Bulletin of Environmental Contamination and Toxicology (2024) 113:30
https://doi.org/10.1007/s00128-024-03938-2

Spatio-temporal Trends of Mercury and Stable Isotopes in Lower Food Web of Winam Gulf, Lake Victoria

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Received: 7 March 2024 / Accepted: 2 August 2024

Abstract
Components of the lower food web (mussels, Caridina and Omena) were collected from stations from Winam Gulf, Lake Victoria, Kenya in 2022 and 2023 to analyze for stable isotopes and total mercury (THg). Temporal comparisons were made with data generated for the same species in 1998. Values of δ^{15}N in mussels and Caridina were similar (6.89‰ vs. 6.78 ± 0.13‰), while Omena occupied an elevated trophic position (9.97 ± 0.24‰) with minor shifts in δ^{15}N over time. All species had elevated δ^{13}C values in 2022–2023 versus 1998 supportive of enhanced eutrophication in the Gulf. THg concentrations exhibited modest spatial differences between sites (< 2.6 fold), but not between Caridina and Omena. Larger temporal differences were apparent relative to spatial patterns with THg concentrations decreasing in study species by 2.8 to 4.1-fold between years. An exposure assessment indicated that Omena, commonly found in local markets, can be consumed up to 0.74 kg/month without generating excess THg exposures.

Keywords Bioaccumulation · Bioavailability · African Great Lakes · Mercury

Introduction

Lake Victoria provides up to 90% of the total fish landed in Kenya and is a vital source of high-quality protein for its surrounding populations (Njiru et al. 2014; Obuya et al. 2023). Mercury (Hg) is an important contaminant, particularly in aquatic ecosystems, owing to its ability to build up in fish tissues due food web biomagnification that can ultimately lead to human health risks due to the consumption of contaminated fish (Basu et al. 2022). However, determining cause-effect linkages of fish Hg contamination over spatial or temporal scales can be difficult because of the many complicating physical, chemical, and biological factors that contribute to Hg bioaccumulation, i.e. the bioavailable component of Hg residues in a given system, to upper trophic level fish (Grillitsch and Schiesari 2010). In addition, changes to and/or between site differences in Hg bioavailability at the base of the food web often show poor relationships with water or sediment total Hg (THg) residues across weak to moderate environmental gradients owing to many environmental factors that regulate microbially mediated mercury methylation and the complexity of mercury species-sediment interactions (Campbell et al. 2003a; Jonsson et al. 2012; Mason and Lawrence 1999). Therefore, uncovering spatial and/or temporal differences in mercury bioavailability is arguably best determined by measuring THg in baseline lower trophic level organisms within a food web.

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Published online: 23 August 2024

The Lake Victoria food web, while previously highly diverse with more than 500 species of endemic haplochromines (Witte et al. 2000; Njiru et al. 2005), became substantially altered and the food web simplified following introduction of the top predator Nile perch (*Lates niloticus*) in the 1950's (Njiru et al. 2005). This coupled with additional introductions of non-native tilapia (*Oreochromis niloticus*), water hyacinth and other lake stressors including increasing eutrophication shifting the plankton community towards cyanobacteria generated major perturbations to the lake's biodiversity and food web structure (Njiru et al. 2005; Otieno et al. 2022; Simiyu et al. 2018). Following the haplochromine crash in the 1970's, Nile perch now dominate the large fish biomass of the lake and continues to exert strong top-down control on many of the remaining fish (Kishe-Machumu et al. 2012). The loss of pelagic haplochromines, which previously were controlling abundance of the freshwater shrimp (*Caridina nilotica*) through predation on its juveniles resulted in dramatic increases of the abundance this species and it now forms a major component to the diet of juvenile and medium sized Nile perch (Cornelissen et al. 2018). *C. nilotica*, henceforward referred to as Caridina, is the only species of shrimp in Lake Victoria with littoral populations that feed on epibenthos and detritus while offshore populations engage in facultative planktivory (Lehman et al. 1996). The small pelagic cyprinid Omena (*Rastrineobola argentea*) also showed major increases in abundance following the haplochromine crash and this species now constitutes one of the largest fisheries in the lake, primarily supporting local markets as a high quality source of protein while Nile perch catches are more often directed towards high value export markets. Omena largely feed on zooplankton and small invertebrates (Witte et al. 2000; Wandera 2000). Despite the importance of lower food web items, Caridina and Omena, to Lake Victoria biomass, trophic transfer and mercury dynamics, there is limited information about the feeding ecology and contaminant concentrations in these lower food web species (Campbell et al. 2004; Mwirigi et al. 2022).

The present study examined trophic position and THg content in lower food web organisms from Winam Gulf, Lake Victoria, Kenya. Campbell et al. (2003a) provided the first combined stable isotope and THg trophodynamic description for Winam Gulf in 1998. Since that time, Winam Gulf has undergone additional changes including high human population growth (Aloo et al. 2013), landscape alterations (Juma et al. 2014), further changes in water quality (Kundu et al. 2017), increased eutrophication (Sitoki et al. 2010), biodiversity loss (Renaut and Owen 2023), additional species invasions (Otieno et al. 2022; Aloo et al. 2013) and other stressors that can alter Hg sources and loads and/or Hg bioavailability. The objective of this work was to characterize spatial and temporal patterns in carbon source, trophic position and THg residues of lower food web organisms inclusive of unionid mussels, Caridina and Omena of Winam Gulf.

Materials and Methods

Study Area and Fisheries

Winam Gulf is a large embayment of Lake Victoria with a surface area of 1400 km^2 and average depth of 6 m (Fig. 1). The gulf's waters encompass Kenya's jurisdiction of this African Great Lake which is considered the most important waterbody supporting the country's commercial fishing activities (Njiru et al. 2018). Portions of the southern catchment of Winam Gulf is characterized by artisanal gold mining known to be sources of mercury (Kola et al. 2019) coupled with industrial and domestic inputs arising from its proximity to Kisumu, a city located to the northeast (Kenya's third largest city). The remainder of the catchment is strongly influenced by agriculture with four major rivers draining into Winam Gulf (Gikuma-Njuru et al. 2018). The Gulf waters are semi-isolated from the main lake via the narrow constrictions at the Mbita Channel and the Rusinga Channel generating an estimated water residence time of 3 years within the embayment (Gikuma-Njuru et al. 2018). Commercial fisheries of Winam Gulf are dominated by the native pelagic cyprinid minnow (Omena), introduced Nile perch (*Lates niloticus* L.) and to a decreasing extent introduced Nile tilapia (*Oreochromis niloticus*). Nile perch is the top predator of the system and feeds extensively on Caridina at sizes below 50 cm shifting to predominantly fish diets inclusive of Omena, native cichlids, Nile tilapia and cannibalism with increasing size (Kishe-Machumu et al. 2012). Both Caridina and Omena play important roles in the early exposure and bioaccumulation of THg by Nile perch, a species considered the most important fishery export for the country. Furthermore, extensive consumption of Omena in dried form by the local population can play a direct role in human Hg exposures.

Study Design

Mussels (*Mutela bourguignati*) were collected by trawl in 2022 at Naya (Fig. 1). Seven live mussels were collected, but owing to the small shucked size, were pooled and analyzed as a single sample. Other food web items were collected in 2023 by trawl. Caridina were sampled in triplicate as pooled samples ($n = 30$ to 50 organisms/pool) at four stations: Bala Rawi, Mirunda, Naya, and Mid Gulf (Fig. 1). Omena were collected as triplicate pooled samples ($n = 5$

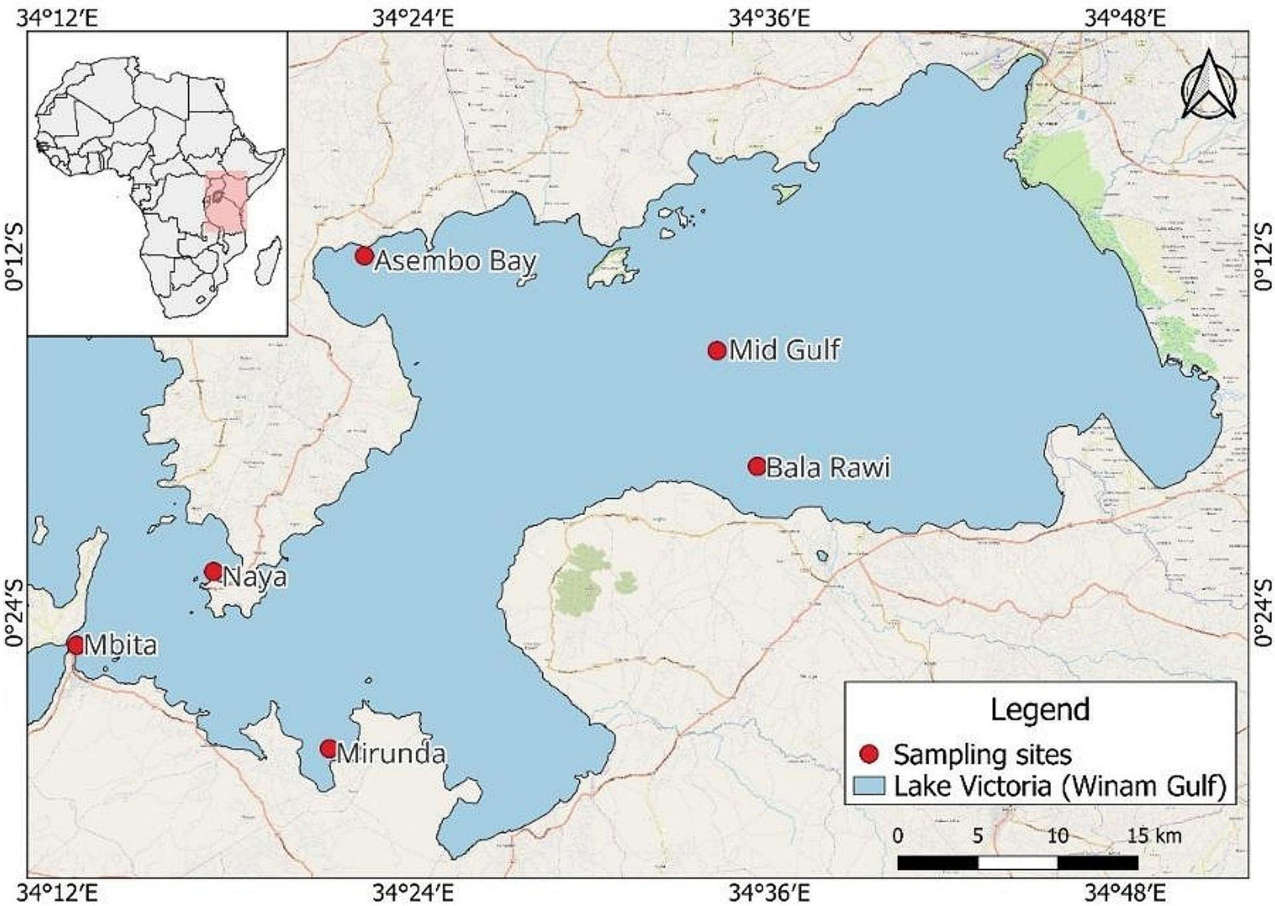

Fig. 1 Map of Winam Gulf, Lake Victoria showing the sampling stations. Continental Africa insert highlights Lake Victoria and surrounding countries (Uganda, Tanzania and Kenya), Winam Gulf (large map) is an embayment in the North Eastern corner of the lake within Kenya

to 15 organisms/pool) from five stations: Bala Rawi, Mbita, Naya, Asembo Bay, and Mid Gulf. Samples were sorted on board the research vessel and frozen until laboratory analysis. At the Kenya Marine Fisheries Research Institute (KMFRI) laboratory in Kisumu, Kenya, samples were dried at 90°C overnight to determine moisture and shipped to the University of Windsor for analytical chemistry. The above sampling was conducted under an approved animal ethics permit from the University of Windsor. Data for equivalent species collected in Winam Gulf from 1998 were obtained from Campbell et al. (2003a).

Analytical Chemistry

Total Hg was analyzed by Direct Mercury Analyzer (DMA-80). Samples were quantified from the signal response generated from a certified liquid mercury standard (High-Purity Standards, Charleston, SC, USA) diluted over a 10-point calibration curve. Certified reference materials (CRMs; National Research Council of Canada – Dorm-3 and Dolt-4) were run with samples and had a mean ± standard deviation (STD) recovery of 92.3 ± 0.04%. The THg detection limit was 0.42 ng/g d.w. Mercury data are reported as dry weight concentrations. Stable isotopes of carbon and nitrogen were analyzed by Delta V Plus Thermo-scientific CF-IRMS with a 4010 Elemental Combustion. Isotope ratios were expressed relative to PeeDEE belemnite standards to calculate $\delta^{13}C$ values and nitrogen gas in ambient air for $\delta^{15}N$ calculation. The accuracy of isotopes was determined using CRMs: NIST standard bovine liver (1577c), USGS 40, and IVA33802174 UREA. The mean ± STD recoveries of isotopes in CRMs were: 99.2 ± 0.21% and 99.3 ± 0.03% for $\delta^{13}C$ and $\delta^{15}N$. Analytical chemistry and quality controls for THg and isotopes from 1998 are reported in Campbell et al. (2003a).

Data Analysis

Two-way PERMANOVA (PAST version 4 software) was used to test for differences in isotope space between species, sample locations, and the location x site interaction for 2023 samples and then the analysis was repeated across the 1998 and 2023 time points. For temporal comparisons, given lack of site specific information from 1998, both 1998 and 2023 data were pooled across sampling locations. For THg, data were non-normal and non-parametric Kruskal-Wallis tests were used to test for spatial differences between species and sites coupled with Dunn's post-hoc tests to contrast THg across combinations of species and locations. Given the lack of normality for THg residues, measures of central tendency and variance are reported in the text as median ± quartile values.

Results and Discussion

Spatial differences in isotopes and mercury in lower food web items. Figure 2 (left graphic) presents a biplot of δ^{13}C and δ^{15}N isotope values measured in 2022/2023 samples. PERMANOVA (restricted to Caridina and Omena) revealed highly significant differences in isotopic signatures between species ($p < 0.001$) and across locations ($p < 0.001$). However, the species × location interaction was non-significant ($p > 0.6$). With respect to carbon, both Caridina and Omena exhibited the lowest δ^{13}C values at Naya and their highest values at Bala Rawi. Caridina demonstrated a larger range of δ^{13}C values compared to Omena although within site variation in stable isotope ratios appeared to be greater for Omena compared to Caridina. Ojwang et al. (2004) reported δ^{13}C values for Caridina and Omena to range from −23.2 to -17.8‰ and −21.9 to -18.0‰, respectively that were narrower in their spread than reported here (our study range −23.0 to -11.8‰ and −21.8 to -15.1‰) but the above study observations were restricted to a smaller number of sampling stations. Our results suggests more localized feeding of carbon sources consumed by Caridina compared to Omena, the latter integrating carbon over larger foraging distances or incorporating greater diversity of prey items at a given location. Although little is known about the spatial movements of the two species, biological profiles report Caridina to inhabit littoral zones of rivers and lakes (Leuven et al. 2008) whereas Omena undergo diurnal vertical migrations switching their diet between zooplankton and benthic invertebrate larvae (Wandera 2000). Concerning δ^{15}N, Caridina and mussels were equivalent at 6.78 ± 0.13‰ and 6.89‰, both occupying a similar trophic position (TP). In contrast, Omena had a mean δ^{15}N value of 9.97 ± 0.23‰. Based on a δ^{15}N trophic enrichment factor of 2.4‰ (Kwon et al. 2012) and use of mussels as a baseline organism with an assumed TP of 2 (Post 2002), Omena had an estimated TP of 3.28. These findings are consistent with others reporting Omena to occupy higher trophic status compared to Caridina (Campbel et al., 2004; Nyamweya et al. 2016; Ndegwa et al. 2019). Ojwang et al. (2004) reported mean ± SD δ^{15}N values of 5.4 ± 0.4‰ and 12.1 ± 0.6‰ for Caridina and Omena, respectively. When expressed relative to phytoplankton, Caridina and Omena from Ojwang et al's study yielded TP estimates of 0.54 and 3.33 based on the same calculation as above. Ojwang et al. (2004) concluded Omena to largely consume zooplankton, others report this species as consuming a combination of zooplankton and benthic invertebrates (Witte et al. 2000; Wandera 2000).

Figure 2 (right graphic) presents THg concentrations by species and sample location from 2022/23 collections. Omena from Mbita had the highest THg residues (median THg concentration of 80.6 ng/g d.w.) but THg

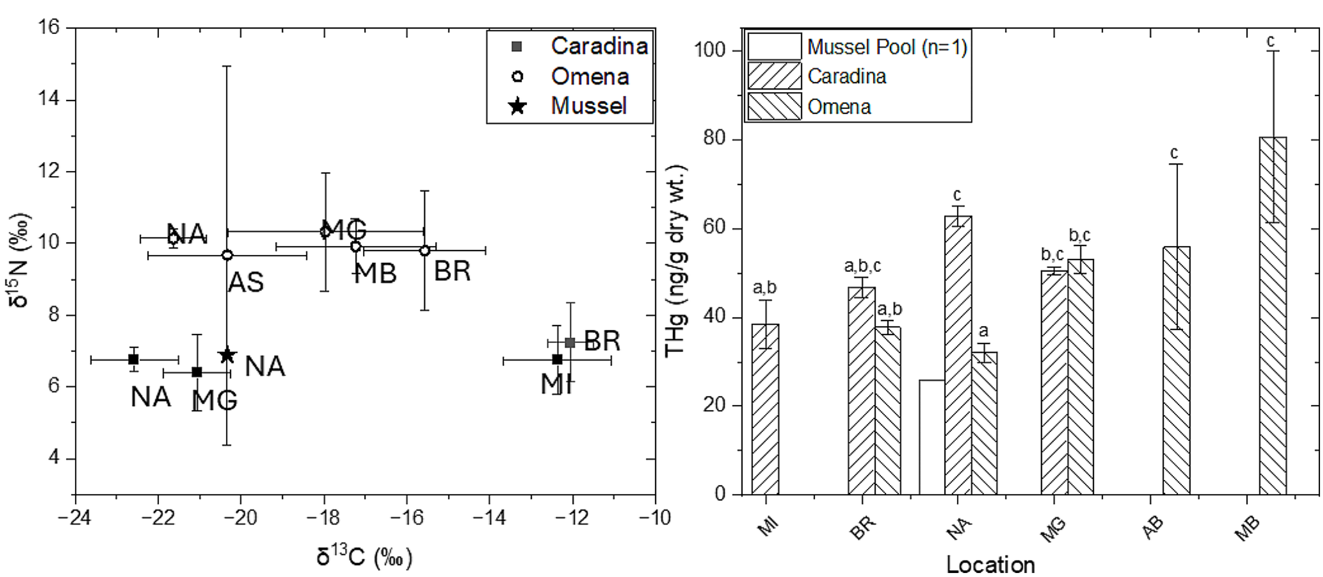

Fig. 2 Median ± 95% confidence intervals of stable isotope of carbon and nitrogen (left graphic) and median ± quartiles of THg (right graphic) of the lower food web mussel pool, Caridina and Omena. Locations: Mirundi (MI), Bala Rawi (BR), Naya (NA), Mid Gulf (MG), Asembo Bay (AB), Mbita (MB)

concentrations at this site did not differ significantly from Asembo Bay ($p > 0.7$; Dunn's test) or the Mid Gulf location ($p > 0.1$; Dunn's test). The lowest THg concentrations in Omena were observed at Naya (median of 32.1 ng/g d.w.) which did not significantly differ from Bala Rawi ($p > 0.5$; Dunn's test). In contrast, Caridina demonstrated its highest median THg concentration at Naya (62.9 ng/g d.w.) where Omena THg was at its lowest and mussels also showed low concentrations at this site. Differences by location in Caridina only occurred between Mirunda and Naya. Despite the above, spatial differences of median THg residues were within factors of 1.6 and 2.6 fold of one another for Caridina and Omena, respectively. These relatively small differences between locations imply a lack of strong point source signals and/or environmental gradients among the stations. Mwirigi et al. (2022) reported a THg concentration in Omena from Asat landing beach of 82 ng/g at the north end of Winam Gulf east of Assembo Bay that was consistent with values reported here. However, another site in proximity to Asat Landing (Bao) had a THg concentration of 258 ng/g d.w. well above the values observed here. Other studies have documented elevated trace metal concentrations near Kisumu Harbour (Kiema et al. 2019) indicative of industrial discharges and metal inputs associated with on-shore vehicle washing locations but did not report on THg residues (Ongeri et al. 2009). THg residues at the Mid Gulf site, having the closest proximity to Kisumu Bay, were intermediate compared to other sites. Major Hg loading sources to the Gulf have been speculated to be mainly of atmospheric origin (Fayiga et al. 2018) along with possible inputs from gold mining entering by river flow in the southwest corner of the Gulf (Kola et al. 2019), potentially explaining elevated residues observed in Omena at Mbita. However, the apparently limited effect of regional gold mining may be due to the large distance of such activities from sampling stations and reduced downstream advective transport from tributary flows into the southern portion of the Gulf. According to Roulet et al. (1999), in the Amazon basin, gold mining does not influence THg concentrations in water further than 50 km downstream. Furthermore, elemental Hg used for amalgamation in gold mining has a low mobility due to its high density (13.6 g cm^3; Lino et al. 2019; Gerson et al. 2018).

Despite the elevated trophic status of Omena relative to Caridina, there was no significant differences in THg concentrations between these species ($p > 0.9$; Kruskall-Wallis Test). There was also no relationship between THg concentration and $\delta^{15}N$ values within a species ($p > 0.5$; linear regression on log Hg vs. $\delta^{15}N$) or across species ($p > 0.05$; linear regression analysis). Many studies have demonstrated strong positive relationships between $\delta^{15}N$ and THg in aquatic food webs (Lavoie et al. 2010; Jones et al. 2014). However, differences in trophic position between the study organisms in this research were small and varied by only one trophic level. Furthermore, the study species were all small bodied organisms living in warm tropical environments, conditions that enhance Hg elimination rates lowering biomagnification potentials compared to larger fish of similar trophic position (Yao and Drouillard 2019). When data from this study are compared against Winam Gulf Nile perch data from 2023 (Drouillard et al. 2024), THg concentrations in Caridina and Omena conformed to expectations of food web biomagnification. Concentrations of THg in 85 cm Nile perch in 2022 ($TP = 4.11$) had a geometric mean THg concentration of 434 ng/g d.w. (based on length vs. THg relationship reported by Drouillard et al. 2024) compared to the median Omena + Caridina THg concentration of 49.8 ng/g d.w.

Omena are commonly consumed by local Kenyan people surrounding Winam Gulf. Replicate samples of this small minnow species had THg concentrations ranging between 28.1 and 88.8 ng/g d.w. Drouillard et al. (2024) compiled Hg tolerable daily intakes from different jurisdictions and reported a lowest tolerable monthly intake (TMI) for total mercury of 3158 ng/kg BW. This TMI is considered protective of human health risks due to Hg exposures for the sensitive population inclusive of women of childbearing age and children. When coupled with an average estimated Kenyan body weight of 60.7 kg (Walpole et al. 2012) and highest THg concentration recorded for the species, a maximum Winam Gulf Omena consumption rate of 2.16 kg per month or 25.9 kg/year (or less) would be considered protective of the THg TMI benchmark. Alternatively, Mwirigi et al. (2022) reported their highest observed dry weight THg concentration of 258 ng/g in Omena at one location in Winam Gulf. Using this value in place of the maximum observed in this study would yield a maximum Omena consumption rate recommendation of 0.74 kg/month or 8.9 kg/year. Estimates of total fish consumption by the Kenyan population vary between 4.5 and 13 kg/year (Cheserek et al. 2022; Drouillard et al. 2024) and while the data from the present research support unrestricted consumption of this species it is clear that additional research to better address spatial variation in Omena THg concentrations throughout the gulf is needed to complete a formal risk assessment.

Temporal patterns of isotopes and mercury in lower food web items. Table 1 presents temporal data (pooled across sites) on isotopes and THg in study species during 1998 and 2022/23. For isotopes, two-way PERMANOVA indicated a highly significant ($p < 0.001$) effect of species (*C. nilotica* and *R. argentea* only), year ($p < 0.001$), and non-significant ($p > 0.9$) species × year interaction. The differences were more strongly evident in relation to temporal change of $\delta^{13}C$ values, which increased by a median value of 5.6‰ across

Table 1 Stable isotope and THg concentrations in mussels, *Caridina nilotica* (Caridina) and *Rastrineobola argentea* (Omena) from Winam Gulf during 1998 and 2022

Year – Species	δ^{13}C (‰) Median (range), *n*	$\delta^{15}N$ (‰) Median (range), *n*	THg (ng/g d.w.) Median (range), *n*
1998 - Mussels	-23.7 (-24.1 – -23.3), 2	7.42 (7.07 – 7.78), 2	71.9 (53.2 – 80.1), 2
1998 – Caridina	-22.4 (-23.6 – -20.6), 11	6.13 (2.98 – 8.36), 11	201.5 (168.1 – 235.0), 2
1998 – Omena	-23.9 (-24.3 – -23.2), 3	12.0 (9.3 – 12.6), 3	191.0 (157.9 – 224.0), 2
2022 – Mussels	-20.3, 1	6.89, 1	25.8, 1
2023 – Caridina	-16.8 (-23.0 – -11.8), 12	6.84 (5.99 – 7.73), 12	48.4 (35.6 – 63.1), 12
2023 - Omena	-18.3 (-21.8 – -15.1), 12	10.1(7.24 – 11.17), 12	50.3 (28.1 – 88.8), 12

species. Relative to Ojwang et al.'s (2004) study, δ^{13}C values in Caridina and Omena increased by 1.9 and 1.7‰ relative to present results. Increased δ^{13}C across years is consistent with eutrophication-induced shifts in carbon isotope signatures (Fogel and Cifuentes 1993). Natugonza et al. (2016) observed 7% increases in primary productivity in Winam Gulf between 1972 and 1986 and others reported increasing eutrophication of the system in recent years (Gikuma-Njuru and Hecky 2005; Hecky et al. 2010). Several other Lake Victoria studies, noting enhanced δ^{13}C values in various food web items across sites, or as shifts in time, have interpreted such changes to be related to increased primary production (Cornelissen et al. 2018; Drouillard et al. 2024). Thus, the temporal shift observed for δ^{13}C in lower food web components provides some support for increasing intensity of eutrophication over time. However, we cannot rule out that the observed δ^{13}C shifts are an artifact of different sampling sites across the different surveys.

Nitrogen isotopes showed smaller changes between years with conflicting trends between species. Mussels and Omena decreased their δ^{15}N values by 0.53 and 1.89‰ whereas Caridina increased its median δ^{15}N by 0.70‰ (Table 1). These trends are consistent with Ojwang et al.'s (2004) Winum Gulf study in comparison to our 2022 data, with Omena δ^{15}N values decreasing by 2.10‰ and Caridina increasing by a larger amount of 1.44‰. Drouillard et al. (2024) reported a similar decrease of δ^{15}N values for Winam Gulf Nile tilapia (by 1.43‰) between 1998 and 2022. The authors interpreted the shift in tilapia δ^{15}N values to be due to a combination of change in nitrogen sources entering the Gulf and shift in feeding behavior of Nile tilapia towards greater reliance on phytoplankton and periphyton over time. Changes in nitrogen sources associated with increased sewage inputs or due to increased N-fixation by cyanobacteria will lower δ^{15}N values incorporated into phytoplankton (Cabana and Rasmussen 1996; Casey and Post 2011). Given the magnitude of difference in nitrogen signals between sewage and chemical fertilizers (between 9 to 18‰; Cormier et al. 2021), even minor shifts among these sources could explain the observed decreases. However, the opposing increasing trend for Caridina is inconsistent with change to nitrogen source. Mussels, being filter feeders, are limited to a narrow range of consumed particle sizes (Mistry and Ackerman 2018) and are commonly assigned as baseline phytoplankton consumers with a TP of 2 (Cabana and Rasmussen 1996; Post 2002). Caridina are epi-benthic detritovores, and given their gape-limited raptatory feeding, are capable of consuming a wider range of particles inclusive of microcrustaceans (Lehman et al. 1996). On the one hand, increases in δ^{15}N values for this species may have been due to greater incorporation of zooplankton in the diet of Caridina between years. However, in 2022 when Caridina δ^{15}N values were heightened, this species occupied a similar δ^{15}N value as mussels suggesting both organisms were consuming phytoplankton. Given that the number of mussel samples analyzed was limited in each year ($n=2$ for 1998 and 1 pooled sample of $n=7$ in 2022) it is likely that the decrease in mussel δ^{15}N value is an artifact of low sample size.

Table 1 compares median THg concentrations across species sampled in 1998 and 2022/2023. Given that spatial sub-sampling was not part of the 1998 sampling design, 2022/2023 samples were combined across sites to generate overall median concentration values representative of the gulf. In addition, limited replication from 1998 prevented temporal statistical contrasts to be made. Reductions in median THg residues between years by factors of 2.8, 4.2 and 3.8 fold for mussels, Caridina and Omena, were observed but given similar magnitude of spatial differences observed earlier and high THg reported for Omena in different locations by Mwirigi et al. (2022), it is not known if the apparent declines reported here represent true temporal trends or not. There are no comparable studies reporting decreases in THg in the same species from other Lake Victoria locations highlighting the paucity of information about these important but understudied lower food web components. Other studies have demonstrated recovery of Hg in Lake Victoria sediments and in some larger fish species. Campbell et al. (2003b), describing sediment core research in a different embayment of Lake Victoria, indicated that Hg loadings increased between 1960 and 1980 followed by declines thereafter. Poste et al. (2012) contrasted Nile perch THg concentrations in Napolean Gulf collected fish from 2008/2009 with Campbell et al. (2003a) data from the same location in 1998 and inferred decreases in Nile perch THg concentrations at this location through time. Drouillard et al. (2024) reported a 1.6-fold decrease in THg residues of Nile tilapia (20–30 cm in length) from Winam gulf between 1998 and 2022 but a contradicting increase in THg of large Nile perch that was interpreted to be due to change in fish growth as opposed to differences in THg loadings. At least some of the declines in THg concentrations for lower food web components can be explained as biodilution due to enhanced eutrophication (Karimi et al. 2007; Walters et al. 2015) as inferred from changes in $\delta^{13}C$ values. However, changes to overall loads or change in mercury methylation efficiencies cannot be ruled out as alternative reasons for the differing THg trends through time. Overall, this study noted modest spatial differences in THg concentrations in lower food web components of Winam Gulf and somewhat larger temporal declines over the past 25 years. Finally, risk calculations indicate that Omena, which are consumed regularly by the Kenyan population and are used as livestock feed are not a major source of excess mercury exposures to humans but may facilitate food web biomagnification and trophic transfer to the top predator fish although further research to better characterize spatial differences in THg concentrations of Omena from Winam Gulf are needed.

Acknowledgements The authors would like to thank Mr. J.C. Barrette, University of Windsor for assistance with Hg analysis and Dr. A.T. Fisk and Ms. L. Paulic, University of Windsor, for performing stable isotope analyses. Funding for this study was provided by a U.S. National Science Foundation Grant (NSF-IRES Project# 1953468) to GSB, KMO and RMM. Additional funding in support was provided by a Natural Sciences and Engineering Research Council grant (NSERC-Discovery Grant) to KGD.

Declarations

Competing Interests The authors have no competing interests directly or indirectly related to this work.

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