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Bioaccumulation of persistent organic pollutants (POPs) in fish species from Lake Koka, Ethiopia: The influence of lipid content and trophic position

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ABSTRACT

The concentrations and bioaccumulation of persistent organic pollutants (POPs) were determined in four fish species from Lake Koka, Ethiopia, representing 2–3 levels in the food chain of the lake. Dichlorodiphenyltrichloroethanes (DDTs), endosulfans, polychlorinated biphenyls (PCBs) and chlorpyrifos were identified, with DDTs as the most predominant pesticide, with concentration ranging from 0.05 to 72.53 ng g^{-1} wet weight (ww). All fish tissue samples collected from different species of the lake contained residues of DDTs. The maximum level of DDTs was found in the fattiest, African sharptooth catfish (*Clarias gariepinus*) sampled from the lake, with a mean concentration of 15.15 ng g^{-1}ww. The significant ($P<0.05$) relationship between concentrations of DDTs and $\delta^{15}N$ indicates that DDTs biomagnified in the food web of the lake. The 4,4′-DDE to 4,4′-DDT ratio in *Oreochromis niloticus* (0.6) and *Cyprinus carpio* (0.5) were below 1, indicating ongoing use of DDTs in the study area and recent exposure of these fish species.

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1. Introduction

Persistent organic pollutants (POPs) are synthesized organic compounds which include polychlorinated biphenyls (PCBs), most of the compounds referred to organochlorine pesticides (OCPs), and other by-product substances originating from human activities (for instance, dioxins and furans). Since the publication of the first classical book 'Silent Spring' by Rachel Carson in 1962, a lot of scientific evidences have revealed that POPs are among the most hazardous pollutants released into the environment. This is due to their toxic, lipophilic and persistent nature (Burreau et al., 2004; Holden, 1966; Jones and de Voogt, 1999), coupled with their ability to bioaccumulate and magnify in the food chain (Alexander et al., 2007; Burreau et al., 2004; Mackay and Fraser, 2000; Rognerud et al., 2002), and their marked impact on top predator species, including humans. There is also a great global concern about the long range transport of these pollutants to cooler environments, both in the southern and northern hemispheres (Blais et al., 1998; Rognerud et al., 2002; Rosseland et al., 1999; Simonich and Hites, 1995; Wania and Mackay, 1993).

POPs are known to persist in the environment for a long period and gradually accumulate in the fatty tissue of living organisms causing for instance, cancer and birth defects in rats (Randi et al., 2003; Stewart et al., 1989), and can disrupt the immune and reproductive systems and even affect the nervous system in birds and fish (King et al., 2003; Longcore and Stendell, 1977; Misumi et al., 2005; Ratcliffe, 1967).

It is well established that lipophilic compounds preferentially accumulate in lipids of fishes and other animals (Mackay and Fraser, 2000), and the extent of accumulation is greater in fish that have high lipid content (Muir et al., 1990). There are two main routes by which fish can bioaccumulate POPs in their natural aquatic habitat: from water via gills and other body surfaces, and from the diet (Burreau et al., 2004; Campbell et al., 2000; Holden, 1966). Stomach content analysis and stable nitrogen, $\delta^{15}N$ levels, provide complementary information which can be used to evaluate the trophic transfer and biomagnification of POPs along the food chain (Kidd et al., 1998a, 1995; Rognerud et al., 2002; Sharma et al., 2009). Therefore, the lipid content and (or) the trophic position of the aquatic organism, are good predictors of concentrations of POPs in aquatic habitats (Kidd et al., 2000, 1998b).

Africa is burdened with an estimated amount of 50,000 tonnes of obsolete pesticides (Curtis and Olsen, 2004), i.e. pesticides that can no longer be used, and therefore require disposal. According to the obsolete pesticide inventory report of Food and Agricultural Organization (FAO, 2001), Ethiopia is one of the many African countries

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burdened by obsolete pesticide stocks, which have accumulated since they were first imported in the 1960s. The pesticide inventory conducted in 1995 and 1999, led by FAO in collaboration with the government of Ethiopia, identified over 1500 t of obsolete pesticides. These were mostly organochlorine compounds such as chlordane, DDT, dieldrin and lindane that are banned or restricted in most countries (Alemayehu et al., 2006). A number of activities, including disposal processes, are being carried out to prevent future pesticide accumulation in Ethiopia. However, the 12 years (1996–2007) of pesticide import database produced by the Ethiopian Ministry of Agriculture shows that pesticide use in the country is increasing, and reached 8302 t during the period 2005–2007 (Haylamicheal and Dalvie, 2009).

The Ethiopian Rift Valley Lakes Region (ERVLR) is a densely populated area with various agricultural activities. Intensive agricultural and deforestation activities in the catchments have been reported to be serious environmental problems in most Ethiopian Rift Valley Lakes (Zinabu, 2002; Zinabu and Elias, 1989). However, the level and extent of contamination and bioaccumulation of POPs in fish species at various trophic levels have not been studied. The objective of this study is, therefore, to determine the concentration levels of POPs and the degree of bioaccumulation in relation to the trophic position and the lipid content in fish species collected from one of the Ethiopian Rift Valley Lakes — the Lake Koka.

2. Methods

2.1. Study area

The study area (Fig. 1), Lake Koka, is located in the southeastern part of Ethiopia, at 1590 m.a.s.l. It is a man-made lake formed by the construction of a concrete dam in the Awash River for the production of hydroelectricity. There are two inflowing rivers: Awash (major) and Modjo (minor), which flow into the lake in its western part, while one outlet river (Awash River), which flows out of the lake from its east part. The surface area of the lake is 220 km^2, with maximum and mean depths of 14 and 9 m, respectively (Wood and Talling, 1988). The climate of the region is characterized by a sharp increase in precipitation and relative humidity between mid-June and mid-September, although mean monthly temperatures are less variable. The conductivity of the lake water is measured 251 μS/cm at 25 °C and a pH of 7.4 during the study period which is low compared to most Ethiopian Rift Valley Lakes.

The general composition of the biota of Lake Koka appears similar to the other Rift Valley Lakes (Von Damm et al., 1984). A total of 72 phytoplankton taxa, with the dominant phytoplankton species being the diatom *Aulacoseira granulat*, have been identified from the lake (Kebede and Willén, 1998). The zooplankton was dominated by the calanoid *Tropodiaptomus processifer* and the cladoceran *Daphnia barbata* (Mesfin et al., 1988). The cladoceran *Moina micrura*, and the rotifer *Keratella tropica* were also present, but in small numbers. As described by Mesfin et al. (1988), the benthic fauna of the lake is composed of mainly nematodes, followed by Chironomidae, Ostracods and Oligochaetes. The principal fish species in the lake are Nile tilapia (*Oreochromis niloticus*), African sharptooth catfish (*Clarias gariepinus*), African big barb (*Barbus intermedius*), common carp (*Cyprinus carpio*), and the cyprinodont minnow (*Aplocheilichthyes antinorii*). According to the Lake Fisheries Development Project's final report (FLDP, 1998), the average annual harvest during the period 1994–1998 was 249 tonnes of *O. niloticus*, 235 tonnes of *C. gariepinus*, 101 tonnes of *C. carpio*, and 10 tonnes of *B. intermedius*.

2.2. Sampling

Sampling was carried out between February and April 2008. To obtain a wide range of fish sizes, fish samples were partly purchased from the local fishermen, partly by own gillnetting, using experimental gill nets with mesh sizes from 5 to 45 mm (bar mesh). The total length (cm), weight (g), sex and maturation stage of each fish were recorded. Stomach contents from the fish were taken and preserved in 96% ethanol. Muscle samples were taken from each specimen, following the procedures in the EMERGE protocol, as described by Rosseland et al. (2007), and frozen. The frozen samples were transported to Norway for analysis of POPs and stable isotopes of carbon (^{13}C and ^{12}C) and nitrogen (^{15}N and ^{14}N).

2.3. Stomach content analysis

The stomach content analysis was done at Hawassa University, Department of Applied Biology. The relative importance and contribution of each food item to the diet of each fish species was determined using the frequency of occurrence method and the relative composition by volume (%) — volumetric analysis (Hyslop, 1980). The identification of the stomach contents was done by using either a dissecting microscope for larger items, or by a compound light microscope for smaller items such as phytoplankton. The following food items were identified: phytoplankton (blue green algae, green algae and diatoms), macrophytes, detritus, aquatic insects, zooplankton, fish and fish eggs.

2.4. Lipid content analysis

The relative lipid content (%) in the fish muscle tissue was determined by a gravimetric method adopted from Lee et al. (1996). The method was tested using Standard Reference Material (SRM) 1946: Lake Superior fish tissue from USA National Institute Standard and Technology (NIST). The standard reference material was analyzed according to the proposed method and the result was within the certified range. About 5 g of fish tissue was defrosted, chopped into pieces, weighted and transferred into a 50 mL centrifuge tube. Each sample was completely homogenized using 25 mL of methyl *tert*-butyl ether (MTBE) and blended for 2 min with Polytron (Kinematica AG) at a moderate speed. To maintain a moderate and constant speed during blending is important because the high speed resulted in solvent vaporization and temperature rise. The extraction was repeated on the remaining, using 25 mL of MTBE to extract the fat completely. The extract was transferred to the same flask, and the volume was adjusted to 50 mL. Five milliliters of the extract (triplicate) was taken into a pre-weighted beaker. The sample was allowed to evaporate completely over night before the weight was determined, and finally the percentage of relative lipid content was calculated.

2.5. Stable isotope analyses

Stable isotope analyses were carried out at the Environmental Chemistry Section, Department of Plant and Environmental Sciences, Norwegian University of Life Sciences (UMB). The stable isotopes of nitrogen (^{15}N and ^{14}N) and carbon (^{13}C and ^{12}C) were analyzed from homogenized and freeze-dried muscle samples subjected to combustion in a Flash Elemental Analyzer (EA) as described in Desta et al. (2007) and Sharma et al. (2009). The isotopic ratios ($^{15}N/^{14}N$, $^{13}C/^{12}C$) were expressed in delta-values as follows:

$$\delta^{15}N \text{ and } \delta^{13}C(‰) = [(R_{Sample}/R_{Standard}) - 1] * 1000$$

where, $R = {}^{15}N/{}^{14}N$ for $\delta^{15}N$ or $R = {}^{13}C/{}^{12}C$ for $\delta^{13}C$.

2.6. POP analyses

The POPs were analyzed at the laboratory of Norwegian Institute for Agricultural and Environmental Research, Plant Health and Plant Protection Division, Pesticide Chemistry Section. Pesticides were prioritized and selected for the analysis, based on the ecotoxicological

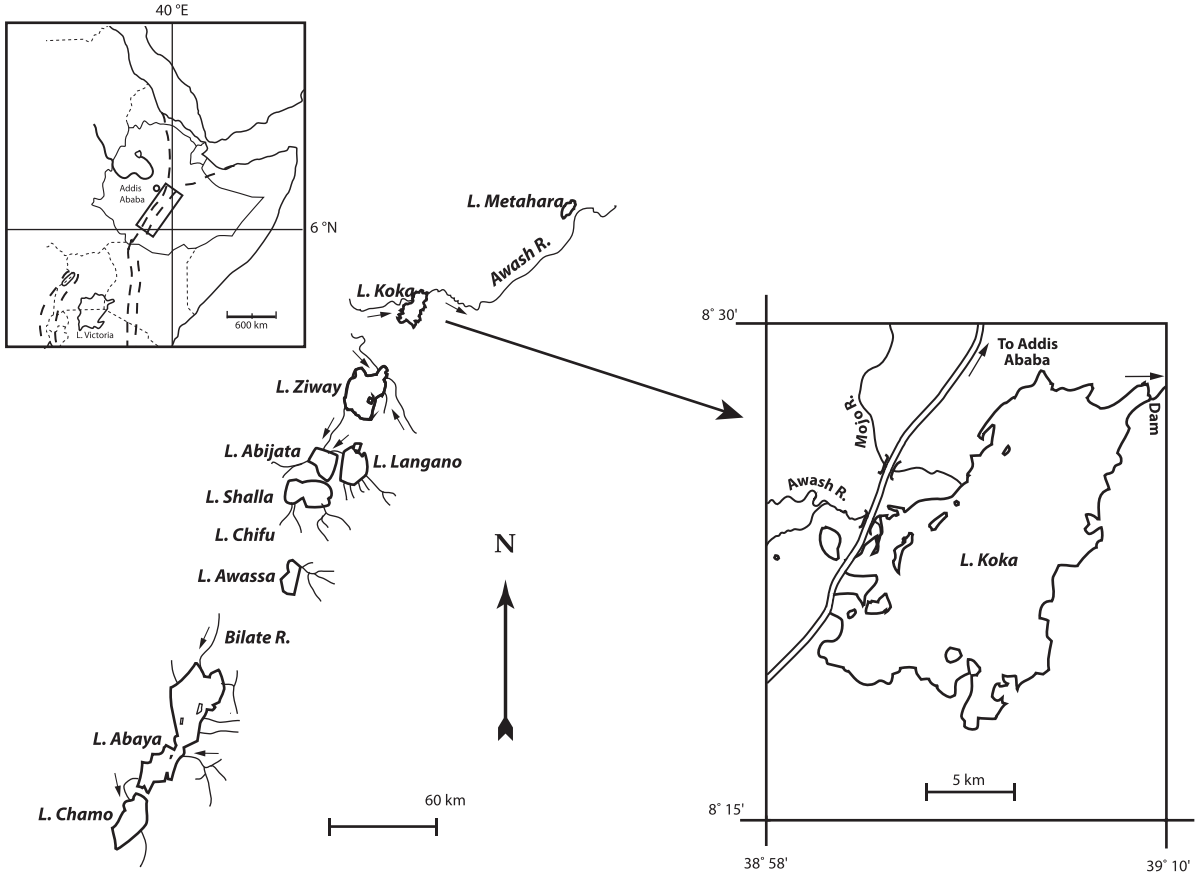

Fig. 1. The geographical position of Lake Koka.

nature of the pesticides, together with published and registered information for Ethiopia where the samples were obtained. Pesticides outside the list of Stockholm Convention (SC) were also included for analysis. List of pesticides targeted for analysis were: Aldrin, Chlordane, Oxychlordane, Dieldrin, 4,4′-DDT, 2,4′-DDT, 4,4′-DDE, 2,4′-DDE, 4,4′-DDD, 2,4′-DDD, Endosulfan-α, Endosulfan-β, Endosulfan-sulfate, Endrin, Heptachlor, Heptachlor-epoxide, Hexachlorboenzene (HCB), Lindane (γ-HCH), α-HCH, β-HCH, Methoxychlor, and Chlorpyrifos. Moreover, 7-PCB congeners were also included for the analysis.

The fish tissue samples were extracted with acetonitrile as described in Norli et al. (2011). Fish tissue was defrosted and chopped into pieces, and thereafter 5.00 ± 0.05 g was accurately weighted and transferred into a 50 mL centrifuge tube. Milliq water (10.0 mL), acetonitrile (10.0 mL) and 0.1 μg each of the internal standards, decachlorobiphenyl, triphenylphosphate and 2-bromobiphenyl, were added to each sample. Each sample was completely homogenized by using Polytron (Kinematica AG). To achieve phase separation between water and acetonitrile, the contents of a tube (Supelco 55227-U) containing pre weighed amounts of salts was added and shaken vigorously for 1 min, and centrifuged for 5 min at 3000 rpm. A combination of freezing and drying of the acetonitrile phase was used to remove the lipids. The sample was aspirated into 5 mL disposable syringe with a 20 μm polyethylene filter in the bottom and placed in the freezer (−20 °C) for 1.5 h. The frozen extract was quickly dispensed in a tube with 1 g of water-free calcium chloride, shaken 30 s by hand and centrifuged 5 min at 3000 rpm. The last step included dispersive solid phase extraction (dSPE) with a primary-secondary amino resin (PSA) which removes fatty acids. The sample was decanted in a tube containing 150 mg of PSA and 1 g of $MgSO_4$, shaken for 5 min with "end over end", and centrifuged for 5 min at 3000 rpm. A part of the extract was transferred to a gas chromatography (GC) vial and injected to the GC–MS (gas chromatography coupled with mass spectrometry). For each sample setup, the residues were quantified against a four level calibration curve together with reagent blanks, and spiked controls of cod (50 ng g^{-1}).

The measurements were carried out on an Agilent 6890 N gas chromatograph connected to an Agilent 5973 mass spectrometer with an inert ion source. The gas chromatograph was equipped with a Gerstel (Mühlheim Ruhr, Germany) Programmable Temperature Vaporising Injector (PTV) with a sintered liner. The separation column was a fused silica J & W Scientific HP-5MSI 30 m with 0.25 mm internal diameter and 0.25 μm film thickness. A 2.5 m methyl deactivated pre column (Varian inc. Lake Forest CA, USA) of the same internal diameter was connected to the analytical column. The temperature program was 70 °C held for 3 min, 25 °C/min to 150 °C, held for 0 min, 3 °C/min to 200 °C, held for 0 min, 20 °C/min to 280 °C, held for 6 min, 50 °C/min to 325 °C, held for 1 min. Total time 34.77 min. The PTV program was as follows: Injection volume 15 μL. The solvent temperature was kept at 50 °C in 1.8 min with a solvent flow at 200.0 mL/min. After 1.89 min the split valve was closed, and the injector temperature was raised by 720 °C/min to 280 °C and held for 1.2 min. Transfer line temperature was set at 280 °C, ion source temperature at 230 °C and quadrupole temperature at 150 °C. The mass spectrometer was operated in selected ion monitoring mode (SIM mode).

Spiking experiments in *O. niloticus* at 5 and 50 ng g^{-1} and extracted with acetonitrile show recovery range from 70 to 115% for all POPs. To further test the method using acetonitrile as extraction solvent, we analyzed the standard reference material SRM 1946®, Lake Superior Fish tissue homogenate. This is a certified trout material containing incurred residues of pesticides, PCBs, heavy metals and other compounds in a broad concentration range. The result from

certified trout material was also within the certified range. Estimation of limit of quantification (LOQ) and limit of detection (LOD) were made by spiking *O. niloticus* with 5 ng/g 7-PCB, chlorpyrifos, endosulfanes and DDTs. The samples were analyzed according to the procedure and the LOQ and LOD values were estimated by use of the root mean square signal to noise ratio (RMS S/N) parameter in the Chem Station software.

2.7. Statistical analysis

Comparisons of concentrations of each POP in different fish species were performed by ANOVA using MINITAB 16. Log transformed values of each POP concentrations ($ng\,g^{-1}$) were regressed against total fish length (L_T) in cm, and fish species (*O. niloticus*, *C. gariepinus*, *B. intermedius*, and *C. carpio*) as a categorical variable to find the relationship between fish age (assume older fish tend to be larger) and POP concentrations. Total length was used instead of weight to avoid bias due to the variation in gut fullness (in weight differences). Biomagnification rate of POPs was determined by regressing the log-transformed POP concentrations against $\delta^{15}N$ values and against relative lipid levels (%). Sigma Plot 10.0 was used to create the graphs.

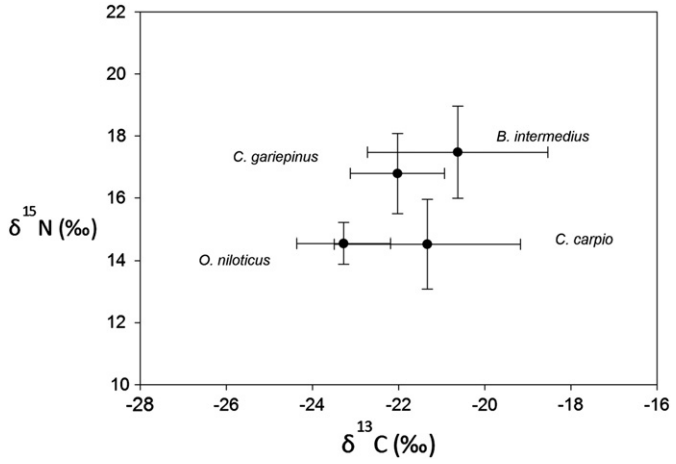

Fig. 3. Mean and standard deviation of stable isotope ratio of nitrogen and carbon ($\delta^{15}N$ and $\delta^{13}C$) of the four fish species, *C. carpio*, *O. niloticus*, *C. gariepinus*, and *B. intermedius*, from Lake Koka in 2008.

3. Results

3.1. Diet and trophic position

The diet of *C. carpio*, *O. niloticus*, *C. gariepinus* and *B. intermedius* was composed of plants, animals, and detritus. The consumed plants consisted of both phytoplankton (blue green algae, green algae and diatoms) and macrophytes. Items of animal origin were quite diverse and included fish, insects, and zooplankton. Fish eggs and unidentified fragments were also encountered (Fig. 2).

The stomach contents of *C. carpio* were diverse, mostly composed of macrophytes, detritus, phytoplankton, aquatic insects, fish eggs and zooplankton. Most of the stomach volume was however dominated by macrophytes and detritus, with a mean volume of 40.5 and 18.2%, respectively (Fig. 2).

Regardless of size, algae were the most important diet of *O. niloticus* in Lake Koka, and contributed to 86.2% by volume of the total stomach content, of which diatoms, blue green algae and green algae accounted for 50.5%, 33.3% and 2.4%, respectively. Zooplankton contributed with 13.9% of the mean volume (Fig. 2), and was found in 16.7% of the stomachs.

The diet of *C. gariepinus* had a very diverse composition that included food items extending from detritus to fish. Aquatic insects, and fish and fish eggs made up 24.2% and 36.4% of the stomach contents by volume, respectively (Fig. 2). The insects recorded as food items of *C. gariepinus* were mainly Hemiptera, Coleoptera and Diptera. Zooplankton (copepods and cladocerans) contributed to 16.9% of the stomach contents by volume. Macrophytes and detritus were also observed frequently in the diet of *C. gariepinus*.

The stomach contents of *B. intermedius* were composed of aquatic insects, fish, detritus and macrophytes (Fig. 2). Aquatic insects, represented by Diptera, Anisoptera, and Coleoptera were found in all individuals, and contributed to 33.6% of the total food contents, while fish contributed to 17.4% of the total food contents by volume.

The $\delta^{15}N$ values of the four fish species ranged from 10.7‰ to 19.8‰ (Fig. 3). The mean values of the $\delta^{15}N$ were relatively larger for *B. intermedius* and *C. gariepinus* than for *C. carpio* and *O. niloticus*, indicating a higher trophic level of the two species, *B. intermedius* and *C. gariepinus* in the food chain of the lake. According to the $\delta^{13}C$ values, ranging from 26.5 to 17.0‰, the fish species of the lake utilized different carbon sources, both with littoral and a pelagic origin. *O. niloticus* had the most negative $\delta^{13}C$, indicating a pelagic origin of its

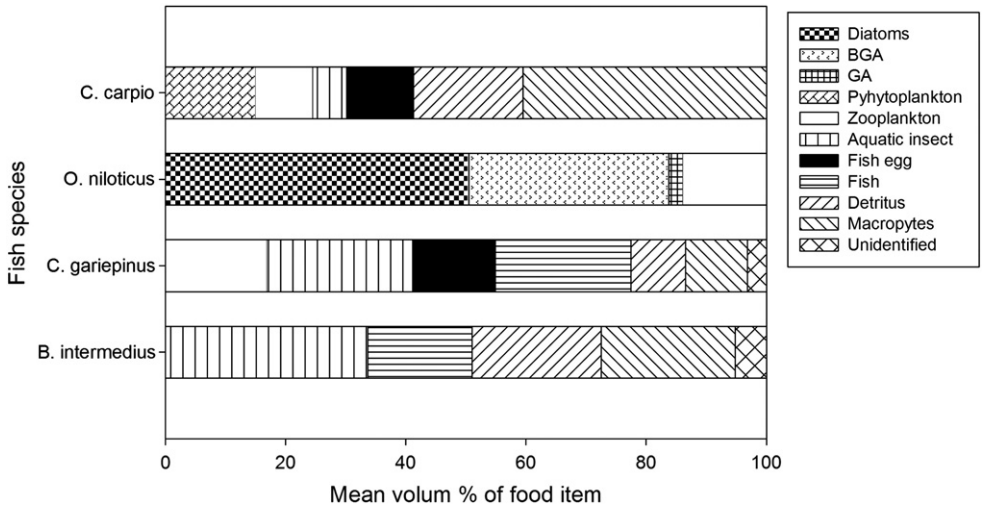

Fig. 2. Relative volume (%) of food items in the four fish species *C. carpio*, *O. niloticus*, *C. gariepinus*, and *B. intermedius*, from Lake Koka, sampled in 2008. BGA = Blue green algae, GA = Green algae.

Table 1
Mean values of total length (cm) and total weight (g), with min–max values in parentheses, and relative (%) fat content ± standard deviation, for four fish species from Lake Koka, sampled in 2008.

Fish species	Sample size (n)	Total length	Total weight	Fat content (%)
C. gariepinus	22	59.73 (31.8–126)	2523 (220–14,000)	3.5 ± 0.80
B. intermedius	20	35.76 (25.2–43.3)	445.3 (227–800)	0.69 ± 0.18
O. niloticus	20	30.2 (21.2–52.2)	508 (175–1150)	0.55 ± 0.08
C. carpio	20	35.84 (20.4–55.7)	881 (110–3279)	0.22 ± 0.09

food, whereas *B. intermedius* had the least negative $\delta^{13}C$, indicating a more littoral origin of the food.

3.2. Lipid content

The mean relative lipid content (%) of the fish species from Lake Koka was in the range 0.22–3.5% (Table 1). The highest lipid content was recorded in *C. gariepinus*, followed by *B. intermedius*, *O. niloticus*, and *C. carpio*.

3.3. POP residues

Most of the values of the concentration of POPs reported in our study are above LOQ but there are also few values reported below LOQ because of the clear chromatogram peaks in our result. DDTs, endosulfans, PCBs and chlorpyrifos were determined in the fish samples, with concentrations varying between 0.05 ng g^{-1} and 72.53 ng g^{-1} (Table 2). DDTs were identified in all fish tissue samples analyzed, with mean concentration of 15.15 ng g^{-1}, 6.90 ng g^{-1}, 6.20 ng g^{-1}, and 4.53 ng g^{-1} for *C. gariepinus*, *B. intermedius*, *O. niloticus* and *C. carpio*, respectively. Significantly higher concentrations of DDTs were found in *C. gariepinus* than in *B. intermedius* (Table 2).

Endosulfans (endosulfan α and endosulfan β) were identified in 68% of *C. gariepinus*, 40% of *B. intermedius*, 15% of *O. niloticus*, and 5% of the *C. carpio* tissue samples. The maximum level of endosulfans was found in *C. gariepinus*, with a mean concentration at 4.3 ng g^{-1}. The concentration of endosulfans ranged from not detectable (ND) to 16.3 ng g^{-1}.

The concentration of PCBs ranged from not detectable (ND) to 0.33 ng g^{-1}. The PCB congeners (PCB-28, PCB-52, PCB-101, PCB-138, PCB-153 and PCB-180) were found, with the highest frequency of detection in *B. intermedius* (75%) and the lowest in *C. carpio* (10%). The maximum level of PCBs was found in *C. gariepinus*, with a mean concentration of 0.33 ng g^{-1}.

During the field data collection, local farmers were observed using chlorpyrifos in their farms, but chlorpyrifos was only identified in 23% of the *C. gariepinus* tissues and 10% of the *O. niloticus* tissues analyzed. The concentration of chlorpyrifos among all fish samples ranged from not detectable to 1.12 ng g^{-1}, with highest concentration found in *C. gariepinus*. The concentrations of chlorpyrifos in *B. intermedius* and *C. carpio* were below the detection limit.

With all data pooled for the four fish species, the concentration of DDTs ($F_{7,82} = 5.87$, $P<0.05$) and PCBs ($F_{7,38} = 3.63$, $P<0.05$) were significantly correlated to total length of the fish, whereas there was no significant relationship between total length and the concentration of endosulfans (Fig. 4). The relationship between the concentration of chlorpyrifos and the size of the fishes was not tested due to the few samples with detectable concentrations (Fig. 4).

The relationship between log transformed DDT and $\delta^{15}N$ was significant ($F_{1,82} = 5.58$, $P<0.05$), where the slope of the regression equation was 0.04 when all data were pooled together (Fig. 5). This was the only case for which significant relationships between POPs and $\delta^{15}N$ could be observed.

With all data pooled for the four fish species, the relationships between log transformed DDTs ($F_{1,82} = 24.5$, $P<0.05$), and the relative lipid levels (%) as well as between the log transformed PCBs ($F_{1,38} = 7.6$, $P<0.05$) and the relative lipid levels (%) were significant where the slope of the regression equation was 0.25 and 0.07, respectively (Fig. 6).

3.4. DDT and its metabolites

The DDTs identified in the fish tissue samples were 4,4′-DDT, 2,4′-DDT, 4,4′-DDE, 2,4′-DDE, 4,4′-DDD, and 2,4′-DDD (Fig. 7). The 4,4′-isomers (4,4′-DDT, 4,4′-DDE, and 4,4′-DDD) were the predominant group, accounting for 85.8% of the sum of DDT. The proportion of 4,4′-DDE in *B. intermedius* and *C. gariepinus* was higher than in *O. niloticus* and *C. carpio*, and accounted for 78% and 54% of the mean DDT concentrations, respectively. On the other hand, the proportions of 4,4′-DDT were higher in *C. carpio* and *O. niloticus* than in *B. intermedius* and *C. gariepinus*, comprising 41% and 34% of the mean DDT concentrations, respectively. The ratios of 4,4′-DDE to 4,4′-DDT in *B. intermedius* and *C. gariepinus* were 16.7 and 2.8, respectively, while the ratios for *C. carpio* and *O. niloticus* were 0.5 and 0.6, respectively.

4. Discussion

The diets of the four fish species, *O. niloticus*, *C. carpio*, *C. gariepinus* and *B. intermedius* from Lake Koka are in good accordance with studies in other lakes. *O. niloticus* in Lake Koka predominantly feeds on algae, and is thus an herbivore. This species has a similar diet in other Ethiopian Rift Valley Lakes such as Lake Awassa (Desta et al., 2007; Getachew, 1987, 1989; Getachew and Fernando, 1989), and

Table 2
Mean concentrations (ng g^{-1} ± standard variation) of POPs in four fish species from Lake Koka, sampled in 2008. Ranges of concentrations are given in parentheses. (% = percent of detection, ND = not detected).

Fish species	ΣDDT		ΣEndoselfan		ΣPCB		Chlorpyrifos	
	%	Mean	%	Mean	%	Mean	%	Mean
B. intermedius	100	[a]6.2 ± 5.82 (0.05–21.03)	40	[a]2.2 ± 1.56 (ND–5.321)	75	[a]0.05 ± 0.05 (ND–0.14)	0	ND
C. carpio	100	[a]4.53 ± 2.24 (2.05–11.26)	5	[a]1.2 ± 0 (ND–1.2)	15	[a]0.17 ± 0.08 (ND–0.22)	0	ND
C. gariepinus	100	[b]15.15 ± 16.89 (1.8–72.53)	68.18	[a]4.3 ± 4,7 (ND–16.28)	68.18	[a]0.33 ± 0.53 (ND–2.03)	22.73	[a]0.67 ± 0.14 (ND–1.12)
O. niloticus	100	[a]6.90 ± 2.72 (2.32–12.16)	15	[a]1.86 ± 1.39 (ND–3.43)	35	[a]0.05 ± 0.24 (ND–0.63)	10	[a]0.11 ± 0.01 (ND–0.11)

Means with different letters within a column are significantly different ($P<0.05$).

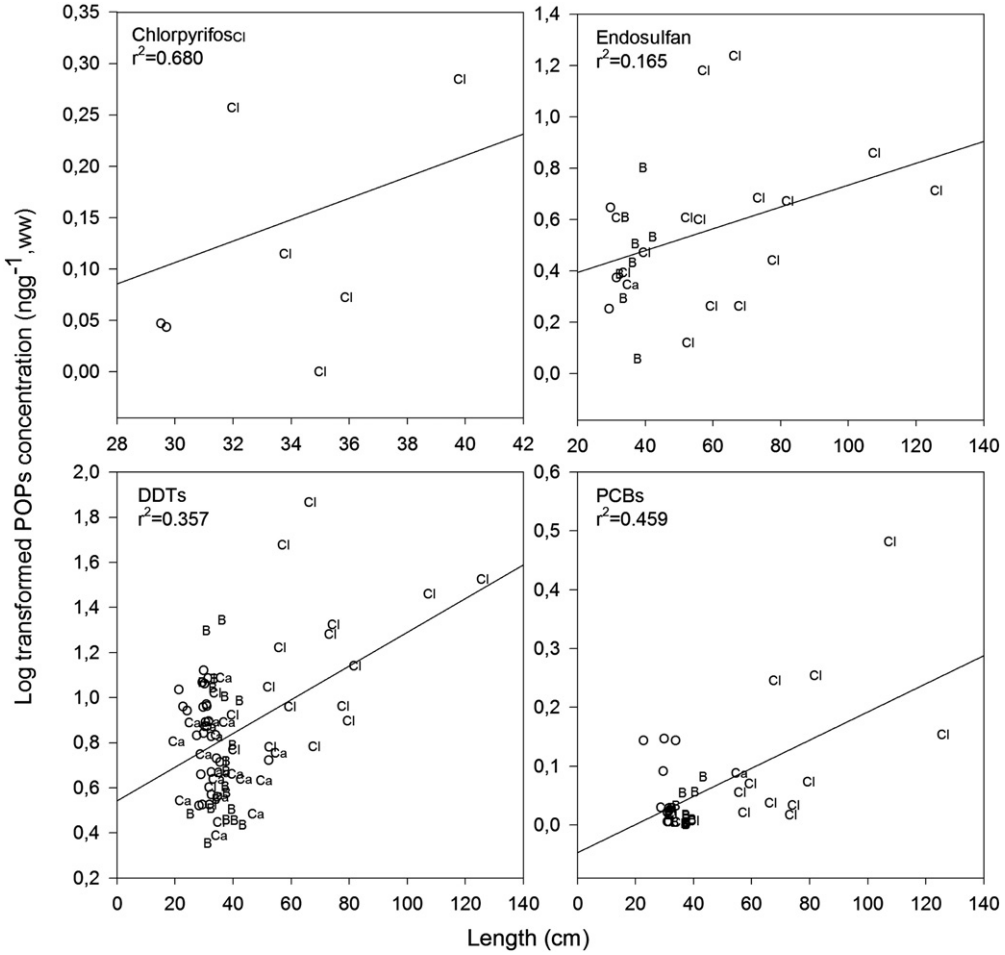

Fig. 4. The relationship between POP (chlorpyrifos, endosufans, DDTs and PCBs) concentrations in ng g^{-1} ww and length in cm of individual fish sampled from Lake Koka in 2008. (O = *O. niloticus*, Cl = *C. gariepinus*, Ca = *C. caprio* and B = *B. intermedius*).

Lake Chamo (Getachew, 1993). A similar finding was also reported from other East African lakes (Lowe-McConnell, 1958). According to the diet, *C. carpio* is omnivorous, as its diet was found to be composed of macrophytes and detritus, as well as zooplankton and insects.

The top consumers in Lake Koka are *C. gariepinus* and *B. intermedius*. Both species are omnivorous, but the large individuals include fish in their diet. Other studies (Bruton, 1978; Dadebo, 2000; Desta et al., 2007) have also shown that the larger individuals of *C. gariepinus* were carnivorous in Lake Awassa. Similar findings were also reported for *B. intermedius* in Lake Awassa (Admassu and Dadebo, 1997; Desta et al., 2007), in Lake Victoria, Uganda (Corbet, 1961), and in Lake Kimret, Israel (Spataru et al., 1987).

The diet compositions of the fish species from Lake Koka are further substantiated by the $\delta^{15}N$ signals — *B. intermedius* and *C. gariepinus* occupying the top trophic level, whereas *C. carpio* and *O. niloticus* were occupying the lower trophic levels in Lake Koka. A range in $\delta^{15}N$ values of greater than 9.5‰ indicates that there are at least 2–3 trophic levels in the fish community of the lake, following a mean enrichment value of $\delta^{15}N$ from 3 to 4‰ per successive trophic level (Vander Zanden and Rasmussen, 2001). The signatures of $\delta^{13}C$ in general indicate that the studied fish species of the lake have a wide range of carbon sources that extend from pelagic to littoral.

The high levels of DDTs observed in fish from Lake Koka, and DDT being the predominant pesticide, with mean concentrations a factor of 10 times higher than other POPs, may be attributed to its intensive and continuous use in vector control like mosquitoes in this region. Reports from other African lakes also indicate much higher levels of DDT in aquatic organisms compared to other POPs: In Lake Malawi up to 60 times higher concentration of DDT than other organochlorine pesticides has been reported for the biota (Kidd et al., 2000). Although the sum of DDTs found in Lake Koka was below the Extraneous Maximum Residue Levels (EMRLs) of 5000 ng g^{-1} set by FAO/WHO Codex Alimentarius Commission for meat from mammals other than marine mammals, the presence of DDT in all tissue samples collected from the four fish species in Lake Koka indicates the magnitude of the problem.

Persistent lipophilic organic compounds bioaccumulate and biomagnify with increasing trophic levels (Connell, 1995), and accordingly fatty carnivorous fish species are expected to have higher concentrations of POPs than lean fish at lower trophic levels. The significantly higher concentration of DDTs in *C. gariepinus* in the present study is in accordance with this, and consistent with results in fish species from Qiantang River in East China (Kong et al., 2005; Zhou et al., 1999, 2007). According to the $\delta^{15}N$ and the stomach content analysis, *B. intermedius* is positioned at the same trophic level as *C. gariepinus*, but has lower concentration of DDTs compared to *C. gariepinus*. This can be explained by its lower relative lipid content than in *C. gariepinus*.

Length and weight of fish are important variables for explaining the variation in concentrations of POPs (Rognerud et al., 2002; Vives et al., 2005). In the present study, both DDT and PCB concentrations progressively increased with fish size. This is presumably due to the bioaccumulation of DDTs and PCBs with age as older fish tend to be large.

The significant relationship between the log transformed concentrations of DDTs and $\delta^{15}N$ indicates that DDTs are biomagnified in the food web of the lake, and thus increases as the trophic level increases.

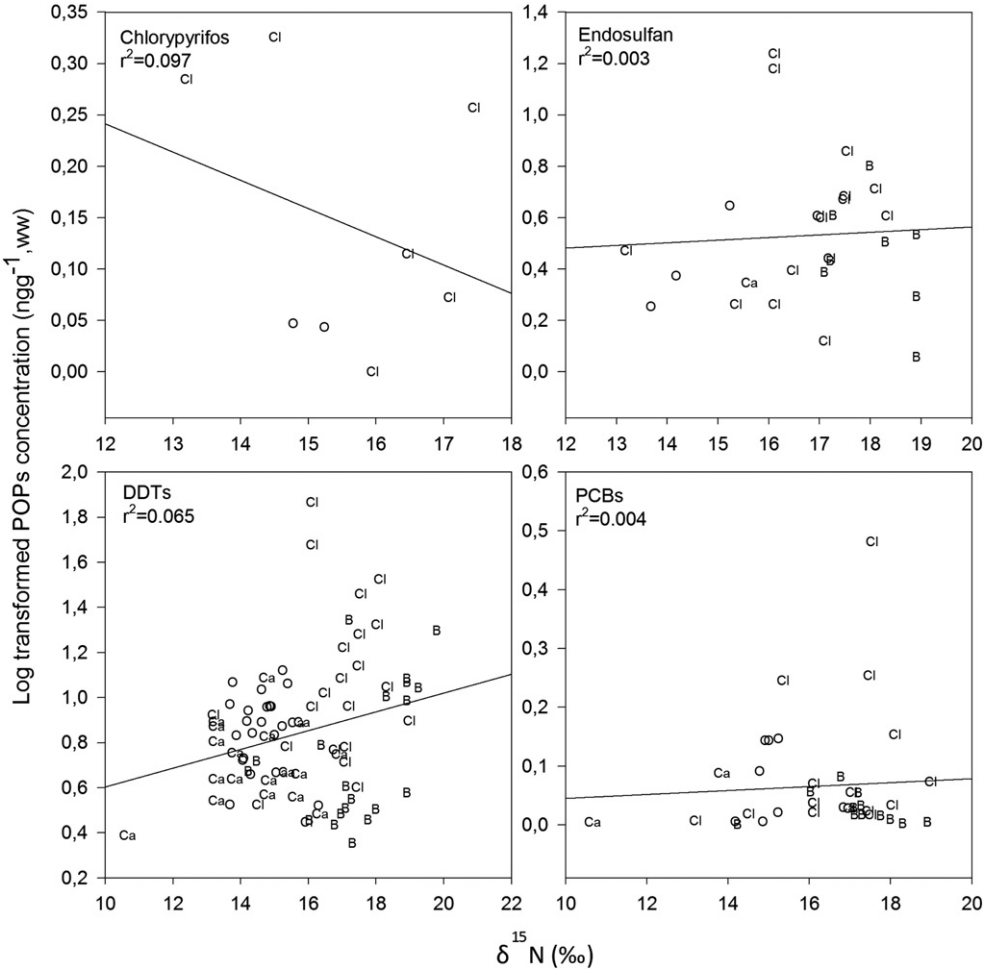

Fig. 5. The relationship between POPs in ng g^{-1} ww (chlorpyrifos, endosufans, DDTs and PCBs) and δ^{15}N signatures of individual fish sampled from Lake Koka in 2008. (O = *O. niloticus*, Cl = *C. gariepinus*, Ca = *C. caprio* and B = *B. intermedius*).

However, the biomagnification rate of DDTs in the food web of Lake Koka is lower than what has been reported from other areas, like the Barents Sea (Hop et al., 2002) and the southern Beaufort–Chukchi Seas, in the Arctic (Hoekstra et al., 2003), European Alpine and Arctic lakes (Rognerud et al., 2002; Vives et al., 2005), Mekong Delta, South Vietnam (Ikemoto et al., 2008) and Lake Malawi, East Africa in the tropical region (Kidd et al., 2000). Nevertheless, other POPs (chlorpyrifos, endosulfans and PCBs) did not follow the biomagnification of persistent contaminants in Lake Koka, such low effectiveness of δ^{15}N in describing organochlorine pesticide accumulation has also been observed in some other studies (Campbell et al., 2000; Guo et al., 2008). Moreover, tropical data on POPs shows rapid degradation pattern, low residual levels, and wide distribution because the tropics are characterized by different climate conditions (high sunshine radiation throughout the year, a high load of microorganisms, tropical rains and various soil types) (Wandiga, 2001).

Although the scatter plot in Fig. 6 shows lipid content of *C. gariepinus* contributed to a higher extent than the other species, we found that lipid content is an important variable for explaining the concentrations of DDTs and PCBs in the fish from Lake Koka when all data pooled for the four fish species. Campbell et al. (2000) in the food web of subalpine Bow Lake and Kidd et al. (2000) in fish from Lake Malawi also found the lipid content as an important variable for explaining concentrations of organochlorine compounds.

The ratio of DDE to DDT is a helpful tool in revealing the significance of the degradation of DDT and to evaluate the current use of DDT in the region (Strandberg and Hites, 2001) although this method is limited in regions where dicofol is used since it contains high level of DDT as impurity (Qiu and Zhu, 2010). The mean 4,4′-DDE to 4,4′-DDT ratio in fish species of Lake Koka is much lower than the ratios found in a study conducted by Erdogrul et al. (2005) in fish from Kahramanmaras, Turkey, but marginally higher than the ratios reported by Ssebugere et al. (2009) in fish from Lake Edward, Uganda. The observed mean 4,4′-DDE to 4,4′-DDT ratio below 1 in *C. carpio* (0.5) and *O. niloticus* (0.6), suggests recent exposure of these fish to DDT. This is most likely due to its recent use in vector control like mosquitoes in the region, as well as contamination from obsolete pesticides. Ethiopia signed SC in May 2002 and ratified it in January 2003, but the Ethiopian government decided to continue its use of DDT because of the high level of sickness and fatalities from malaria.

The mean ratio of 4,4′-DDE to 4,4′-DDT in fish at the bottom of the food web, i.e. *C. carpio* and *O. niloticus*, was below 1.0, whereas it was above 1.0 in the top consumers, i.e. *B. intermedius* and *C. gariepinus*. Similar results were also obtained by Zhou et al. (2007) in fish species from Qiantang River, East China, probably as a result of more efficient transfer of DDT to phytoplankton and macrophyte consuming fish, while the carnivore fish species feeding on these species accumulate a more degraded form, as DDT is transformed to DDE. In addition, this may be attributed to the more persistent nature of 4,4′-DDE, and to its rate of biomagnification along the food chain, as it has been explained by the food web magnification factors (FWMFs) of 4,4′-DDE and 4,4′-DDT in a marine Arctic ecosystem (Hoekstra et al., 2003), in freshwater ecosystems in temperate and Arctic areas (Rognerud et al., 2002), as well as in the tropical Lake Malawi, East Africa (Kidd et al., 2000).

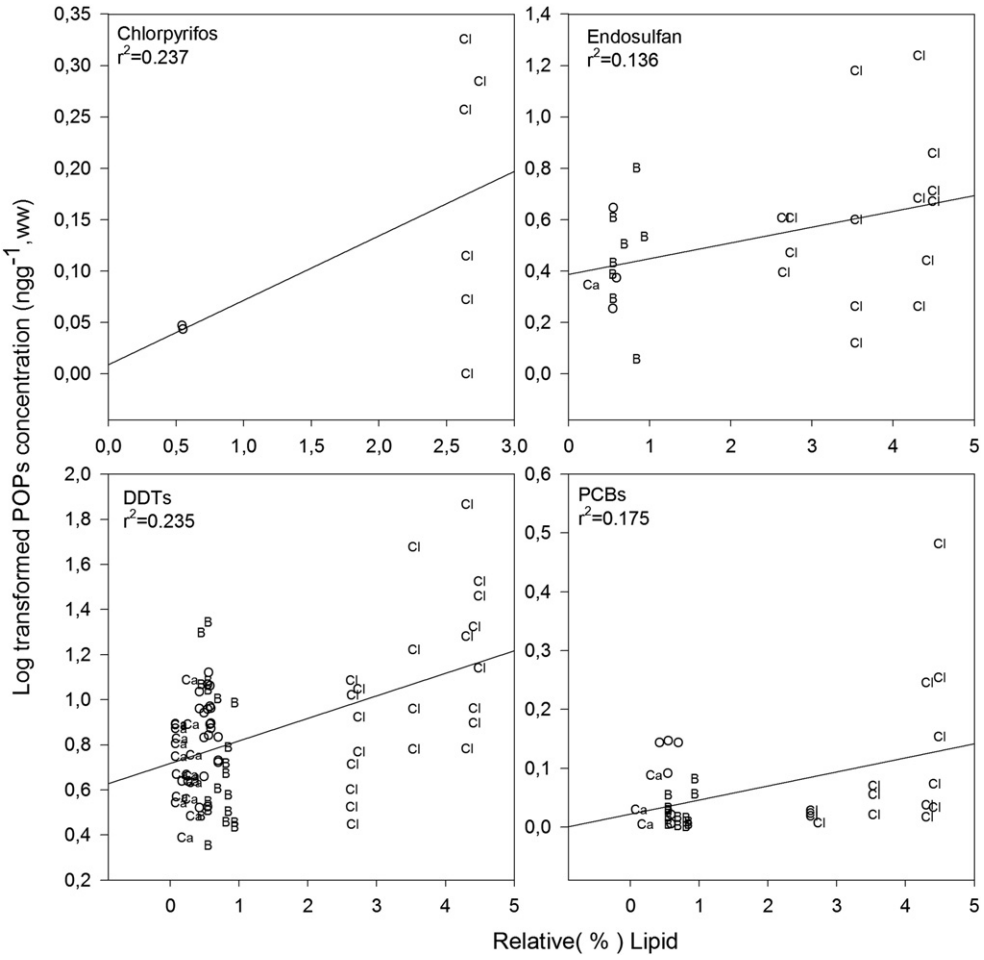

Fig. 6. Relationship between POPs in ng g^{-1} ww (chlorpyrifos, endosufans, DDTs and PCBs) and the relative lipid levels (%) of individual fish sampled from Lake Koka in 2008. (O = *O. niloticus*, Cl = *C. gariepinus*, Ca = *C. caprio* and B = *B. intermedius*).

5. Conclusion

All fish tissue samples collected from four fish species of the lake contained DDT residues, however, in low concentrations found to be safe for human consumption, according to the present EMRLs set by FAO/WHO Codex Alimentarius Commission for mammals other than marine mammals. The lower mean 4,4′-DDE to 4,4′-DDT ratio observed, in the fish species, suggests recent exposure of fish attributed to the current use of DDTs in the study area. The maximum level of DDTs was found in the relatively the most fatty, African sharptooth catfish (*C. gariepinus*). The present study demonstrates a biomagnification of persistent contaminants (e.g. DDTs) in tropical lakes, but with a lower extent of biomagnification rate than other areas. The results from the present study, albeit limited, call for a further study on the level and

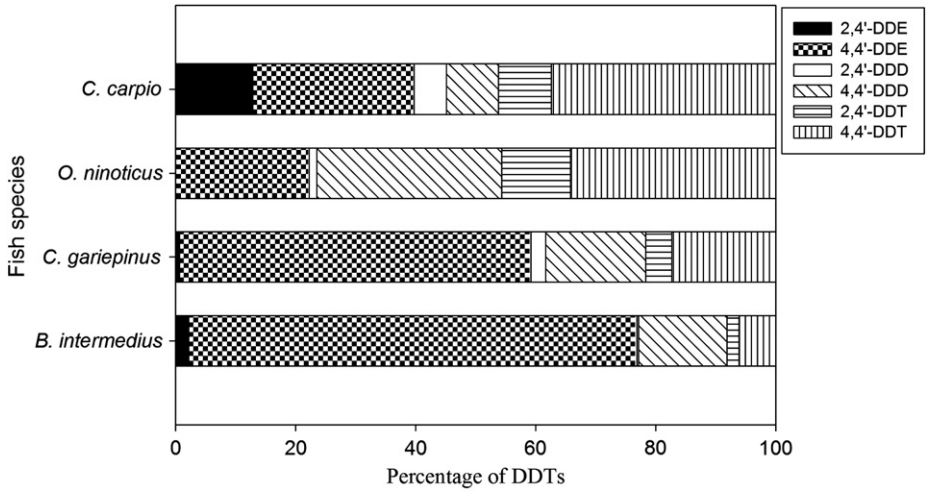

Fig. 7. The relative (%) of DDTs in the fish species *O. niloticus*, *C. gariepinus*, *B. intermedius* and *C. carpio* sampled from Lake Koka in 2008.

the extent of POP contamination in fish species at various trophic levels in the rest of the Ethiopian Rift Valley Lakes. This is very important given the intensive agricultural and deforestation activities in the catchments of the Ethiopian Rift Valley Lakes, and hence the possibility of chemical pollution from fertilizers and pesticides, which could be of a big public health concern caused by consuming, POPs in fish.

Acknowledgments

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