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RESEARCH ARTICLE

Trace elements in sediments, blue spotted tilapia *Oreochromis leucostictus* (Trewavas, 1933) and its parasite *Contracaecum multipapillatum* from Lake Naivasha, Kenya, including a comprehensive health risk analysis

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Abstract This study presents the distribution of 15 major and trace elements in sediments and fish and their pericardial parasites from Lake Naivasha, Kenya. The lake is one of the few freshwater lakes in the Great Rift Valley and is under strong anthropogenic pressure mainly due to agricultural activities. Its fish provide a valuable protein source for approximately 100,000 people in the area. Fish and their parasites have been acknowledged as indicators of environmental quality due to their accumulation potential for both essential and nonessential trace elements. A total of 34 specimens of the blue spotted tilapia *Oreochromis leucostictus* and pooled samples of their pericardial parasite, the anisakid nematode *Contracaecum multipapillatum* (larvae 3), were examined. Element concentrations were determined by inductively coupled plasma-optical emission spectroscopy (ICP-OES) and graphite furnace atomic absorption spectrometry (GF-AAS). The concentrations of elements in the sediments reflected the geology of the area and did not point to pollution: none of the investigated trace elements, including Pb, Cd, Cu, and Zn, showed elevated values. In contrast, concentrations in the fish muscle were elevated for Li, Sr, Cd, and Zn, with high target hazard quotients (THQ>0.1) indicating a potential health risk to the consumers of this fish. Fish liver showed significantly higher concentrations of the trace elements Fe, Mn, Cd, and Cu compared to the muscle and *C. multipapillatum*. In the parasite, Zn had the highest concentration, but the worms only minimally accumulated trace elements in relation to their fish host.

Keywords Trace elements · *Contracaecum multipapillatum* · *Oreochromis leucostictus* · Bioindicator · THQ · Lake Naivasha

Introduction

Trace elements are natural components of the aquatic environment (Ochieng et al. 2007), but their levels have significantly increased in some areas due to domestic, industrial, mining, and agricultural activities, leading to pollution. In a recent report of the world's worst pollution problems, heavy metals are among the top threats; they jeopardize the health of tens of millions of people (McCartor and Becker 2010). The report further states that more than 100 million people worldwide are estimated to be at risk from toxic pollution at levels

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above the international health standards. In Kenya, the need for studies on pollutants in aquatic ecosystems and their biota was recognized more than three decades ago as a result of rapid population expansion, growing industries, and increasing urbanization (Onyari 1981). Since then, several studies have been conducted on trace elements including heavy metals in different lakes in Kenya, among them Lake Naivasha. Tarras-Wahlberg et al. (2002), Ochieng et al. (2007), Kamau et al. (2008), and others have already investigated the levels and sources of heavy metals such as Cu, Cd, Zn, Fe, Pb, and Ni in Lake Naivasha. In addition, Njogu et al. (2011) and Mutia et al. (2012) showed that the most important sources of heavy metal pollution in the Lake Naivasha basin are its major tributary, the Malewa River, and some flower farms surrounding the lake. Therefore, the studies of the latter and other authors on trace elements and heavy metals in the lake have focused on the anthropogenic sources of these metals and concentrations in the muscle of fish. No studies from Lake Naivasha have partitioned trace elements in different fish organ tissues or examined the accumulation of trace elements in fish parasites; trace elements such as Li, Mo, Sr, Rb, and others have also been neglected.

Fish parasites have been recognized for their bioindicative potential regarding pollution in the last decades. Among other properties, certain parasites have shown a high potential for metal accumulation in relation to their fish hosts. Studies have mostly involved endo-helminthes, of which acanthocephalans and cestodes appear to be the best investigated taxa. Among others, Sures (2001, 2003), Jirsa et al. (2008), and Oyoo-Okoth et al. (2010) have shown the impressive ability of those intestinal worms to accumulate heavy metals to a high degree, showing bioaccumulation factors (BAF) up to several thousand for parasites compared to fish muscle, but varying considerably from species to species.

Only a few studies are available on nematodes. In Africa, little research has been done, linking parasitism and pollution. Regarding heavy metal accumulation, Retief et al. (2006) and Oyoo-Okoth et al. (2010) have examined cestodes and their fish hosts from South African river systems.

The blue spotted tilapia *Oreochromis leucostictus* is a very important commercial fish in Lake Naivasha. It is benthopelagic and occupies the inshore zone, being common in lagoons. It feeds on phytoplankton and detritus. The parasite chosen for this study was an anisakid nematode, *Contracaecum multipapillatum* (L3), from the pericard of the fish host because it was the only helminth parasite occurring in sufficient numbers and quantity. Anisakid nematodes are widespread and abundant in many regions around the world, including Africa, Europe, Asia, Australia and North and South America (Barson and Marshall 2004). They therefore meet the basic criteria of sentinel organisms. Larval stages of *Contracaecum* spp. usually occur in the body cavity and mesenteries of fish, whereas the adults inhabit the intestine of piscivorous birds, especially pelicans, cormorants, herons, and darters.

The current study was designed to determine a wide spectrum of trace element concentrations in sediments and in the muscle and liver of the blue spotted tilapia and in its pericardial parasite *C. multipapillatum*. The aim was to study bioaccumulation in both the fish host and its parasite and therefore gain new insights into this issue. In addition, a comprehensive health risk analysis with parameters adapted to Kenyan realities was undertaken to show possible risks for human fish consumers, including some elements that have been neglected so far in this area. This is a step forward in shedding new light on potential problems arising from fish consumption from Lake Naivasha.

Materials and methods

Study site

The study was conducted in Lake Naivasha, Kenya between February and August 2011. The lake is situated at 00°45′S and 36°20′E (Kamau et al. 2008) in a closed basin at an altitude of 1,890 m above sea level and covers approximately 160 km^2 in the eastern Rift Valley of Kenya (Electronic supplementary material—ESM Fig. 1). It is the only freshwater lake in the Rift Valley without a surface outlet but with a substantial exchange with groundwater (Clarke et al. 1990). It is shallow (approximately 6 m mean depth), with a volume of 4.6 km^3. It is bordered by papyrus *Cyperus papyrus* in some sections, and the overall composition of aquatic macrophytes is in a state of change (Tarras-Wahlberg et al. 2002), probably due to anthropogenic influences such as destruction of littoral vegetation, eutrophication along with plant, and animal introductions. Most of its freshwater inflow (approximately 80–90 %) comes from the Malewa River (Kamau et al. 2008) with an estimated mean annual flow of 153 million m^3 and a catchment area of 1,730 km^2, followed by the Gilgil River with an estimated average annual flow of 24 million m^3 and a catchment area of 420 km^2; an additional river, the Karati River, flows only intermittently. The basin area is generally semiarid, receiving a mean annual rainfall of 620 mm, while the mean annual evaporation is estimated at 1,735 mm. Evaporation generally exceeds precipitation throughout the year except at peak rainfall, with the rainfall trend being bimodal with a major peak in April-May and a minor peak in October-November. The water from Lake Naivasha is used extensively for agriculture (horticultural farms: approx. 77 million m^3/year), geothermal power generation (approx. 1 million m^3/year), domestic water supply, commercial fishing, tourism, and recreation as well as ranching and game farming. The key environmental problems facing the lake are water abstraction, leading to changes in water level, eutrophication, pollution, and invasive species as well as the decline in fish stocks and biodiversity (Harper et al. 2011).

Water and sediment sampling

In situ measurements of the pH, dissolved oxygen, conductivity, and temperature were performed 10 cm below the water surface at the point where the fishing nets were set using a portable Hach Field Case multiparameter meter (Model Multi HQ40d, USA). A total of 10 water samples (about 20 ml each) were obtained biweekly from February to August 2011 from the same depth using a syringe and were immediately filtered with 0.2-μm nylon filters into pre-cleaned (acid washed) high-density polyethylene (HDPE) bottles and acidified with 0.1 mL concentrated nitric acid (TraceSELECT®, Fluka). Similarly, 10 sediment samples were obtained biweekly from February to August 2011 using stainless Ekman grab samplers; caution was taken not to obtain the samples from sediment directly in contact with the surface of the Ekman grab sampler. The samples were placed into 50-mL polypropylene (PP) centrifuge tubes. In the laboratory, samples were first sieved through a 1-mm colander to remove pebbles, and the small fraction (<1 mm) was dried at 105 °C to a constant weight and used for further analyses.

Fish sampling and parasitological examination

In total, 34 fish were caught by fishermen using gill nets of 2.5-in. mesh size (2–4 fish obtained biweekly from February to August 2011). Fish were transported alive in aerated tanks to a laboratory at the Biological Sciences Department, Egerton University, Kenya where they were killed by cervical dislocation and dissected following standard procedures used in parasitological analyses. The fish were weighed and their total length (TL) determined. Where found, *C. multipapillatum* was collected from the pericardium of the fish; some were preserved in 4 % formaline, others in absolute (95 %) ethanol for further identification purposes, and still others were thoroughly rinsed with double-distilled water and dried in the oven to weight constancy at 60 °C for trace element determinations. Fish tissues were carefully removed using a ceramic knife and plastic tweezers. Approximately 1 g of dorsoventral muscle and the identical mass of liver was washed with double-distilled water and then dried in the same way as parasite samples. The water content of fish muscle was determined by weighing samples before and after drying, yielding a mean of 80 % (*n*=10).

Trace elements determination

The water, dried sediment, dried fish tissues, and parasites were transported to the Institute of Inorganic Chemistry, University of Vienna. The sediment samples were first homogenized with mortar and pestle. Samples were then split, with about 1 g weighed into Teflon bombs for acid leaching, using 8 mL of HNO_3 34 % (*Trace*SELECT® Fluka) in a microwave MARS XPRESS system (CEM Corporation), and another 1 g into crucibles for the determination of organic matter (TOC) content using the loss on ignition (LOI) method determined as percent AFDW (ash-free dry weight). After treatment in the microwave oven, samples were transferred quantitatively into 15- or 20-mL flasks and brought to volume with Millipore water. Before measurement, samples were filtered through 0.2-μm PTFE syringe filters (VWR) and, where necessary, diluted.

Fish tissues and parasites (where possible 0.2 g dry weight) were digested in 8 ml of 34 % HNO_3 (*Trace*SELECT® Fluka). Reference samples comprising 0.2 g (dry weight) of fish protein DORM-3 and 0.2 g (dry weight) of marine sediment PACS-2 obtained from the National Research Council Canada (NRCC) were digested and diluted in the same manner as described above for fish tissues and sediments, respectively. To determine the detection limits, analytical blanks were prepared without insertion of a sample. Elements were determined using inductively coupled plasma-optical emission spectrometry (ICP-OES) using an Optima 5300DV (Perkin Elmer). When necessary, graphite furnace atomic absorption spectrometry (GF-AAS) using a PinAAcle 900Z (Pelkin Elmer) was used. The results for the reference samples showed recovery rates between 95 and 104 %, demonstrating the appropriateness of the sample preparation used. A table showing detection limits and recovery rates is given in the ESM Table 1.

Risk assessment

The target hazard quotients (THQ) were determined for the trace elements in order to assess the risk to people who consume *O. leucostictus* in the area. THQ is the ratio between the potential exposure to a substance and the reference dose (level at which no adverse effects are expected) (USEPA 2012). A $THQ \leq 1$ means no significant health risk for fish consumers, and a $THQ > 1$ means a possible risk regarding the respective metals.

Furthermore, a THQ of 0.1 was later suggested for noncarcinogens to account for additive effects (USEPA 2012). Even though the standard fish equation inputs for fish consumption are given (USEPA 2012), we adapted certain variables in order to reflect the local reality because of the wide discrepancies. For example, the USEPA body weight (BW_a) is given as 70 kg, whereas the average body weight in Africa is 60.7 kg (Walpole et al. 2012); the per capita fish consumption in Kenya is 5.2 kg (Lokuruka 2009) which translates to a daily consumption of 14.2 g/day, whereas USEPA (2012) set the value at 54 g/day. The equation for determining THQ according to USEPA (2012) is the following:

$$\mathrm{THQ} = \frac{\mathrm{EFr} \times \mathrm{EDr} \times \mathrm{IRFa} \times C}{\mathrm{RfDo} \times \mathrm{BWa} \times \mathrm{AT}}$$

where the variables are defined as follows: $\mathrm{EF_r}$ is the exposure frequency (350 days/year), $\mathrm{ED_r}$ is the exposure duration (30 years), $\mathrm{IRF_a}$ is the fish consumption per day (0.0142 kg/day) since the per capita is 5.2 kg/year, in Kenya (Lokuruka 2009), C is the metal concentration in the edible portion of fish (milligrams per kilogram wet weight (ww)), RfDo is the reference dose, oral (milligrams per kilogram per day, according to the updated 2012 Regional Screening Level (RSL) in the fish ingestion table (USEPA 2012), $\mathrm{BW_a}$ is the body weight, adult 60.7 kg, for Kenya (Walpole et al. 2012), and AT is the averaging time for noncarcinogens (365 days/year). The results for metal concentrations in milligrams per kilogram dry weight were multiplied by 0.2 to refer to wet weight based on the calculated water content of 80 % (see above).

Additionally, the mean concentrations of the trace elements were also compared with FAO/WHO recommended values. In all these comparisons, an average body weight of 60.7 kg (adult) and a daily fish consumption of 14.2 g (5.2 kg per year) were adhered to as defined above.

Statistical analysis

Data were analyzed using Predictive Analytics Software (PASW statistics 18, SPSS). A nonparametric Friedman's test was used to analyze differences for Si, Al, Fe, Cu, Zn, Mn, Sr, Pb, and Cd because data were not normally distributed. Multiple comparisons were performed using the Wilcoxon's signed-rank test with a Bonferroni correction of the level of significance ($\alpha=0.05$). A Wilcoxon's signed-rank test was used in analyses for Rb and Mo differences.

Descriptive statistics for Li were also determined. All graphs were drawn using SigmaPlot 10.0. The BAF were determined using the formula:

$$\mathrm{BAF}(x\ y) = \frac{\text{concentration of trace element in } x}{\text{concentration of the trace element in } y}$$

in which the variables x and y stand for matrices that are compared to each other, such as sediment, fish muscle, fish liver, and parasites.

Results

Physicochemical parameters

A summary of the physicochemical parameters monitored during the study is presented in Table 1.

Table 1 Physicochemical parameters of water (10 cm below surface) measured in situ during the study period (February-August 2011) in Lake Naivasha and dissolved major cations (0.2 μm filtered), $n=10$

Parameter	Mean	Min-max
T (°C)	21.4	19.6–24.4
pH	9.0	8.4–9.6
Dissolved oxygen (mg/L)	6.6	5.5–8.8
Dissolved oxygen (% sat)	93.9	70.7–126
Conductivity (μS/cm)	355	238–426
Na (mg/L)	25.4	22.9–29.5
K (mg/L)	10.9	10.0–13.4
Ca (mg/L)	25.8	20.9–31.6
Mg (mg/L)	8.4	7.8–10.6

Characteristics of the fish samples

The average length (TL) of the *O. leucostictus* sampled was 15.6±0.3 cm (6.7–28.4) and weight 69.0±6.9 g (5.0–418) (mean±SE). The prevalence of *C. multipapillatum* was 51.8 %, with a mean intensity of 2.3 nematodes per fish. The parasite was located in the pericardium exclusively.

Trace element concentrations in sediment

The order of concentration of trace elements in sediment was Al>Fe>Mn>Zn>Rb>Sr>Li>Pb>Cu>Cd. Mo was below the detection limit. Most of the trace elements were positively correlated ($p<0.05$) with the organic matter content of the sediment, with the exception of Sr (Table 2).

Trace element concentrations in *O. leucostictus*

The liver had the highest concentration of the trace elements Fe, Mn, Cu, and Cd compared to the muscle and *C. multipapillatum* ($p<0.05$) (Table 3). The concentration was, in descending order, as follows: liver>*C. multipapillatum*>muscle for Mn, Fe, Cu, and Cd (Fig. 1). Although Al and Si concentrations were not statistically different between the muscle and *C. multipapillatum* ($Z=-0.744$, $p>0.05$ and $Z=-0.180$, $p>0.05$, respectively), they were both significantly lower than in the liver ($p<0.05$). Zinc (Zn) was highest in the muscle compared to the liver and *C. multipapillatum* ($p<0.05$) (Fig. 1). Strontium (Sr) concentration did not different significantly between muscle and liver ($Z=-1.222$, $p>0.05$) but was higher in both than in *C. multipapillatum* ($p<0.05$) (Table 3). Zn and Sr showed the pattern: muscle>liver>*C. multipapillatum* in a descending order of concentration. Mo was below the detection limit for the muscle but was higher in the liver versus *C. multipapillatum* ($p<0.05$). Li was below the detection limit in liver and in *C. multipapillatum*. Rb was below the detection

Table 2 Concentrations of elements in sediment samples from Lake Naivasha and organic matter content (OM) as compared to literature data of other Rift Valley lakes and shale (milligrams per kilogram dw, $n=10$)

Element	Mean	SE	r (TOC)	Lake Nakuru[a]	Lake Naivasha[b]	Lake Bogoria[a]	TEF[c]	Shale[d]
Li	17	6	0.85					66
Na	908	140		98,100		79,200		9,600
Mg	3,210	220						15,000
Al	31,700	2,770	0.85					80,000
K	6,460	504		32,800		28,700		26,600
Ca	8,310	595						22,100
Mn	633	306	0.76		1,118			850
Fe	24,800	6,780	0.86	46,300		53,300		47,200
Cu	11.5	6	0.90		10.33		35.7	45
Zn	138	51	0.89	236	229.6	222	123	95
Rb	82.9	32	0.89	120		108		140
Sr	63.3	10	0.17 ns	48		39.4		300
Mo	<1.25							2.6
Cd	0.34	0.15	0.95		0.73		0.6	0.3
Pb	12.5	4	0.84		25.34		35	20
OM (%)	17.9	2.8						

r correlation coefficient for organic matter and the trace elements, *ns* correlation not significant, *TEF* threshold effect level as a sediment quality guideline, *shale* the Earth's crust geochemical background value of sedimentary rocks

[a] Jirsa et al. (2013)

[b] Ochieng et al. (2007)

[c] MacDonald et al. (2000)

[d] Turekian and Wedepohl (1961)

limit in *C. multipapillatum* but was not significantly different between the muscle and liver ($Z=-2.009$, $p>0.05$). The order of trace element concentration in the muscle was Zn>Li>Si>Rb>Fe>Al>Sr>Mn>Cd>Cu>Pb, while that of the liver was Fe>Cu>Si>Zn>Mo>Mn>Rb>Al>Cd>Sr>Pb; in *C. multipapillatum*, the order was Zn>Fe>Si>Al>Mo>Cd>Mn>Cu>Sr>Pb.

Table 3 Trace element concentrations in muscle, liver, and *C. multipapillatum* from *O. leucostictus* in Lake Naivasha: values are means (milligrams per kilogram dw)

Element	Muscle	SE	Liver	SE	*C. multipapillatum*	SE
Li	299	31	<0.5		<0.5	
Al	5.24*	0.74	11.1	1.8	3.91*	0.99
Si	34.4*	2.3	129	15	32.8*	6.15
Mn	2.43	0.3	20.5	1.6	1.14	0.09
Fe	14.3	1.5	1,930	272	40.5	1.6
Cu	0.54	0.03	470	46	1.06	0.13
Zn	604	82	73.1	2.1	45.6	1.6
Rb	18.2*	0.8	15.6*	1.2	<2.9	
Sr	2.67*	0.52	2.38*	0.19	0.66	0.08
Mo	<0.4		48.5	3.1	2.06	0.41
Cd	0.74	0.09	2.44	0.24	1.17	0.26
Pb	0.024	0.015	0.108*	0.052	0.169*	0.117

SE standard error

*$p>0.05$ (do not differ significantly)

Bioaccumulation factors for trace elements in *O. leucostictus*

Only a few trace elements showed BAF values >1. These included Li, Cu, Cd, and Zn for the fish tissues and Cd for *C. multipapillatum* when comparing element concentrations in biota samples to the respective levels in sediment (Table 4).

Target hazard quotients

The THQ values are shown in Table 5. Lithium (Li) and Zn had the highest values among the other elements.

Discussion

Physicochemical parameters of water

The values of the physicochemical parameters agreed well with earlier studies from the lake (Ochieng et al. 2007; Mutia et al. 2012) and did not fluctuate highly during the study

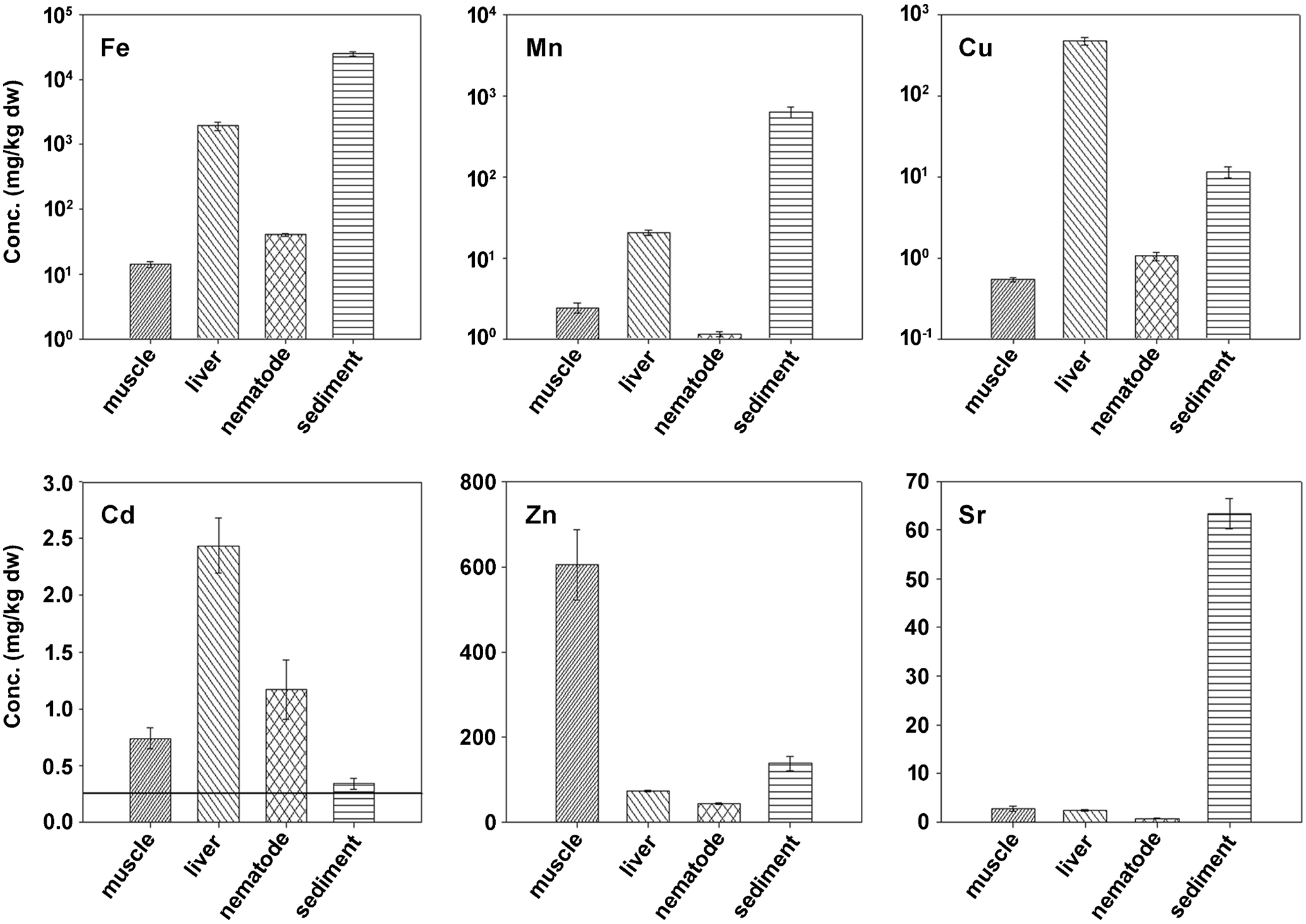

Fig. 1 Concentration patterns of selected trace elements in various matrices from Lake Naivasha, *Horizontal line* maximum allowance for fish used for human consumption in the EU for Cd

period. In comparison to "average fresh water systems," a particularly high K content was recorded, which implies effects in overall K metabolism and helps explain the relatively high concentrations of Rb in fish, as discussed under Rb.

Table 4 Bioaccumulation factors (BAF) of selected trace elements in *O. leucostictus* and *C. multipapillatum* from Lake Naivasha (mean)

Element	Muscle/ sediment	Liver/ sediment	*C. multipapillatum*/ sediment	*C. multipapillatum*/ muscle
Li	17.9			
Al	<0.001	<0.001	<0.001	0.746
Mn	0.004	0.032	0.002	0.500
Fe	0.001	0.075	0.002	2.94
Cu	0.047	40.9	0.092	1.96
Zn	4.37	0.528	0.326	0.075
Rb	0.217	0.183		
Sr	0.041	0.038	0.01	0.250
Cd	2.16	7.08	3.41	1.58
Pb	0.002	0.009	0.014	7.04

Trace element concentrations in sediment

Of the analyzed trace elements, the mean concentration of Al was highest in the sediment, followed by Fe, Mn, Zn, and Rb. Table 2 shows comparisons of the mean concentrations in sediments from Lake Naivasha and other studies. Cu, Mn, and Zn levels compared well with earlier studies from the same lake, whereas Cd and Pb levels were lower than those reported by Ochieng et al. (2007) and Mutia et al. (2012). To our knowledge, this is the first report of Li, Rb, and Mo from the lake. Aluminum (Al), Li, Fe, Cu, Rb, Sr, Mo, and Pb in sediment were below the average concentration for shale of sedimentary rocks, which is considered to be the normal

Table 5 Target hazard quotients (THQ) for various trace elements in *O. leucostictus* from Lake Naivasha (RfDo, reference dose, oral according to USEPA (2012))

	Li	Al	Mn	Fe	Cu	Zn	Sr	Cd
RfDo	0.002	1	0.14	0.7	0.04	0.3	0.006	0.001
THQ	198	0.007	0.023	0.027	0.018	2.67	0.590	0.980

background level in the Earth's crust (Turekian and Wedepohl 1961). These results reflect the lake's geology. Manganese (Mn) and Cd were within the range of normal background levels, but Zn was elevated compared to the average shale (Turekian and Wedepohl 1961). An earlier study reported higher Cd, Fe, and Zn concentrations in the Lake Naivasha catchment and attributed this to the volcanic rocks or erosion of iron-enriched lateritic soils in the catchment (Tarras-Wahlberg et al. 2002). Cu, Cd, and Pb were below the threshold effect level (TEL) estimated by MacDonald et al. (2000), so no adverse effects are expected; Zn approximated the TEL.

Most trace elements, except Sr, showed a positive significant correlation with the organic matter content of the sediment (Table 2). These findings agree with other studies, e.g., Yang et al. (2010). As Lake Naivasha is considered eutrophic (Kitaka et al. 2002), we expect nitrates to play a role in reducing the formation of Sr-organic matter complexes, which could partly explain the insignificant correlation obtained in this study.

Variation in trace element concentrations in fish tissues

The liver of *O. leucostictus* had a significantly higher concentration of most trace elements than the muscle. This result reflects the general trend, but there were exceptions. For instance, whereas Zn and Sr concentration were highest in *O. leucostictus* muscle, Li was below the detection limit in the liver, although highly prevalent in the muscle. This is in agreement with Mohamed and Gad (2008) on Zn but contradicts findings by Uysal et al. (2009), Gül et al. (2011), and others, who found Zn highly concentrated in the liver. Zn is an essential element and fish actively regulate its concentration in the muscle (Hofer et al. 1995) but are only able to do so up to a certain concentration in the water. In contrast, Sr is a biochemical analog of Ca, and its uptake and regulation are through the Ca pump system. Since Ca plays a role in muscle function triggering contraction, the muscle concentration of Sr may be related to that of Ca, hence higher in muscle versus liver.

Each trace element concentration in the fish tissues is briefly discussed below.

Li

We detected Li in high concentration only in *O. leucostictus* muscle, supporting the findings by Chassard-Bouchaud et al. (1984) on marine fish species, but contrasting with the conclusion by Aral and Vecchio-Sadus (2008) that Li can occur in all organs and tissues. To the best of our knowledge, no comparable literature is available for Li concentration in freshwater fish.

The THQ for Li was 198 (Table 5), which points to a very high risk for fish consumers here. Furthermore, a provisional adult recommended daily intake of 14.3 μg/kg body weight of Li has been suggested by Aral and Vecchio-Sadus (2008). On this basis too, our study found very high concentrations in the edible part of the fish, potentially posing a threat to fish consumers here.

Al

The Al concentration was higher in the liver than the muscle. This agrees with the findings by Moiseenko and Kudryavtseva (2001) in white fish *Coregonus lavaretus* from the Kola region of Russia, who recorded 4.8±0.6 μg/g dry weight (dw) in the muscle and 19±2.9 μg/g dw in the liver; this area was considered polluted by metals due to mining and other activities. Even higher Al values have been reported by Budambula and Mwachiro (2006) for redeye labeo *Labeo cylindricus* from the polluted Nairobi River, Kenya (70.0 μg/g ww for the muscle and 150 μg/g ww for the liver).

The THQ for Al was 0.007, i.e., no consumer risk. Additionally, FAO/WHO (2011) reduced the provisional tolerable weekly intake value for Al from 7 to 1 mg/kg body weight/week: even on this basis, our results show no potential risk exposure.

Mn

The Mn concentration was higher in the liver than the muscle. These results generally agree with those of Budambula and Mwachiro (2006) from the Nairobi River. The Mn levels in *O. leucostictus* muscle compare well with literature values: 1.75±0.05 μg/g dw in white fish *C. lavaretus* and 2.27±0.5 μg/g in brown trout *Salmo trutta* from the metal-polluted Kola region (Moiseenko and Kudryavtseva 2001). Lower values include 1.0±0.1 μg/g dw in rainbow trout *Oncorhynchus mykiss* from the Yesilirmak River in Tokat, Turkey (Mendil et al. 2010). Higher values than we reported were found by Silva and Shimizu (2004): 6.62 μg/g dw in Nile tilapia *O. niloticus* from a hydropower reservoir in Sri Lanka.

The THQ of Mn was 0.023, i.e., no risk to fish consumers here.

Cu

The Cu concentration was higher in the liver than the muscle, which generally agrees with many findings (Avenant-Oldewage and Marx 2000; Budambula and Mwachiro 2006; Jirsa et al. 2008). This reflects the livers' important role in storing and detoxifying Cu. The muscle levels we found compare well to those reported for largemouth bass *Micropterus salmoides* from Lake Naivasha (0.31±0.05 μg/g dw) by Njogu et al. (2011). Lower Cu values but still in the same order of magnitude as in our study have been reported from the same lake: in *O. leucostictus* (0.27±0.08 μg/g dw) and in common carp *Cyprinus carpio* 0.28±0.11 μg/g dw (Njogu et al. 2011).

The THQ of Cu was 0.018. FAO/WHO (2011) has set a toxicological guidance value, a provisional maximum tolerable daily intake (PMTDI) of 0.05–0.5 mg/kg body weight. These values exceed the accumulation levels we found for the muscle, demonstrating no risk due to the consumption of this fish.

Zn

In contrast to most trace elements discussed above, the Zn concentration was higher in the muscle than the liver. The reasons have been described above. The muscle levels were tenfold higher than those reported by Mendil et al. (2010) from chub *Squalius cephalus* (63.5±6.5 μg/g dw). From Kenya, the following authors have reported lower Zn concentrations than we found: Njogu et al. (2011)—7.31±0.89 μg/g ww in *O. leucostictus* from Lake Naivasha, Budambula and Mwachiro (2006)—70 μg/g ww in the redeye labeo *L. cylindricus* from Nairobi River, and Mavura and Wangila (2003)—238 μg/g dw in *Tilapia grahami* from Lake Nakuru.

The THQ of Zn was 2.67, demonstrating a clear risk to fish consumers here.

Rb

Rb was higher in the muscle than the liver, but not significantly different ($p>0.05$). In *O. leucostictus* muscle, we found Rb concentrations comparable to those of Campbell et al. (2005) for several fish species from Lake Erie and two Arctic lakes (Lake Hazen and Resolute Lake). They reported 4.5±0.4 μg/g ww in freshwater drum *Aplodinotus grunniens* as the highest concentration. Conversely, the concentrations we found are much lower than those reported by Guevara et al. (2006): mean value of 57.6(±5.7) μg/g ww for brown trout *S. trutta* and 45.9(±6.2) μg/g ww for creole perch *Percichthys trucha* in lakes of Patagonia, Argentina. Similarly, Silva and Shimizu (2004) reported a range of 20.90–70.75 μg/g dw in the flesh of nine fish species in Sri Lanka. The latter authors also reported 30.10 μg/g dw in Mozambique tilapia *O. mossambicus*, 21.60 μg/g dw in Nile tilapia *O. niloticus*, and 70.75 μg/g dw in redbreast tilapia *Tilapia rendalli*.

We found Rb in both muscle and liver, which agrees partly with the findings of Peters et al. (1999), showing muscle to be the main location of Rb. Note, however, that no literature is available on Rb in fish livers. Although there is some disagreement on the importance of Rb as an ultra-trace essential element to humans and other biota, even small concentrations in combination with other metals such as Pb, Mo, and As can be toxic to fish and other organisms (Yamaguchi et al. 2007). Neither safe concentrations nor the RfDo (reference dose) are known, preventing the determination of the THQ for Rb. Nonetheless, experimental toxic responses linked with elevated Rb have been observed in mammals fed low-K high-Rb diets; these responses are linked to physiological interference with K and Na (Kosla et al. 2002).

Sr

More Sr was present in the muscle than in the liver of *O. leucostictus*, in agreement with, for example, Moiseenko and Kudryavtseva (2001). Although those authors reported the highest concentrations in the skeleton and gills of white fish *C. lavaretus* and brown trout *S. trutta*, the values were notably higher in the muscle versus in the liver, similar to this study. Fish take up Sr and its analog Ca from water, food, and sediment. The uptake is through the Ca transport system located in the chloride cells of gills and enterocytes of the intestine. Sr is accumulated in the bone and other calcareous tissues (Chowdhury and Blust 2002) and is considered a nonessential element in fish. The THQ value for Sr is 0.590, and it could therefore pose a risk (additive effects for noncarcinogens) for fish consumers here.

Mo

In this study, Mo was below the detection limit (0.4 μg/g dw) in *O. leucostictus* muscle but was detected in the liver. Values for freshwater fish have not been reported before, to our knowledge, but published concentrations from marine fish species never exceed 0.5 mg/kg fresh weight in edible muscle (Eisler 2010). Since Mo was below the detection limit in the muscle, no THQ value was determined.

Cd

The Cd concentration was higher in the liver than in the muscle, which generally agrees with literature, e.g., (Jirsa et al. 2008; Uysal et al. 2009; Wang et al. 2010; Gül et al. 2011). The levels we found compare well with those (0.75±0.03 μg/g dw) reported by Njogu et al. (2011) in largemouth bass *M. salmoides* from Lake Naivasha. Mutia et al. (2012) reported even higher values (1.59±0.002 mg/kg ww) in *C. carpio* from Hippo point in Lake Naivasha, and much higher values were reported by Budambula and Mwachiro (2006): 52.0 μg/g in the muscle and 57.0 μg/g in the liver of *L. cylindricus* from Nairobi River.

Cd is a contaminant in phosphate fertilizers (Tarras-Wahlberg et al. 2002). The intense agricultural activities in the Lake Naivasha catchment area could therefore represent a significant source of easily soluble Cd compounds, which are taken up via the gills of fish and have a very long biological half life (Hofer et al. 1995) and therefore accumulate in all fish species, regardless of their diet.

The THQ value for Cd was close to 1 (0.980), which represents a risk for additive effects. The European Commission regulation set a maximum level of 0.05 mg/kg wet weight for fish consumed by humans (EU 2001). Therefore, based on the THQ value and EU regulation, Cd poses a clear risk to consumers of fish from the lake.

Pb

Our Pb results were unique and unexpected, differing greatly from earlier studies which reported very high levels in various fish species from Lake Naivasha (Njogu et al. 2011; Mutia et al. 2012). The reasons for these differences remain unknown. For example, Njogu et al. (2011) reported a range of 5.12–58.11 mg/kg ww for *C. carpio* muscle, while Mutia et al. (2012) reported 3.22±0.15, 1.49±0.1, and 1.56±0.19 mg/kg ww for *M. salmoides*, *O. leucostictus,* and *C. carpio*, respectively. The levels reported in our study are safe and below the maximum permissible level according to the European Commission regulation (EU 2001).

Accumulation of trace elements in *C. multipapillatum*

The trace elements with the highest concentration in *C. multipapillatum* were Zn, Fe, and Si. Accordingly, *C. multipapillatum* strongly accumulates certain essential elements. This supports the findings by Nachev et al. (2013) on the nematode *Eustrongylides* sp.: it accumulates essential metals (Co, Cu, Fe, Se, Zn) better than its host fish, the barbel *Barbus barbus*. In our study, however, *C. multipapillatum* had significantly more Pb than *O. leucostictus* muscle. Also, *C. multipapillatum* seemed to accumulate more Pb than reported from another nematode, *Anguillicola crassus*, which infects the swim bladder of eels (*Anguilla anguilla*) (Sures et al. 1994). The concentrations of Mn, Fe, Cu, and Zn in *C. multipapillatum* were lower than those reported in the literature on other anisakid nematodes (Table 6).

Compared to the host's muscle, the concentrations of Fe, Cd, Cu, and Pb were 2.94, 1.58, 1.96, and 7.04 times higher, respectively, in *C. multipapillatum*. Mo was unique because it was below the detection limit in the muscle, but was detected in the parasite. Barus et al. (2001) found Pb bioaccumulation factors between *C. rudolphii* tissue and the cormorant *Phalacrocorax carbo* muscle of 2.35 in male nematodes and 1.81 in females, whereas the Cd values were 0.52 and 0.22 in males and females, respectively—lower than those found in the present study. Overall, these bioaccumulation capacities of *C. multipapillatum* are much lower compared to acanthocephalans and cestodes (Sures 2003). This points to an inefficient accumulation indicator, supporting the conclusion of Sures (2001) and Barus et al. (2001) that nematodes in general are not useful as accumulation indicators. Exceptions to this have been shown by Azmat et al. (2008), Dural et al. (2011), Morsy et al. (2012), and Nachev et al. (2013). For example, Dural et al. (2011) found that *Hysterothylacium aduncum* accumulates more Cd, Cu, and Pb than the muscle of its final host, the sea bream *Sparus aurata*. Interestingly, their findings of no significant differences in the mean Cd and Cu concentrations in *H. aduncum* and the liver of sea bream corroborate our results.

Three probable factors influence the trace element concentrations of in *C. multipapillatum*: (1) the parasite's development stage, (2) parasite location within the host, and (3) parasite feeding and excretion. The parasites obtained from *O. leucostictus* in our study were larval-stage (L3) nematodes, potentially contributing to the low trace element concentrations. Barus et al. (2001) however, who examined very closely related adult nematodes and their definitive hosts, also found poor bioaccumulation factors. This suggests that other complex factors play a role in influencing trace element uptake by nematodes. We hypothesize that, because *C. multipapillatum* inhabits the pericardial cavity where it primarily gets nutrition from blood and other host body fluids, the trace element concentrations could be determined by the levels in the immediate muscle tissues. For example, Si and Al levels were not significantly different between the muscle of *O. leucostictus* and *C. multipapillatum*. Accordingly, the regulation of these trace elements in *C. multipapillatum* is probably tightly linked to their concentration in the host fish muscle. Finally, *C. multipapillatum* has a complete digestive system, setting it apart from cestodes and acanthocephalans,

Table 6 Trace element concentrations in *C. multipapillatum* from *O. leucostictus* in Lake Naivasha compared with published data on other nematode species: mean values (milligrams per kilogram ww)

Nematode species ex fish host	Al	Si	Mn	Fe	Cu	Zn	Sr	Mo	Cd
C. multipapillatum ex *O. leucostictus* (this study)	1.96	18.7	0.57	20.23	0.53	21.5	0.33	1.03	0.59
Anisakis sp.[a] *ex Dicentrarchus labrax*			25.9	234	21.0	43.2			4.28
Echinocephalus sp. ex *Liza vaigiensis*[b]				72		30			12
Ascaris sp. ex *Liza vaigiensis*[b]				80		32			15
Hysterothylacium aduncum[c] ex *Sparus aurata*			3.60	102	28.0	62.6			2.05
Eustrongylides sp. ex *Barbus barbus*[d]			1	250	80	50			0.05

[a] Morsy et al.(2012)

[b] Azmat et al. (2008)

[c] Dural et al. (2011)

[d] Nachev et al. (2013)

which absorb nutrients through their tegument. Therefore, the uptake, deposition, and excretion of trace elements are different, although, like cestodes and acanthocephalans, nematodes cannot synthesize their own steroids and fatty acids and must ingest sterol precursors from their hosts.

Conclusions

The mean concentration of trace elements in the sediment reflected normal background levels due to the geology of the basin; Zn is the exception. The liver accumulated the highest concentration of most trace elements except Zn, Sr, Rb, and Li. Our findings therefore support most studies that recommend considering the liver of fish when conducting studies on trace elements: it proved to be a good biomarker of most trace elements examined. Although *C. multipapillatum* showed potential to accumulate trace elements, its values were far lower than in the *O. leucostictus* liver and comparable to muscle levels. The parasite is therefore not an efficient accumulation bioindicator. Li, Sr, Cd, and Zn had high THQ values and could represent a health risk to the local community that depends on fish for regular food.

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