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Review

Harmful algae: Effects of cyanobacterial cyclic peptides on aquatic invertebrates-a short review

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ABSTRACT

Cyanotoxins are secondary metabolites produced by cyanobacteria. Cyclic peptides, microcystins and nodularin commonly detected in water reservoirs of different parts of the world may induce various detrimental effects in a wide range of organisms from bacteria to humans. This paper presents the current state of knowledge on the effects of microcystins and nodularin on aquatic invertebrates: zooplankton, decapods and mollusks. Accumulation of microcystins and nodularin in these organisms and possible transfer of the cyanotoxins through the food web and possible threat to humans as consumers are also discussed.

Contents

1. Introduction

Cyanobacteria, (*Cyanophyta, Cyanoprocaryota*) also known as blue-green algae are phototrophic, prokaryotic microorganisms well adapted to a wide range of temperature and various, sometimes extreme environments, such as tropical regions or arctic ice. These organisms play an important role as oxygen producers and possess the ability to fix atmospheric nitrogen. (Lesser, 2008). However, intensive influx of biogenic elements such as nitrogen and phosphorus to shallow eutrophic water reservoirs with no thermal stratification and specific meteorological conditions (warm days and nights) may promote massive proliferation of strains producing cyanotoxins which induce various negative changes in

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aquatic and terrestrial animals (Xu et al., 2010). On the basis of the effect induced in organisms, these toxins can be divided into: hepatotoxins, neurotoxins, cytotoxins, dermatotoxins and irritant toxins. Hepatotoxins include a well-known, common heptapeptide microcystins (MCs) and pentapeptide nodularin (NOD), neurotoxins: anatoxin-a, anatoxin-a(s), homoanatoxin-a, saxitoxins formerly known as paralytic shellfish poisons (PSPs), cytotoxins such as cylindrospermopsin targeting a variety cells and irritant cyanotoxins such as lipopolysaccharides (LPSs) which are constituents of cyanobacterial cell wall (Sivonen, 2009).

Cyanotoxins may enter the organism after dermal or oral contact with the scum or contaminated water containing the toxins released from the decaying cyanobacterial cells after a bloom collapse or after inhalation of these peptides dispersed in aerosol. Various accidental intoxications caused by cyanotoxins have been documented in mammals: humans, cows, cattle, dogs, fish, invertebrates and in other organisms including bacteria, algae and higher plants (Carmichael, 1997; Griffiths and Saker, 2003; Wiegand and Pflugmacher, 2005). Aquatic invertebrates play many important roles in freshwater and marine ecosystems, such as clearing the water out of organic particles, serving as food source and oxygenating the sediment and mineralizing of the organic matter. (Lopez and Levinton, 1987; Covich et al., 1999; Wojtal et al., 2004). However, increased water concentrations of cyanotoxins may affect biochemical, physiological and behavioral parameters of these animals and, in a consequence, destabilize the ecological balance and nutritional relations in aquatic reservoirs. The purpose of this paper is to review the current state of knowledge on the effects and accumulation of cyclic peptides produced by cyanobacteria, MCs and NOD in aquatic invertebrates and consequences to other animal species and humans as consumers.

2. Effects of MCs on aquatic invertebrates

MCs are hepatotoxic cyanotoxins produced by cyanobacterial genera such as *Microcystis, Anabaena, Planktothrix, Anabaenopsis, Nostoc, Hapalosiphon* (Carmichael, 1992; Wiegand and Pflugmacher, 2005). These cyanotoxins are cyclic heptapeptides consisting of seven amino acids including two characteristic amino acids: methyldehydroalanine (MDHA) and 3-amino-9-methoxy-2,6,8-trimethyl-10-phenyldeca-4,6-dienoic acid (ADDA) (Fig. 1). The ADDA side chain is a structural amino acid necessary for the toxic action of the peptide. On the basis of two variable amino acids present in the structure of MCs, more than 85 congeners are currently distinguished (Rastogi et al., 2014). The best known and widely distributed variant, MC-LR has leucine (L) and arginine (R) as variable structural amino acids. Other common analogues of MC are MC-RR and MC-LA (Harada et al., 1990; Sivonen, 2009).

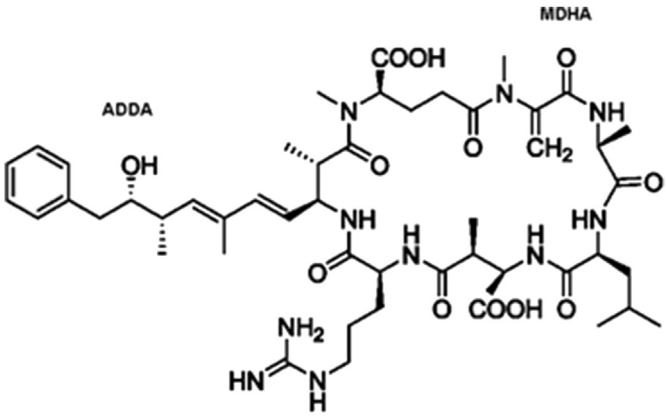

Fig. 1. Structure of microcystin-LR.

Results from the studies on the vertebrates indicate that the main target organ for MCs is the liver. In an intoxicated organism the peptides, are transported from the gastrointestinal tract to the cells via bile acid carrier transport system and enter hepatocytes inducing disruption of their cytoskeleton. Although inhibition of protein phosphatases type 1 (PP1) and 2A (PP2A) was recognized as the mechanism responsible for MC hepatotoxicity (Toivola et al., 1994; Runnegar et al., 1995), the cyanotoxin is also a causative agent of oxidative stress leading to cytoskeleton disruption and cell apoptosis (Huang et al., 2015). Studies in rats and fish, mussels and daphnids showed that MC-LR is eliminated from the organism by conjugation with glutathione which increases water solubility (Pflugmacher et al., 1998; Takenaka, 2001).

2.1. Zooplankton

Although zooplankton animals possess specific feeding mechanism based on water filtration which is ecologically beneficial for clearing water out of algae, efficacy of these animals to eradicate cyanobacterial blooms may be diminished when they are exposed to MCs. Results obtained from the field and laboratory studies suggest that MCs induce various deleterious effects in freshwater zooplankton ranging from subtle biochemical changes, growth and fecundity disturbances to increased mortality (Kaebernick el al., 2001; Ghadouani et al., 2003; Chen and Xie, 2003; Vilar et al., 2014). A number of studies showed lethal effects in *Daphnia* spp. exposed to *Microcystis aeruginosa* strains producing MCs (Nizan et al., 1986; Lürling and van der Grinten, 2003; Rohrlack et al., 2001, 2005). However, mortality rate may be reduced in the presence of green algae (Chen and Xie, 2004). It was demonstrated that sublethal concentrations of the cyanotoxins induce physiological changes in the alimentary system in daphnids manifested by alteration of ingestion rate, histopathological changes in the midgut epithelia and lowered activity of digestive enzymes: trypsin and chymotrypsin (Agrawal et al., 2001, 2005; Chen et al., 2005). Other physiological dysfunctions induced by MCs include reduction of heartbeat and thoracic limb movement, motility of mandibles and second antennae, decreased activity of the foregut and stimulation of the midgut muscles (Rohrlack et al., 2005). Although in mammals MCs toxicity is associated with the inhibition of protein phosphatases P1 and P2A activity, the results obtained in the studies on zooplankton invertebrates do not clearly show the same mechanism of action. The reduced activity of the phosphatases was reported in *Daphnia pulex*, *Daphnia pulicaria* and *Diaptomus birgei* (DeMott and Dhawale, 1995) but, on the contrary, studies with the use of P1 and P2A inhibition assay and a biotest with zooplankton crustacean *Thamnocephalus platyurus* revealed no correlation between P1 and P2A inhibition and acute toxicity of six MC congeners (Blom and Jüttner, 2005). Species-dependent toxicity of MCs in crustaceans may be also a result of different detoxification pathways.

MCs may induce developmental changes and growth disturbances of some zooplankton species. Studies on *Daphnia magna* showed that low concentrations of MC-LR may reduce the growth rate (Lürling and Van der Grinten, 2003; Dao et al., 2010). Exposure of parent daphnids to the cyanotoxin results in the decreased survival of the offspring or cessation of the eggs and reduced number of neonates and deformations of neonates such as incomplete development of the antennae may also occur (Dao et al., 2010; Cerbin et al., 2010). Although MC-LR does not affect egg hatching of estuarine calanoid copepod *Eurytemora affinis*, it was noted that survival of parents was decreased (Reinikainen et al., 2002). The mechanisms of the developmental toxicity of MCs on zooplankton