Feeding habits and trace metal concentrations in the muscle of lapping minnow *Garra quadrimaculata* (Rüppell, 1835) (Pisces: Cyprinidae) in Lake Hawassa, Ethiopia

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ABSTRACT

Diet composition and trace metal concentration in the muscle of the lapping minnow *Garra quadrimaculata* (Rüppell, 1835) was investigated to study the trophic status of the species as well as to assess the level of bioaccumulation of heavy metals in the body of the fish. The study was conducted based on 328 gut samples collected from February to March (dry months) and from August to September (wet months) of the year 2011. Frequency of occurrence and volumetric methods were employed in this study. Detritus, fish eggs, macrophytes, phytoplankton and insects occurred in 54.9%, 16.2%, 43.9%, 56.4% and 26.6% of the guts, respectively and comprised 27.1%, 22.2%, 18.2%, 18.2% and 14.1% of the total volume of food, respectively. The proportions of different food items consumed varied during the dry and wet months. Fish eggs and detritus were the dominant food items during the dry months. Macrophytes and insects were also common in the diet. During the wet months, phytoplankton was the most dominant food item (33.5% by volume). Macrophytes, detritus and insects were also important in the diet. The mean volume of fish eggs and phytoplankton decreased as the size of fish increased while the contributions of detritus, insects and macrophytes increased with fish size. The present study clearly indicated that *G. quadrimaculata* in Lake Hawassa is omnivorous in its feeding habits. Out of the seven analyzed trace metals (Cr, Mn, Zn, Cu, Cd, Pb and Ni), Cd, Pb, and Ni were below detection limits. There was a significant difference ($p<0.05$) between the mean concentrations of Zn and Cr in the muscle of the fish in the dry and wet months. The concentration of Zn in the dry month was significantly lower than its concentration in the wet months ($p<0.05$). By contrast, the concentration of Cr in the dry months was significantly higher than its concentration in the wet months. There were no significant variations in the concentrations of Mn and Cu between dry and wet months. There was no sign of bioaccumulation of the investigated trace metals as the fish grew older. The detected elements were compared with different standards and found to be below the maximum permissible limits for human consumption.

Keywords: Feeding habits, *G. quadrimaculata*, Ontogenetic diet shift, heavy metals, Lake Hawassa.

1. INTRODUCTION

The genus *Garra* is widely distributed in Ethiopia and contains about 12 species of small barbs that belong to the family Cyprinidae. The minnow *G. quadrimaculata* is mainly found in the Arabian

Peninsula (Saudi Arabia and Yemen), south-eastern Eritrea, south-eastern Ethiopia, and possibly also in Somalia (Froeser and Pauly, 2000). The species is indigenous and it is found in many of the Ethiopian Rift Valley lakes including Lake Hawassa. It is benthopelagic, non-migratory, freshwater fish that thrives in tropical climate (Froeser and Pauly, 2000). *G. quadrimaculata* grows to a maximum of 15 cm (TL) and a weight of 40 g (Elias Dadebo, unpublished data). Different *Garra* species eat foods of animal and plant origin. The proportion of these food items is usually correlated with the gut length of the fish (Froeser and Pauly, 2000).

Hawassa is a fast growing city and has shown rapid expansion and industrialization in recent years. In this regard, there is a growing concern about pollution of the lake from discharges released from domestic, industrial and agricultural activities. Accordingly, several studies have been conducted to investigate the level of heavy metal accumulation in the body of some species of fish in Lake Hawassa (Abayneh Ataro et al., 2003; Aweke Kebede and Tadesse Wondimu, 2004; Zerihun Desta, 2007; Diskowitzky et al., 2012). However, such studies have not been done on *G. quadrimaculata*.

Garra quadrimaculata does not have any commercial importance in the fishery of Lake Hawassa because of its small size. However, it is ecologically important, since it is used as a prey fish for *Clarias gariepinus* (Burchell, 1822) and *Labeobarbus intermedius* (Rüppell, 1836), which are commercially important fish species in Lake Hawassa (Elias Dadebo, 2000). Accordingly, knowledge on the feeding habits of *G. quadrimaculata* helps to understand energy flow among the different trophic levels of organisms in the lake. Furthermore, knowledge on heavy metal concentrations in the body of *G. quadrimaculata* is vital to understand the level of bioaccumulation of heavy metals in the body of large fish species that consume it. In particular, *G. quadrimaculata* being benthic fish, it may have important contribution in the bioaccumulation of heavy metals in the food chain. Therefore, this study was conducted to fill the gap by providing information on the feeding habit and trace metal concentrations in the body of *G. quadrimaculata* in Lake Hawassa.

1.1. The Study Area

Lake Hawassa (6^0 33’ - 7^0 33' N and 38^0 22’ – 38^0 29' E) is located at an altitude of 1,680 m in the central part of the Ethiopian Rift Valley 275 km south of the capital, Addis Ababa (Fig. 1). Lake Hawassa has a surface area of 90 km^2, a catchment area of 1,250 km^2, a maximum depth of

22 m and a mean depth of 11 m (Elias Dadebo, 2000). The area has a dry, sub-humid climate and receives a mean annual rainfall of 1,154 mm in the long eight rainy months (March to October). According to Zinabu Gebre-Mariam and Elias Dadebo (1989), Lake Hawassa mixes during the main rainy months (June and July) and in December. The lake is topographically a closed basin, and there is no known outflow. It is primarily fed by a small river named Tikur-Wuha that stems from Shallo swamp and also from the rivers (streams) in the north and west caldera walls, which are ephemeral (Tadesse Fetahi, 2005).

The limnology and plankton of Lake Hawassa have been studied by several authors. The predominant ions in the lake water are sodium and bicarbonates and the lake is freshwater with a mean conductivity of 850 $\mu S\ cm^{-1}$ (Elizabeth Kebede, 1996). The major taxa of phytoplankton in the lake are *Botryococcus braunii*, *Microcystis spp* and *Lygnbbya nyassa* (Elizabeth Kebede, 1996). The dominant groups of zooplankton include cyclopoid copepods (*Mesocyclops aequatorialis similis and Thermocyclops consimilis*) and Cladocera (*Diaphanosoma excisum*) (Seyoum Mengistu and Fernando, 1991). Benthic diatoms (such as *Synedra*, *Cymbella*, *Nedium* and *Gomphonema*), Ostracods, Chironomidae larvae and other insect larvae constitute the benthic community (Tudorancea et al., 1988).

There are six fish species in Lake Hawassa amongst of which three are commercially important namely the Nile Tilapia (*Orechromis niloticus* L., 1758), the African catfish (*Clarias gariepinus*, Burchell 1822), and the African big barb (*Labeobarbus intermedius* Rüppell 1836). The three minnow fish that are not fished due to their small size are the straightfin barb (*Barbus paludinosis* Peters 1852), the black lampeye (*Aplocheilichthys antinorii* Vinciguerra, 1883) and the stone lapping minnow (*G. quadrimaculata*) (Elias Dadebo, 2000). The commercial fishery mainly rests on *O. niloticus* that accounts for over 85 % by weight to the total annual landings followed by *C. gariepinus* (14%) and *L. intermedius* (<1%). Gillnetting is the major method used by the commercial fishery although long lines are used to catch *C. gariepinus*. The annual fish landing ranges between 450 and 600 t/year (Yosef Tekle-Giorgis, 2002).

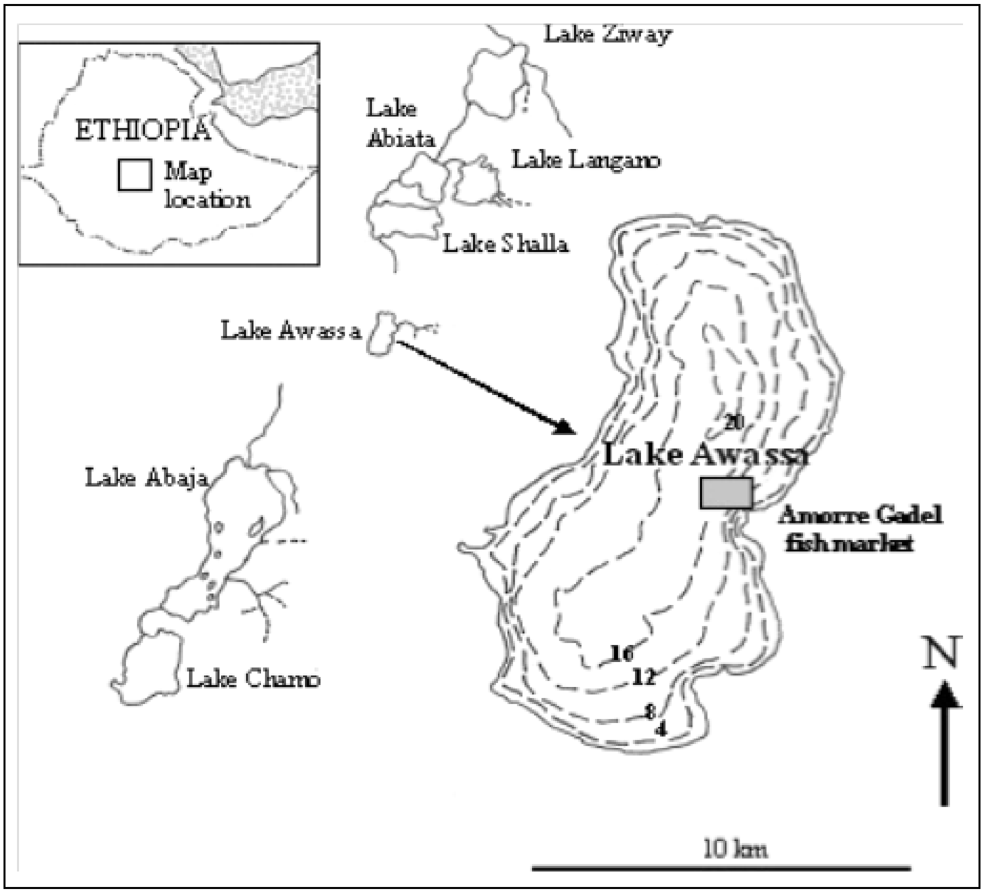

Figure 1. Lake Hawassa in relation to the other Ethiopian Rift Valley lakes, and its bathymetric map; the sampling area is indicated by a rectangle (Zerihun Desta et al., 2006).

There are five factories located in close proximity to Lake Hawassa. These are Hawassa Textile Factory, Hawassa Soft Drink Factory, Hawassa Ceramic Factory, Hawassa Flour Factory and Brewery. Except the flour factory, the rest discharge their effluent into Tikur Wuha River and eventually into Lake Hawassa (Behailu Berhanu et al., 2015). A referral hospital built close to the lake also releases its liquid waste directly into the lake (Zerihun Desta et al., 2008).

2. METHODOLOGY

2.1. Fish sampling and measurements

Samples of *G. quadrimaculata* were collected from the littoral area close to the Amorra Gedel site of the lake (Fig 1) using multi-mesh monofilament nylon gillnets from February - March 2011 (dry months) and August - September 2011 (wet months). Five hundred eleven fish samples were collected during the dry and wet months using gill nets that were set around 7:00 am in the morning and pulled around 3:00 pm in the afternoon. Fish were removed from gillnets and the total length (TL), fork length (FL) and standard length (SL) were measured to the nearest millimeter. Total weight (TW) of each fish was measured to the nearest 0.1 g. The sex and stage of gonad maturation of each fish was determined by visual examination of the gonads and the fish were categorized into five maturity stages as described by Holden and Raitt (1974).

2.2. Food and feeding

Since *G. quadrimaculata* has no distinct stomach, contents of the gut up to the first bend were taken and preserved in plastic vials containing 5% formalin solution and each sample was labeled and brought to laboratory for further analysis. The gut contents were examined and prey items were identified under a dissecting (LEICA MS5) and compound (LEICA DME) microscopes at the Biology Laboratory of Hawassa University. The relative importance of food items was determined using the following methods:

2.2.1. Frequency of occurrence

The number of stomach samples containing one or more of a given food items was expressed as a percentage of all non-empty stomachs examined (Bagenal and Braum, 1978). The proportion of the population that fed on certain food items was estimated by this method.

2.2.2. Volumetric analysis

The volume of the gut from which the food was taken was determined by measuring the length and the diameter of the gut. Then the food items that were found in the intestine were stored into different taxonomic categories and the number of food items was counted using a dissecting or a compound microscope. The volume of food items in different taxonomic categories was then estimated by multiplying the number of food items in each category by the mean volume of the organism (Bowen, 1983). The volume of each category of food items was expressed as a percentage of the total volume of the gut contents (Bowen, 1983). Stomach content analysis was conducted at the Fishery Laboratory of Biology Department, Hawassa University.

2.3. Ontogenetic dietary shift

The presence of ontogenetic dietary shift in *G. quadrimaculata* was investigated based on the volumetric contribution of each food item within each length group. Accordingly for studying ontogenetic dietary shift, fish were classified into three size classes based on the changes in the proportion of major food items as the fish increased in length. The relative importance of each food item in each size class was determined by taking the mean percentage volume of each category of food items to the total volume in that size class.

2.4. Heavy metal concentration analysis

2.4.1. Collection and drying of fish samples

Among the 511 fish samples of *G. quadrimaculata* collected, 13 subsamples were randomly taken from each of the two sampling seasons, representing the dry and wet months. In each case, the sub-sampled fish contained all size ranges from the smallest to the largest fish. Muscle samples were taken from each of the 26 fish for heavy metal analysis, following the procedures in the EMERGE protocol (Rosseland et al., 2003). The muscle samples were removed from each fish using surgical blade, forceps and scissors. The dissecting kits were washed with distilled water and rinsed with alcohol before taking the samples. The muscle samples were quickly wrapped with aluminum foil, placed in plastic bags and kept in a deep freeze. The muscle samples were then used for heavy metal analysis, which was carried out at the Chemistry Laboratory of Hawassa University.

2.4.2. Digestion of fish samples

A blending device (mortar and pestle to protect metal contamination) was used for grinding and homogenizing the fish samples. Analytical digital balance was used to weigh the fish samples. Measuring cylinders and micropipettes were used for measuring different volumes of acid reagents and standard solutions. Volumetric flasks (25 ml, 50 ml and 100 ml) were used for dilution of the sample solutions and preparation of standards. Flame Atomic Absorption Spectrophotometer (FAAS) was used for the determination of concentrations of the metals (Mn, Cu, Zn, Cd, Cr, Ni and Pb) (Sneddon et al., 2006).

Stock standard solutions of concentration 1,000 mg/l in 2% HNO_3 of the metals (Mn, Zn, Cu, Cd, Cr, Ni and Pb) was used for preparing an intermediate standard solution containing 10 mg/l. Working standards were prepared from the intermediate standards of each metal. Deionized water (chemically pure <1.5 µs/cm) was used for cleaning of the glassware and dilution of the samples and other purposes (Sneddon et al., 2006). All glassware and polyethylene flasks used were cleaned with detergent solutions, soaked in 10% (v/v) nitric acid for 24 h, rinsed with deionized water and dried with hot air blower (Sneddon et al., 2006).

In this study, a method developed by Berghof (2011) Microwave Digestion Application was followed. Accordingly, a mass of 250 mg of each fish sample was transferred to digestion vessels followed by addition of 10 ml of concentrated (69-70%) nitric acid. The vessels were carefully shaken and placed in a fume hood for about 20 minutes for pre-digestion and to avoid foaming before they were placed on the turntable of the microwave system. Then the pre-digested samples in the digestion vessels were closed and heated in a microwave oven following the optimized procedure. In the first step the temperature was linearly increased to 160^{o}C in 15 min by rotating the magnetron to 70 watt power. In the second step, the temperature was kept at 205^{o}C for 15 min by rotating the power to 90 W and in the third step, the temperature was linearly decreased to 50 ^{o}C in 10 min by rotating the power to zero W. Similarly, to avoid splashing or spraying which can cause sample lose, the digestion vessel was cooled to room temperature (about 20 min) in a fume hood wearing hand, eye and body protection since a large amount of gas was produced during the digestion process. Finally, by opening the digestion vessel carefully, the digested solutions were transferred to 25 ml volumetric flasks and they were filled with deionized water up to the mark. The digested samples were then kept in a refrigerator until analyzed by FAAS.

2.4.3. Analytical procedure

The detection limit values of elements (mg/l) in FAAS for Cu, Cd, Mn, Zn, Pb, Co and Ni, were 0.005, 0.01, 0.03, 0.005, 0.04 and 0.05, respectively. The calibration curve for each element was done using the standards of each element at four different concentrations. The calibration curve relates the standard concentration with absorbance so as to determine the unknown concentration of the particular heavy metal in the sampled fish tissue.

2.5. Data analysis

Descriptive statistical procedure (frequencies, percentages, means and standard errors) were used to summarize and describe data obtained from gut content as well as heavy metal analysis. Single factor ANOVA test was employed to compare differences in trace metal concentration in the muscle of *G. quadrimaculata* between the dry and wet month samples. A linear regression relationship was also established between heavy metal concentration and body length of fish and the slope of the relationship was tested if significantly higher than zero or not using a t test procedure. Accordingly a relationship that gave a slope significantly higher than zero was considered to indicate that the particular trace element has been accumulated in the body of the fish as the fish grew older.

3. RESULTS

3.1. Food and feeding habits

3.1.1. Diet composition

Of the total 511 fish samples, 328 (64.2%) fish guts contained food whereas 183 (35.8%) were empty. The results of this study indicated that *G. quadrimaculata* in Lake Hawassa fed on a variety of food items including, phytoplankton, detritus, macrophytes, insects, fish eggs and zooplankton (Table 1). Amongst these detritus, fish eggs, macrophytes, phytoplankton and insects contributed most of the food volume and were found more frequently in the guts of *G. quadrimaculata*. On the other hand, zooplankton were relatively small in their contribution to the food volume and they were found in few of the guts examined (Table 1).

Table 1. Frequency of occurrence (%) and volumetric contribution (%) of various food items in the diet of 328 samples of *Garra quadrimaculata* in Lake Hawassa.

Food items	***Frequency of occurrence***		***Volumetric analysis***	
	Frequency	***Percent***	***Volume (ml)***	***Percent***
Phytoplankton	185	56.4	3.6	18.2
Diatoms	59	18.0	1. 9	9.4
Green algae	101	31.0	1.4	6.9
Blue green algae	31	9.5	0.4	2.0
Zooplankton	7	2.1	0.05	0.22
Copepods	5	1.5	0.041	0.2
Rotifers	1	0.3	0.004	0.02
Insects	87	26.6	2.8	14.1
Diptera	58	17.7	1.5	7.4
Ephemeroptera	42	12.8	1.3	6.4
Coleoptera	3	0.9	0.06	0.3
Hemiptera	1	0.3	0.002	0.0
Detritus	180	54.9	5.5	27.1
Macrophytes	144	43.9	3.7	18.2
Fish eggs	53	16.2	4.5	22.2

Detritus was the most dominant food item and it occurred in 54.9 % of the guts and accounted for 27.1 % of the total volume. Fish eggs were the second important food items occurring in 16.2% of the guts and comprising 22.2 % of the food volume. Macrophytes occurred in 43.9 % of the guts and accounted for 18.2 % of the total volume. Phytoplankton occurred in 56.4 % and contributed 18.2% of the volume of the food (Table 1). Among the identified phytoplankton, diatoms and green algae were plentiful and frequent food. They accounted for 9.4% and 6.9% of the total food volume, respectively. Among insects, Diptera (7.4% by volume) and Ephemeroptera (6.4% by volume) were the most important while Coleoptera and Hemiptera had minor contribution as food items. Again among identified zooplankton in the gut, copepods had relatively more contribution (0.2%) than rotifers (0.02%).

3.1.2. Variation in the diet compositions during dry and wet months

The results of this study showed that there was variation in the contribution of the different food items to the diet of *G. quadrimaculata* during the dry and wet months (Table 2). During the dry months, the bulk of the food was composed of fish eggs and detritus. Macrophytes and insects were of intermediate importance while the contribution of phytoplankton and zooplankton was insignificant in the diet. Fish eggs occurred in 24.3% of the guts and comprised 34.3 % of the total volume. The frequency of occurrence of detritus was 64.1% of the guts and its volumetric contribution was 34.0% of the total volume. Macrophytes and insects occurred in 39.8% and 27.7% of the guts examined, respectively and their volumetric contributions were 16.0% and 12.8% of the total volume, respectively. Phytoplankton occurred in 34.5% of the stomachs and accounted for 2.9% of the total volume while zooplankton, although present it was insignificant as food (Table 2).

In wet months, the volumetric contributions of food items in descending order were phytoplankton > macrophytes > detritus > insects > fish eggs > zooplankton. Phytoplankton occurred in 93.4% of the stomachs and comprised 33.5% of the total volume (Table 2). Among phytoplankton groups, diatoms were the most frequent food items and their volumetric contribution was also high (36.9% frequency and 18.3 % volume). Within diatom groups *Navicula, Cymbella and Mellosira* were abundant and frequent in the gut samples. Similarly, volumetric contributions of green algae (11.5 %) was also large (*Botrycocccus* was most identified species). *Merismopedia and Planktolyngbya* species were blue green algae, present in the gut contents.

Next to phytoplankton, macrophytes were important source of food for *G. quadrimaculata* in Lake Hawassa during the wet months. They occurred in 50.8% of the guts and constituted 20.4% of the total volume of food items (Table 2). Detritus occurred in 39.3% of the guts and accounted for 20.2% of the total volume of food items. The contribution of insects, macrophytes and detritus was relatively high and comparable during both dry and wet months.

Table 2. Relative contribution of different food items in the diet of *Garra quaimaculata* in Lake Hawassa during the dry and wet months.

Food items	***Frequency of occurrence***		***Volumetric analysis***	
	Dry months	***Wet months***	***Dry months***	***Wet months***
Phytoplankton	34.5	93.4	2.9	33.5
Diatoms	6.8	36.9	0.5	18.3
Green algae	23.8	37.7	2.2	11.5
Blue green algae	3.9	18.9	0.2	3.7
Zooplankton	1.0	4.1	0.01	0.50
Copepods	-	3.0	-	0.45
Rotifers	-	0.8	-	0.05
Insects	27.7	24.6	12.8	15.4
Diptera	16.5	19.7	3.9	10.9
Ephemeroptera	17.0	5.7	8.5	4.3
Coleoptera	1.0	0.8	0.4	0.3
Hemiptera	0.5	-	0.02	-
Detritus	64.1	39.3	34.0	20.2
Macrophytes	39.8	50.8	16.0	20.4
Fish eggs	24.3	12.5	34.3	10.1

3.1.3. Ontogenetic diet shift

The diet composition of *G. quadrimaculata* was found to vary with its size (Fig. 2). At the smallest size class (4.0-6.5 cm TL), fish eggs were the most important food items constituting 52.0% of the volume. Detritus was also an important source of food for the smallest size class constituting 24.4% of the food volume in the size class. The remaining food items, namely insects, macrophytes and phytoplankton comprised 7.4%, 6.7% and 9.6% of the volume in the size class, respectively.

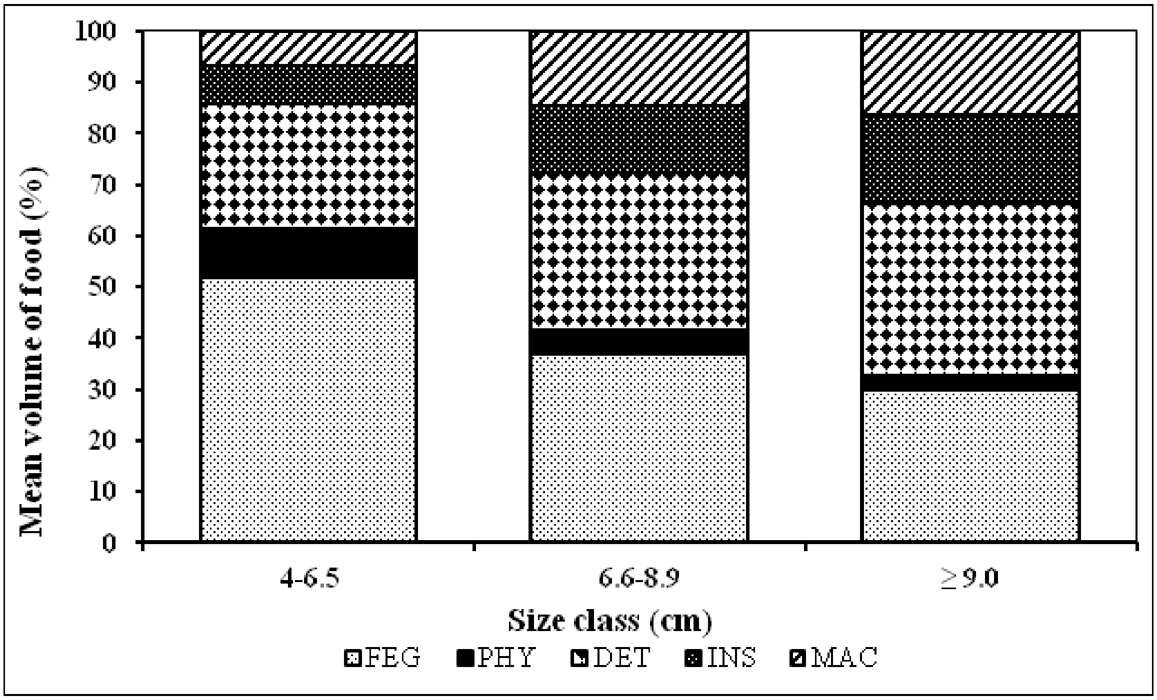

Figure 2. Relative volume of food items consumed by different size classes of *G. quadrimaculata* sampled from Lake Hawassa (FEG = fish egg, DET = detritus, INS = insects, MAC = macrophytes, PHY = phytoplankton).

In the size class 6.6- 8.9 cm, the volumetric contributions of fish eggs (37.1%) and phytoplankton (4.5%) declined while the contributions of detritus (30.7%), insects (13.0%) and macrophytes (14.6%) increased (Fig. 2). In the largest size class (≥9 cm TL), the volumetric contributions of fish eggs (29.8%) and phytoplankton (3.1%) further declined, while the contributions of detritus (33.5%), insects (17.3%) and macrophytes (16.3%) increased. In general, the importance of fish eggs and phytoplankton declined while the contributions of detritus, insects and macrophytes increased with fish size.

3.2. Concentrations of some trace metals in the muscle of G. quadrimaculata

Zn, Cr, Mn and Cu were found in the muscle of *G quadrimaculata* while Pb, Cd and Ni were below the detection limit of the instrument (Table 3). The concentrations of Cr and Zn varied in dry and wet month samples. Zn concentration in the dry month samples (0.76 mg/kg dry mass) was significantly lower than its concentration in the muscle samples taken during the wet months (1.14 mg/kg dry mass). By contrast, Cr concentration in the dry month samples (0.31 mg/kg dry mass) was significantly higher than in the samples collected during the wet months (0.07 mg/kg dry mass). Mn and Cu concentrations were considerably low and did not show seasonal variations.

Table 3. Mean (±SE) concentration of trace elements (mg/kg dry mass) in the muscle of *G. quadrimaculata* species sampled during dry and wet seasons from Lake Hawassa.

Element	***Concentration (mg/kg dry mass)****		
	Dry season (n = 13)	***Wet season (n = 13)***	***Overall (n = 26)***
	Mean (SE)	***Mean (SE)***	***Mean (SE)***
Zn	0.76 (0.039)[a]	1.14 (0.078)[b]	0.92 (0.054)
Cr	0.31 (0.055)[b]	0.07 (0.019)[a]	0.21 (0.04)
Mn	0.06 (0.006)[a]	0.06 (0.003)[a]	0.06 (0.003)
Cu	0.04 (0.004)[a]	0.03 (0.001)[a]	0.04 (0.002)
Pb	ND	ND	ND
Cd	ND	ND	ND
Ni	ND	ND	ND

**Note: ***Dry and wet season mean values of respective metal given different superscript letters (a, b) are significantly different from each other ($\alpha \leq 0.05$).

A relationship was established between metal concentration level in muscle and body size of *G. quadrimaculata* (Fig 3). The analysis showed that metal concentration level in muscle was not significantly related to body length of fish. The value of the slope was not significantly different from zero for all the metals analyzed indicating that metal concentration level was not significantly related to the body length of fish. This is also seen from the low R^2 value ($R^2 < 0.10$) exhibited by the relationships (Fig 3). In the present study, concentration level of the respective metal was compared with the corresponding value of maximum allowable concentration in fish flesh set by international standards (Table 4). Nonetheless, none of the metals were concentrated in the body of *G. quadrimaculata* more than the permissible level in the diet.

Table 4. Comparison of concentrations of heavy metals in the muscle of *G. quadrimaculata* with different dietary standards and guidelines (mg/kg dry weight).

Cu	***Cd***	***Zn***	***Mn***	***Pb***	***Cr***	***Reference***
4	0.04	10	-	0.4	-	MAFF(1995)
2	0.04	8	-	0.2	-	Zauke et al.(1999)
-	0.02	-	-	0.1	-	Zauke et al.(1999)
6	0.1	6	-	-	-	FAO(1983)
6	0.2	-	-	-	-	WHO(1996)
4	0.02	-	-	-	-	EU(2001)
0.04	-	0.92	0.06	-	0.21	Present study

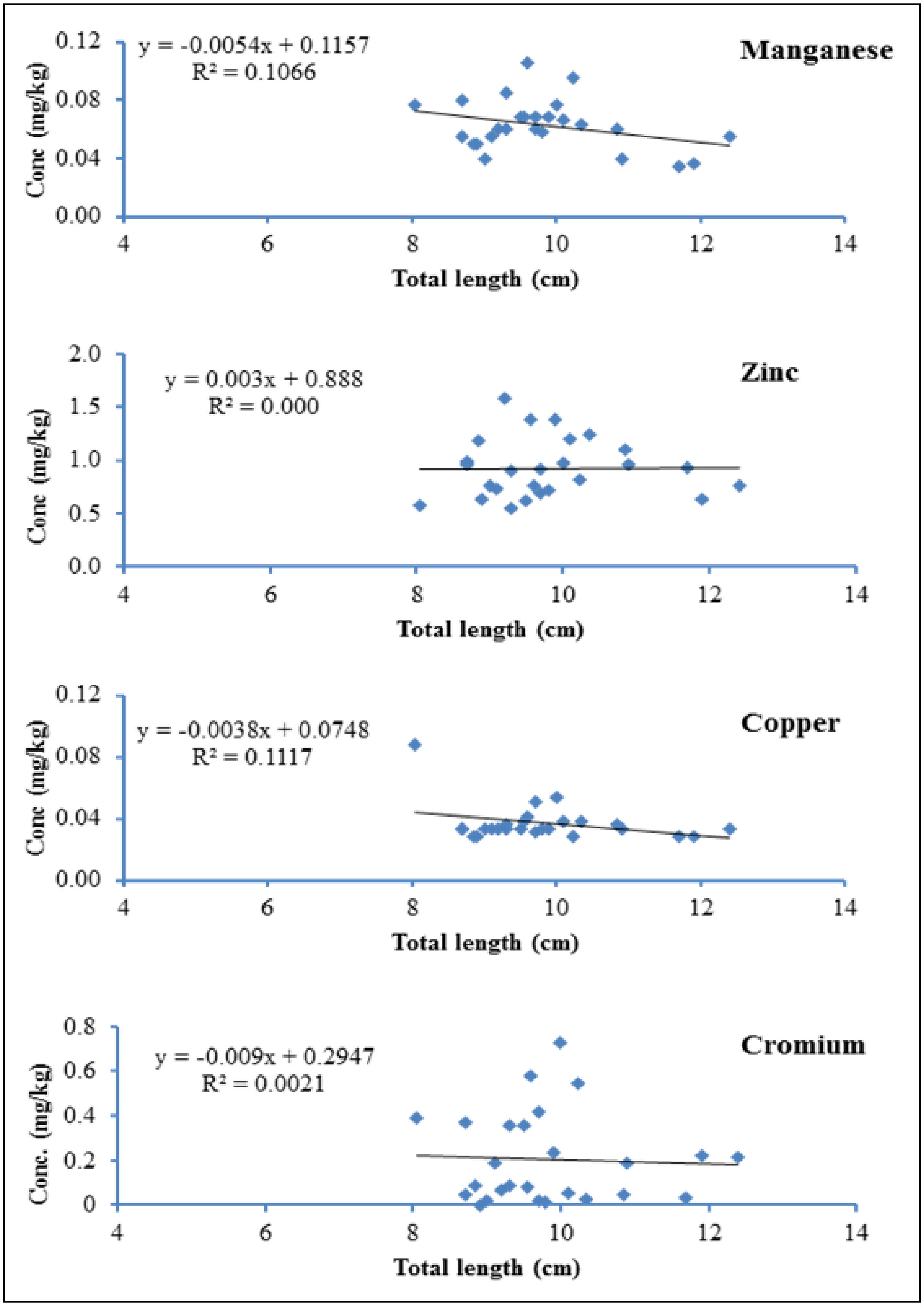

Figure 3. The relationship between heavy metal concentration in the muscle of *G. quadrimaculata* and body length.

4. DISCUSSION

Fishes differ greatly in the types of food they consume. The same species living in different ecological niches may not have the same food items in their gut (Shrestha and Shrestha, 2008). Most fish species in Lake Hawassa intensively utilize littoral habitat as their spawning, nursery, feeding and resting sites. Small fish prefer sheltered habitats because the physical complexity of these habitats often reduces both prey encounter rates and swimming speed of large predatory fish (Zerihun Desta et al., 2008).

The results of this study indicated that *G. quadrimaculata* in Lake Hawassa fed on a variety of food items including, phytoplankton, detritus, macrophytes, insects, fish eggs and zooplankton. Except zooplankton, the rest food items were important to the nourishment of *G quadrimaculata* in Lake Hawassa. No published work is available on the food and feeding ecology of *G. quadrimaculata* in Lake Hawassa and the present result is compared with other related species in different water bodies. Accordingly, compared to *G. quadrimaculata* in Lake Hawassa, the food item of *G. rufa* in Asi River and its tributaries (Turkey) is dominated by benthic algae (*Cyanobacteria*, *Chrysophyta* and *Chlorophyta*, with *Chrysophyta* being the most common),as well as rotifers and protozoans (Yalçin-Özdilek and Ekmekçi, 2006). Similarly *G. flavatra* in Rakhine State is herbivorous and mainly feeds on algae and detritus (Yalçin-Özdilek and Ekmekçi, 2006). Thus unlike the results of Yalçin-Özdilek and Ekmekçi (2006), the present findings clearly indicated that *G. quadrimaculata* in Lake Hawassa is an omnivorous species.

Seasonal variation has great effect on the contributions of different food items. The most important food items that constituted the bulk of food during dry season were fish eggs, detritus, insects and macrophytes. During this time, the consumption of fish eggs was high probably due to the spawning activities of the tiny cyprinidont, *Aplocheilichthys antinorii* in Lake Hawassa. Detritus was also important source of food during the dry season. The ventrally positioned (ventro-terminal) mouth of *G. quadrimaculata* is particularly suited to feeding on the bottom of the lake. Many bottom feeding fish living in mud flats, feed in this way and examination of the diet of these species showed that there was high percentage of detritus in their stomachs (Oso et al., 2006). Insects were important food sources during both seasons. The contribution of zooplankton was insignificant to the bulk of food consumed in both seasons. It is probable that the fish might have ingested zooplankton incidentally as it pursues its prey in the littoral region.

During the wet season, phytoplankton (diatoms, green algae and blue green algae) were the most dominant food items. Compared to the dry season, the contribution of phytoplankton was very high during the wet season. The most probable reason for high contribution of phytoplankton during the wet season could be the floods that drain large quantities of nutrients into the lake from the surrounding catchment areas causing phytoplankton bloom. In some environments large quantities of clay and other inorganic substances that drain from the catchment area during the wet season can be responsible for low productivity of phytoplankton, because the inorganic particles attenuate solar radiation from reaching the lower parts of the lake. However, in Lake Hawassa, Shallo Swamp is used to settle silt and other inorganic compounds that come from the surrounding areas.

Ontogenetic diet shifts have been shown to occur during the life of many fish species. This is because prey size is generally positively correlated with fish size (Zerihun Desta, 2007). A fundamental characteristic of fish is that growth in size is usually associated with changes in food resource use (Zerihun Desta et al., 2006). Generally, in case of *G. quadrimaculata*, the importance of detritus, insects and macrophytes increased from smaller to larger size class, while phytoplankton and fish eggs somehow declined in their importance in larger size class. As the age of the fish increases, the gape of the mouth and its swimming speed also increases and this enables the fish to feed on progressively large food items.

The concentration of Zn increased during the wet months probably due to runoff draining large quantities from the surrounding agricultural areas. The runoff consist point source and non point sources. Zinc is found in high concentration in the water of Lake Hawassa (i.e., 80 mg/l) (Behailu Berhanu et al., 2015), which could be the possible explanation for the high Zn in the muscle of *G. quadrimaculata* more than the other trace elements. According to Aweke Kebede and Tadesse Wondimu (2004), the Zn distribution in the tissue of *O. niloticus* in Lakes Ziway and Hawassa followed the order: liver > gill > bone > muscle. It ranged from 34.6–38.6 mg/kg in muscle, 61.9–78.8 mg/kg in bone, 82.3–97.1 mg/kg in gill and 85.6–115.9 mg/kg dry mass in liver. In the present study, Zn concentration of 0.92 mg/kg dry mass in the muscle of G. *quadrimaculata* was much lower than the concentration in the muscle of *O. niloticus* in Lake Hawassa. Similarly the concentration found in the present study was lower than the values reported in the muscle of *Barbus bynni*, *O. niloticus* and *Synodontis schall* in Lake Abaya, (Abnet Woldesenbet, 2011; Bishaw Tadele, 2011; Bizualem Gutema, 2011), But lower concentration than the present result was reported for *Hydrocynus forskahalii* in Lake Abaya (Seble Bancha, 2011).

Chromium was also one of the four detected elements in the muscle of *G. quadrimaculata.* This element is an essential nutrient in the diets of humans and is needed for many important functions, including lipid, protein, and fat metabolism. The present result indicated relatively higher concentration of Cr in the dry month samples than the wet month samples. Cr is present everywhere and can be found in three forms: metal ore, trivalent chromium (Cr III), and hexavalent chromium (Cr VI). The mean Cr concentration of 0.21 mg/kg dry mass in the present study was much lower than the maximum permitted level (13 mg/kg) in fish flesh for human consumption (FDA, 2001).

The third element in the muscle of *G. quadrimaculata* was Mn. There was no significant difference in its concentration during the wet and dry months. Mn is a trace mineral that is present in tiny amounts in the body. It is found mostly in bones, liver, kidneys, and pancreas (Atayese et al., 2009). Manganese can cause a poisoning syndrome in mammals with neurological damage, which is sometimes irreversible (Normandin, 2002). According to Aweke Kebede and Tadesse Wondimu (2004), the concentration of Mn in the muscle of *O. niloticus* in Lakes Hawassa and Ziway was 1.03 and 6.78 mg/kg dry mass, respectively. In the present study the concentration of Mn in the muscle of *G. quadrimaculata* was 0.06 mg/kg dry mass which is much lower than its concentration in the muscle of *O. niloticus* from Lakes Hawassa and Ziway. Manganese concentration in the present study was comparable to the concentrations found in the muscle of *B. bynni,* 0.067 mg/kg (Bishaw Tadele, 2011), *S. schall* , 0.07 mg/kg (Bizualem Gutema, 2011) and *O. niloticus*, 0.051 mg/kg (Abnet Woldesenbet, 2011) in Lake Abaya. But a lower concentration than the present result was reported in the muscle of *H. forskahalii* in Lake Abaya, 0.006 mg/kg (Sebele Bancha, 2011).

Copper was also found in the muscle of *G. quadrimaculata* in small concentration and its concentration was similar in the dry and wet month samples. Copper concentration in the muscle of *G. quadrimaculata* in the present study was much lower than the findings of Aweke Kebede and Tadesse Wondimu (2004) in the muscle of *O. niloticus* in Lakes Hawassa and Ziway. Cu concentration found in the present study was comparable with the value reported in the muscle of

B. bynni in Lake Abaya, 0.052 mg/kg (Bishaw Tadelle, 2011). On the other hand higher values were reported than the present finding in the muscle of *S. schall*, 0.944 mg/kg (Bizualem Gutema, 2011), *H. forskahalii*, 0.182 mg/kg (Sebele Bancha, 2011) and *O. niloticus*, 0.15 mg/kg in Lake Abaya (Abnet Woldesenbet, 2011).

In the present study, the relationship between metal concentration in the muscle and total length of fish was not significant for all trace metals detected. Thus there is no sufficient evidence to say that trace metal concentration increased as the fish grew older. That is to say, there is no strong evidence indicating more trace metal concentration in older fish than in younger fish. Hence there is no sufficient evidence to say that the fish bioaccumilates the investigated trace metals as it grows older. The reason for this may be the low level of the investigated trace metals in the food of the fish and the lake water.

5. ACKNOWLEDGEMENTS

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