

### **Status of cyanotoxin contamination of animals in Africa**

This work is an original research article carried out in collaboration between the authors. The authors hereby declared that there is no any competing interests as regard this study.

#### **Abstract**

This study reviews the work done on the accumulation of cyanotoxins in fish and other animals in Africa. Microcystins (MC-LR, MC-RR and MC-YR) appear to be the main contaminating toxin in fish and other animals. In fish, the highest concentration was detected in the whole body of a small fish species (*Rastrineobolaargenta*) consumed whole by the populations. In big species, the intestine (followed by the liver) is the organ that accumulates the most cyanotoxins. The fish muscle, accumulates little or no cyanotoxins in most publications. And, *Oreochromis niloticus*, a phytoplanktonophagous species widely consumed by African populations, remains the most studied species. In other animal species, mortality due to cyanotoxins has been recorded in most publications in southern and central Africa and only one publication in northern Africa. No studies on this topic have been referenced in other parts of Africa.

**Key words:** accumulation, cyanotoxins, fish, other animals, Africa

#### **Introduction**

The massive introduction of organic matter and nutrients (nitrogen, phosphorus) through direct discharges of effluents (domestic, industrial and agricultural), contaminated runoff after passing over agricultural and non-agricultural surfaces, and atmospheric deposition into surface waters, disrupts the natural balances of aquatic ecosystems [1]. These effluent discharges added to the effects of global warming lead to massive primary production or eutrophication of aquatic environments [2; 3; 1]. Freshwater algae, a natural component of the aquatic ecosystem, can grow uncontrollably under appropriate environmental conditions, such as warm, nutrient-rich, stagnant water, and sometimes produce toxins that are harmful to the lives of other organisms such as fish, aquatic mammals, birds, and even humans that use these

waters for their life needs [4; 5]. Thus, the cyanotoxins produced by cyanobacteria or blue-green algae are metabolites that according to their effects on vertebrates are classified as hepatotoxins (microcystins, nodularins), cytotoxins (cylindrospermopsin), neurotoxins (saxitoxins, anatoxins), dermatotoxins, and general irritants (lipopolysaccharides) [6].

The first studies carried out in the context of cyanotoxin contamination of aquatic environments were often triggered by dermatological diseases, gastroenteritis or the death of contaminated people as in Zimbabwe [7] and Brazil [8], or the death of animals frequenting contaminated water bodies [9;10].

The aspect of bioaccumulation of cyanotoxins in aquatic animals has been addressed much more by North African authors, and those from the Lake Victoria region and South Africa (e.g. [11]; [12]; [13]; [14]). This work is a review of the work carried out in Africa in the context of vertebrate contamination by cyanotoxins in order to assess the current state of research in this area.

This bibliographic review took into account the theses and articles published on the work carried out in Africa. This work was conducted using Google scholar and PubMed search engines.

## **1. Different types of cyanotoxins analyzed in aquatic animals in Africa**

In the majority of the work done on animal mortality due to cyanotoxin exposure, microcystins (MC) appear to be the main toxin responsible for contamination of water consumed by animals and therefore for the death of these animals. Three variants of microcystins (MC-LR, MC-RR and MC-YR) were detected in most cases ([15]; [16]; [17]). In addition, nodularin has been incriminated in South Africa in the death of dog and cattle ([18] and [19]). Finally, a study revealed anatoxin-a in the tissues of flamingo carcasses on Lake Bogoria in Kenya [9].

## **2. Cyanotoxin contamination of fish in Africa**

The study of cyanotoxin contamination of fish in Africa is not sufficiently well documented due to the number of studies carried out on this subject. The vast majority of the work conducted on this subject concerns North Africa and the countries around Lake Victoria. As in water, microcystins are the most studied and detected variants in fish tissue.

### **2.1. North Africa**

Amrani (2016) did his thesis work on the bioaccumulation of cyanotoxins in fish from an aquaculture production on Lake Oubeira in Algeria. This study takes place from April 2010 to March 2011 on two fish species *Cyprinus carpio* (common carp) and *Anguilla anguilla* (European eel). During the study period the results obtained showed that the genus *Microcystis* is the most dominant and the concentrations of microcystins vary between 0.028 to 13.4 µg equivalent MC-LR.L<sup>-1</sup> [12].

The detection of microcystins in the different organs of the two fish, by the PP2A test, shows that the common carp has a high accumulation of toxins in the intestines while the European eel accumulates more in the liver. She has written an article on this subject (see [20]).

Mohamed and collaborators (2003) have previously carried out work on the same theme of bioaccumulation on *Oreochromis niloticus* in Egypt. The highest amount of microcystins determined by ELISA (Enzyme-linked immunosorbent assay) was recorded in the intestines (821 ng.g<sup>-1</sup> fresh weight (fw)), followed by the liver (531.8 ng.g<sup>-1</sup>fw) and kidneys (400 ng.g<sup>-1</sup>fw). The smallest amounts of MCYST were detected in muscle (102 ng.g<sup>-1</sup>fw) [11].

## 2.2 Sub-Saharan Africa

Furthermore, in Uganda, Nyakairu and collaborators (2010) studied the distribution of microcystins in the tissues of two fish *Oreochromis niloticus* and *Lates niloticus* from Lake Victoria. The presence of MC-RR, MC-LR and MC-YR in different organs (intestine, muscle and liver) was determined by liquid chromatography coupled to a mass spectrometry detector (LC-MS / MS). The MC content in *Oreochromis niloticus* from Lake Mburo was 1312.08 ng.g<sup>-1</sup>fw for intestine, 208.65 ng.g<sup>-1</sup>fw for muscle and 73.10 ng.g<sup>-1</sup>fw for liver and in Murchison Bay it was 1479.24 ng.g<sup>-1</sup>fw for intestine, 9.65 ng.g<sup>-1</sup>fw for muscle and 48.07 ng.g<sup>-1</sup>fw for liver while for *Lates niloticus* from Murchison Bay it was 27.78 ng.g<sup>-1</sup>fw for intestine, 1.86 ng.g<sup>-1</sup> for muscle and 3.74 ng.g<sup>-1</sup>fw for liver. They showed that the intestine accumulates more followed by the liver and muscle [15].

Poste and collaborators (2011) made a comparative study of the distribution of microcystins in tissues of several fish species of different trophic levels from Uganda and USA. The concentrations of microcystins assayed by ELISA ranged from 0.5 to 1917 µg.kg<sup>-1</sup>fw and from 4.5 to 215.2 µg.kg<sup>-1</sup>fw in muscle tissues and whole fish, respectively. And they showed that *Rastrineobola argentea*, a small zooplankton-eating cyprinid from Lake Victoria consumed whole, records the highest concentration of microcystins [13].

In *Oreochromis grahami*, a major consumer of cyanobacteria, the concentration of microcystin-RR in whole fish ranged from 0.41-0.79  $\mu\text{g}\cdot\text{g}^{-1}$  fw [21]. This work was diligent after the discovery of flamingo carcasses in Lake Bogoria, Kenya. This study shows that microcystins would probably be the cause of the death of these birds.

Semyalo and collaborators (2010) also conducted work on the bioaccumulation of microcystins in the tissues of Nile tilapia (*Oreochromis niloticus*). This study was conducted on two water bodies in Uganda (Lake Mburo and Murchison Bay of Lake Victoria). The concentration of MCs (LR, RR and YR) in the digestive tract (stomach, intestine with their contents), muscle and liver was determined by liquid chromatography coupled with a mass spectrometry detector (LC/MS/MS). They found that the concentration of MCs in the fish digestive tract was positively correlated with that of the water with a maximum of 300 and 390  $\mu\text{g}\cdot\text{kg}^{-1}$  fw respectively for Lake Mburo and Murchison Bay. The maximum for liver was 87.89  $\mu\text{g}\cdot\text{kg}^{-1}$  fw in Murchinson Bay and for muscle 5 and 6  $\mu\text{g}\cdot\text{kg}^{-1}$  fw in Murchinson Bay and Lake Mburo respectively [16].

In Africa, Nchabeleng and collaborators (2014), studied the bioaccumulation of microcystins in liver and muscle of two fish species, *Labeorosae* and *Oreochromis mossambicus* caught in the Loskop Dam. ELISA analyses revealed microcystin concentrations of 1.72  $\mu\text{g}$  MC-LReq  $\text{kg}^{-1}$  in liver and 0.19  $\mu\text{g}$   $\text{kg}^{-1}$  in muscle of *Labeorosae*, and 2.14  $\mu\text{g}$  MC-LReq  $\text{kg}^{-1}$  in liver and 0.17  $\mu\text{g}$   $\text{kg}^{-1}$  in muscle of *Oreochromis mossambicus*. These concentrations, in their opinion, indicate that consumption of the flesh of these fish could pose a health risk to humans [22].

Just recently, Simiyu and collaborators (2018) conducted work also on Lake Victoria in the Nyanza Gulf. They quantified the concentration of microcystin in the phytoplankton biomass of the lake and in small fish composed mostly of *Rastrineobolaargenta*. They used three assay methods: ELISA, PPIA and LC-MS/MS. The concentrations obtained by the PPIA method ranged from 25 to 109  $\text{ng}\cdot\text{g}^{-1}$  dry weight (dw) of fish compared to 14  $\text{ng}$  MC  $\text{g}^{-1}$  dw in lake water [14].

In Ethiopia, in Koka reservoir, Zewde and collaborators (2018) detected and quantified by LC-ESI-HRMS, three microcystin variants (MC-LR, -RR, -YR) in the liver of four wild fish the Nile tilapia (*Oreochromis niloticus*), Common carp (*Cyprinus, carpio*) and African sharp-toothed catfish (*Clariasgariepinus*). The concentrations ranged from 2.23  $\mu\text{g}\cdot\text{g}^{-1}$  dw of MC-RR to 591.60  $\mu\text{g}\cdot\text{g}^{-1}$  dw of MC-LR for liver and below the limit of detection for muscle [23]. This

study is the first in Ethiopia on this topic and suggest further work to prevent health risks from cyanotoxins in fish.

### 3. Cyanotoxin contamination of other animals in Africa

Following the death of a bull terrier after drinking water from Lake Zeekoevlei near Cape Town in South Africa, Harding and collaborators (1995) carried out investigations that led to the discovery of cyanobacterial blooms in the lake, dominated by *Nodularia spumigena*, where the presence of nodularin was confirmed [18]. Other cases of sheep and cattle deaths were studied by van Halderen and collaborators (1995). Three outbreaks of mortality of these animals were found; the water consumed by the animals in the first two outbreaks was dominated by *Nodularia spumigena* and that of the last one dominated by *Microcystis* sp. The presence of microcystin-LR was demonstrated in the third outbreak by high pressure liquid chromatography [19].

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Krienitz and collaborators (2003) sought to understand the cause of death of several flamingos migrating on different lakes in East Africa (Kenya, Tanzania and Uganda). Their work demonstrated the presence of microcystins (MC-LF, MC-LR, MC-RR and MC-YR) and Anatoxin-a in the waters at various concentrations and in flamingos at  $0.196 \mu\text{g}\cdot\text{g}^{-1}$  fw for microcystins and  $4.34 \mu\text{g}\cdot\text{g}^{-1}$  fw for Anatoxin-a [9]. Nonga and collaborators (2011) also identified cyanobacteria and their toxins as the cause of death of many flamingos in Tanzania. Three microcystin variants (MC-LR, MC-RR, and MC-YR) were detected in flamingo carcass tissues with a concentration of  $22 \pm 16 \mu\text{g}\cdot\text{g}^{-1}$  fw of MC-LR in the liver [17].

In October 2005, mortality of 12 turtles of the species *Emys orbicularis* and *Mauremys leprosa* was observed in Lake Oubeira in Algeria. The death of these two turtle species was observed during a *Microcystis* spp. bloom. The total microcystin content of the phytoplanktonic biomass was  $1.12 \text{ mg MC-LReq}\cdot\text{g}^{-1}\text{dw}$  while it was  $1192.8 \text{ mg MC-LReq}\cdot\text{g}^{-1}\text{dw}$  in the liver of *M. leprosa* and  $37.19 \text{ mg MC-LReq}\cdot\text{g}^{-1}\text{dw}$  in the viscera of *E. orbicularis* [24]. Thus, according to these authors, this mortality is due to exposure to microcystins.

Comment [U4]: phytoplankton

In the Kruger National Park in South Africa, mortality of zebra, wildebeest and white rhinoceros has been attributed to exposure to microcystins [25]. From the point of view of these authors, in addition to the climate, hippopotamuses contribute to the eutrophication of the ponds and pools of the park by adding nutrients through their excreta (urine and faeces)

[25; 26]. This leads to the development of cyanobacteria (especially *Microcystis* spp.) and the production of toxins that cause the death of these animals.

Most recently, Wang and collaborators (2021) incriminated cyanotoxins as the cause of death of 330 African elephants (*Loxodonta africana*) in Botswana between May and June 2020. According to them, the climate (low rainfall and high evaporation) would be the basis for the development of cyanobacterial blooms and the production of their toxins, notably MCs, in ponds and pools in hot and dry weather (Figure 1). Thus, the eastern and southern zones of Africa would be areas of high risk of exposure to cyanotoxins for megafauna and even for humans due to global warming [10].

Comment [U5]: and collaborators

### Conclusion

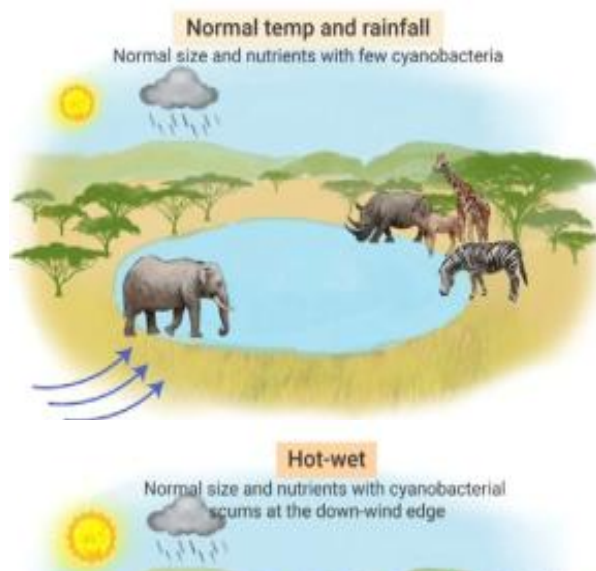
In fish, the highest concentration was detected in the whole body of a small fish species (*Rastrineobolaargenta*) consumed whole by the populations [12]. In large species, the gut (followed by the liver) is the organ that accumulates the most cyanotoxins (e.g. [11], [15], [16], [20]). The muscle or flesh of fish, the organ consumed by the populations, accumulates little or no cyanotoxins in most publications (e.g. [11], [15], [16], [23]). We had these same conclusions in a study in progress of publication. In addition, *Oreochromis niloticus*, a phytoplanktonophagous species widely consumed by African populations, remains the most studied species (e.g. [11], [15], [16], [23]).

In other animal species, mortality due to cyanotoxins has been recorded for most publications in the area around Lake Victoria for birds and in South Africa for domestic animals and wild animals. Only one publication in North Africa mentioned turtle mortality. No studies on this topic have been referenced in other parts of Africa.

The increase in temperature, salinity and anthropogenic activities leads to a massive development of cyanobacteria at the expense of other phytoplanktonic taxa [2]. Thus, cyanobacteria forming blooms and producing toxins in water bodies constitute a health risk for animal populations and even humans. In natural areas, such as parks, large herbivorous mammals also contribute nutrients to water bodies through their urine and feces.

Comment [U6]: lead

Comment [U7]: phytoplankton



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Figure 1: Schematic representation of changes in vegetation, water surface, and cyanobacteria concentrations and their poisoning risks to elephants under combined changing temperature and rainfall conditions [10].

In addition, global warming is an adjunct to both of these causes of eutrophication in African water bodies. Finally, the monitoring of water bodies must become a necessity for African governments to prevent health risks due to exposure to cyanotoxins.

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