homoanatoxin-a, jamaicamide, kalkitoxin and saxitoxins. The neurotoxic and cytotoxic effects of the cyanotoxins discussed here are of particular interest because of their pharmacological potential. This review also discusses the potential of these compounds to serve as drugs for cancer and central nervous system failure.

ABBREVIATIONS

Ach	acetylcholine
AchE	acetylcholinesterase
AldH	aldehyde dehydrogenase
ANTX-a	anatoxin-a
ANTX-a(s)	anatoxin-a(s)
ATX	antillatoxin
BMAA	L-β-N-methylamino-L-alanine
CoA	coenzyme A
CYN	cylindrospermopsin
EGFR	epidermal growth factor receptor
HSP	heat shock protein
MC	microcystin

© UNIVERSITY OF WARMIA AND MAZURY IN OLSZTYN

mWBC	murine whole brain cells
NADH	nicotinamide adenine dinucleotide
NMDAR	N-methyl D-aspartate receptor
NOD	nodularin
Oatp/OATP	organic anion transporting polypeptide
PPs	protein phosphatases
PSP	paralytic shellfish poisoning
ROS	reactive oxygen species
SLCO	organic anion transporting family
STX	saxitoxin
VGSC	voltage gated sodium channels

INTRODUCTION

Cyanobacteria are important component of phytoplankton. They occur in almost every freshwater and marine environment. Cyanobacteria constitute up to 70% of the total phytoplankton biomass and they produce more than 30% of total free O_2 and account for more than 30% of total primary production (Paerl et al. 2001).

Accumulation of cyanobacteria in eutrophic conditions may lead to toxicity of affected waters. Among phytoplankton, cyanobacteria have the greatest share in the overall production of toxins in aquatic ecosystems (Paerl et al. 2001). Cyanobacteria toxins may affect organisms by direct absorption from water or via ingestion (Cazenave et al. 2005) (Figure 1). Several cyanobacterial toxins can be accumulated by and transferred through the members of food webs (Figure 1), which may lead to short- or long-term destabilization of natural systems (Christoffersen 1996). These toxins also cause organ damage, cancers or even human deaths. The problem of cyanobacterial blooms is a growing concern worldwide as the abundance of cyanotoxin producing bacteria continues to expand with each summer (Merel et al. 2013). This review focuses on the mechanisms and effects of neurotoxic cyanotoxins, such as microcystins. It describes the results of recent research and what still remains to be determined.

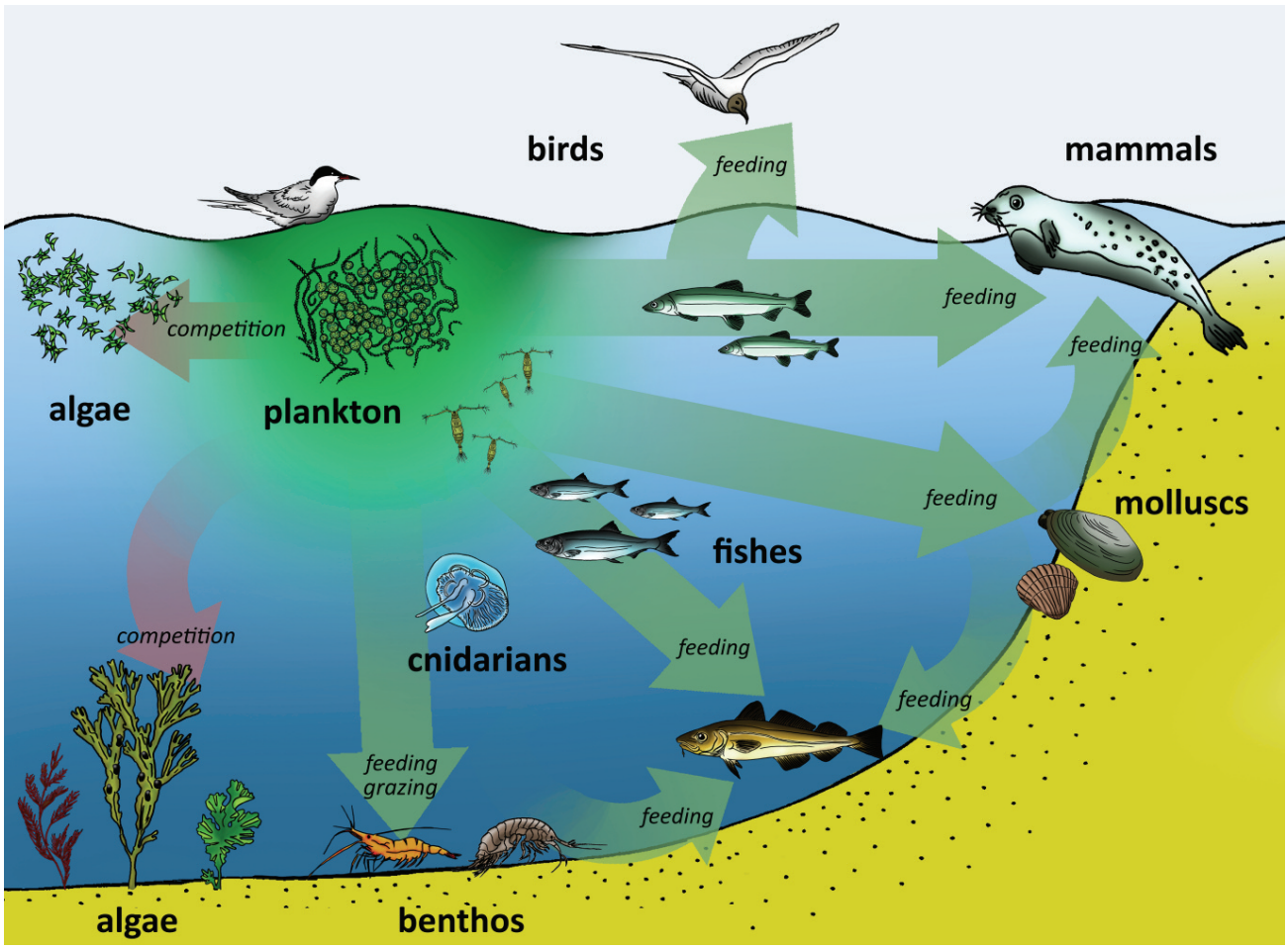


Figure 1. Cyanotoxins in aquatic environment. Cyanotoxic strains and toxic secondary metabolites affect different aquatic organisms. The cyanobacteria compete with other algae species (red arrows), and their toxins are harmful (green arrows) to plankton-feeding organisms (e.g. zooplankton, benthos, fishes), as well as predators feeding on intoxicated organisms. Presence of cyanotoxins in drinking water may pose a risk for humans.

CYANOTOXINS

Cyanotoxins are produced within cells and are only released into water during cell senescence or cell death and lysis (Sivonen and Jones 1999). The latter may be caused by water treatment processes such as algaecide application (Babica et al. 2006; Gupta et al. 2001; James and Fawell 1991). It is known that blooms of different cyanobacterial species (e.g. *Anabaena circinalis* and *Microcystis aeruginosa*) have been observed to occur in the same water body but also may overlap when occurring in rapid succession to one after another (Dietrich et al. 2008). Moreover, many cyanobacterial species have been observed to be capable of producing several different toxin types as well as different toxin congeners.

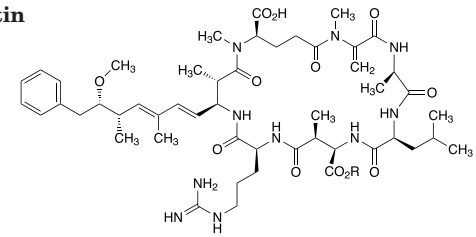
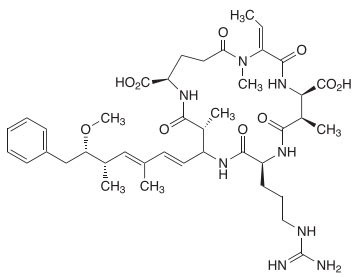
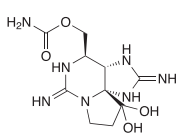
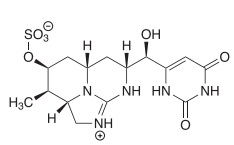
Numerous bioactive compounds produced by cyanobacteria have been described: non-ribosomal peptides, alkaloids, lipopeptides, esters, amino acids and polyketides. They all present a wide diversity of structures, variants and include potent toxins. Cyanotoxins are usually divided into groups according to the effect that they provoke. Hepatotoxins cause liver damage, cytotoxins are responsible for cell dysfunction and damage, neurotoxins cause damage to nervous tissue, whereas dermatotoxins are linked with allergenic reactions (Stewart et al. 2006).

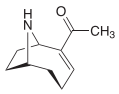
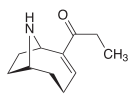
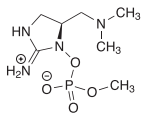
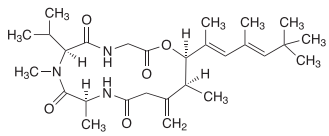
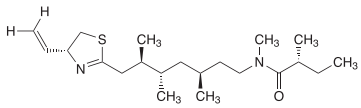
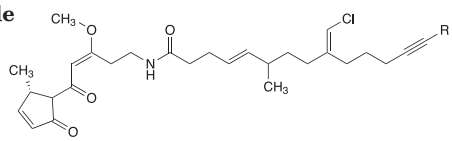
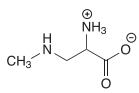
Nonetheless, the toxic effects of cyanotoxins are not straightforward. For example, exposure to microcystins may cause hepatotoxic effects (Gupta and Guha 2006; Li et al. 2005; Solter et al. 1998), as well as oxidative stress (Angeles et al. 2005), growth inhibition (Bury et al. 1995; Zhao et al. 2006), reproductive disorders (Ding et al. 2006) and kidney damage (Fischer and Dietrich 2000; Kotak et al. 1996). Many authors describe hematological and biochemical consequences (Malbrouck et al. 2003; Vajcova et al. 1998; Zhang et al. 2007) including apoptosis in a variety of cell types (McDermott et al. 1998). In case of sufficiently high doses, microcystins may cause even death (Lindholm et al. 1999). A detailed examination of Caruaru case in Brazil (1996) after which 60 patients died after severe intoxication

with diverse cyanobacterial toxins showed that affected humans not only suffered from liver failure but also exhibited symptoms of neurological disorders: dizziness, vertigo, tinnitus, mild deafness, visual disturbance, blindness, and grand mal convulsion (epilepsy), nausea, vomiting (Pouria et al. 1998).

This review focuses on cyanotoxins which are capable of inducing adverse effects in nervous tissue. For this purpose, we classified them by similarities in their chemical structure: cyclic peptides, alkaloids, phosphate esters, lipopeptides, amino-acids (Table 1). Here, we present cyanotoxins of proven neurotoxicity and those whose neurotoxicity has been suggested.

Table 1. Examples of adverse effects and stability in environment of the cyanobacteria toxins discussed in the article.

Toxin	Chemical structure	Organism (genera)	Adverse effects	Stability in the environment
Microcystin		*Microcystis* *Anabaena* *Oscillatoria* *Planktothrix* *Chroococcus*	Behavioral changes; Neuronal loss and morphology changes; Oxidative stress; Cytoskeletal changes: microtubule depolymerization, growth inhibition, axonal damage; Enchanced protein phosphate activity; Affection of aminopeptidase activity; Reduced level of apoptosis preventing proteins; Hyperphosphorylation of Tau protein; Disturbed energy generation and organic acid metabolism.	Stable compound, could persist even several months or years; UV and degrading bacteria speeds up removal.
Nodularin		*Nodularia*	Inhibition of PP1 and PP2A; Tumors promotion; Oxidative stress; Induction of apoptosis.	May persist in aquatic systems for weeks; Photodegradation speed up in the presence of turbidity and humic acid or humic substances.
Saxitoxin		*Alexandrium* *Pyrodinium* *Gymnodinium* *Anabaena* *Aphanizomenon,* *Cylindrospermopsis* *Lyngbya*	Blockage of sodium ion channels in nerve axon membrane; Blockage of calcium channels; Prolonged gating of the potassium channels; Nerve dysfunction; Paralysis; Damage of cellular homeostasis; Embryos edema and body curvature; Sensorimotor impairments and paralysis in herring larvae; Abnormal growth of embryos with high neurogenesis and cellular proliferation.	In water could persist over 3 months; In high temperatures it is degraded into more toxic variants.
Cylindrospermopsin		*Cylindrospermopsis* *Aphanizomenon* *Umezakia* *Rhaphdiopsis* *Anabaena* *Lyngbya*	Affection of protein synthesis, inhibition of glutathione and cytochrome P450; DNA damage; Potentially affection of nicotinergic ACh receptors of the neurones.	More than 90% of total toxin volume degrades within few days in sunlight and in the presence of cell pigments. Stable in the dark.

Toxin	Chemical structure	Organism (genera)	Adverse effects	Stability in the environment
Anatoxin-a		*Anabaena* *Cylindrospermum* *Microcystis* *Oscillatoria* *Raphidiopsis* *Planktothrix* *Aphanizomenon*	Potent agonists of the muscular and neuronal nicotinic acetylcholine receptor; Membrane depolarization and desensitization; Paralysis; Intoxication resulting in vomiting, respiratory difficulties, convulsion.	Unstable in alkaline water, where it undergoes rapid photochemical degradation into non-toxic form.
Homoanatoxin-a		*Oscillatoria*	Enhanced release of acetylcholine from peripheral cholinergic nerves through opening of endogenous voltage dependent neuronal L-type calcium channels.	No data
Anatoxin-a(s)		*Anabaena*	Irreversible inhibition of acetylcholinesterase from degradation of acetylcholine; Constant stimulation of muscle; Muscular paralysis.	No data
Antillatoxin		*Lyngbya*	Activation of the mammalian voltage-gated sodium channels; Up-regulating NMDAR function; Respiratory irritation; Eye inflammation; Contact dermatitis.	No data
Kalkitoxin		*Lyngbya*	Inhibition of the voltage-gated sodium channels.	No data
Jamaicamide		*Lyngbya*	Inhibition of the voltage-gated sodium channels.	No data
BMAA		*Cycas* *Trichodesmium*	Bind to glutamate receptors; Cause of neurodegenerative syndrome; Incorporation into proteins.	Could persist and accumulate in the environment due to incorporation into proteins.

STABILITY OF CYANOTOXINS IN AQUATIC ENVIRONMENT

The chemical stability of a toxic compound in the water environment is the key factor determining both its persistence in aquatic reservoirs and distribution within an organism. Cyclic peptide cyanotoxins are highly stable compounds, and after being released from cell, they may persist in aquatic systems for weeks (Edwards et al. 2008), several months or even years (Sivonen and Jones 1999). However, some studies report that sunlight UV or biodegradation may speed up their removal from the environment (Sivonen and Jones 1999). Results also demonstrate that the duration of photochemical degradation of microcystin and nodularin may vary from two to six weeks depending on the presence of water-soluble cell pigments (Tsuji et al. 1994; Welker and Steinberg 2000). However, the photolysis is an important

process only in shallow lakes, as the half-life of microcystins in deep lakes is longer than the season of cyanobacteria growth (Welker and Steinberg 2000). Recent studies by Thirumavalavan et al. (2012) which were conducted in laboratory conditions, revealed that the presence of turbidity and humic acid affected the photodegradation process of nodularin. Another study has shown, that in sea water the rate of nodularin photolysis could be accelerated by the presence of some cell components and humic substances (Welker and Steinberg 1999).

Alkaloidal cytotoxins exhibit various chemical stability. It is known, that saxitoxins are difficult to degrade and highly stable even after heating (Falconer and Humpage 2005). They can persist over 90 days in water, but they are degraded in high temperatures into more toxic variants (Sivonen and Jones 1999). A cylindrospermopsin, another cytotoxic alkaloid, is relatively stable in the dark. However, in sunlight and in the presence of cell pigments degradation occurs rapidly. More than 90% of total cylindrospermopsin volume degrades within 2–3 days (Chiswell et al. 1999). Another alkaloid, anatoxin-a, is unstable in alkaline water, where it is transformed by sunlight into a non-toxic form. This neurotoxin is relatively stable in the dark, but it undergoes rapid photochemical degradation in sunlight, particularly in alkaline conditions and even in the absence of cell pigments (Smith and Sutton 1993; Stevens and Krieger 1991). Unfortunately, even though they can affect the health of the organisms, data on the chemical stability of many other cyanobacterial toxins are scarce.

GENERAL MECHANISMS OF CYANOBACTERIAL NEUROTOXICITY

Aquatic animal health problems are most likely related to chronic exposure to variable concentrations of diverse cyanobacterial toxins through drinking of contaminated waters and eating contaminated food (e.g. fish, and molluscs; Figure 1). Besides the liver (a common target organ for many toxins), exposure to the cyanotoxins also affects other organs (Nagarajan et al. 2012). In recent years much attention has been paid to adverse effects of cyanotoxins on the nervous system of animals. The nervous system (NS) is a prime target for toxins which are known to rapidly disrupt functioning of whole organism. NS functions as a master communication network. Rapid communication within NS relies on the generation of two types of electrical signal. Dendrites respond to neurotransmitters released from adjacent neurons by generating slow depolarizing junctional potential which elicits an action potential. Then, it rapidly travels to the end of the axon where the neurotransmitter is released again (Kem 2000). Postsynaptic membrane of the muscle cell is able to generate an action potential in response to the acetylcholine (Ach) induced depolarization. An action potential involves activating and then deactivating two different ion channels: sodium-selective and potassium channels. In smooth muscles and many neurons sodium channels are substituted by voltage-gated calcium channels. Additionally calcium ions flowing through channels mediate the exocytotic release of neurotransmitter into the synaptic cleft at nerve terminals (Kem 2000).

Toxicity can only occur when toxins are transported into cells or interact with specific receptors or channels present on cells membrane (Stillwell 2013) (Figure 2). Thus almost every ion channel seems to be a potential target for natural toxins (Kem 2000). A known example involves Na^+ and K^+ channels (Figure 2), which are both facilitated diffusion carriers that conduct the cation down the ion's electrochemical gradient (Stillwell 2013). Neurotoxicity of saxitoxins (STXs) depends on blocking sodium ion channels in the nerve axon membrane (van Apeldoorn et al. 2007). In addition STXs are known to block calcium channels (Su et al. 2004). Ca^{2+} channels significantly coordinate with Na^+ channels in regulating signal transduction pathways, including cell cycle and apoptosis (Belkacemi et al. 2005). STXs also affect heart muscle cells by prolonging gating of the potassium channels, which can in turn lead to alteration in the influx of ions to the cell (Wang et al. 2003). In addition, the Na^+ channel blockage may alter the selective permeability of the membrane and may change the flow of ions, leading to damage of cellular homeostasis (Hille 1992). Jamicamide and kalkitoxin are another toxins possessing neurotoxic properties in sodium channel-blocking activity (Edwards et al. 2004; LePage et al. 2005). In contrast to above-mentioned toxins, antillatoxin is an activator of the mammalian voltage-gated sodium channels (VGSCs) that elevates intracellular Na^+ concentration in intact neurons (Cao et al. 2008; Li et al. 2001). However, its precise recognition site on the channel protein remains to be defined (Jabba et al. 2010).

Acetylcholine (Ach) is the most common neurotransmitter at synapses. It initiates an action potential, although it may also act as inhibitor of the signal at some synapses. Then activity of the Ach is terminated by the acetylcholinesterase (AchE). Other signals are transmitted by newly Ach released from presynaptic neuron. Anatoxin-a and homoanatoxin-a are potent agonists of the muscular and neuronal nicotinic acetylcholine receptor (Aráoz et al. 2010). Anatoxin-a has an affinity to the acetylcholine receptors without being degraded by acetylocholinesterase. The receptor channel remains active and in consequence a constant inflow of sodium ions to cells take place (Valério et al. 2010). Homoanatoxin-a enhance the release of acetylcholine from peripheral cholinergic nerves through opening of endogenous voltage dependent neuronal L-type calcium channels (Aas et al. 1996; Lilleheil et al. 1997). Anatoxin-a(s) irreversibly inhibits acetylcholinesterase from degradation of acetylcholine (Molica et al. 2005).

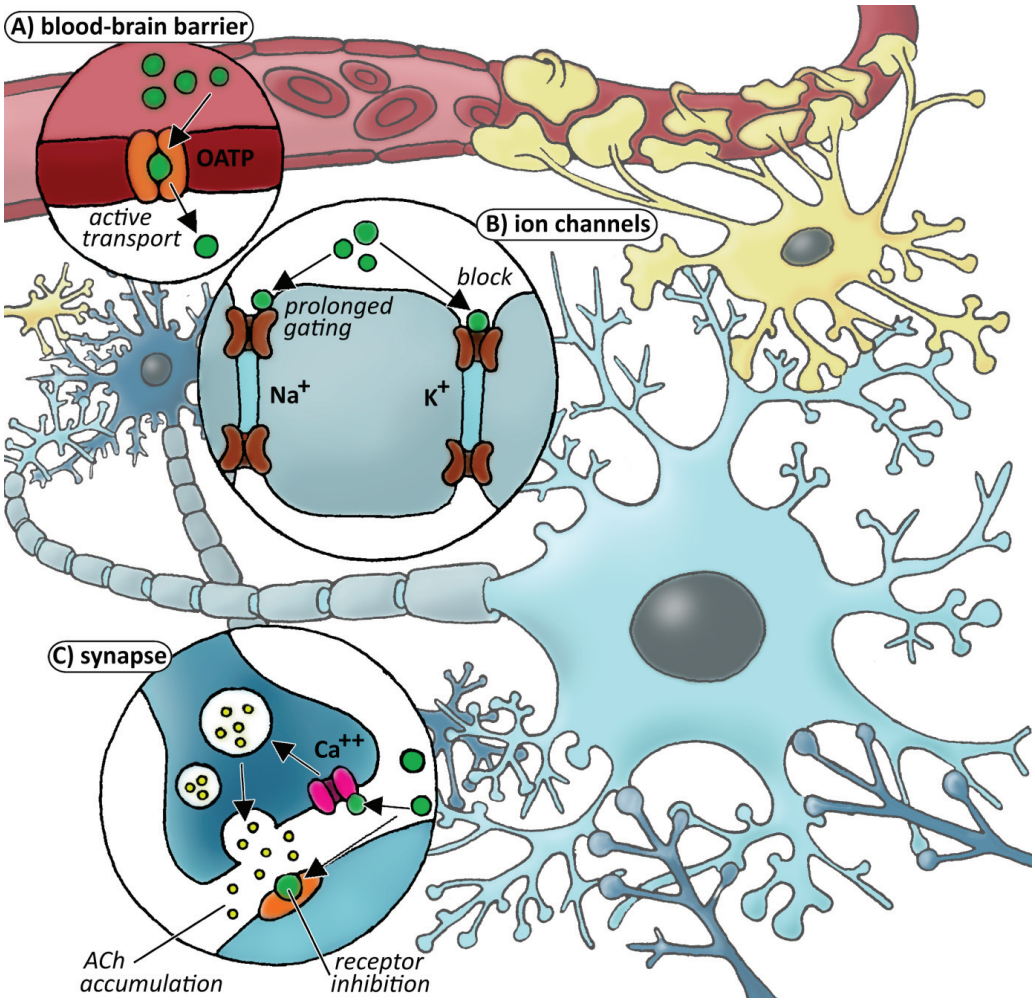


Figure 1. Schematic representation of targets of the central nervous system affected by cyanobacterial toxins (illustrated as green circles). A) blood-brain barrier; neurotoxicity of microcystins and probably nodularin depends on active transport from vessels into the brain carried by organic anion transporting polypeptides (OATPs). B) ion channels in cell membrane of neuron; many cyanotoxins (i.e. jamicamide, kalkitoxin, saxitoxin) may disturb flow of ions responsible for proper functioning of the nervous system. C) synapse; various locations and types of toxic actions triggered by synapse-targeting toxins (i.e. anatoxin-a, anatoxin-a(s), homoanatoxin-a). See text for more details.

Microcystins and nodularins which are similar in structure require active transport through cell membranes. In case of neurotoxicity another prerequisite is toxin being capable of passing blood-brain barrier/blood-cerebrospinal fluid barrier (Feurstein et al. 2010) e.g. via MC-transport competent Oatp/OATP or other yet unknown transporters. Organic anion transporting polypeptides (Oatps/OATPs) are members of the solute carrier organic anion transporting family (SLCO). Oatps/OATPs mediate uptake of a wide diversity of amphipathic organic compounds (bile salts, anionic peptides, steroid conjugates, thyroid hormones and an increasing number of pharmaceutical drugs and xenobiotics) which is independent of sodium ions (Hagenbuch and Meier 2004). It is known, that some of the Oatps/OATPs are expressed in various tissues, some occur just in one type of organ, and some occur in many, e.g. in the liver as well as in the brain (Fischer et al. 2005). The work of Feurstein et al. (2009) on primary murine whole brain cells (mWBC) showed the expression of five mOatps at the mRNA level. Oatp1b2 was thought to be specific to the liver in mice and rats but its transcripts (as well as proteins) are also present in the brain homogenates (Fischer et al. 2005). A study confirming that mOatps are involved in the neuronal uptake of MC was done by Feurstein et al. (2009, 2010).

Their research group tested for six mOatps in neuronal membranes and found that two of the proteins were present, mOatp1b2 and mOatp1a5.

NEUROTOXIC EFFECTS OF CYCLIC PEPTIDES

Microcystins

The group of cyclic peptides includes a wide number of toxins. One of these is microcystin (MC), which forms the most structurally diverse group among cyanotoxins (Table 1). Microcystins are described as monocyclic heptapeptides containing D-amino and L-amino acids plus N-methyldehydrolamine and a unique B-amino acid side-group (ADDA) (Neilan et al. 2010). The peptide rings contain seven amino acids, two of which are used to distinguish different structural variants of the microcystins.

The MCs are potent hepatotoxins produced by the cyanobacteria of the genera *Planktothrix*, *Microcystis*, *Aphanizomenon*, *Nostoc* and *Anabaena* (Kaasalainen et al. 2012; Neilan et al. 2010), and microcystin-LR, -RR or –YR are the most dominant variants found in the environment (Ettoumi et al. 2011). These cyclic heptapeptides have strong affinity to serine/threonine protein phosphatases (PPs) thereby acting as an inhibitor of this group of enzymes. Through this interaction a cascade of events responsible for the MC cytotoxic and genotoxic effects in animal cells may take place (reviewed in Campos and Vasconcelos 2010). Moreover, MCs induce oxidative stress in animal cells and together with the inhibition of PPs this pathway is considered to be one of the main mechanisms of MC toxicity. Microcystin-LR (MC-LR) is both the most toxic and the most commonly encountered member of MC family (Bogo et al. 2011).

Toxic effects of MCs are compound-specific. In general, MC-LR causes greater cytotoxicity than MC-RR (Huguet et al. 2013). The effects of the acute toxicity of microcystin-LR are different to chronic exposure where formation of reactive oxygen species (ROS) dominates, instead of the PP inhibition pathway (Wang et al. 2010a). A recent study done by Niedermeyer and coworkers (2014) has proven differences in potency between MC variants and above-mentioned studies, suggesting that *in vivo* toxicity is dependent on OATP transport kinetics rather than on differences in PP inhibition (Blom and Jüttner 2005; Fischer et al. 2010). However, Oatps/OATPs may differ in their ability to transport structurally varied MC congeners. There are suggestions that the Oatp/OATP which is not capable of transporting one given MC (e.g. MC-LR) may still be able to transport other types of MCs (Feurstein et al. 2009). Thus, the potential neurotoxicity of individual MC congeners should depend on functional expression of Oatps/OATPs in the blood-brain barrier and in the neuronal cell membrane.

Behavioral changes (such as different swimming habits and uncommon daily activity) are the first line of defense, when the animal is exposed to a hazardous environment, and indicate how the animal feels and responds (Begout Anras and Lagard`ere 2004). Possible neurotoxic effects have been reported in fish exposed to microcystin: changes in behavior, swimming activity or swimming performance (Baganz et al. 1998, 2004; Bogo et al. 2011; Cazenave et al. 2008). Baganz et al. (2004) compared zebrafish (*Danio rerio*) and belica (*Leucaspius delineatus*), using dissolved MC-LR in an environmental range between $0.5\mu g \cdot L^{-1}$ (which is the lowest observed effect concentration of MC-LR; Oberemm et al. 1997) and $50\mu g \cdot L^{-1}$. In both species, higher microcystin concentration caused significantly decreased locomotion during daytime, whereas lower concentration led to increased mobility (Baganz et al. 1998, 2004). Results showed different nocturnal activity in both species. The activity of *D. rerio* at higher, as well as lower concentrations was decreased, while that of *L. delineatus* was increased. Furthermore Baganz et al. (2004) found the latter species to be more sensitive to MC-LR: it reacted earlier and for a longer time, than zebrafish. These results show, that MC is capable of altering behavioral day and night rhythms, as *L. delineatus* is predominantly a diurnal fish.

In another study, Cazenave and coworkers (2008) examined the impact of microcystin-RR on a small omnivorous–planktivorous fish *Jenynsia multidentata*, and showed increased nocturnal activity at lower doses ($0.01\mu g \cdot g^{-1}$), whereas higher dosage ($1\mu g \cdot g^{-1}$) elicited no changes or it decreased swimming activity only during the last hours of exposure. Cazenave proposed that the hyperactivity of *J. multidentata* could be the consequence of fish attempting to escape from a contaminated environment, however it also could be a reaction caused by the presence of MC in the brain. In the work on a nematode *C. elegans* exposed to MC-LR, Ju et al. (2013) observed changes in locomotion behavior. They found neuronal loss and morphology changes in GABAergic neurons, but not in cholinergic, serotonergic, dopaminergic, nor glutamatergic neurons. These results showed potent neurotoxicity of MC and suggested design of further research to study more aspects of the responses that MC causes.

The experimental proof that MCs are actually responsible for causing adverse effects in neurons may come from studies at the proteomic level. Wang et al. (2010a) showed striking accumulation of MC-LR and enhanced PP activity in zebrafish brains after 30 d exposure. The treatment resulted in alteration of levels of proteins involved in the cytoskeleton assemblage, signal transduction, protein degradation, metabolism, transport, apoptosis and translation. Such a response indicates that MC-LR toxicity in the fish brain is complex and diverse, and involves several molecular pathways. Authors suggest that the chronic neurotoxicity of MCLR might initiate the PP pathway via an upregulation of PP2C in the zebrafish brain, in addition to the reactive oxygen species and endocrine signaling pathways (Wang et al. 2010a).

Indications of the complexity of neurotoxic effects by MC-LR exposure have also come from the measurements of antioxidant activity in the affected tissues. Chen et al. (2006) showed, that the microsystin-LR in the liver especially attacks mitochondrial aldehyde dehydrogenase 2 (AldH2). In the work on zebrafish, Wang et al. (2010a) showed significant decrease in expression of the AldH2 in the brain, which was concurrent with the expression of the aldehyde dehydrogenase 9A1a (Wang et al. 2010a). This led to interrupted metabolism of aldehydes, which is an indicator of DNA damage, enzyme deactivation or protein modifications (Lindahl 1992; O'Brien et al. 2005).

Data from the study of Li et al. (2012) indicate, that microcystin-LR causes oxidative stress and apoptosis response in brain cells. Six proteins (GFAP, Hsp70, Hsp75, Prdx2, SOD, and Stip1) involved in oxidative stress and apoptosis responses in rats' hippocampi have been identified (Li et al. 2012). Induction of HSP (marker of oxidative stress) as well as SOD and Prdx2 (antioxidant enzymes) are defense systems against reactive oxygen species, therefore this suggests oxidative stress caused by MC-LR. Moreover, the research group has showed the induction of septin 5, a-internexin, and a-synuclein, which are known to be related to neurodegenerative diseases and could be involved in the progression of Alzheimer's disease.

Cytoskeletal changes result in loss of cell shape and also create obstacles in mitosis due to a lack of chromosome separation and creation of binucleated cells (Mezhoud et al. 2008). It has been shown that MC-LR treatment of zebrafish affects expression of diverse cytoskeletal proteins in the fishes' brain (Wang et al. 2010a). In this study, cellular damage caused by MC-LR has been related to cytoskeletal disruptions linked with accumulation of tubulin folding cofactor B. Accumulation of this protein leads to microtubule depolymerization, growth inhibition, axonal damage and, in consequence, neuronal decay (Lopez-Fanarraga et al. 2007). Moreover, Wang et al. (2010a) have shown that transient ischemia in gebril brains decreased the mRNA expression of β-actin. They concluded that decreased β-actin RNA may be due to oxidative stress caused by MC-LR, as other studies consider actin to be a direct target for oxidative modifications (Fiaschi et al. 2006; Lassing et al. 2007). β-actin is a major protein of actin filaments which are present in synaptic areas, it plays a role in cell adhesion, neurite growth, formation of synapse and is involved in exocytosis of neurotransmitters (Asanuma et al. 1993; Sobue and Kanda 1989). Thus, decreased mRNA levels of β-actin may suggest severe physiological consequences: oxidative stress, cytoskeletal disruptions or even neuronal death (Asanuma et al. 1993).

Protein phosphatase activity in the brain of zebrafish was enhanced in the study of Wang et al. (2010a), which could be the consequence of upregulation of PP2Cα2 after MC-LR treatment. However, lethal doses of MC-LR inhibit PP activity in the nuclear compartment. In another study, MC-RR was responsible for upregulation of A subunit of PP2A in human amniotic epithelial cells (Fu et al. 2009). PP activity is also increased after higher doses of the toxin (Wang et al. 2010a), although this is inconsistent with other studies (Guzman et al. 2003; Malbrouck et al. 2003, 2004). Overexpression of PP2Cα2 leads to G2/M cell cycle inhibition and apoptosis through p53 protein kinase pathway activation (Ofek et al. 2003). Overexpression of PP2Cα2 might be involved in the interruption of the protein kinase pathway (e.g. MAPK and AMPK). On the other hand, it could be a compensatory effect of the cells responding to the toxin attack (Wang et al. 2010a), which, however, deserves further investigation.

Wang et al. (2010b) also observed another change in the brain's proteome after MC-LR treatment. It has been shown, that microcystin could reduce levels of Ywhai protein (belonging to the 14-3-3 family, a phosphoserine specific adapter protein, which binds Bad protein and, as a result, prevents apoptosis). Down-regulation of Ywhai protein leads to a release of phosphorylated Bad protein into cytoplasm, where it could induce pro-apoptotic activity.

In a more recent paper, Li et al. (2012) have shown that MC-LR hyperphosphorylates tau protein. Tau protein is a principal neuronal microtubule protein associated with assembly, stabilization and maintaining normal morphology of axonal microtubules (Goode et al. 1997). Studies have shown that PPs are involved in the regulation of the phosphorylation of tau (Feurstein et al. 2011). Inhibition of PPs in neurons causes hyperphosphorylation and aggregation of tau protein, leading to neuronal degenerative changes and apoptosis which were similar to those observed in the brains of Alzheimer patients (Li et al. 2012). Li and coworkers (2012) also observed the impairment of memory and cognitive function in MC-treated rats which is also associated with Alzheimer's disease as a result of inhibition of PPs.

Microcystins may destroy energy-generating pathways and organic acid metabolism in the zebrafish brain (Wang et al. 2010b). MC-LR was shown to decrease protein levels of NADH (ubiquinone) dehydrogenase I alpha subcomplex subunit 10 (NDUFA10 is a subunit of NADH ubiquinone oxidoreductase), which may decrease energy production from the respiratory chain. But it is also known that the complex I of the respiratory chain is a potent source of ROS (Murphy 2009), so decreased levels of NDUFA10 may prevent the brain from producing ROS. Overexpression of propionyl-CoA carboxylase, beta polypeptide and 3-oxoacid CoA transferase 1a, and 3-hydroxyisobutyrate dehydrogenase b is thought to be a compensatory mechanism in facing the decreasing energy production caused by MC-LR induced oxidative stress, as they are involved in catabolism processes (Wang et al. 2010b).

Nodularin

Nodularin (NOD) is one of the most frequently encountered toxins found in the Baltic Sea during summer months. This is because of *Nodularia spumigena*, whose blooms occur there annually (Sivonen et al. 1989). Toxin production by *N. spumigena* tends to be highest in conditions that also

promote its growth (Lehtimäki et al. 1997). During recent decades, the intensity of blooms has increased considerably in the Baltic Sea region (Finni et al. 2001; Suikkanen et al. 2007), which has led to increased exposure to the toxin for animals in the aquatic environment, as well as domestic animals (Simola et al. 2012). Nodularin induces similar toxic effects as microcystin and it is associated with the same transporters (Runnegar et al. 1995a).

Due to similarity of NOD structure with microcystin (Table 1), the toxin is considered to present molecular mechanisms of up-take and toxicity similar to those of MC. Simola et al. (2012) studied mechanisms of NOD toxicity in dogs. They found that nodularin is absorbed from the ileum by bile acid carriers; the toxin enters the liver with portal blood flow and is transported into hepatocytes via bile acid carriers (Simola et al. 2012). Nodularin inhibits PP1 and PP2A and is a potent tumor promoter that may also act as a carcinogen or tumor initiator (Valério et al. 2010). However, it does not covalently bind to serine/threonine protein phosphatase-1 and 2A (Bagu et al. 1997). It has been reported that the toxin induces harmful effects on wide rage of organisms (fish, invertebrates), but some species may be resistant to the NOD's harmful effects (Karjalainen et al. 2007). Zhang et al. (2012) found that nodularin at low doses can induce apoptosis in fish lymphocytes *in vitro* and that the main mechanism in nodularin-induced apoptosis is oxidative stress. He also showed the accumulation of ROS in fish cells after exposure to nodularin and found that NOD can destroy intracellular antioxidant enzymes and cause oxidation damage in fish immune cells. The effect was reversed with natural antioxidants, which improved the activity of enzymes and inhibited cellular lesions (Zhang et al. 2012).

Although only several data on neurotoxic effects of non-ribosomal peptides, such as MCs or NOD, have been gathered so far, the molecular background of underlying processes remains obscure. The successful elucidation of the aberrant signaling pathways, by exploiting new types of experimental tools and new thinking about the organization of complex signaling circuits in nervous tissues of diverse animal systems, should shed light on neurotoxic effects caused by the natural heptapeptides.

ALKALOIDS

Saxitoxin

Alkaloids constitute an important group of cyanotoxins which occur in the aquatic environment. One of them is Saxitoxin (STX), which is a trialkyl tetrahydropurine produced by cyanobacteria of different genera (Neilan et al. 2010; Table 1). Saxitoxins differ structurally at four positions which can be hydroxylated, sulfated or carbamyolated (Sivonen and Jones 1999). Saxitoxins are part of a wider group of toxins - Paralytic Shellfish Poisoning (PSP) toxins, which is produced by harmful dinoflagellates (Tian et al. 2014). PSPs form a group of neurotoxins that accumulate mainly in shellfish, and STX being the most common and the most lethal toxin amongst them (Wong et al. 2011). STX can specifically accumulate in mussels, gastropods, fish, copepods, as well as many other water organisms which all have a world-wide geographical distribution (Kumar-Roiné et al. 2011; Neilan et al. 2010).

Expanding blooms of STX producing cyanobacteria have led to mass deaths of fish, as well as other animals, including livestock (Negri et al. 1995; Reyero et al. 1999). Because STXs accumulate through the food chain, intoxication with these compounds is not necessarily associated with drinking water (Shumway 1995). STXs have been linked to many poisoning events, including humans (Merel et al. 2013). STXs induce nerve dysfunction and paralysis. In lethal doses, they cause respiratory failure due to muscle paralysis (van Apeldoorn et al. 2007). Adverse effects after direct exposure to STXs such as fever, eye irritation, abdominal pain, and skin rashes, have been reported in infants and children (Rapala et al. 2005). These effects are related to STXs ability to modulate activity of ion channels (Belkacemi et al. 2005; Su et al. 2004).

There are several reports documenting adverse effects of STXs on fish. For example, studies conducted on *D. rerio* embryos show that exposure to STX results in edema and body curvature. Sensorimotor impairments and paralysis in herring larvae were also noted (Lefebvre et al. 2004, 2005). Tian et al. (2014) found that treated marine medaka (*Oryzias melastigma*) embryos exhibited abnormal growth with high neurogenesis and cellular proliferation after exposure to STX.

Cylindrospermopsin

Cylindrospermopsin (CYN) is another biologically active alkaloid found in several cyanotoxin genera (Neilan et al. 2010; Table 1). In contrast to a highly differentiated group of microcystins the structural variability of CYN is much lower. So far, only three variants of the cylindrospermopsin molecule have been described (Valério et al. 2010). It was first recognized in 1979 when 148 people were hospitalized with symptoms of hepatoenteritis on Palm Island (Queensland, Australia). Since then, the toxin has been linked to a bloom of *Cylindrospermopsis raciborskii* in a drinking water source (Bourke et al. 1983; Byth 1980), and has been found responsible for the death of domestic animals (e.g. Saker et al. 1999). *C. raciborskii* is a highly adaptive cyanobacterium capable of inducing considerable physiological and morphological changes, which presents a major problem for water management on a global scale (Neilan et al. 2003). *C. raciborskii* is one of the most invasive cyanobacterial species in the world (Kling 2009; Sinha et al. 2012). In contrast to other cyanotoxins CYN is more often found at higher levels in dissolved form than within cells as it readily leaks from cells under normal growth conditions (Falconer and Humpage 2005; Norris et al. 2001; Wormer et al. 2008).

The presence of guanidino and sulfate groups makes CYN a zwitterionic molecule and hence more soluble in water. Moreover, small compounds are likely to be taken by the cells through diffusion (Valério et al. 2010). After ingestion from water, the toxin mainly bears upon the liver, affecting protein synthesis and inhibition of glutathione and cytochrome P450 (Froscio et al. 2003, 2008; Metcalf et al. 2004; Runnegar 1995b). CYN can also covalently bind to DNA and there is evidence that CYN causes DNA breakage (Shen et al. 2002). It exerts general cytotoxic, hepatotoxic and neurotoxic effects (Kiss et al. 2002) and it is considered as a carcinogen due to DNA damaging activity. However, the exact mode of action has yet to be determined (Falconer and Humpage 2001; Valério et al. 2010). The only study related to neurotoxicity of cylidrospermopsin (Kiss et al. 2002) was performed on crude extract from *C. raciborskii*. It showed that the extract had a component that affected nicotinergic ACh receptors of the neurones. However, these effects are more similar to effects caused by anatoxin-a, therefore authors of the paper linked them with anatoxin-like substances. In support of this view a recent pharmacological study confirmed the presence of some unidentified anatoxin-a like neurotoxins produced by *C. raciborskii* (Vehovszky et al. 2013). Further studies are necessary to confirm or deny neurotoxic properties of the toxin.

Anatoxin-a and homoanatoxin-a

Anatoxin-a (ANTX-a) and its homologue homoanatoxin-a are highly water-soluble alkaloids, synthesized by species of the genera: *Anabaena*, *Cylindrospermum*, *Microcystis*, *Oscillatoria*, *Raphidiopsis*, *Planktothrix* and *Aphanizomenon* (Valério et al. 2010; Table 1). Neurotoxic effects of anatoxin-a have been proven in many studies (reviewed by Osswald et al. 2007). ANTX-a causes overstimulation of the muscle cells as a result of membrane depolarization and desensitization (Figure 2). This leads to paralysis or even death when respiratory muscles are affected (Osswald et al. 2007). ANTX-a is considered to be responsible for poisoning different animal species resulting in vomiting, respiratory difficulties and convulsion (Gugger et al. 2005; Henriksen et al. 1997; Krienitz et al. 2003; Wood et al. 2007).

PHOSPHATE ESTERS

Anatoxin-a(s)

Anatoxin-a(s) (ANTX-a(s)) is a phosphate ester of cyclic N-hydroxyguanine (Sivonen and Jones 1999; van Apeldoorn et al. 2007), which is synthesized by *Anabaena lemmermanni* (Henriksen et al. 1997) and *Anabaena flos-aquae* (Mahmood and Carmichael 1986) (Table 1). Despite the similarity in names it is not structurally related to anatoxin-a (Table 1) and it exhibits different physiological properties. ANTX-a(s) irreversibly inhibits hydrolysis of acetylcholine by acetylcholinesterase (Molica et al. 2005). As a result, muscles become constantly stimulated (Matsunaga et al. 1989) (Figure 2). Exposure to anatoxin-a(s), as with anatoxin-a, could lead to muscular paralysis with possible respiratory arrest leading to death (Molica et al. 2005).

LIPOPEPTIDES

Antillatoxins

First reported in 1995, *Lyngbya majuscula* metabolite, antillatoxin (ATX; Table 1) is an extremely potent ichthyotoxic compound. *L. majuscula* grows on solid or sandy substrates or epiphytically on seagrass, macroalgae and corals in the coastal zones of many sub-tropical and tropical oceans (e.g. it has been observed seasonally in Moreton Bay, Queensland, Australia). It has been noted that after periods of high light and temperatures combined with calm weather, bubbles from rapid photosynthesis by the *L. majuscula* are trapped within the filament matrix and cause the *L. majuscula* to eventually float to the water surface to form large surface aggregations. This may enable *L. majuscula* to spread to other regions (Albert et al. 2005).

Antillatoxin is a lipopeptide with a high degree of methylation. Antillatoxin is one of the lipid-soluble gating modifier toxins of voltage-gated sodium channels (Figure 2). This group includes brevetoxins, batrachotoxin, veratridine and gambierol (Cao et al. 2008). It is among the most ichthyotoxic metabolites isolated and is considered to be the second most potent compound obtained from marine waters after brevetoxin (Orjala et al. 1995). Studies show that exposure to blooms of *L. majuscula* may cause adverse human health effects like irritation of the respiratory system, eye inflammation and also contact dermatitis (Jabba et al. 2010). In addition ATX is neurotoxic in cerebellar granule cells. As a consequence of glutamate release, ATX has been shown to be an indirect activator of N-methyl-D-aspartate receptors (NMDARs) (Li et al. 2001, 2004). Jabba et al. (2010) found that ATX was capable of mimicking activity-dependent neuronal development by up-regulating NMDAR function.

Kalkitoxin and jamaicamide

Kalkitoxin is another toxin produced by marine cyanobacterium *Lyngbya majuscula*. It is a lipid derivative containing thiazoline (Table 1) and it was first discovered from samples collected from Curaćao (Wu et al. 2000). This study reported that kalkitoxin was toxic to the *Carassius auratus*, as well as to the aquatic crustacean brine shrimp (*Artemia salina*). Data collected by LePage et al. (2005) confirmed, that kalkitoxin acted like an inhibitor to the voltage-gated sodium channels in the cerebellar granule cell culture (Figure 2).

Jamaicamide (Table 1) is also produced by *L. majuscula*, a strain found growing in low abundance in Hector's Bay, Jamaica. There are three forms (A, B, C) of jamaicamide, which differ structurally. All three forms showed similar cytotoxicity to mouse neuroblastoma cell lines. Edwards and coworkers (2004) revealed, that all jamaicamide isomers showed channel-blocking activity (Figure 2). However, none of the toxin isomers exhibited sodium channel-activating activity (Edwards et al. 2004). This study also included toxicity assay on *C. auratus*, which concludes that different jamaicamide isomers display different lethal dose levels.

NEUROTOXIC AMINO ACIDS

L-β-N-methylamino-L-alanine (BMAA)

L-β-N-methylamino-l-alanine (BMAA; Table 1) is a neurotoxic amino acid produced by cyanobacteria that were originally isolated from cycad seeds (*Cycas micronesica*) used by the Chamorro people of Guam to prepare food (Cox et al. 2005). Its structure basically consists of alanine with an added methylated amino group (Banack et al. 2007). BMAA was thought to be linked only with neurodegenerative illness among the Chamorro people of Guam (Ince and Codd 2005). It was proven, that the toxin accumulates in food webs, making Chamorro people diets rich in BMAA (Cox et al. 2003). BMAA has been considered as a "slow toxin" which produces delayed symptoms. This theory was confronted with another, more probable one: incorporation of the toxin into proteins. Recently BMAA has been found incorporated into protein fraction of cyanobacteria, cycads and animals that forage on cycad seeds. Furthermore, it was found that toxin incorporated into protein could easily be released due to protein catabolism (Cox et al. 2003; Murch et al. 2004). Following this observation Cox and co-workers examined BMAA production by cyanobacteria, including 30 laboratory strains of free-living cyanobacteria, representing five major microbiological taxonomic groups. Results showed that 29 strains had free or protein-bound BMAA, including a sample of an ocean *Trichodesmium* bloom (Cox et al. 2005).

Storage of BMAA through incorporation into proteins could explain observed biomagnification of a water-soluble molecule. It also explains disease latency, indicating the intermittent release of the endogenous neurotoxic reserves stored in the proteins (Murch at al. 2004). Moreover, the finding that BMAA occurs with other cyanotoxins in contaminated water sources suggests that people may be exposed to the toxin in many parts of the world. For example, it was found in brain tissues of Canadian patients with Alzheimer's disease (Metcalf et al. 2008). According to Aráoz et al. (2010) monitoring for the presence of BMAA in water supplies should be recommended. Data collected so far indicate the need for more research on BMAA to be done.

PHARMACEUTICAL POTENTIAL OF CYANOTOXINS

Cyanobacteria are one of the richest known sources of potent bioactive compounds. These compounds include toxins, which may have wide pharmaceutical applications. Extracting drugs from the sea environment started in the late 1960s. Between 1977-87, about 2500 new compounds were reported from a variety of marine organisms. These early studies clearly demonstrated a novel source of potentially useful chemicals not found in freshwater sources (Zanchett and Oliveira-Filho 2013). For example, extracts isolated from cyanobacteria strains showed antimicrobial activity against the fungus *C. albicans*, Gram-positive and Gram-negative bacteria. Natural products from cyanobacteria seemed to possess strong growth-inhibitory potential against several prokaryotic pathogens and eukaryotic parasites (Nagarajan et al. 2012).

Metabolites of cyanobacteria are potent drugs in cancer treatment (Zanchett and Oliveira-Filho 2013). An increasing number of compounds (such as curacin A and dolastatin 10) were found to target tubulin or actin filaments in cells (Jordan and Wilson 1998). The above-mentioned compounds have undergone clinical or preclinical trials as potent drugs (Gerwick et al. 2001) and were used in drug development of synthetic analogues where they were found to be superior over currently used anticancer drugs (Uzair et al. 2012). *In vitro* studies of a synthetic analog of dolastatin 10, soblidotin, showed promising results against human colon adenocarcinomas. A synthetic compound derived from dolastatin 15 (sythadotin) has shown promising results in phase II clinical trials of inoperable, locally advanced or metastatic melanoma (Liu and Rein 2010). Cryptophycin I, a natural compound isolated from *Nostoc* sp. GSV224, showed anticancer abilities against nasopharyngeal cancer cells and human colorectal cancer cells. It was 100-1000 times more potent than the currently available drugs (Singh et al. 2011). Anticancer activity against cell lines of human breast cancer was shown by *Oscillatoria boryana* (Nair and Bhimba 2013). Further anti-cancer activity was observed in cyanobacteria strains isolated from the Portuguese coast. Six selected strains were able to induce cytotoxic effects in human cancer cell lines and they were marked for the future as promising compounds with potential anticancer activity (Costa et al. 2014).

Bioinformatic molecular approaches have shown that 33 cyanobacterial compounds from 9 species have potential to be used in treating lung cancer (Vijayakumar and Menakha 2014). All of these compounds have the potential to block the epidermal growth factor receptor (EGFR). The EGFR regulates cell proliferation, apoptosis, angiogenesis, and tumor invasion. Mutations and amplification of EGFR are common in non-small cell lung cancer (Salgia and Skarin 1998). Of the analyzed compounds, the tiglicamide A, derived from *Lyngbya confervoides*, had the strongest interaction and binding with the EGFR (Vijayakumar and Menakha 2014). This finding has encouraged further study of tiglicamide A for potential therapeutic use.

Cyanobacteria metabolites also include neurotoxins which could be used as therapeutic drugs. They seem to be particularly advantageous in the field of neurology and diseases correlated with nervous system failure. In recent years they have also been studied as potent pharmaceutical drugs involved in pain management. Antillatoxin, as a modifier of sodium channels, is a source of important molecular information about characterization of VGSC, as well as potential analgesics and neuroprotectants (Uzair et al. 2012). PSP toxins such as STX and gonyautoxins, may provide a new source of selective ion channel blockers which have proved to be effective analgesics (Zhang et al. 2013). In 2004, US Food and Drug Administration approved the first marine sourced natural product – Ziconotide. The drug reduces chronic pain in spinal cord injury by selectively blocking N-type voltage-gated Ca^{2+} channels. Research done on VGSC has demonstrated that these channels are also involved in pain management. STX and its analogs may have pharmacological potential, possibly a novel class of analgesics in neuropathic and inflammatory pain (Zhang et al. 2013).

A study by Feurstein et al. (2010) confirming that organic anion-transporting peptides are involved in the neuronal uptake of MC, has raised questions and hopes about microcystin being suitable as leads for drug substances against cancer. OATP1B1 and OATP1B3 are the most efficient microcystin transporters (Hagenbuch and Gui 2008). As the OATP1B3 has been found to be expressed in a number of cancer tissues: colon tumors, breast tumors, lung tumors, pancreatic and hepatocellular tumors, the peptide is a discussed target for cancer therapy (Sainis et al. 2010). A challenge of using MCs as a lead for a drug is based on the finding that MC is selectively transported by OATPs only by cancer cells (Buxhofer-Ausch et al. 2013). On the other hand even with selective MC variants, side effects of the treatment, such as toxicity to healthy tissues, may also be challenging (Niedermeyer et al. 2014).

Although a wide range of cyanobacterial metabolites may have anti-cancer properties, thus being useful for human medicine, they have not been used in clinical trials due to difficulties in synthesizing them in laboratory. To minimize risk of various activities of toxins it may be necessary to identify and synthesize only the active parts of metabolites (Nagarajan et al. 2012). Further research and clinical trials are essential to solve the problem of delivering therapeutic compounds at the right time to the right cells.

SUMMARY

Cyanotoxins are a diverse group of natural compounds, including non-ribosomal peptides, alkaloids, lipopeptides, esters, amino acids and polyketides. These toxins have been implicated in the deaths of wild and domestic animals as well as in incidents of human illness. The liver is the most affected organ in humans but the exposure to the toxin is likely to affect organs such as the kidney, colon, gonads, and brain as evidenced by *in vivo* and *in vitro* studies. The neurotoxicity of cyanotoxins is a multi-pathway process, and despite recent achievements, the molecular mechanisms underlying neurotoxic effects remain elusive. There remains a lack of knowledge regarding the specific target/interacting proteins, the signaling pathways triggering the cell responses and the downstream pathways of toxicity and cell injury. The neurotoxic and cytotoxic effects of the cyanotoxins discussed here are of particular interest because of their pharmacological potential. However, further detailed studies of the mode of action of cyanobacterial toxins, their targets and possible side effects are essential.

ACKNOWLEDGEMENTS

We thank Dr. Maciej Góra (Jagiellonian University, Poland) for consultation regarding chemistry of the toxins. The authors also wish to thank Trev Hill, M.Sc. for proofreading the manuscript. The study was supported by research grant from National Science Centre (Poland) No. DEC-2012/07/B/NZ9/01320.

REFERENCES

Aas, P., S. Eriksen, J. Kolderup, P. Lundy, J.E. Haugen, O.M. Skulberg, F. Fonnum. 1996. Enhancement of acetylcholine release by homoanatoxin-a from *Oscillatoria formosa*. Environmental Toxicology and Pharmacology 2: 223–232.

Albert, S., J.M. O'Neil, J.W. Udy, K.S. Ahern, C.M. O'Sullivan, W.C. Dennison. 2005. Blooms of the cyanobacterium *Lyngbya majuscula* in coastal Queensland, Australia: disparate sites, common factors. Marine Pollution Bulletin 51: 428–437.

Angeles, J., S. Pichardo, A.I. Prieto, G. Repetto, C.M. Vázquez, I. Moreno, A.M. Cameán. 2005. Toxic cyanobacterial cells containing microcystins induce oxidative stress in exposed tilapia fish (*Oreochromis* sp.) under laboratory conditions. Aquatic Toxicology 72: 261–271.

Aráoz, R., J. Molgó, N. Tandeau de Marsac. 2010. Neurotoxic cyanobacterial toxins. Toxicon 56: 813-828.

Asanuma, M., N. Ogawa, H. Hirata, H. Chou, Y. Kondo, A. Mori. 1993. Ischemia-induced changes in a-tubulin and b-actin mRNA in the gerbil brain and effects of bifemelane hydrochloride. Brain Research 600: 243–248.

Babica, P., L. Bláha, B. Maršálek. 2006. Exploring the natural role of microcystins – a review of effects on photoautotrophic organisms. Journal of Phycology 42: 9–20.

Baganz, D., G. Staaks, C. Steinberg. 1998. Impact of the cyanobacteria toxin, microcystin-LR on behaviour of zebrafish, *Danio rerio*. Water Research 32: 948–952.

Baganz, D., G. Staaks, S. Pflugmacher, C.E.W. Steinberg. 2004. Comparative study of microcystin-LR-induced behavioral changes of two fish species, *Danio rerio* and *Leucaspius delineatus*. Environmental Toxicology 19: 564–570.

Bagu, J.R., B.D. Sykes, M.M. Craig, C.F.B. Holmes. 1997. A molecular basis for different interactions of marine toxins with protein phosphatase-1. The Journal of Biological Chemistry 272: 5087–5097.

Banack, S.A., H.E. Johnson, R. Cheng, P.A. Cox. 2007. Production of the neurotoxin BMAA by a marine cyanobacterium. Marine Drugs 5: 180-196.

Begout Anras, M.L., J.P. Lagard`ere. 2004. Measuring cultured fish swimming behaviour: first results on rainbow trout using acoustic telemetry in tanks. Aquaculture 240: 175–186.

Belkacemi, L., I. Bédard, L. Simoneau, J. Lafond. 2005. Calcium channels, transporters and exchangers in placenta: a review. Cell Calcium 37: 1-8.

Blom, J.F., F. Jüttner. 2005. High crustacean toxicity of microcystin congeners does not correlate with high protein phosphatase inhibitory activity. Toxicon 46: 465–470.

Bogo, M.R., L.W. Kist, A.L. Piato, J.G. Santos da Rosa, G. Koakoski, L.J.G. Barcellos, J.S. Yunes, C.D. Bonan. 2011. Acute exposure to microcystin-producing cyanobacterium *Microcystis aeruginosa* alters adult zebrafish (*Danio rerio*) swimming performance parameters. Journal of Toxicology, Volume 2011, article ID 280304, 9 pages.

Bourke, A.T.C., R.B. Hawes, A. Neilson, N.D. Stallman. 1983. An outbreak of hepato-enteritis (the Palm Island mystery disease) possibly caused by algal intoxication. Toxicon 21: 45–48.

Bury, N.R., F.B. Eddy, G.A. Codd. 1995. The effects of the cyanobacterium *Microcystis aeruginosa*, the cyanobacterial hepatotoxin microcystin-LR, and ammonia on growth rate and ionic regulation of brown trout. Journal of Fish Biology 46: 1042–1054.

Buxhofer-Ausch, V., L. Secky, K. Wlcek, M. Svoboda, V. Kounnis. 2013. Tumor-specific expression of organic anion-transporting polypeptides: transporters as novel targets for cancer therapy. Journal of Drug Delivery 2013: 1–12.

Byth, S. 1980. Palm Island mystery disease. The Medical Journal of Australia 2: 40–42.

Campos, A., V. Vasconcelos. 2010. Molecular mechanisms of microcystin toxicity in animal cells. International Journal of Molecular Sciences 11: 268-287.

Cao, Z., J. George, W.H. Gerwick, D.G. Baden, J.D. Rainier, T.F. Murray. 2008. Influence of lipid-soluble gating modifier toxins on sodium influx in neocortical neurons. The Journal of Pharmacology and Experimental Therapeutics 326: 604–613.

Cazenave, J., D.A. Wunderlin, M. de los Ángeles Bistoni, M.V. Amé, E. Krause, S. Pflugmacher, C. Wiegand. 2005. Uptake, tissue distribution and accumulation of microcystin-RR in *Corydoras paleatus*, *Jenynsia multidentata* and *Odontesthes bonariensis*: A field and laboratory study. Aquatic Toxicology 75: 178–190.

Cazenave, J., M.L. Nores, M. Miceli, M.P. Díaz, D.A. Wunderlin, M.A. Bistoni. 2008. Changes in the swimming activity and the glutathione S-transferase activity of *Jenynsia multidentata* fed with microcystin-RR. Water Research 42: 1299–1307.

Chen, T., J. Cui, Y. Liang, X.B. Xin, D.O. Young, C. Chen, P.P Shen. 2006. Identification of human liver mitochondrial aldehyde dehydrogenase as a potential target for microcystin-LR. Toxicology 220: 71–80.

Chiswell, R.K., G.R. Shaw, G. Eaglesham, M.J. Smith, R.L. Norris, A.A. Seawright, M.R. Moore. 1999. Stability of cylindrospermopsin, the toxin from the cyanobacterium, *Cylindrospermopsis raciborskii*: effect of pH, temperature, and sunlight on decomposition. Environmental Toxicology 14: 155–161.

Christoffersen, K. 1996. Ecological implications of cyanobacterial toxins in aquatic food webs. Phycologia 35: 42-50.

Costa, M., M. Garcia, J. Costa-Rodrigues, M.S. Costa, M.J. Ribeiro, M.H. Fernandes, P. Barros, A. Barreiro, V. Vasconcelos, R. Martins. 2014. Exploring bioactive properties of marine cyanobacteria isolated from the Portuguese coast: high potential as a source of anticancer compounds. Marine Drugs 12: 98-114.

Cox, P.A., S.A. Banack, S.J. Murch. 2003. Biomagnification of cyanobacterial neurotoxins and neurodegenerative disease among the Chamorro people of Guam. Proceedings of the National Academy of Sciences of the United States of America 100: 13380–13383.

Cox, P.A., S.A. Banack, S.J. Murch, U. Rasmussen, G. Tien, R.R. Bidigare, J.S. Metcalf, L.F. Morrison, G.A. Codd, B. Bergman. 2005. Diverse taxa of cyanobacteria produce b-N-methylamino-l-alanine, a neurotoxic amino acid. Proceedings of the National Academy of Sciences of the United States of America 102: 5074–5078.

Dietrich, D.R., A. Fischer, C. Michel, S.J. Hoeger. 2008. Toxin mixture in cyanobacterial blooms – a critical comparison of reality with current procedures employed in human health risk assessment. In: Cyanobacterial Harmful Algal Blooms: State of the Science and Research Needs (ed. H.K. Hudnel), pp. 887-914. New York.

Ding, X.S., X.Y. Li, H.Y. Duan, L.K. Chung, J.A. Lee. 2006. Toxic effects of *Microcystis* cell extracts on the reproductive system of male mice. Toxicon 48: 973–979.

Edwards, C., D. Graham, N. Fowler, L.A. Lawton. 2008. Biodegradation of microcystins and nodularin in freshwaters. Chemosphere 73: 1315–1321.

Edwards, D.J., B.L. Marquez, L.M, Nogle, K. McPhail, D.E. Goeger, M.A. Roberts, W.H. Gerwick. 2004. Structure and biosynthesis of the jamaicamides, new mixed polyketide-peptide neurotoxins from the marine cyanobacterium *Lyngbya majuscula*. Chemistry & Biology 11: 817–833.

Ettoumi, A., F. El Khalloufi, I. El Ghazali, B. Oudra, A. Amrani, H. Nasri, N. Bouaïcha. 2011. Bioaccumulation of cyanobacterial toxins in aquatic organisms and its consequences for public health. In: Zooplankton and Phytoplankton: Types, Characteristics and Ecology (ed. G. Kattel), pp. 1–34. Nova Science Publishers Inc., New York.

Falconer, I.R., A.R. Humpage. 2001. Preliminary evidence for in vivo tumour initiation by oral administration of extracts of the blue-green alga *Cylindrospermopsis raciborskii* containing the toxin cylindrospermopsin. Environmental Toxicology 16: 192–195.

Falconer, I.R., A.R. Humpage. 2005. Health risk assessment of cyanobacterial (blue–green algal) toxins in drinking water. International Journal of Environmental Research and Public Health 2: 43–50.

Feurstein, D., K. Holst, A. Fischer, D.R. Dietrich. 2009. Oatp-associated uptake and toxicity of microcystins in primary murine whole brain cells. Toxicology and Applied Pharmacology 234: 247–255.

Feurstein, D., J. Kleinteich, A.H. Heussner, K. Stemmer, D.R. Dietrich. 2010. Investigation of microcystin congener–dependent uptake into primary murine neurons. Environmental Health Perspectives 118: 1370–1375.

Feurstein, D., K. Stemmer, J. Kleinteich, T. Speicher, D.R. Dietrich. 2011. Microcystin congener- and concentration-dependent induction of murine neuron apoptosis and neurite degeneration. Toxicological sciences: an official journal of the Society of Toxicology 124: 424-431.

Fiaschi, T., G. Cozzi, G. Raugei, L. Formigli, G. Ramponi, P. Chiarugi. 2006. Redox regulation of beta-actin during integrin-mediated cell adhesion. Journal of Biological Chemistry 281: 22983–22991.

Finni, T., K. Kononen, R. Olsonen, K. Wallström. 2001. The history of cyanobacterial blooms in the Baltic Sea. AMBIO: A Journal of the Human Environment 30: 172–178.

Fischer, W.J., D.R. Dietrich. 2000. Pathological and biochemical characterization of microcystin-induced hepatopancreas and kidney damage in carp (*Cyprinus carpio*). Toxicology and Applied Pharmacology 164: 73–81.

Fischer, W.J., S. Altheimer, V. Cattori, P.J. Meier, D.R. Dietrich, B. Hagenbuch. 2005. Organic anion transporting polypeptides expressed in liver and brain mediate uptake of microcystin. Toxicology and Applied Pharmacology 203: 257–263.

Fischer, A., S.J. Hoeger, K. Stemmer, D.J. Feurstein, D. Knobeloch, A. Nussler, D.R. Dietrich. 2010. The role of organic anion transporting polypeptides (OATPs/SLCOs) in the toxicity of different microcystin congeners in vitro: A comparison of primary human hepatocytes and OATP-transfected HEK293 cells. Toxicology and Applied Pharmacology 245: 9-20.

Froscio, S.M., A.R. Humpage, P.C. Burcham, I.R. Falconer. 2003. Cylindrospermopsin-induced protein synthesis inhibition and its dissociation from acute toxicity in mouse hepatocytes. Environmental Toxicology 18: 243–251.

Froscio, S.M., A.R. Humpage, W. Wickramasinghe, G. Shaw, I.R. Falconer. 2008. Interaction of the cyanobacterial toxin cylindrospermopsin with the eukaryotic protein synthesis system. Toxicon 51: 191–198.

Fu, W., Y. Yu, L. Xu. 2009. Identification of temporal differentially expressed protein responses to microcystin in human amniotic epithelial cells. Chemical Research in Toxicology 22: 41–51.

Gerwick, W.H., L.T. Tan, N. Sitachitta. 2001. Nitrogen-containing metabolites from marine cyanobacteria. In: The Alkaloids: Chemistry and Biology. 57, pp. 75–184. Academic Press, San Diego, Calif, USA.

Goode, B.L., P.E. Denis, D. Panda, M.J. Radeke, H.P. Miller, L. Wilson, S.C. Feinstein. 1997. Functional interactions between the proline-rich and repeat regions of tau enhance microtubule binding and assembly. Molecular Biology of the Cell 8: 353–365.

Gugger, M., S. Lenoir, C. Berger, A. Ledreux, J. Druart, J.-F. Humbert. 2005. First report in a river in France of the benthic cyanobacterium *Phormidium favosum* producing anatoxin-a associated with dog neurotoxicosis. Toxicon 45: 919–928.

Gupta, N., A.S.B. Bhaskar, R.S. Dangi, G.B.K.S. Prasad, P.V.L. Rao. 2001. Toxin production in batch cultures of freshwater cyanobacterium *Microcystis aeruginosa*. Bulletin of Environmental Contamination and Toxicology 67: 0339–0346.

Gupta, U.S., S. Guha. 2006. Microcystin toxicity in a freshwater fish, *Heteropneustes fossilis* (Bloch). Current Science 91: 1261–1271.

Guzman, R.E., P.F. Solter, M.T. Runnegar. 2003. Inhibition of nuclear proteinphosphatase activity in mouse hepatocytes by the cyanobacterial toxin microcystin-LR. Toxicon 41: 773–781.

Hagenbuch, B., C. Gui. 2008. Xenobiotic transporters of the human organic anion transporting polypeptides (OATP) family. Xenobiotica 38: 778–801.

Hagenbuch, B., P.J. Meier. 2004. Organic anion transporting polypeptides of the OATP/SLC21 family: phylogenetic classification as OATP/SLCO superfamily, new nomenclature and molecular/functional properties. Pflugers Archiv-European Journal of Physiology 447: 653-665.

Henriksen, P., W.W. Carmichael, J. An, Ø. Moestrup. 1997. Detection of an anatoxin-a(s)-like anticholinesterase in natural blooms and cultures of cyanobacteria/blue-green algae from Danish lakes and in the stomach contents of poisoned birds. Toxicon 35: 901–913.

Hille, B. 1992. Potassium channels and chloride channels. In: Ionic Channels of Excitable Membranes (ed. B. Hille), pp. 115–139. Sinauer Associates, Inc., Sunderland, MA.

Huguet, A., J. Henri, M. Petitpas, K. Hogeveen, V. Fessard. 2013. Comparative cytotoxicity, oxidative stress, and cytokine secretion induced by two cyanotoxin variant, microcystin LR and RR, in human intestinal Caco-2 Cells. Journal of Biochemical and Molecular Toxicology 27: 253-258.

Ince, P.G., G.A. Codd. 2005. Return of the cycad hypothesis - does the amyotrophic lateral sclerosis/parkinsonism dementia complex (ALS/PDC) of Guam have new implications for global health? Neuropathology and Applied Neurobiology 31: 345-353.

Jabba, S.V., A. Prakash, S.M. Dravid, W.H. Gerwick, T.F. Murray. 2010. Antillatoxin, a novel lipopeptide, enhances neurite outgrowth in immature cerebrocortical neurons through activation of voltage-gated sodium channels. The Journal of Pharmacology and Experimental Therapeutics 332: 698–709.

James, H., J.K. Fawell. 1991. Detection and removal of cyanobacterial toxins from freshwaters. Report No. FR0211 Foundation for Water Research, Marlow, UK.

Jordan, M.A., L. Wilson. 1998. Microtubules and actin filaments: dynamic targets for cancer chemotherapy. Current Opinion in Cell Biology 10: 123–130.

Ju, J., Q. Ruan, X. Li, R. Liu, Y. Li, Y. Pu, L. Yin, D. Wang. 2013. Neurotoxicological evaluation of microcystin-LR exposure at environmental relevant concentrations on nematode *Caenorhabditis elegans*. Environmental Science and Pollution Research International 20: 1823-1830.

Kaasalainen, U., D.P. Fewer, J. Jokela, M. Wahlsten, K. Sivonen, J. Rikkinen. 2012. Cyanobacteria produce a high variety of hepatotoxic peptides in lichen symbiosis. Proceedings of the National Academy of Sciences of the United States of America 109: 5886-5891.

Karjalainen, M., J. Engstrom-Ost, S. Korpinen, H. Peltonen, J.P. Paakkonen, S. Ronkkonen, S. Suikkanen, M. Viitasalo. 2007. Ecosystem consequences of cyanobacteria in the northern Baltic Sea. Ambio 36: 195–202.

Kem, W.R. 2000. Properties and effects of natural toxins and venoms. In: Principles of Toxicology: Environmental and Industrial Applications (ed. P.L. Williams, R.C. James, S.M. Roberts), pp. 409-432. John Wiley & Sons, New York.

Kiss, T., A. Vehovszky, L. Hiripi, A. Kovacs, L. Voros. 2002. Membrane effects of toxins isolated from a cyanobacterium, *Cylindrospermopsis raciborskii*, on identified molluscan neurones. Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology 131: 167–176.

Kling, H.J. 2009. *Cylindrospermopsis raciborskii* (Nostocales, Cyanobacteria): A brief historic overview and recent discovery in the Assiniboine River (Canada). Fottea 9: 45–47.

Kotak, B.G., S. Semalulu, D.L. Fritz, E.E. Prepas, S.E. Hrudey, R.W. Coppock. 1996. Hepatic and renal pathology of intraperitoneally administered microcystin-LR in rainbow trout (*Oncorhynchus mykiss*). Toxicon 34: 517–525.

Krienitz, L., A. Ballot, K. Kotut, C. Wiegand, S. Pütz, J.S. Metcalf, G.A. Codd, S. Pflugmacher. 2003. Contribution of hot spring cyanobacteria to the mysterious deaths of lesser flamingos at lake Bogoria, Kenya. FEMS Microbiology Ecology 43: 141-148.

Kumar-Roiné, S., M. Matsui, S. Pauillac, D. Laurent. 2011. Ciguatera fish poisoning and other seafood intoxication syndromes: a revisit and a review of the existing treatments employed in ciguatera fish poisoning. The South Pacific Journal of Natural and Applied Sciences 28: 1–26.

Lassing, I., F. Schmitzberger, M. Björnstedt, A. Holmgren, P. Nordlund, C.E. Schutt, U. Lindberg. 2007. Molecular and structural basis for redox regulation of b-actin. Journal of Biological Chemistry 370: 331–348.

Lefebvre, K.A., V.L. Trainer, N.L. Scholz. 2004. Morphological abnormalities and sensorimotor deficits in larval fish exposed to dissolved saxitoxin. Aquatic Toxicology 66: 159–170.

Lefebvre, K.A., N.E. Elder, P.K. Hershberger, V.L. Trainer, C.M. Stehr, N.L. Scholz. 2005. Dissolved saxitoxin causes transient inhibition of sensorimotorfunction in larval Pacific herring (*Clupea harengus pallasi*). Marine Biology 147: 1393–1402.

Lehtimäki, J., P. Moisander, K. Sivonen, K. Kononen. 1997. Growth, nitrogen fixation, and nodularin production by two Baltic Sea cyanobacteria. Applied and Environmental Microbiology 63: 1647–1656.

LePage, K.T., D. Goeger, F. Yokokawa, T. Asano, T. Shioiri, W.H. Gerwick, T.F. Murray. 2005. The neurotoxic lipopeptide kalkitoxin interacts with voltage-sensitive sodium channels in cerebellar granule neurons. Toxicology Letters 158: 133-139.

Li, L., P. Xie, J. Chen. 2005. In vivo studies on toxin accumulation in liver and ultrastructural changes of hepatocytes of the phytoplanktivorous bighead carp i.p.-injected with extracted microcystins. Toxicon 46: 533–545.

Li, G., F. Cai, W. Yan, C. Li, J. Wang. 2012. A proteomic analysis of MCLR-induced neurotoxicity: implications for Alzheimer's disease. Toxicological Sciences 127: 485–495.

Li, W.I., B.L. Marquez, T. Okino, F. Yokokawa, T. Shioiri, W.H. Gerwick, T.F. Murray. 2004. Characterization of the preferred stereochemistry for the neuropharmacologic actions of antillatoxin. Journal of Natural Products 67: 559 –568.

Li, W.I., F.W. Berman, T. Okino, F. Yokokawa, T. Shioiri, W.H. Gerwick, T.F. Murray. 2001. Antillatoxin is a marine cyanobacterial toxin that potently activates voltage-gated sodium channels. Proceedings of the National Academy of Sciences of the United States of America 98: 7599–7604.

Lilleheil, G., R.A. Andersen, O.M. Skulberg, J. Alexander. 1997. Effects of a homoanatoxin-a-containing extract from *Oscillatoria formosa* (cyanophyceae/cyanobacteria) on neuromuscular transmission. Toxicon 35: 1275–1289.

Lindahl, R. 1992. Aldehyde dehydrogenases and their role in carcinogenesis. Critical Reviews in Biochemistry and Molecular Biology 27: 283–335.

Lindholm, T., P. Ohman, K. Kurki-Helasmo, B. Kincaid, J. Meriluoto. 1999. Toxic algae and fish mortality in a brackishwater lake in Aland, SW Finland. Hydrobiologia 397: 109–120.

Liu, L., K.S. Rein. 2010. New peptides isolated from *Lyngbya* species: A review. Marine Drugs 8: 1817–1837.

Lopez-Fanarraga, M., G. Carranza, J. Bellido, D. Kortazar, J.C. Villegas, J.C. Zabala. 2007. Tubulin cofactor B plays a role in the neuronal growth cone. Journal of Neurochemistry 100: 1680–1687.

Mahmood, N.A., W.W. Carmichael. 1986. The pharmacology of anatoxin-a(s), a neurotoxin produced by the freshwater cyanobacterium *Anabaena flos-aquae* NRC 525-17. Toxicon 24: 425–434.

Malbrouck, C., G. Trausch, P. Devos, P. Kestemont. 2003. Hepatic accumulation and effects of microcystin-LR on juvenile goldfish *Carassius auratus* L. Comparative Biochemistry and Physiology part C: Toxicology & Pharmacology 135: 39–48.

Malbrouck, C., G. Trausch, P. Devos, P. Kestemont. 2004. Effect of microcystin-LR on protein phosphatase activity and glycogen content in isolated hepatocytes of fed and fasted juvenile goldfish *Carassius auratus* L. Toxicon 44: 927–932.

Matsunaga, S., R.E. Moore, W.P. Niemczura, W.W. Carmichael. 1989. Anatoxin-a(s), a potent anticholinesterase from *Anabaena flos-aquae*. Journal of the American Chemical Society 111: 8021–8023.

McDermott, C.M., C.W. Nho, W. Howard, B. Holton. 1998. The cyanobacterial toxin, microcystin-LR, can induce apoptosis in a variety of cell types. Toxicon 36: 1981–1996.

Merel, S., D. Walker, R. Chicana, S. Snyder, E. Baurès, O. Thomas. 2013. State of knowledge and concerns on cyanobacterial blooms and cyanotoxins. Environment International 59: 303-327.

Metcalf, J.S., A. Barakate, G.A. Codd. 2004. Inhibition of plant protein synthesis by the cyanobacterial hepatotoxin, cylindrospermopsin. FEMS Microbiology Letters 235: 125–129.

Metcalf, J.S., S.A. Banack, J. Lindsay, L.F. Morrison, P.A. Cox, G.A. Codd. 2008. Co-occurrence of β-N-methylamino-L-alanine, a neurotoxic amino acid with other cyanobacterial toxins in British waterbodies, 1990-2004. Environmental Microbiology 10: 702-708.

Mezhoud, K., A.L. Bauchet, S. Château-Joubert, D. Praseuth, A. Marie, J.C. Franćois, J.J. Fontaine, J.P. Jaeg, J.P. Cravedi, S. Puiseux-Dao, M. Edery. 2008. Proteomic and phosphoproteomic analysis of cellular responses in medaka fish (*Oryzias latipes*) following oral gavage with microcystin-LR. Toxicon 51: 1431–1439.

Molica, R.J.R., E.J.A. Oliveira, P.V.V.C. Carvalho, A.N.S.F. Costa, M.C.C. Cunha, G.L. Melo, S.M.F.O. Azevedo. 2005. Occurrence of saxitoxins and an anatoxin-a(s)-like anticholinesterase in a Brazilian drinking water supply. Harmful Algae 4: 743–753.

Murch, S.J., P.A. Cox, S.A. Banack. 2004. A mechanism for the slow release of biomagnified cyanobacterial neurotoxins and neurodegenerative disease in Guam. Proceedings of the National Academy of Sciences of the United States of America 101: 12228-12231.

Murphy, M.P. 2009. How mitochondria produce reactive oxygen species. Biochemical Journal 417: 1–13.

Nagarajan, M., V. Maruthanayagam, M. Sundararaman. 2012. A review of pharmacological and toxicological potentials of marine cyanobacterial metabolites. Journal of Applied Toxicology 32: 153–185.

Nair, S., B.V. Bhimba. 2013. Bioactive potency of cyanobacteria *Oscillatoria* spp. International Journal of Pharmacy & Pharmaceutical Sciences 5: 611–612.

Negri, A.P., G.J. Jones, M. Hindmarsh. 1995. Sheep mortality associated with paralytic shellfish poisons from the cyanobacterium *Anabaena circinalis*. Toxicon 33: 1321–1329.

Neilan, B.A., M.L. Saker, J. Fastner, A. Torokne, B.P. Burns. 2003. Phylogeography of the invasive cyanobacterium *Cylindrospermopsis raciborskii*. Molecular Ecology 12: 133–140.

Neilan, B., L. Pearson, T. Mihali, M. Moffitt, R. Kellmann. 2010. On the chemistry, toxicology and genetics of the cyanobacterial toxins, microcystin, nodularin, saxitoxin and cylindrospermopsin. Marine Drugs 8: 1650-1680.

Niedermeyer, T.H.J., A. Daily, M. Swiatecka-Hagenbruch, J.A. Moscow. 2014. Selectivity and potency of microcystin congeners against OATP1B1 and OATP1B3 expressing cancer cells. PLoS ONE 9: e91476.

Norris, R.L.G., G.K. Eaglesham, G.R. Shaw, P. Senogles, R.K. Chiswell, M.J. Smith, B.C. Davis, A.A. Seawright, M.R. Moore. 2001. Extraction and purification of the zwitterions cylindrospermopsin and deoxycylindrospermopsin from *Cylindrospermopsis raciborskii*. Environmental Toxicology 16: 391–396.

O'Brien, P.J., A.G. Siraki, N. Shangari. 2005. Aldehyde sources, metabolism, molecular toxicity mechanisms, and possible effects on human health. Critical Reviews in Toxicology 35: 609–662.

Oberemm, A., J. Fastner, C. Steinberg. 1997. Effects of microcystin-LR and cyanobacterial crude extracts on embryo-larval development of zebrafish (*Danio rerio*). Water Research 31: 2918–2921.

Ofek, P., D. Ben-Meir, Z. Kariv-Inbal, M. Oren, S. Lavi. 2003. Cell cycle regulation and p53 activation by protein phosphatase 2Calpha. Journal of Biological Chemistry 278: 14299–14305.

Orjala, J., D. Nagle, V. Hsu, W.H. Gerwick. 1995. Antillatoxin: an exceptionally ichthyotoxic cyclic lipopeptide from the tropical cyanobacterium *Lyngbya majuscula*. Journal of the American Chemical Society 117: 8281– 8282.

Osswald, J., S. Rellán, A. Gago, V. Vasconcelos. 2007. Toxicology and detection methods of the alkaloid neurotoxin produced by cyanobacteria, anatoxin-a. Environment International 33: 1070–1089.

Paerl, H.W., R.S. Fulton, P.H. Moisander, J. Dyble. 2001. Harmful freshwater algal blooms with emphasis on cyanobacteria. Scientific World Journal 4: 76-113.

Pouria, S., A. de Andrade, J. Barbosa, R.L. Cavalcanti, V.T.S. Barreto, C.J. Ward, W. Preiser, G.K. Poon, G.H. Neild, G.A. Codd. 1998. Fatal microcystin intoxication in haemodialysis unit in Caruaru, Brazil. Lancet 352: 21–26.

Rapala, J., A. Robertson, A.P. Negri, K.A. Berg, P. Tuomi, C. Lyra, K. Erkomaa, K. Lahti, K. Hoppu, L. Lepistö. 2005. First report of saxitoxin in Finnish lakes and possible associated effects on human health. Environmental Toxicology 20: 331–340.

Reyero, M., E. Cacho, A. Martínez, J. Vázquez, A. Marina, S. Fraga, J. Franco. 1999. Evidence of saxitoxin derivatives as causative agents in the 1997 mass mortality of monk seals in the Cape Blanc Peninsula. Natural Toxins 7: 311–315.

Runnegar, M., N. Berndt, N. Kaplowitz. 1995a. Microcystin uptake and inhibition of protein phosphatases: effects of chemoprotectants and self-inhibition in relation to known hepatic transporters. Toxicology and Applied Pharmacology 134: 264–272.

Runnegar, M.T., S.M. Kong, Y.Z. Zhong, S.C. Lu. 1995b. Inhibition of reduced glutathione synthesis by cyanobacterial alkaloid cylindrospermopsin in cultured rat hepatocytes. Biochemical Pharmacology 49: 219–225.

Sainis, I., D. Fokas, K. Varel, A.G. Tzakos, V. Kounnis. 2010. Cyanobacterial cyclopeptides as lead compounds to novel targeted cancer drugs. Marine Drugs 8: 629–657.

Saker, M.L., A.D. Thomas, J.H. Norton. 1999. Cattle mortality attributed to the toxic cyanobacterium *Cylindrospermopsis raciborskii* in an outback region of North Queensland. Environmental Toxicology and Water Quality 14: 179–182.

Salgia, R., A.T. Skarin. 1998. Molecular abnormalities in lung cancer. Journal of Clinical Oncology 16: 1207-1217.

Shen, X., P.K.S. Lam, G.R. Shaw, W. Wickramasinghe. 2002. Genotoxicity investigation of a cyanobacterial toxin, cylindrospermopsin. Toxicon 40: 1499–1501.

Shumway, S.E. 1995. Phycotoxin-related shellfish poisoning: Bivalve molluscs are not the only vectors. Reviews in Fisheries Science 3: 1–31.

Simola, O., M. Wiberg, J. Jokela, M. Wahlsten, K. Sivonen, P. Syrjä. 2012. Pathologic findings and toxin identification in cyanobacterial (*Nodularia spumigena*) intoxication in a dog. Veterinary Pathology 49: 755–759.

Singh, R.K., S.P. Tiwari, A.K. Rai, T.M. Mohapatra. 2011. Cyanobacteria: An emerging source for drug discovery. Journal of Antibiotics 64: 401–412.

Sinha, R., L.A. Pearson, T.W. Davis, M.A. Burford, P.T. Orr, B.A. Neilan. 2012. Increased incidence of *Cylindrospermopsis raciborskii* in temperate zones – is climate change responsible? Water Research 46: 1408–1419.

Sivonen, K., G. Jones. 1999. Cyanobacterial toxins. In: Toxic Cyanobacteria in Water: a Guide to their Public Health Consequences, Monitoring and Management (ed. I.Chorus, J. Bartram), pp. 41-112. London, UK: E & FN Spon.

Sivonen, K., K. Kononen, W.W. Carmichael, A.M. Dahlem, K.L. Rinehart, J. Kiviranta, S.I. Niemela. 1989. Occurrence of the hepatotoxic cyanobacterium *Nodularia spumigena* in the Baltic Sea and structure of the toxin. Applied and Environmental Microbiology 55: 1990–1995.

Smith, C., A. Sutton. 1993. The persistence of anatoxin-a in reservoir water. In: UK Report No. FR0427. Foundation for Water Research, Buckinghamshire (UK).

Sobue, K., K. Kanda. 1989. Alpha-actinins, calspectrin (brain spectrin or fodrin), and actin participate in adhesion and movement of growth cones. Neuron 3: 311–319.

Solter, P.F., G.K. Wollenberg, X. Huang, F.S. Chu, M.T. Runnegar. 1998. Prolonged sublethal exposure to protein phosphatase inhibitor microcystin-LR results in multiple dose-dependent hepatotoxic effects. Toxicological Sciences 44: 87–96.

Stevens, D.K., R.I. Krieger. 1991. Stability studies on the cyanobacterial nicotinic alkaloids anatoxin-A. Toxicon 29: 167–179.

Stewart, I., P. Webb, P. Schluter, G. Shaw. 2006. Recreational and occupational field exposure to freshwater cyanobacteria - a review of anecdotal and case reports, epidemiological studies and the challenges for epidemiologic assessment. Environmental Health 5: 6.

Stillwell, W. (ed.). 2013. An Introduction to Biological Membranes: From Bilayers to Rafts. 355 p. Elsevier B.V.

Su, Z., M. Sheets, H. Ishida, F. Li, W.H. Barry. 2004. Saxitoxin blocks L-type ICa. Journal of Pharmacology and Experimental Therapeutics 308: 324–329.

Suikkanen, S., M. Laamanen, M. Huttunen. 2007. Long-term changes in summer phytoplankton communities of the open northern Baltic Sea. Estuarine, Coastal and Shelf Science 71: 580–592.

Thirumavalavan, M., Y.-L. Hu, J.-F. Lee. 2012. Effects of humic acid and suspended soils on adsorption and photo-degradation of microcystin-LR onto samples from Taiwan reservoirs and rivers. Journal of Hazardous Materials 217–218: 323–329.

Tian, L., J. Cheng, X. Chen, S.H. Cheng, Y.L. Mak, P.K.S. Lam, L.L. Chan, M. Wang. 2014. Early developmental toxicity of saxitoxin on medaka (*Oryzias melastigma*) embryos. Toxicon 77: 16-25.

Tsuji, K., S. Naito, F. Kondo, N. Ishikawa, M.F. Watanabe, M. Suzuki, K. Harada. 1994. Stability of microcystins from cyanobacteria: effect of light on decomposition and isomerization. Environmental Science & Technology 28: 173–177.

Uzair, B., S. Tabassum, M. Rasheed, S.F. Rehman. 2012. Exploring marine cyanobacteria for lead compounds of pharmaceutical importance. The Scientific World Journal 2012: 179782.

Vajcová, V., S. Navrátil, M. Palíková. 1998. The effect of intraperitoneally applied pure microcystin LR on haematological, biochemical and morphological indices of silver carp (*Hypophthalmichthys molitrix* Val.). Acta Veterinaria 67: 281–287.

Valério, E., S. Chaves, R. Tenreiro. 2010. Diversity and impact of prokaryotic toxins on aquatic environments: A review. Toxins 2: 2359-2410.

van Apeldoorn, M.E., H.P. van Egmond, G.J.A. Speijers, G.J.I. Bakker. 2007. Toxins of cyanobacteria. Molecular Nutrition & Food Research 51: 7–60.

Vehovszky, Á., A.W. Kovács, A. Farkas, J. Győri, H. Szabó, G. Vasas. 2013. Pharmacological studies confirm neurotoxic metabolite(s) produced by the bloom-forming *Cylindrospermopsis raciborskii* in Hungary. Environmental Toxicology [in press].

Vijayakumar, S., M. Menakha. 2014. Prediction of new cyanobacterial drug for treating lung cancer. Biomedicine & Aging Pathology 4: 49-52.

Wang, J., J.J. Salata, P.B. Bennett. 2003. Saxitoxin is a gating modifier of HERG K^+ channels. Journal of General Physiology 121: 583–598.

Wang, M., D. Wang, L. Lin, H. Hong. 2010a. Protein profiles in zebrafish (*Danio rerio*) brains exposed to chronic microcystin-LR. Chemosphere 81: 716–724.

Wang, M., L.L. Chan, M. Si, H. Hong, D. Wang. 2010b. Proteomic analysis of hepatic tissue of zebrafish (*Danio rerio*) experimentally exposed to chronic microcystin-LR. Toxicological Sciences 113: 60–69.

Welker, M., C. Steinberg. 1999. Indirect photolysis of cyanotoxins: one possible mechanism for their low persistence. Water Research 33: 1159–1164.

Welker, M., C. Steinberg. 2000. Rates of humic substance photosensitized degradation of microcystin-LR in natural waters. Environmental Science & Technology 34: 3415–3419.

Wong, C.K., P. Hung,, H.C.C. Ng, S.-Y. Lee, K.-M. Kam. 2011. Cluster analysis of toxins profile pattern as a tool for tracing shellfish contaminated with PSP-toxins. Environmental Research 111: 1083–1090.

Wood, S.A., J.P. Rasmussen, P.T. Holland, R. Campbell, A.L.M. Crowe. 2007. First report of the anatoxin-a from *Aphanizomenon issatschenkoi* (cyanobacteria). Journal of Phycology 43: 356–365.

Wormer, L., S. Cires, D. Carrasco, A. Quesada. 2008. Cylindrospermopsin is not degraded by co-occurring natural bacterial communities during a 40-day study. Harmful Algae 7: 206–213.

Wu, M., T. Okino, L.M. Nogle, B.L. Marquez, R.T. Williamson, N. Sitachitta, F.W. Berman, T.F. Murray, K. McGough, R. Jacobs, K. Colsen, T. Asano, F. Yokokawa, T. Shioiri, W.H. Gerwick. 2000. Structure, synthesis, and biological properties of kalkitoxin, a novel neurotoxin from the marine cyanobacterium *Lyngbya majuscula*. Journal of The American Chemical Society 122: 12041–12042.

Zanchett, G., E.C. Oliveira-Filho. 2013. Cyanobacteria and cyanotoxins: from impacts on aquatic ecosystems and human health to anticarcinogenic effects. Toxins 5: 1896-1917.

Zhang, F., X. Xu, T. Li, Z. Liu. 2013. Shellfish toxins targeting voltage-gated sodium channels. Marine Drugs 11: 4698-4723.

Zhang, H., D. Shao, Y. Wu, C. Cai, C. Hu, X. Shou, B. Dai, B. Ye, M. Wang, X. Jia. 2012. Apoptotic responses of *Carassius auratus* lymphocytes to nodularin exposure in vitro. Fish and Shellfish Immunology 33: 1229-1237.

Zhang, X., P. Xie, D. Li, Z. Shi. 2007. Hematological and plasma biochemical responses of crucian carp (*Carassius auratus*) to intraperitoneal injection of extracted microcystins with the possible mechanisms of anemia. Toxicon 49: 1150–1157.

Zhao, M., S. Xie, X. Zhu, Y. Yang, L. Gan, L. Song. 2006. Effect of inclusion of blue-green algae meal on growth and accumulation of microcystins in gibel carp (*Carassius auratus gibelio*). Journal of Applied Ichthyology 22: 72–78.