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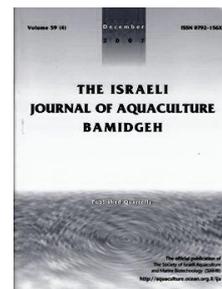
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## Low Mercury Levels in Lake Kinneret Fish

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### Abstract

Human exposure to the neurotoxin, methylmercury (MeHg), occurs primarily through the consumption of contaminated fish that are present even in pristine freshwater ecosystems. Lake Kinneret is the sole freshwater lake in Israel and supports an important commercial fishery. We examined total mercury (THg; often equivalent to MeHg in fish muscle tissue) for nine fish species in Lake Kinneret. Concentrations were low for all fish species, 6-409 ng/g, and below 500 ng/g MeHg, the level generally considered safe for human consumption. Of particular relevance are the very low levels of THg (<60 ng/g) in Galilee St. Peter's fish (*Sarotherodon galilaeus*) and lavnun (*Acanthobrama terrascantae*), the most commercially important species in Lake Kinneret. Biomagnification was apparent among trophic guilds, with THg concentrations increasing from primary to secondary to tertiary consumers. This study suggests that consumption of commercial species, especially primary consumers, will result in low MeHg exposure to humans.

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## Introduction

Consumption of contaminated fish is the main route through which humans are exposed to the potent neurotoxin methylmercury (MeHg; Mergler et al., 2007). Concern for human exposure to MeHg has prompted widespread fish and seafood consumption advisories, especially directed at top predatory species in marine and freshwater environments. The World Health Organization advises that fish for human consumption should not exceed 500 ng/g MeHg (WHO, 1990), but fish are commonly found at concentrations well about this established limit, even in pristine areas that do not receive direct industrial inputs of mercury (Wiener et al., 2003).

The widespread occurrence of unacceptable levels of MeHg in fish and seafood is due, in part, to the fact that human activities such as coal-power generation have greatly increased the global pool of inorganic mercury released into the atmosphere (Lindberg et al., 2007). Atmospheric mercury has a lengthy residence time, allowing for long-range dispersal before it is eventually deposited to aquatic ecosystems and their watersheds. Inorganic mercury from atmospheric deposition is converted to MeHg by sulfur-reducing bacteria, a process that occurs primarily in anoxic environments such as lake sediments and hypolimnia of stratified lakes (Gilmour et al., 1992). MeHg readily bioaccumulates and biomagnifies in aquatic food webs, resulting in MeHg concentrations in top predatory fish that can be a million-fold greater than the concentration in the water (Bowles et al., 2001). Subsequently, dietary exposure and food chain length can be important determinants of MeHg concentration among fish species. For example, fish that eat phytoplankton (primary consumers) have lower MeHg concentrations than fish that feed at higher trophic positions on zooplankton, benthos, or fish (Wiener et al., 2003). MeHg binds to proteins in fish muscle and is eliminated very slowly from this tissue (Van Walleggem et al., 2007), further contributing to high MeHg concentrations in the fish that humans consume.

Lake Kinneret, also known as the Biblical Sea of Galilee or Lake Tiberias, is the sole freshwater lake in Israel and has been a valuable source of fish for local consumption in the Galilee region since ancient times (Reich, 1978). Lake Kinneret has undergone a series of natural and anthropogenic changes over the last century, including marked changes in the composition of the fish community through stocking of non-native species and variable harvest practices (Ben-Tuvia et al., 1992; Hambright et al., 2008). Historically, Lake Kinneret has supported an important commercial fishery that has exceeded 1000 t annually since the 1960s (Reich, 1978; Ben-Tuvia et al., 1992), but in recent years has become considerably reduced. Annual harvest of lavnun (*Acanthobrama terrascantae*), also known as Kinneret sardine or bleak, is ~1000 t and constitutes about half of the total catch in Lake Kinneret, but only 6% of its commercial value (Ben Tuvia et al., 1992). Galilee St. Peter's fish (*Sarotherodon galilaeus*), popular at local restaurants and the most important commercial species, accounts for ~50% of the value of the Lake Kinneret fishery. Over the past 40 years, almost two-thirds of the ~5 million fish stocked annually into Lake Kinneret have been *S. galilaeus* reared in local hatcheries (G. Rubinstein, Israel Ministry of Agriculture and Rural Development, Ginosar). Introduced fish species, such as thin-lipped grey mullet (*Liza ramada*) and silver carp (*Hypophthalmichthys molitrix*), are also captured for local consumption.

Although there is a long history of fishing on Lake Kinneret, surprisingly and to the best of our knowledge, there have been no measurements of fish MeHg concentrations, even though several features of the lake may make it prone to the production of MeHg and subsequent transfer to fish species. First, the geographic location of Lake Kinneret in northern Israel, approximately 60 km inland from the Mediterranean coast, receives prevailing westerly winds from Europe that dominate the summer months. Recent atmospheric measurements made close to the nearby coastal city of Haifa, Israel, frequently reveal elevated mercury concentrations compared to other coastal Mediterranean areas as a result of wind masses passing overland before reaching the coast (Wängberg et al., 2008). Because of the large surface area of Lake Kinneret (168 km<sup>2</sup>), the strong sea breezes produce internal seiches that occur throughout the stratified period (May to December) and allow for extensive horizontal movement of

hypolimnetic water (Ostrovsky et al., 1996). In addition, the potential for mercury methylation is high because anoxic hypolimnetic waters constitute the lower 15-20 m of Lake Kinneret during stratification (Ostrovsky and Yacobi, 2010). Organic rich sediments and warm water temperatures promote intensified sulfate reduction in the hypolimnion of this productive, subtropical lake (Hadas and Pinkas, 1995), which should also serve to enhance mercury methylation. Taken together, these features suggest that Lake Kinneret may have inherent conditions that favor MeHg production and transfer to fish.

The main objective of this study was to measure mercury concentrations in a variety of Lake Kinneret fish species. An understanding of the levels of MeHg in Lake Kinneret fish species is warranted given the continued reliance upon the fish resources of this lake for human consumption. We selected fish species that feed at different trophic levels to understand the potential range of MeHg exposure to humans, as well as determine whether feeding patterns correspond to MeHg levels in fish species, as in other systems.

### Materials and Methods

*Fish collection.* Fish were collected from the pelagic region of Lake Kinneret by purse seining from a commercial vessel during the winter and spring of 2010 (Table 1). The net used was 20-mm stretch mesh with a 28-mm stretch bunt capable of capturing fish as short as 80 mm. After each seine haul, the catch was sorted and fish were immediately placed on ice. A random sample of the fish was selected, taken to the Kinneret Fisheries Laboratory in Tiberias, and frozen for later analysis. In total, we captured nine fish species from Lake Kinneret.

Table 1. Number and size of fish used in composite samples to analyze total mercury concentration in muscle tissue of Lake Kinneret fish species.

<i>Species</i>	<i>Common name</i>	<i>Major diet components (source)</i>	<i>n</i>	<i>Total length in cm (range)</i>	<i>Weight in g (range)</i>
1 <i>Tristramella simonis</i>	Simon's tristramella	Organic material (Spataru and Gophen, 1986)	6	17.5 (17-19)	134.9 (110-160)
2 <i>Liza ramada</i>	Thin-lipped grey mullet	Organic material (Shapiro, 1998)	7	32.8 (15-43)	398.0 (26-803)
3 <i>Sarotherodon galilaeus</i>	Galilee St. Peter's fish	Phytoplankton (Spataru, 1976)	6	19.1 (15-20)	133.1 (108-147)
4 <i>Hypophthalmichthys molitrix</i>	Silver carp	Phytoplankton, zooplankton (Spataru and Gophen, 1985a)	2	65.0 (50-80)	3545.5 (1220-5871)
5 <i>Acanthobrama terrascantae</i>	Lavnun, bleak, Kinneret sardine	Zooplankton (Landau et al., 1988)	9	15.4 (14-20)	35.6 (21-95)
6 <i>Tilapia zilli</i>	Redbelly tilapia	Benthos, zooplankton (Spataru, 1978)	4	20.0 (19-22)	162.8 (132-222)
7 <i>Barbus longiceps</i>	Long headed barbel	Benthos (Spataru and Gophen, 1987)	3	25.0 (21-30)	212.0 (143-340)
8 <i>Barbus canis</i> (small) <i>Barbus canis</i> (large)	Large-scaled barbel	Benthos, fish (Spataru and Gophen, 1985b)	4	24.2 (22-27)	160.4 (110-230)
			4	29.9 (29-34)	314.3 (241-481)
9 <i>Clarias lazera</i>	Catfish	Fish, benthos (Spataru et al., 1987)	4	49.5 (19-74)	992.3 (60-2081)

*Sample preparation and mercury analyses.* In the laboratory, we again sampled the fish catch to represent a range in fish size for each species, when possible. We measured the total length (nearest mm) and weight (nearest 0.5 g) of each fish and extracted a piece of the dorsal muscle using a stainless steel blade and trace metal clean techniques. We specifically selected muscle tissue, as this is the main storage site for MeHg in fish and because this is generally what humans consume. The pieces of muscle tissue were individually sealed in plastic bags and immediately frozen prior to transfer to the Kinneret Limnological Laboratory for further preparation. Skin and bone were removed from the muscle tissue pieces, which were then weighed and freeze-dried (Heto Lyolab 3000) for

48 h. The muscle tissue was individually weighed a second time, after drying, then we made a composite sample by homogenizing all tissue. For *Barbus canis*, we constructed two size-structured composite samples: small and large fish, which were analyzed separately.

We measured total mercury (THg) in the muscle tissue samples since THg is often equivalent to MeHg in many species, even in small fish (Van Walleggem et al., 2007). Mercury analysis by cold-vapor atomic fluorescence spectrophotometry (CVAFS) was conducted at the National Institute of Oceanography, Haifa, following established protocols. The homogenized dry tissue composite samples were approximately 0.5 g, digested with 5 ml concentrated nitric acid (65%) in Uniseal, Teflon-lined, high-pressure decomposition vessels. The digests were placed into a preheated oven at 140°C for 4 h. The cooled digests were diluted to 25 ml in volumetric flasks with double deionized water. Next, ionic mercury was reduced to an elemental state with a stannous chloride (2%) agent. The sample was aerated with argon to separate the volatile and the insoluble elemental mercury and the vapor was analyzed in a spectrophotometer at 254 nm (Millennium system, PS Analytical with a fluorescence detector). Accuracy of the procedure was verified by certified reference material (Dorm-3, NRCC) with the same batch of samples. Recovery of the reference material was 99.7%. Mercury concentration data were converted to wet weight measurements using the average percent moisture loss for each species based on the wet and dry weights of all individuals.

*Data analysis.* We broadly classified fish species according to published reports of their diet composition. Fish that consume organic material and phytoplankton were classified as primary consumers; those that consume zooplankton and benthos as secondary consumers; and those for which fish is a significant dietary component as tertiary consumers. Previous research using stable isotopes of carbon and nitrogen confirmed the feeding relationships and trophic position of the two most important commercial species in Lake Kinneret: Galilee St. Peter's fish, an omnivorous member of the *Tilapia* species feeds primarily on phytoplankton (i.e., it is a primary consumer), and lavnun that feeds on zooplankton as a secondary consumer (Zohary et al., 1994). Differences between trophic guilds in log-transformed THg concentrations were tested with ANOVA and LSD post-hoc tests (Statistica v6.1, StatSoft Inc.).

## Results

Mercury (THg) concentrations in all nine species were well below the safe limit for human consumption (500 ng/g wet wt). With the exception of catfish, THg concentrations were very low, below 200 ng/g (Fig. 1). There were large differences in THg muscle concentration between species, with values ranging 6-409 ng/g, likely related to the trophic guild within the Lake Kinneret food web. The three primary consumers, with diets consisting largely of organic material or phytoplankton, had the lowest THg levels ranging 6-20 ng/g. Included in this category is the Galilee St. Peter's fish with a low THg of 14 ng/g. The four secondary consumers, with diets comprised primarily of zooplankton or benthos, had THg concentrations ranging 24-126 ng/g. The two species known to consume fish as part of their diet, barbel and catfish, had the highest muscle THg concentrations: 132-409 ng/g. Large barbel averaged twice the mass of their small-bodied conspecifics and had 36% higher THg concentrations.

The significant differences in THg concentrations among the Lake Kinneret fish species was consistent with the trophic guild classification (ANOVA:  $F_{2,6} = 13.1$ ,  $p = 0.0065$ ). The mean THg concentration in primary consumers was  $13.5 \pm 7.1$  ng/g, significantly lower than for fish at higher trophic levels (LSD post-hoc: secondary consumers  $p = 0.025$ ; tertiary consumers  $p = 0.0023$ ). A similar difference was found between secondary ( $63.4 \pm 43.5$  ng/g) and tertiary ( $282.4 \pm 178.4$  ng/g) consumers ( $p = 0.034$ ). THg concentrations averaged 5-fold higher for secondary consumers than primary consumers. Likewise, the THg concentrations of tertiary consumers were 2.5 to 8-fold greater than lavnun, their main fish prey.

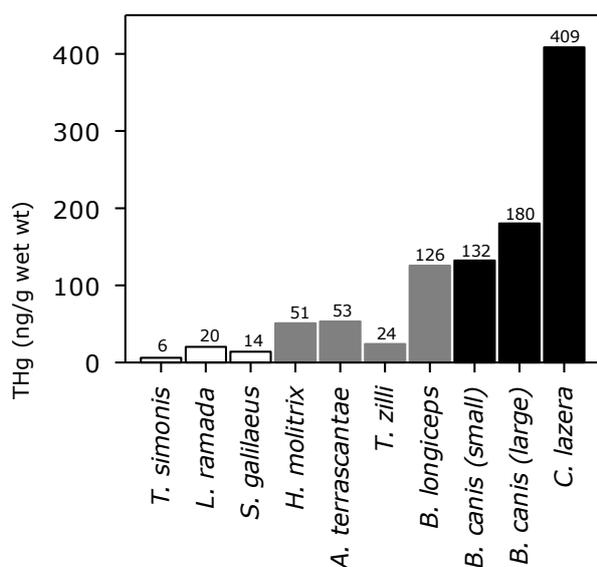


Fig. 1. Total mercury concentrations (THg) in muscle tissues of nine fish species from Lake Kinneret, Israel. Open bars represent primary consumers, shaded bars secondary consumers, and solid bars tertiary consumers.

Lake Kinneret supports a commercial fishery that supplies local markets in the Galilee, the findings from this study provide a strong basis for understanding potential MeHg exposure to humans from consumption of local fish.

The mercury concentrations in Lake Kinneret fish species were surprisingly low, especially given the suite of local conditions that could favor MeHg production and transfer to fish such as extensive anoxia and frequent mixing of hypolimnetic water from seiches (Ostrovsky et al., 1996). Low mercury levels such as those found in Lake Kinneret fishes appear to be a common feature of tropical African lakes (reviewed in Black et al., 2011). For example, the THg concentration of Galilee St. Peter's fish in Lake Kinneret was comparable to Lake Chad, <10 ng/g (Kidd et al., 2004). Likewise, the low THg concentration in *T. zillii* from Lake Kinneret was similar to Lake Malawi, 19 ng/g (Campbell et al., 2003) and lakes in the Alexandria region of Egypt, 12 ng/g (El Nabawi et al., 1987).

While the reasons for low THg levels in fish from tropical African systems remain unclear (Black et al., 2001), a number of features of tropical and subtropical lakes may limit the biomagnification of MeHg to upper trophic levels: a short food web, rapid fish growth, dilution of MeHg at the base of the food web in productive lakes, and lake-specific biogeochemistry may play roles in reducing biomagnification of MeHg (Campbell et al., 2003). Intensive fish harvest reduces MeHg levels in fish by promoting higher growth efficiency (Verta, 1990), which may also be an important factor regulating MeHg levels in Lake Kinneret fish. Another important feature of Lake Kinneret is the intensive stocking of fish from local hatcheries, especially *S. galilaeus* where up to 6 million are added on an annual basis. Mercury concentrations in freshwater fishponds in Israel were below the level of detection for several fish species, irrespective of whether they consumed pelleted feed or naturally-occurring organisms (Feldlite et al., 2008), suggesting that local hatchery practices do not contribute to the THg burden of fish stocked in Lake Kinneret, at least for those species examined. While this study did not focus on the mechanisms of MeHg transfer to fish, this area should receive future research attention in Lake Kinneret.

The wide variation in THg concentrations among Lake Kinneret fish species appears to be related to diet. Biomagnification of MeHg in aquatic food webs results in higher

## Discussion

The THg muscle tissue concentrations in the nine examined fish species ranged 6-409 ng/g, below the guideline considered a concern for human consumption (500 ng/g). For most of the species, the THg level was <200 ng/g, the level considered safe for frequent consumption (WHO, 1990). The only exception was catfish, which had a THg concentration that approaches the guideline value for limited consumption.

The THg concentrations were very low in Galilee St. Peter's fish, which is very popular in local restaurants, and in lavnun, which constitutes a large portion of the annual harvest from Lake Kinneret (Ben Tuvia et al., 1992). To put these concentrations into perspective, a 70-kg person could eat 8 kg of Galilee St. Peter's fish per week and still be within the limit recommended for the most sensitive portion of the population (1.6 ug MeHg per kg body weight; WHO, 2006). Because

concentrations of this contaminant with increasing trophic position (Bowles et al., 2001). Hence, we broadly partitioned the fish into three feeding guilds – primary, secondary, and tertiary consumers – based on published dietary data on Lake Kinneret. Fish THg concentrations increased significantly with increasing trophic guild. The THg concentrations of tertiary consumers, for which fish are an important dietary component, were approximately 4-fold higher than for secondary consumers and 22-fold higher than for primary consumers. The mercury concentration in St. Peter's fish, which feed primarily on the algae *Peridinium* but also on some zooplankton, was 4-fold lower than lavnun, which feed on zooplankton and are one trophic level higher (Zohary et al., 1994). Large-scaled barbel and catfish, the two tertiary consumers examined in this study, ingest a wide range of plants and animals (>50 species) in Lake Kinneret, however, fish is the most important dietary component of both species, and lavnun is the main consumed fish (Spataru and Gophen, 1985b; Spataru et al., 1987). Mercury concentrations in barbel and catfish were ~3-fold and 8-fold higher, respectively, than in lavnun. The mercury biomagnification between prey and predatory fish generally ranges 3-10x, suggesting that trophic transfer of MeHg in Lake Kinneret is similar to tropical and temperate freshwater ecosystems (Bowles et al., 2001; Wiener et al., 2003).

The variation in THg concentrations within Lake Kinneret fish species is consistent with known patterns of increasing accumulation with age. For example, in the large-scaled barbel, for which we produced size-stratified composite samples, large (and presumably older) fish had a 36% higher muscle THg concentration than their smaller conspecifics. Age may explain the size-related difference in THg, as older fish accumulate more mercury (Wiener et al., 2003). In addition, dietary preference may also play a role. Large-scaled barbel consume a wide range of taxa in Lake Kinneret, but the proportion of fish consumed (primarily lavnun) can markedly differ between the two size classes. Fish comprise 40% of the diet of small barbels and 70% of the diet of large barbels (Spataru and Gophen, 1985b). We were unable to distinguish between age and diet-related factors influencing THg concentrations in this study, but our data suggest that these factors are important for MeHg accumulation by Lake Kinneret fish species.

In conclusion, in all species examined from Lake Kinneret, the THg concentration was low and below the level considered a concern for consumption (WHO, 1990). Many of the fish species we examined are caught commercially and sold for local consumption. The findings from this study suggest that there is likely little health concern due to MeHg from consumption of Lake Kinneret fish. The one exception is catfish, perhaps, but this species is unpopular locally and has historically contributed <1% of the annual catch from Lake Kinneret (Ben Tuvia et al., 1992). Our data collection did not allow for assessment of factors influencing MeHg concentrations in fish. However, given the low levels of fish mercury contamination in spite of factors specific to Lake Kinneret that may enhance MeHg production and transfer, we believe further examination of MeHg cycling would be an important area of future study.

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