Determination of water quality, trace elements contamination in *Oreochomis niloticus* and *Clarias gariepinus* reared in 2 types of ponds and health risk assessment in Cameroon

Evaluation de la qualité de l'eau, de la contamination par les éléments traces métalliques d'Oreochomis niloticus et Clarias gariepinus élevés dans 2 types d'étangs et évaluation des risques sanitaires au Cameroun.

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ABSTRACT:

Fish farming in Cameroon is growing very rapidly and fish available in the markets is mostly coming from fish farms, but domestic fish production is still low to meet demand. Intensification of production could lead to the occurrence of various types of contaminants that can affect the sanitary quality of farmed fish and consumer health. This study aimed to assess the quality of farmed fish collected in fish farms located in four regions of Cameroon (Center, South, West, and Littoral). Concentrations of arsenic, cadmium, lead, chromium, cobalt, and nickel were determined in pond water and muscles of *Oreochromis niloticus* and *Clarias gariepinus* raised in concrete and earthen ponds to evaluate health risk for consumers. Trace elements were determined using Inductively Coupled Plasma Optical Emission spectrometry (ICP-OES). Metals in water for concrete and earthen ponds shows there are no significant differences (p<0.05) between the rearing systems except for cobalt and nickel. Metal concentrations in water were lower when compared to the WHO recommended limits except for cadmium in all the ponds. For concrete ponds, heavy metal concentrations decreased in water in the sequence of Cd > As > Co > Ni > Cr > Pb. For earthen ponds the concentrations decreased as Cd > Co > As > Ni > Cr > Pb. Concentrations recorded in the muscles of the 2 species were above the safety limits recommended by FAO/WHO. The target hazard quotient of As was highest compared to other metals and higher than the acceptable limits according to US Environmental Protection Agency guidelines in all fish species. The level of each investigated metal, highlighted a very low health risk for consumers.

Keywords: Fish farming, Metals, Concrete ponds, Earthen ponds, Target hazard quotient.

RÉSUMÉ :

La pisciculture au Cameroun se développe très rapidement et le poisson disponible sur les marchés provient en partie des exploitations piscicoles, mais la production nationale de poisson est encore faible pour répondre à la demande. L'intensification de la production pourrait entraîner l'apparition de divers types de contaminants susceptibles d'affecter la qualité sanitaire des poissons d'élevage et la santé des consommateurs. Cette étude visait à évaluer la qualité des poissons d'élevage collectés dans les exploitations piscicoles situées dans quatre régions du Cameroun (Centre, Sud, Ouest et Littoral). Les concentrations d'arsenic, de cadmium, de plomb, de chrome, de cobalt et de nickel ont été déterminées dans l'eau des étangs et les muscles d'*Oreochromis niloticus* et de *Clarias gariepinus* élevés dans des étangs en béton et en terre afin d'évaluer les risques pour la santé des consommateurs. Les éléments traces ont été déterminés en utilisant la spectrométrie d'émission optique à plasma à couplage inductif (ICP-OES). Les métaux dans l'eau pour les étangs en béton et en terre montrent qu'il n'y a pas de différences significatives (p <0,05) entre les systèmes d'élevage, à l'exception du Cobalt et du Nickel. Les concentrations d'éléments traces dans l'eau étaient inférieures aux limites recommandées par l'OMS, sauf pour le cadmium dans tous les étangs. Pour les étangs en béton, les concentrations d'éléments traces dans l'eau décroissent dans l'ordre Cd> As> Co> Ni> Cr> Pb. Pour les étangs en terre, les concentrations décroissent dans l'ordre Cd> Co> As> Ni> Cr> Pb. Les concentrations enregistrées dans les muscles des 2 espèces étaient supérieures aux limites de sécurité recommandées par la FAO / OMS. Le quotient de danger cible de l'As était le plus élevé par rapport aux autres métaux et supérieur aux limites acceptables selon les lignes directrices de l'Agence de Protection de l'environnement des États-Unis pour toutes les espèces de poissons. Le niveau de chaque élément traces étudié, a mis en évidence un risque sanitaire très faible pour les consommateurs.

Mots clés : Pisciculture, éléments traces, étangs en béton, étangs en terre, quotient de danger.

1. INTRODUCTION

During the past years the global consumption of fish has increased due to growing population and rapid urbanization (Anderson et al., 2017). Furthermore, fish provides a healthy, low cholesterol source of protein and other nutrients including omega 3 fatty acid that reduce cholesterol levels and the incidence of heart disease (FAO, 2018, Golden et al., 2016). In addition to directly providing high-quality food, fisheries and aquacultures create economic value through the production, trade, and marketing of wild and farmed fish (Cai et al., 2019). By 2030, annual fish consumption is likely to rise to some 150-160 million tonnes, or between 19-20 kg per person (FAO, 2018). Considering that 33% of the wild fish stocks are overexploited, fish farming appears to be a great solution to the massive demand for animal protein (Kobayashi et al., 2015; Chan et al., 2019). Many production systems exist for fish farming, in Cameroon it's mainly earthen ponds and concrete ponds that are used to cultivate fishes. It's well known that aquatic organisms are characterized by accumulation of many harmful elements in their body amongst them heavy metals. The water used for fish farming contains very high concentrations of heavy metals which can affect adversely the quality of the final product cultivated fish and consumer health (Aderinola et al., 2012; Das et al., 2017). The accumulation of heavy metals in fish mainly depends on their concentrations in food and water, in this aspect the production system has an essential role. According to many authors high concentrations of heavy metals in water may have devastating effect on the ecological balance in the environment and in the diversity of aquatic organisms (Kamaruzzaman, 2011; Kundu et al., 2017; Chatta et al., 2016). The consequence of this is the occurrence of diseases on fish farms and a large loss on fish production.

The presence of contaminating heavy metals in freshwater ponds at concentrations above natural loads has become a growing concern. Recent studies have investigated for instance fish contamination by lead, cadmium and mercury which are very toxic (Zaza et al., 2015; Zheng Zhang et al., 2007; Stancheva et al., 2013).Various studies in different countries reported the contamination of different farmed fish species with a different type of pollutants, trace metals are of particular concern, due to their potential toxic effect and ability to bioaccumulate in aquatic ecosystems (Vutukuru, 2005; Dirilgen, 2001; Voegborlo et al., 1999; Canli et al., 1998). In many developing countries, metals pollution problems in the fish farming sites were serious as reflected by high metal concentration recorded in Malawi (Puchase and Jamu, 2009), Zimbabwe (Teta, Ncube and Naik, 2017). Other studies conducted in India and Nigeria (Nath and Bhoumik, 2013; Ibemenuga et al., 2013; Kumari et al., 2017) indicated high level of cadmium, arsenic and lead in water and fish tissues. Some regulatory agencies have categorized them as pollutants of high importance (US Enviromental Protection Agency, 2006). Trace metals occur naturally in the environment and are present in low concentrations in freshwater (VanLoon and Duffy, 2011). Rapid population growth, increased urbanization, the expansion of industrial activities, the exploration and exploitation of natural resources, the extension of irrigation and the spread of modern agricultural practices should be blamed for the occurrence of heavy metals. Increases use of fertilizers, which contain metals could also result in high concentration of trace metal in fish ponds. Fishes have been widely used as bio-indicators of pollution by metals in aquatic system because they occupy different trophic levels (Burger et al., 2002).

Cameroon like many other countries, is facing severe problem of freshwater pollution as almost 99% of the industrial waste water is discharged into streams and rivers without any treatment (Khan *et al.,* 2012) which

is undesirably distressing natural resources of the country (Rauf *et al.,* 2009). Domestic fish production is still low to meet demand. Fish farming appears as a solution to fill this gap. These last years there is a tendency to increase the number of fish farm and intensify the production to fill the gap. Intensification of production could lead to the occurrence of various types of contaminants that can affect the quality of farmed fish and consumer health. Few studies exist concerning fish ponds contamination by heavy metals in Cameroon. The studies have mainly concerned wild fish species commonly found in rivers and lake. For instance, Fonge et al., *(*2011) assessed the contamination of Dibamba river and *Arius heudelottii* by Co, Cd, Pb, Fe, Cu. Another study reported the contamination of heavy metals (Cr, Cd, Hg, Pb and Zn) in fish, mud and water from two urban lakes in Yaoundé (Demanou and Brumett, 2010). Nkwelle et al., (2012) works on the determination of water quality, and trace metals in endemic *Sarotherodon linellii, Pungu maclareni* and *Clarias maclareni*, in crater lake Barombi Mbo in the south west region of Cameroon. There is no report on water quality in fishponds and heavy metals contamination in farmed species; furthermore, no data exist on the potential health risk related to the consumption of contaminated fishes with respect to the rearing system. There is a need to investigate on the influence of intensification of production system on farmed fish in order to limit the risks and to improve the quality of water and final product. *O. niloticus* and *C. gariepinus* are amongst the most common fishes reared and consumed in Cameroon. Therefore, metal accumulation in these fish species presents serious public health. The fact that metals accumulate in the environment necessitates their continuous monitoring and assessment in respect of both ecological and human health impact. The purpose of the present study was to determine the level of heavy metal contamination (As, Cd, Cr, Pb, Co, Ni) in water, and muscle tissues of *O. niloticus* and *C. gariepinus* reared in earthen and concrete ponds. Then, assess the ecological and public health risks and to identify possible sources of the metals for future remedial action. Additionally, to compare the two systems with regards to heavy metals contamination and health risks.

2. MATERIAL AND METHODS

2.1. Study area

The study was conducted in four regions of Cameroon: the center (Yaoundé, Mfou) the south (Sangmelima), the littoral (Douala) and the west regions (Dschang). The different regions were selected as they represent a high potential for fish farming due to favourable resources and climatic conditions. The center region covers 69.000 km^2 and is composed of rolling hills on a vast plain with a mean altitude of 700–800 m, with lowered mounds. The climate has two wet seasons. The population density is low, with about 36 inhabitants/km^2 (BUCREP 2010; NIS 2006). The south region covers an area of 47.110 km^2, with a population of about 534.900 inhabitants and a density of 13.4 inhabitants per km^2 (NIS 2006). The Littoral region is covering an area of 20.239 km^2 and housing more than 2.202.340 inhabitants. The west region covers 13.872 km^2 and is mountainous, marked by highlands with a mean altitude of 1600 m and narrow valleys with catchments separating them. The climate has a unimodal wet season. The population density is relatively high, with about 143 inhabitants/km^2 (BUCREP 2010). The population density is 124 inhabitants per km^2.

2.2. Sample collection

All the samples, water and fishes were collected between April and August 2017, a total of 70 fish samples (35 samples of *O. niloticus* and 35 of *C. gariepinus*) and 35 water samples were randomly collected in 14 ponds respectively 8 earthen ponds (plate 1) and 6 concrete ponds (plate 2). The pond sizes vary between 200 and 500 m^2, each pond was sampled twice directly from fish farmers based in four regions of Cameroon as mentioned in Table 1. These regions were selected according to their importance in farmed fish production in the country. They have the largest proportion of fish farms in Cameroon.

Table 1: Number of fish and water samples per region

Region	Town	Fish farms	Production system	Pond type	Water(n)	Fish (n)	Number of ponds sampled
Centre	Yaoundé	Etokoss	Extensive	Earthen	5	10	2
-	-	odja	Semi intensive	Concrete	5	10	2
-	Mbalmayo	Fokou	Semi intensive	Concrete	5	10	2
-	Mfou	Bindo	Extensive	Earthen	5	10	2
Littoral	Douala	Agroworld	Semi intensive	Concrete	5	10	2
Sud	Sangmelima	Mimbo	Extensive	Earthen	5	10	2
Ouest	Dschang	Fokoué	Extensive	Earthen	5	10	2

Plate 1 : Concrete fish ponds   **Plate 2:** Earthen fish ponds

- *Water*

Triplicate 0.5 litre water samples from concrete and earthen were collected from fish farmers in each region. A water sampler was used to collect surface water (0–15 cm depth) at each specified location. A representative sample was transferred into low-density plastic bottles, rinsed thrice with ambient water. One representative 1.5 litres composite of water for each fish pond was then obtained. For trace metals analysis samples were stored in acid-washed low density polyethylene bottles. Raw water samples were

further transported in icebox to laboratory and were filtered through Whattman cellulose acetate membrane filter paper (0.45 mm) and kept in the freezer prior to trace metal analysis.

• *Fish*

Fish were collected to obtain information on the bioaccumulation of trace metals in fish pond area. Two species *Clarias gariepinus and Oreochromis niloticus* were chosen because they are amongst the farmed species mostly reared and consumed in the four regions. High consumption of these species was attributed to the fleshy nature and sweet taste. 70 fish samples, 5 each of *C. gariepinus* and *O. niloticus* were procured in seven fish farms located in each region. *C. gariepinus* samples have a mean weigth of 200 ± 47.68g and mean length of 20 ± 1.5 cm *O. niloticus* have a mean weigth of 130 ± 18g and mean length of 16 ± 1.45 cm. These fish samples were put in sterile polythene bags and taken in icebox to the laboratory where they were washed with running tap water to remove dirt. All the fish samples were then separately stored inside deep freezer at about $-4°C$ and were allowed to thaw, scales were removed and washed with tap water before dissected with sterile scissors to remove gills, operculum, vertebrae, heart, muscles and kidney. Only the muscles tissue were transferred into sterile sample bottles, labeled and kept for digestion and analysis of heavy metals.

2.3. Sample preparation and analysis

• *Determination of physico-chemical parameters of water samples*

The physico-chemical parameters of the water, including the temperature, pH, electrical conductivity (EC), and total dissolved solids (TDS) were determined and analysed, Temperature was measured with glass mercury thermometer, conductivity and TDS was measured using Hanna portable instrument conductivity meter (model HI991300) according to the Standard Methods for the Examination of Water and Wastewater (Clescerl et al., 1998).

• *Digestion of fish samples*

Fish samples were dried in an oven at 80–85 °C overnight. The samples were removed from the oven, allowed to cool, and ground in a clean mortar and pestle. Approximately, 0.5 g of each sample was placed in a teflon microwave digestion bomb with 10 mL of concentrated HNO_3. The samples were allowed to ramp to 180 °C for 5 min, digest at 180 °C for 9.5 min, and cool down for 5 min in a MARS 5 microwave digestion system (CEM, Matthews, NC). The samples were then transferred to clean volumetric flasks, and diluted with H_2O to 10 mL. The samples were stored at 5 °C until ready for analysis of metals (Meche et al., 2010). An aliquot of 1 L water sample was digested with 5 mL concentrated nitric acid to a final volume of about 25 mL. The digest was left to cool. It was then filtered into 50 mL volumetric flask and diluted to 50 mL mark with distilled water.

• *Trace elements determination*

The digested sample was then filtered through Millipore membrane filter of 0.45 µm (Type HV) and diluted up to 25 ml by addition of distilled water. This filtrate of each fish sample was then processed for analysis of fish muscle content of arsenic (As), cadmium (Cd), chromium (Cr), cobalt (Co), lead (Pb), and nickel (Ni), were using Inductively Coupled Plasma (Perkin Elmer, Model: Optima7000 DV ICP-OES) following

the Standard Method (AOAC, 2012). Detection limits for Pb, Cr and Cd were 0.01 ppm and for As, Cr, Co were 0.03. High purity metal standards were used for instrument calibration and also for accuracy checks after analyzing every five samples. Metal levels in fish tissues were expressed as mg kg^{-1} dry weight, while metal levels in water were expressed as mg l^{-1} (ppm). The wavelengths used for the detection and measurement of the trace elements are 193.696 nm; 228.802 nm; 267.716 nm; 228.616 nm; 220.353 nm; 231.604 nm respectively. The levels of metal contamination of water in relation to fish were expressed as bioconcentration factor (BCF) according to the methods of Lau et al., (1998) calculated as:

$$\text{BCF fish} = \frac{\text{Cfish}}{\text{Cwater}} \tag{1}$$

where, C fish is the mean concentration of the heavy metal in muscle of fish, C water is the mean concentration of the metals in water and BCF fish is the bioconcentration factor of the metals in the fish.

2.4. Health risk assessment

- ***Target hazard quotient (THQ)***

The target hazard quotient (THQ) is an estimate of the risk level (non-carcinogenic) due to pollutant exposure. To estimate the human health risk from consuming metal-contaminated fish, the target hazard quotient (THQ) was calculated as per United States Environmental Protection Agency (USEPA) Region III Risk-Based Concentration. The method of estimating estimated daily intake (EDI) was provided by the US-EPA Region III risk based concentration table (US EPA, 2009) and Wang, Sato, Xing, and Tao (2005). EDI of each toxic metal was calculated using the Equation:

$$(E_F \text{ x ED x FIR x } C_f \text{ x C) / (BWx } T_A) \tag{2}$$

where E_F is the exposure frequency (365 days/year); ED is the exposure duration, equivalent to average lifetime, 59 years for Cameroonian population WHO (2017); FIR is the fresh food ingestion rate (g/person/day), which was considered to be 42g/person/day for fish (Ali & Hau, 2001); Cf is the conversion factor (0.208) for fresh weight (FW) to dry weight (DW), Cm is the heavy metal concentration in foodstuffs (μg/ kg FW); BW is the average body weight (average adult body weight was considered to be 75 kg) and TA is the average exposure time for non-carcinogens (equal to E_F . ED) (Saha & Zaman, 2012). Health risk of consumers due to intake of metal in fish was assessed by using THQ in accordance with US-EPA Region III risk-based concentration table (US EPA, 2009) and Bortey-Sam et al., (2015). If THQ is less than 1 then the exposed population is unlikely to experience adverse effects; whereas THQ greater than 1 indicates a likelihood of non-carcinogenic effects, with an increasing probability as the value increase (Wang et al., 2005). THQ was calculated using the Equation :

$$THQ = \frac{(M_c \times \text{IR} \times 10^{-3} \times \text{EF} \times \text{ED})}{(RfD \times BW \times ATn)} \tag{3}$$

2.5. Quality assurance (QA)

The analytical method followed to ensure reliability of the study results was validated by measuring several quality parameters including precision, linearity, detection limits and spike recovery. Precision is describes as the closeness of individual measures of an analyte when the procedure is applied repeatedly to multiple samplings of a homogenous sample. Khan et al., (2013), evaluated precision by using relative standard deviation of 10 repeated determinations of one sample. Following this method, the percent coefficient of variation (CV %) were obtained for all analyte elements. The % RSD was found to be within the specification (% RSD should not be more than 15%). The linearity is the ability to elicit test results that are directly, or by a well-defined mathematical transformation, proportional to the concentration of analyte in samples within a given range. Linearity was established by preparing the calibration curves of all analyte elements. All calibration curves were prepared with six standard solutions including the blank. These were prepared in such a way that the concentrations all analyte elements in the samples were within the linear range of calibration curves and above the established lower linearity limit (Khan et al., 2013). A detection limit is the concentration of an analyte that results in signal intensity that is three times the standard deviation of the background intensity at the measurement wavelength. The limit of quantification (LOQ) is the minimum concentration that can be quantitatively determined with satisfying certainty. The LOQ is normally defined as 10 times the standard deviation (10s) for blank samples, and is thus approximately three times higher than the LOD. The analytical quality control for the all trace elements was also verified via recovery experiments by spiking at two selected concentrations of 1000 µg/kg and 100 µg/kg (Khan et al., 2014).

Table 2: Quality Assurance parameters of method used for elements determination

Element	Detection Limit (µg/g)	LOQ (µg/g)	Coefficient of variation (CV%)	Linear Correlation Coefficient (R^2)	Recovery
As	0.011	0.001	7.90	0.98	85
Cd	0.0052	0.02	2.00	0.98	84
Co	0.0001	0.002	12.82	0.98	85
Cr	0.0006	0.02	12.94	0.99	84
Ni	0.0006	0.083	9.75	0.99	80
Pb	0.006	0.05	10.92	0.99	85

2.6. Statistical analyses

The results obtained were subjected to statistical evaluation. The concentration of trace metals in water and tissue samples were reported as the average means ± standard deviation. From the normality test, the variables (metals in water and fish tissues) were not normally distributed. Consequently, a non-parametric test Kruskal Wallis was used to examine differences in mean values among the two species and rearing systems with significance at $p < 0.05$, using statistical package R.

3. RESULTS AND DISCUSSION

3.1. Physicochemical parameters of pond waters and concentrations of toxic metal in water

Physicochemical parameters (temperature, pH, conductivity, total dissolved solids) and concentration of toxic metals of the pond waters are shown in Table 3. There were no significant differences in the physico-chemical characteristics of the water from earthen and concrete, only for TDS and conductivity ($p < 0.05$).

Table 3: Physicochemical parameters and concentration of trace elements (As, Cd, Cr, Pb, Ni, Co) in water from the earthen and concrete ponds (Mean ± SD).

Parameters	Fish ponds		
	Concrete	**Earthen**	**Permissible levels**
Temperature (°C)	26±0.01	29±0.05	20-30
pH	6.70±0.02	6.76±0.06	6.5-8.5
TDS (mg/L)	483±0.06	56.66±0.03	1000
Conductivity(μs/cm)	980±0.07	123±0.06	20-1000
Hardness	23.31	22.17	
As (mg/L)	0005±0,007	0.007±0,009	-
Cd (mg/L)	0.049±0,08	0.015±0,02	0,01
Co (mg/L)	0.0004±0.0001	0.0003±0.0001	-
Cr (mg/L)	0.0003±0.0004	0.004±0.0007	0.05
Pb (mg/L)	0.0001±0.0003	0.001±0.0001	0.05
Ni (mg/L)	0.0001±0.0004	0.004±0.005	0.02

The physicochemical parameters and level of trace elements in pond water were compared with international standards. According to Madu et al., (2017), water quality parameters influence the heavy metals levels in freshwater bodies and their accumulation in aquatic biota. The findings of the study revealed that the mean temperatures for both earthen (29.5°C) and concrete (26°C) ponds were within the WHO recommended limits. Fish is a cold blooded animal, its temperature is dependent on the temperature of its environment. It changes with the temperature of the surroundings. The optimum water temperature for fish survival has been reported to be between 20-30ºC (Bhatnagar and Devi, 2013; Ntenegwe and Edema, 2008). The temperature changes affect both the metabolism and physiology of fishes, and so its productivity (Agbaire et al., 2015). It was also observed that there is no significant difference in temperature in the culturing facilities used for the experiment. The reasons can be ascribe to the fact that the earthen and concrete facilities are subjected to the same light intensity and maybe the type of holding structure does not influence temperature change. The present results are in line with recent studies conducted by Moshood (2017) and Agbaire et al., (2015) which reported that the temperature of ponds ranges between (30.9±0.1-29.4±0.1°C) and (26.73±1.730°C) respectively.

The pH measurement helps to determine if the water is a proper environment for fish, plants and algae (Olukunle et al., 2017). Fish are known to have an average pH of 7.4 therefore pond water average within this range is optimum. In the current study, the results obtained for pH were respectively in concrete ponds and earthen pond where within the limits. The desirable range for pond pH is 6.5 - 9.5 and acceptable range is 5.5 - 10.0 (Stone and Thomforde, 2003). It has been reported that pH value between 6 and 9 was appropriate for fish production (Bhatnagar and Devi, 2013). Thus, good pond productivity and fish health can be maintained. The findings are slightly lower than those obtained by Olukunle and Oyewumi (2017) in Akure (Nigeria) which reported a pH (7.10 ±0.06 - 8.39±0.01) for concrete ponds and earthen ponds. A comparative study conducted by Moshood (2017) to assess the water quality of four types of aquaculture ponds under different culture systems reported pH values of 7.88 ±0.2 for earthen and 6.75±0.2 for concrete.

The conductivity values of the ponds gave a good estimate of the condition of ponds under the different culture systems. Both TDS and electrical conductivity are usually positively correlated. The earthen pond has a lower conductivity value of 123 ms/cm, and lower value of TDS than concrete ponds. The FAO acceptable limit for conductivity in aquaculture is between 20 and 1500 μs/cm (DWAF. 1996). The conductivity and total dissolved solids in earthen and concrete ponds probably came from the source of water for the ponds, absence of phytoplankton and aquatic vegetation to assimilate the mineral salts and the effects of residual feed in water which on decomposition added some mineral salts into the water. The presence of algae and macrophytes which utilized the salts and rainwater as source of water might be the reason for the slightly lower concentrations of conductivity and TDS in both earthen and natural ponds. The conductivity and TDS range however in the ponds were ideal for fish culture. Fish farmers generally use artificial animal feeds (pellets) to supplement pond nutrients in the study area (Bouelet Ntsama et al., 2018), previous authors (Ehigbonare and Ogunrinde, 2010; Delince, 1992; Ogbeibu and Edutie, 2006) have also reported it to increase total dissolved solids in fish pond.

When compared with recommended limits, all the parameters were within the prescribed water quality standards for fish farming. Furthermore, there was no significant difference (P<0.05) obtained among the parameters, except for TDS and conductivity showing the effects of the different culture systems. All these results in the present study compare reasonably well with other results obtained (Olukunle and Oyewui, 2017). Water quality describes physical, chemical, biological and aesthetic properties of water which determine its fitness for use and its ability to maintain the health of farmed aquatic organisms. It is imperative to have all the physical, chemical and biological factors in the ponds in a balanced proportion and tolerable limit for optimum fish production.

3.2. Concentrations of toxic metal in water

Table 3 shows the concentration of trace element (As, Cd, Cr, Pb, Ni, Co) in water from the earthen and concrete ponds. Heavy metal concentrations decreased in concrete ponds water in the sequence Cd > As >Co > Ni > Cr > Pb. For earthen ponds, the concentrations decreased as Cd > Co > As > Ni > Cr > Pb. The analysis of heavy metals in water showed that the concentrations did not exceed WHO guidelines. The concentrations of each trace element in water did not vary significantly either in concrete or earthen ponds. Comparatively the values obtained were lower than other studies in same type of fish ponds carried out by Echor and Okaliwe (2017) that reported As (1.36-1.30 µg/L), Cr (0.72-0.90 µg/L) and Pb (2.00-1.60 µg/L). Aladasanmi et al., (2014) also reported high values above optimal levels for Pb (3.30 µg/L), Cr (9.11 µg/L), Co (10.53 µg/L) and Ni (90.22 µg/L). Fish performs all their body functions in water therefore the quality of water is very important, the type of ponds influenced the water quality and the absorption of heavy metals in the tissue of fishes, in the present study no significant differences occurs concerning the contamination by heavy metals. Thilza and Muhammad (2010) reported that concrete ponds pose lesser threat to the livehood of the fish as compared to the earthen pond types. This is due to the management practices on fish farms; the constant change of water in concrete pond type increases the chances of heavy metals to be absorbed in the tissues of fishes causing health threat to both fish and fish consumers. Water is the first source of contamination for fish, since all production materials move into the water, the food they consume, their waste products such as faeces and therefore the origin of water should be control, in the present study water comes mainly from borehole for concrete ponds and for earthen ponds from rivers (Bouelet Ntsama et al., 2018). Fish can be affected by contaminants present in these different sources. However, there should be periodic or constant water quality control of fish ponds in order to ensure fish consumer's safety. The knowledge of heavy metal concentrations in water is very important with respect to management, human consumption of these aquaculture products and to determine the useful monitor and remediation of the most polluted area (Pravin et al., 2011). The concentrations of heavy metals in concrete and earthen fish ponds were slightly higher than permissible limits for these metals. This underscores the need to periodically study our fish rearing environment.

3.3. Heavy metal in farmed fishes and bioconcentration factor (BCF) of trace elements

Trace metal concentrations of tested heavy metals (dry weight) found in *Oreochromis niloticus* species raised in concrete and earthen ponds have been presented in Figure 2. All the six tested metals i.e. cadmium (Cd), lead (Pb) and chromium (Cr), Arsenic (As), Nickel (Ni), Cobalt (Co) were present in samples collected from different sites. In the flesh of Nile tilapia *(Oreochromis niloticus)* samples, the following values were obtained as median concentration of trace element in concrete ponds, As (1.45 µg/g) ; Cd (0.155µg/g) ; Pb (0.32 µg/g); Co (0.37 µg/g); Ni (12.75µg/g); Cr (4.133µg/g). Heavy metal concentrations decreased in fish of concrete ponds with the sequence of Ni> Cr >As > Co > Pb > Cd. For earthen ponds

the median concentrations are As (1.39 µg/g); Cd (0.150 µg/g); Co (0.402 µg/g); Cr (2.17 µg/g); Pb (0.137 µg/g); Ni (0.570 µg/g). The concentrations decreased as Cr > As > Ni > Co > Cd > Pb.
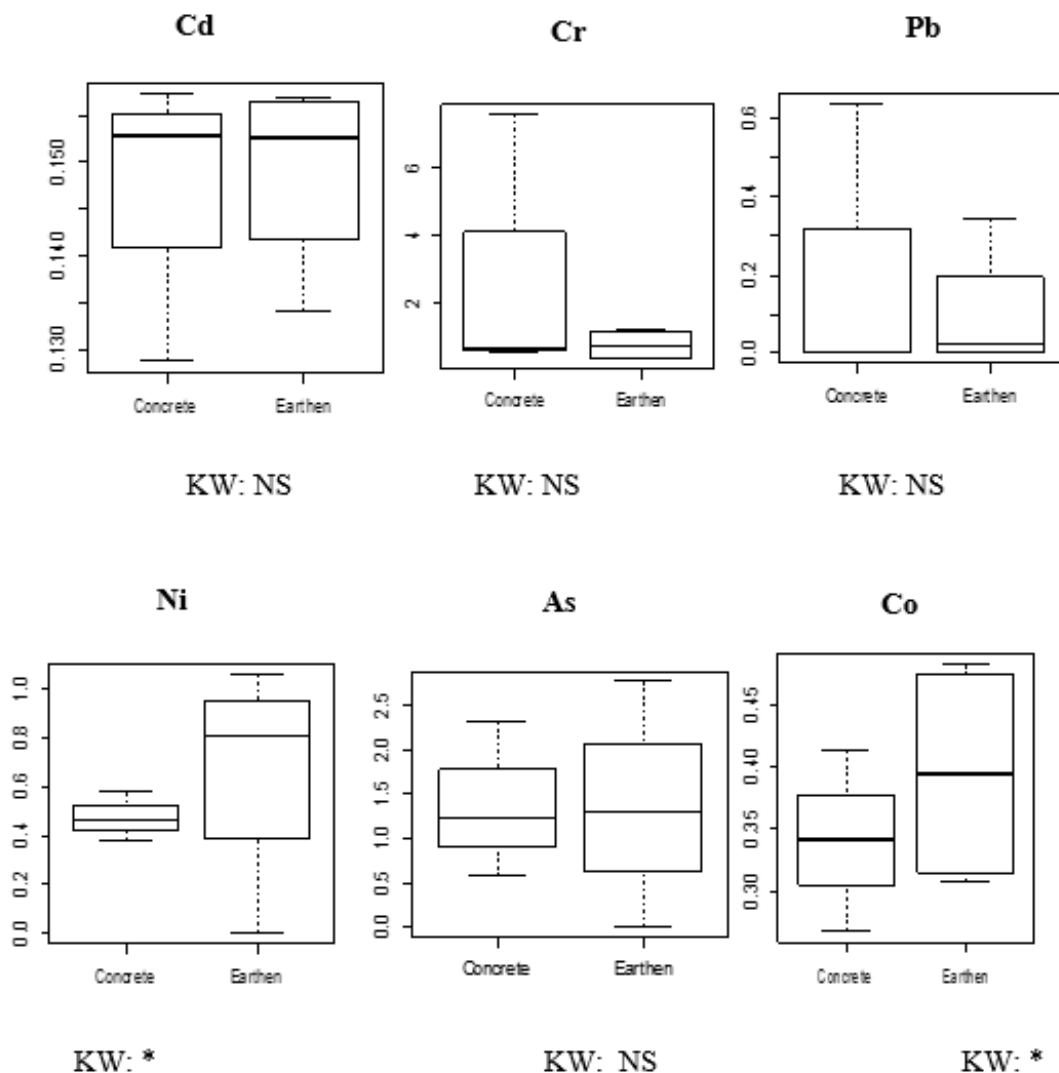

Figure 2: Comparison of Trace metal concentrations (µg.g−1 d.w.) in flesh of Nile tilapia *(Oreochromis niloticus)* reared in concrete and earthen. For each species, the significances of the level differences between both rearing systems are indicated below the boxplots. KW: Kruskal Wallis test; NS: Non significative.

Trace metal concentrations of tested heavy metals (dry weight) found in *Clarias gariepinus* species raised in concrete and earthen ponds have been presented in Figure 3.
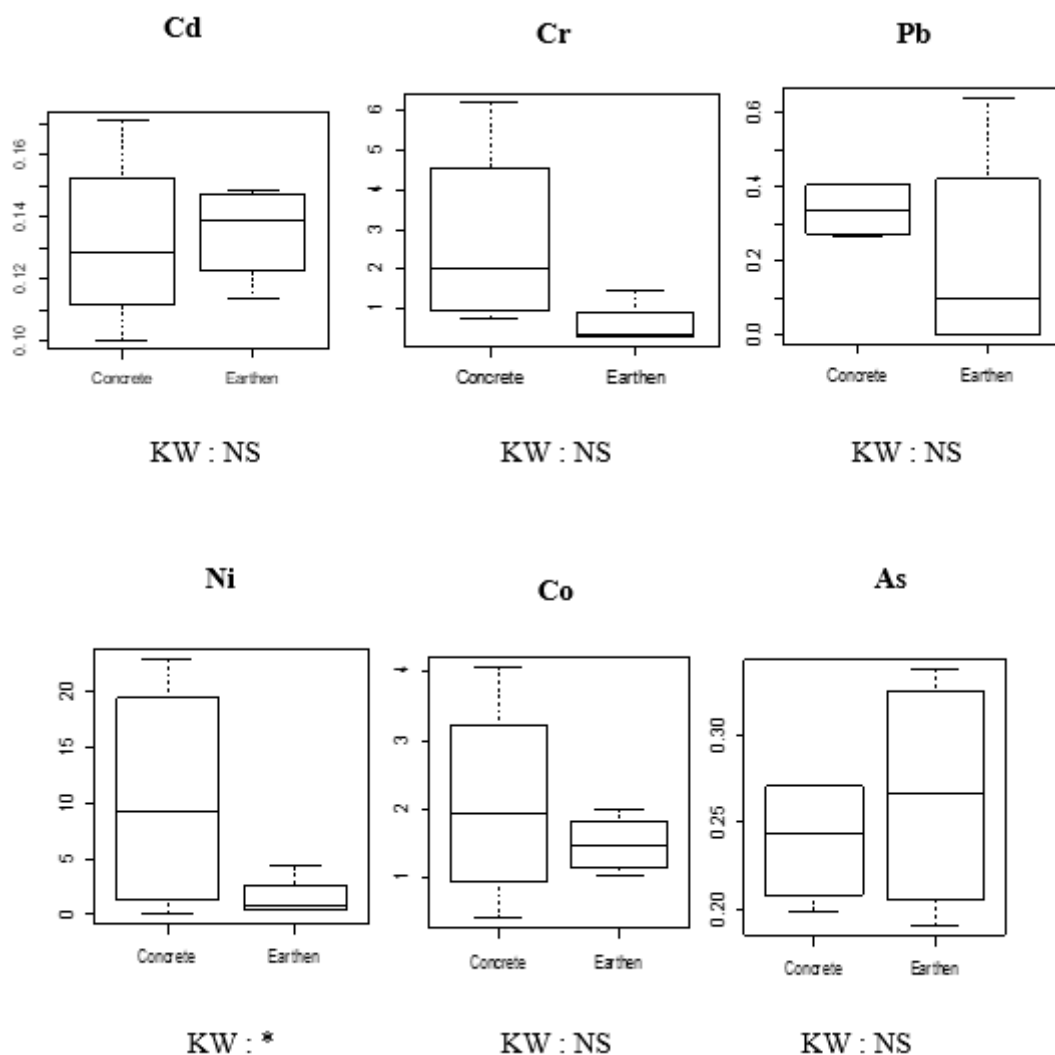
Figure 3: Comparison of Trace metal concentrations (µg.g−1 d.w.) in flesh of   African catfish (*Clarias gariepinus*) reared in concrete and earthen. For each species, the significances of the level differences between both rearing systems are indicated below the boxplots. KW: Kruskal Wallis test; NS: Non-Significative.

In the flesh of African catfish (*Clarias gariepinus*) samples, the following values were obtained as median concentration of trace element in concrete ponds, As (2,77 µg/g); Cd (0.11µg/g); Pb (0.33 µg/g); Co (0.24 µg/g); Ni (0.47µg/g); Cr (4.55µg/g). Heavy metal concentrations decreased in water of concrete ponds the sequence of Cr> As>Ni > Pb> Co > Cd. For earthen ponds the median concentrations are As (1.72µg/g); Cd (0.12µg/g); Co (0.28 µg/g); Cr (2.08µg/g); Pb (0.22 µg/g); Ni (9.23 µg/g). The concentrations decreased as Ni > Cr> As> Co > Pb > Cd.

The present study revealed the presence of heavy metals in both rearing systems. This highlights the fact that pollution could affect concrete fish ponds as well. The results obtained suggest that the presence of heavy metals such as As, Cd, Pb, Cr, Ni, Co and some of which are essential for life at trace levels are well above permissible concentrations making them a significant threat to ecosystems. Furthermore, toxic trace elements, arsenic, lead and cadmium were all present, in some cases at higher levels than acceptable concentrations. The levels of heavy metals concentrations in concrete fish ponds and earthen fish ponds were not significantly different when compared the two species except for Ni in Nile tilapia and cobalt for

African catfish. Concentrations of Arsenic in the flesh of Nile tilapia and African catfish do not varies so. These values were above the maximum limits of FAO/WHO (0.2 µg/g). Cadmium concentrations in all fish samples were above the maximum limits set by the European commission (EC, 2006) of 0.05 mg/kg. They were no significant differences at p<0.05. For Lead, Pb content in all samples from earthen ponds was below the maximum limits (0.3mg/kg), for concrete ponds the mean values slightly above the limit. The permissible limit for cobalt set by EU is 0.04 mg/kg, Co concentrations obtained in the present study were above the limits. The values do not differ for each rearing system, concrete (0.37 µg/g) and earthen (0.402 µg/g) in the flesh of Nile tilapia but for African catfish these values varies significantly respectively concrete (0.24 µg/g) and earthen (2.08 µg/g). Ni Concentrations in all fish samples were significantly different for Nile tilapia, concrete (12.75 µg/g) and earthen (0.570 µg/g). For African catfish, there was no difference regarding the contamination in concrete and earthen (0.47; 0.22 µg/g). The concentration obtained were all within the limits of 0.4 mg/kg set by the European commission and 1 mg/kg set by the USEPA (2000) except for Nile tilapia raised in concrete ponds. Cr concentrations were above the international limits (1 µg/g) for all the fish samples, for Nile tilapia when compare the two system concrete (4.133 µg/g) and earthen (2.17 µg/g) no differences was but for African catfish the difference was significant (concrete: 4.55 µg/g, earthen: 9.23 µg/g). The result obtained from this study was similar to the findings of Daniel and Mathew (2016) and Taweel et al., (2013). Madu et al., (2017) reported higher values compares to this study, for instance Pb (23.20 µg/g) and Ni (17.55 µg/g), these values were above the permissible values. These variations may be attributed to the differences between the localities, and the amount and source of pollution from an area to another, ecological needs, metabolism and feeding patterns of fish, and also the season in which studies were carried out in this study. Trace metals found commonly in fish are attributed to water, sediment or fish feed contaminated by the raw ingredients and by a mineral pack added by the manufacturer. In the current study, the presence of trace elements in fish were related to pond water, fertilizers and animal droppings use frequently in the study area (Bouelet Ntsama et al., 2018) and probably to fish feed, some authors reported the contamination of raw materials use to produce the feed (Das et al., 2017). The source of chromium in this research may have originated primarily from the feeds Cadmium is released to the environment in wastewater, and diffuse pollution is caused by contamination from fertilizers and local air pollution. The potential hazards of metals transferred to humans are probably dependent on the amount of fish consumed by an individual.

The bioconcentration factor of each studied metal was evaluated for muscle tissues of *C. gariepinus*, *O. niloticus (*Table 4). The trend of the bioconcentration factors was similar to the trend of the metals in the tissues of the fish. In addition, the bioconcentration factor of Mn in the gill and liver of the three fish was significantly different ($p < 0.05$). Among the fish, BCF values recorded for tissues were higher than those recorded for the tissues of *H. niloticus* and *C. gariepinus*. The overall highest BCF was recorded for Ni (67.99 ± 9.58) and the overall lowest BCF was recorded for Mn (8.79 ± 1.59) in the muscle of *H. niloticus*. In *H. niloticus,* the BCF of Pb was highest in all tissues studied, whereas in *C. gariepinus*, Pb had the highest BCF in the gills and muscle and Ni had the highest in the liver.

Table 4: Bioconcentration factor (BCF) values of heavy metals in muscle tissues of two fish species raised in concrete and earthen ponds

Fish species	Pond type	As	Cd	Co	Cr	Pb	Ni
C.gariepinus	**Concrete**	435.8	3.08	525	240.6	2000	5600
	Earthen	311.28	10.06	700	180.5	200	140
O.niloticus	**Concrete**	276	3.14	800	106.66	3400	7700
	Earthen	197.14	10.26	1066.66	80	340	192.5

Bioconcentration factors of trace elements give a measure of the metal concentration in an organism relative to its concentration in the medium (Ajima et al., 2015). The BCF of heavy metals in muscle of the fish species in the present study showed that there was appreciable bioaccumulation of the various heavy metals in the fish tissues. Because all BCF values determined for the two fish species studied were above 100, according to USEPA (1991) there is no risk to human health associated with consumption of the flesh of these fish. The results of this study are consistent with the report by Abdallah and Nweeze et al., (2008) in which pelagic fish (herbivores) recorded higher metal concentrations than benthic (carnivorous) fish. Khalid argued that since species such as tilapia are herbivores, they bioaccumulate a higher metal concentration in the flesh than carnivorous catfish. This suggestion is in agreement with the current study, the catfish being generally more contaminated than tilapia. This may be due to dietary habits of carnivorous fish observed during dissection, a low tendency for sequestration (higher BCF) of trace metals and a greater susceptibility of this species to the presence of trace metals pollutants relative to in Tilapia.

3.4. Relationships between trace element levels in fishes

Inter-metal correlations of fish species were assessed and presented in Table 3 and 4 for both species. The correlations between the different metals may result from the similar accumulation behavior of the metals in the fishes and their interactions. Noted significant correlations among metals may reflect a common source of occurrence and indicative of similar biogeochemical pathways for subsequent accumulation in the muscle tissue of fishes. In the present study, significant correlation was observed between studied some heavy metals, in Nile tilapia correlations were between Lead and Nickel, Lead and Arsenic, Nickel and cobalt. In African Catfish, correlations were less strong between Cadmium and Nickel, Chromium and Nickel, Lead and Nickel, Cobalt and lead. Correlation matrixes were used to understand the significant differences between selected heavy metals in fish. Higher correlations between metals may reveal similar contamination levels and primarily originate from a similar source and be transported together (Yi et al., 2011; Maanan et al., 2014). Metals with highly positive correlations are believed to have similar sources, while those with highly negative correlations are believed to have different origins (Al-Alimi and Alhudify, 2016).

Table 5: Pearson's moment correlation coefficients between the heavy metals in Nile tilapia *(Oreochromis niloticus)*

	Cd	Cr	Pb	Ni	As	Co
Cd	0.000	0.434	0.467	0.136	0.453	0.280
Cr		0.000	0.306	0.729	0.595	0.764
Pb			0.000	0.968	0.870	0.195
Ni				0.000	0.330	0.917
As					0.000	0.260
Co						0.000

Table 6: Pearson's moment correlation coefficients between the heavy metals African catfish (*Clarias gariepinus*)

	Cd	Cr	Pb	Ni	As	Co
Cd	0.000	0.475	0.421	0.841	0.687	0.167
Cr		0.000	0.531	0.848	0.516	0.727
Pb			0.000	0.811	0.283	0.814
Ni				0.000	0.296	0.330
As					0.000	0.754
Co						0.000

3.6. Health risk assessment

The mean concentrations of heavy metals in the muscle of the two fish species were used to evaluate the human health risk from fish consumption. The average concentration of each metal in the muscle tissues of the fish was converted from dry weight to wet weight, in order to evaluate the health risk (Table 6). The EDI of Ni was highest in all fish species (8.76 for concrete pond in *C. gariepinus* and 6.34 for earthen ponds*)* followed by that of Cr (3.13 in O. niloticus for earthen ponds). The target hazard quotient of As was highest in all fish species (for *C.* gariepinus 6.36 in concrete ponds and 3.96 for earthen ponds; For O. *niloticus,* 3.33 in concrete ponds and 3.32 for earthen ponds*),* all these values were above 1. The target hazard quotient of Pb was lowest in both species and rearing systems.

The THQ for most metals was less than 1, which indicates that consumers may experience minor health effects by these metals consumption, except for As. Arsenic had THQ values between 3.32 to 6.76 for species and fish ponds. Hazard indices calculated for five metals (Pb, Cd, Cr, and Ni) in samples from both specie were all below 1, indicative of a non-carcinogenic effect on human health for all the four metals. This means that the exposure of these metals to the human population from the consumption of farm – raised will not result in any considerable health risks that may be associated with the metals. The

accumulation of metals in fish depends on equilibrium between absorption and depuration rates, and thus may reflect localized bioavailability of these substances. It also has to do with the concentration of the trace metal in the surrounding water as well as the feeding habits of the fish species. The adverse effects of heavy metals on aquatic and human lives are well known. Fish lose sense of smell in polluted water and consequently this affects their feeding ability (Brian 2013). In humans, acute or chronic exposure to heavy metals can lead to various disorders such as cancer and can also result in excessive damage due to oxidative stress induced by free radical formation (Monisha et al., 2014).

Table 7: Health risks assessment for individual metal by the determination of target hazard quotients (THQ) for fish species

Fish species	Metal	Concrete (µg/g)	EDI	THQ	Earthen (µg/g)	EDI	THQ	(RfD)
C. gariepinus	Cd	0.11	0.07	0.15	0.12	0.08	0.16	0.0005
	Cr	4.55	3.13	0.62	2.088	1.43	0.28	0.005
	Pb	0.33	0.23	0.01	0.228	0.15	0.01	0.14
	Ni	12.75	8.76	0.43	9.23	6.34	0.31	0.02
	As	2.77	1.91	6.36	1.729	1.18	3.96	0.0003
	Co	0.24	0.16	0.55	0.28	0.19	0.64	0.0003
O. niloticus	Cd	0.15	0.1	0.21	0.145	0.09	0.19	0.0005
	Cr	4.13	2.84	0.56	0.6941	0.48	0.09	0.005

4. CONCLUSION

The present investigation reveals that all the physicochemical parameters and heavy metals except for cadmium in surveyed fish ponds were within the recommended limits for good fish production. The concentration in the fish flesh was slightly above the maximum limits and all the hazard quotient were below 1, except for Arsenic. There were no significant differences between concrete and earthen ponds. Thus, fish farming in the different regions of Cameroon investigated demands regular monitoring of physico-chemical parameters and metals status for effective management. Fish therefore, fish farmers should possess basic water quality kits to regularly monitor basic water quality parameters of their ponds. Principles of Hazard Analysis of Critical Control Point (HACCP) should be strictly adhered to in fish farm site location and production processes, so that our aquaculture product can go to the international market. The use of vascular plants that possess abilities to absorb these metals via their roots in ponds contaminated with pollutants can help to remedy such environment. Government should put in place a regulatory body which will control aquaculture production and ensure that licensed fish farmers are in line with the international norms.

5. ACKNOWLEDGEMENTS

6. COMPETING INTERESTS

The authors declare that they have no competing interests.

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