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minireview

Methylmercury in the Gulf of Trieste (Northern Adriatic Sea): From Microbial Sources to Seafood Consumers

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Summary

The Gulf of Trieste (northern Adriatic Sea) is one of the most mercury-polluted areas in the Mediterranean and in the world due to the past mining activity in the Idrija region (western Slovenia). The link between microbial production of toxic methylmercury (MeHg), and its bioaccumulation and biomagnification in marine food webs of the gulf is at present rather poorly characterized but is critical to understanding the links between sources and higher trophic levels, such as fish, that are ultimately vectors of human and wildlife exposure. This overview explores three major topics: (*i*) the microbial biogeochemical cycling of Hg in the area, (*ii*) the trophic transfer and bioaccumulation of MeHg in pelagic and benthic marine food webs, and (*iii*) human exposure to Hg through marine fish and shellfish consumption. These are important goals since the Gulf of Trieste is an area of great economical importance.

Key words: methylmercury, seawater, sediment, methylation, demethylation, food web, northern Adriatic Sea

Introduction

Mercury (Hg) is a toxic pollutant (neurotoxin) and human exposure occurs mostly through the uptake of edible marine organisms, especially fish (*1,2*). Methylmercury (MeHg), contributing >95 % of total Hg in fishes on top of the food web (*3*), is especially severe because it is a potential neurotoxin and represents a risk for humans (*4,5*). Microbial (biogeochemical), physical and ecological processes control the distribution and fluxes of MeHg between the sources and the bioaccumulation in the marine food web, leading to great areal differences in factors controlling Hg methylation in various aquatic systems. Globally, the majority of MeHg in the marine environment is derived mainly from *in situ* methylation of Hg(II), since the MeHg delivered by freshwater discharges and precipitation is small (*6*). The potential internal sources encompass Hg methylation in sediment (benthic) and Hg methylation in the seawater (pelagic) column. Estuaries and coastal waters are shallow marine areas with habitats where conditions, frequently with low oxygen levels, favour MeHg production and accumulation in food webs (*7*). Estuaries and coastal waters are also highly productive ecosystems and support fisheries and mariculture, which are significant food sources for humans and, thus, major vectors for human MeHg exposure (*5*).

The Gulf of Trieste (northern Adriatic Sea) and its local watershed (Fig. 1) represent an interesting case

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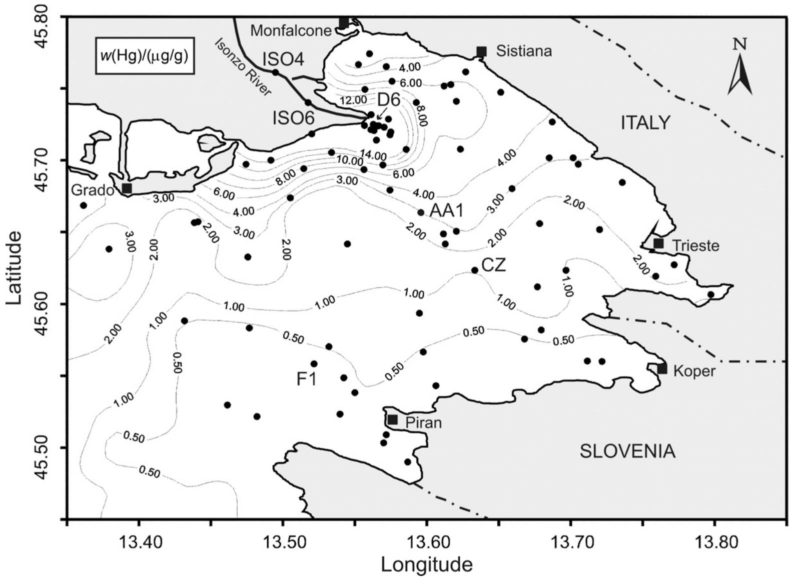

Fig. 1. Areal distribution of total Hg in surface sediments of the Gulf of Trieste (*10*) with location of study sites ISO_4, ISO_6, D_6, AA_1, CZ and F_1

study of anthropogenic Hg contamination of coastal waters. The Isonzo/Soča river system is the most important source of Hg to the Gulf of Trieste due to nearly five centuries of mining and draining of the cinnabar-rich deposits of the Idrija (western Slovenia) mining district (*8,9*). Because of erosion of the Hg-laden riverbank and fluvial deposits, the river remobilizes and transports the Hg remains into estuarine and coastal waters (*10,11*). Despite the fact that high riverine inflows are not frequent, the data indicate that they result in the transport and accumulation of Hg-rich riverine particles into the Gulf of Trieste. Even today about 1.5 tonnes per year of Hg is transported into the coastal area, of which 0.1 % is in the methylated form (*12,13*). When the river plume is oriented SW under the influence of the Bora wind (ENE), tidal currents transport mostly inorganic particulate Hg into the Gulf of Trieste and the adjacent Grado and Marano Lagoon producing highly Hg-contaminated sediments that are among the most contaminated in the Mediteranean and throughout the world (*10,14,15*). Hence, the gulf is a 'natural laboratory' for such study. Elevated Hg concentrations in deep layers of the sediment cores collected in the Gulf of Trieste are associated with sediment layers deposited just before the First World War (1913), corresponding to the period of maximal Hg production in the Idrija mining district (*10,16*).

The objective of this overview is to present, scale and synthesize the processes controlling MeHg production and degradation (bioremediation) and the bioaccumulation of MeHg in food webs in the Gulf of Trieste – an example of Hg-contaminated coastal water system where seafood (higher trophic levels) is the major route for the intake of very toxic MeHg in local population (*9*).

Scaling Hg and MeHg Distribution and Sources

Total, dissolved and reactive Hg in the water column

Analyses of total Hg (THg) and dissolved Hg (DHg) in the water column of the Gulf of Trieste and the Isonzo/Soča estuary (*17*) revealed that the majority is bonded onto suspended particulate matter (SPM). Higher THg (5–25 ng/L) and DHg (1–5 ng/L) concentrations in the surface layer of the gulf are restricted to the area in front of the Isonzo/Soča river plume expanding in a westerly direction. Data demonstrated increased Hg levels in the periods of higher riverine inflow in spring and autumn. The surface waters in front of the river Isonzo/Soča outflow exhibit about 10-fold higher THg concentrations than those found in the central and southern Adriatic (*18*), and in the Mediterranean (*19,20*), while DHg values differ less. The significant relationship between THg and DHg in the surface layer indicates their common origin and dispersion. A great proportion of Hg associated with SPM in the bottom layer is due to resuspension. Higher bottom DHg concentrations observed at some sites are probably due to remobilization from sediments including benthic recycling and resuspension. Relationships between DHg and salinity in the surface layer indicated nonconservative mixing in late spring during higher riverine inflow and nearly conservative behaviour in the late summer during lower riverine inflow, confirming the Isonzo/Soča river as the most important source of THg and DHg in gulf waters. Reactive Hg, readily reducible by $SnCl_2$ and volatile species, is significantly correlated with DHg, suggesting that the majority of DHg is reactive and potentially involved in biogeochemical transformations. The distribution coefficient K_D of Hg between the suspended and dissolved phases in seawater ranges between 10^5 and 10^6, somewhat lower compared to that in the riverine plume (*21*), confirming its strong association with suspended particulate matter (SPM). Seasonal data revealing the dynamics of THg in the water column (Fig. 2) show high concentrations in the surface layer in spring that occur simultaneously with lower salinities due to higher riverine inflow. However, during autumn, a maximum THg can be a consequence of Hg associated with 'fresh' phytoplankton organic matter that exhibits a low C_{org}/N ratio. The higher bottom

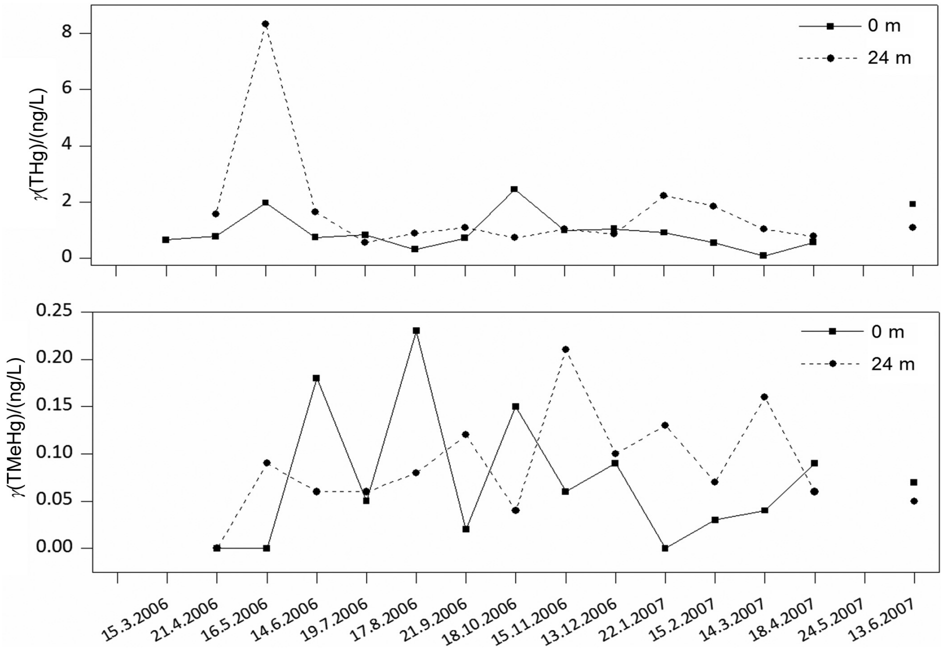

Fig. 2. Temporal variations of total Hg (THg) and total MeHg (TMeHg) concentrations in surface (0 m) and bottom (24 m) water layers at study site CZ (Fig. 1) in the Gulf of Trieste (unpublished results)

THg concentrations noted in winter can be the consequence of sediment resuspension. The waters of the Gulf of Trieste, especially the Isonzo/Soča estuary, are a net source of dissolved gaseous Hg (DGM) to the atmosphere (*22*).

Hg and MeHg in the water column are complexed by marine dissolved organic matter (DOM) that is compositionally and structurally complex (*23–26*). According to Hg analyses in mucilage (macroaggreate) interstitial water colloids of prevalently phytoplankton origin (*27*), representing agglomerated dissolved macromolecules, especially heteropolysaccharides (*28*), nearly all the dissolved Hg in the Gulf of Trieste is macromolecularly bonded (in colloids) prevalently onto higher molecular mass (M_r>30 kDa) fractions (*28*). Macromolecules are important in natural waters since they exhibit a larger surface areas, are abundant and more reactive compared to particles, and they aggregate. This agglomeration proceeds rapidly and metals are binding macromolecules in typical conformations. Despite the fact that all fractions, except lower M_r (<5 kDa), of the produced gels are microbially (*27*) and photochemically labile, the majority of colloidally bonded Hg (and metals) is preserved in the high M_r fraction containing various chelating functional groups, *i.e.* sulphydryl, carboxyl and amino (*27*). Lower K_D values of colloidal Hg (10^2–10^4), when compared with particulate Hg, demonstrate rather weak metal binding compared to that of particulate matter (*28*).

Total and dissolved methylmercury in the water column

Higher total MeHg (TMeHg) concentrations, up to 0.2 ng/L, have been observed only in the bottom layer, which was probably due to benthic recycling including benthic fluxes (*17*). These bottom water concentrations in the gulf were up to 3-fold higher than those reported for open Mediterranean waters (*19,20*). Conversely, concentrations of dissolved MeHg (DMeHg; <0.025–0.07 ng/L) are similar to those reported in other marine basins worldwide (*26*). The percentage of TMeHg as DMeHg is commonly low, ranging mostly between 0 and 1 %, but higher percentages, up to 5 %, are frequently found in the bottom water layer, suggesting the benthic origin of MeHg. While the percentage of TMeHg in THg is low most of the year, at <10 %, it increases in spring and summer to nearly 40 % due to higher microbial (methylation) activity in sediments, especially in the central part of the gulf (*29*). This consideration is further supported by the low percentage, <1 %, found in front of the Isonzo/Soča river mouth indicating lower importance of MeHg originating from the river input. The K_D of MeHg ranges between 10^{-1} and 10 confirming the much weaker binding of MeHg to SPM as compared to Hg, which is most probably present as cinnabar. Also, lower K_D values calculated for marine as compared to riverine MeHg indicate weaker binding of MeHg in the marine environment in the gulf (*21*).

The highest concentrations of surface water TMeHg occur in the warmer period (June-August, Fig. 2) following the algal bloom period that produces high dissolved organic carbon (DOC) (*30,31*) and particulate organic carbon (POC) (*30*) concentrations, which mostly originate from phytoplankton, and high heterotrophic (bacterial) abundance and production (*32*). The phytoplankton biomass exhibits early spring and autumn maxima, while the highest primary production usually occurs throughout the whole of spring until summer (*32*). MeHg production can be counterbalanced by the photochemical reduction of Hg(II) and by demethylation (*33,34*). However, photochemical degradation of MeHg can also be significant in surface waters (*35*) and in the Gulf of Trieste as well, since the euphotic zone is extended to the seabed (*36*). Conversely, bottom water TMeHg concentrations are generally higher due to processes occurring at the sediment-water interface and in surficial sediments, including sediment resuspension in late autumn (Fig. 2).

Sediment

Hg contents are particularly high along the littoral zone of the northern shore of the gulf near the Isonzo/Soča river mouth and the surrounding beaches (*10*), where the metal is present in a detrital form (cinnabar) in sandy sediments (*37*). Concentrations decrease nearly exponentially towards the south (Table 1 (*10,29,38–41*) and Fig. 1), in accordance with local hydrological conditions (*10*). Stable Hg isotope (δ^{202}Hg) analyses (*15*) confirmed that approx. 90 % of the Hg in sediments of the northern part of the gulf (sampling sites D_6, AA_1, CZ; Fig. 1) was of fluvial (Isonzo/Soča) origin, while Hg in sediments in the south (sampling site F_1; Fig. 1) is predominantly (>50 %) marine ('pristine') and similar to other Adriatic sediments. In the central part of the gulf, Hg, as Hg^{2+}, is mostly weakly associated with fine particles (<16 μm) and adsorbed onto the surface of clay minerals, and is partially complexed with colloids. Also, MeHg levels show a decrease with distance from the Isonzo/Soča river mouth, towards the gulf entrance (Table 1). No corrrelation was found between Hg and C_{org} of terrestrial origin, deduced from $\delta^{13}C_{org}$ values, while the link between MeHg and C_{org} of marine origin highlights the *in situ* production of MeHg (*39,42*). This is consistent with findings in the adjacent Grado and Marano Lagoon where an interaction of MeHg with solid-phase humics of marine origin has been reported (*43,44*). It seems that Hg is less reactive within the solid phase as one moves offshore in the gulf, a phenomenon that may be due to lower concentrations of organic matter and to the decreased rate of sulphide production (sulphate reduction) in offshore sediments (*29,40,45*).

Unlike solid-phase Hg concentrations, which decrease appreciably from the north to the south, pore water concentrations of Hg do not vary greatly throughout the gulf (Table 1). In addition, in sediment pore waters of the 'pristine' southern part of the Gulf of Trieste, it appears that there is little relationship between the pore water distribution and that of pore water humics, suggesting that pore water Hg is not strongly associated with dissolved humic compounds in sediments (*39,46*).

Benthic diffusive fluxes of Hg and MeHg in the Gulf of Trieste (Table 1) show similar values at all studied sites. The MeHg desorption from reduced metal (Fe) hydroxides under anoxic conditions and coprecipitation with iron oxides and MeHg demethylation in reoxidation may play a major role in determining MeHg benthic fluxes (*39,44,46*). The benthic fluxes appear, hence, mostly controlled by sedimentary redox conditions (redoxcline) as well as enhanced demethylation influenced by organic matter, microbial activity, sediment bioturbation and local hydrodynamics. Tentative benthic mass balance calculations showed that about 75 % of Hg is buried in sediments while about 25 %, from which about a quarter in the methylated form, is annually recycled and released at the sediment-water interface (*40*).

Microbial MeHg Production and Potential Controls

The Hg delivered by the Isonzo/Soča river is subjected to transformations in the Isonzo/Soča estuary, Gulf of Trieste and adjacent Grado and Marano Lagoon. Most of the Hg is in the form of cinnabar, with some Hg^0,

Table 1. Total Hg (THg) and MeHg in surface sediments, and dissolved Hg (DHg) and dissolved MeHg (DMeHg) in pore waters with respective distribution coefficients (K_D), organic carbon and sulphur contents, C_{org}/N molar ratios in surface sediments, potential methylation (k_{meth}) and demethylation (k_{demeth}) rate constants, and diffusive benthic fluxes (J) of Hg and MeHg in estuarine (ISO_4, ISO_6) and marine (D_6, AA_1, CZ, F_1) sites in the Gulf of Trieste

	ISO_4	ISO_6	D_6	AA_1	CZ	F_1	Ref.
w(THg)/(μg/g)	11.8–15.4	12.6	23.3–31.5	2.4–4.9	0.8–0.9	0.2–0.4	(*10*)
w(MeHg)/(ng/g)	2.1	1.9	4.9	0.2–1.1	0.1–0.4	0.2–0.7	(*29*)
w(MeHg)/%	0.01–0.02	0.02	0.01	0.01	0.02	0.002	(*29*)
γ(DHg)/(ng/L)	300	80	10–40	5–40	<40	3–18	(*29*)
logK_D	4.6	5.2	6.0	6.8	7.7	6.5	
γ(DMeHg)/(ng/L)	10	1.2	0.4	0.3	1.7	0.2–2	(*29*)
logK_D	2.3	2.8	1.7–4.1	2	3.8	3.3	
$w(C_{org})$/%	0.8–1.3	1.0	0.8	0.9–1.4	1.0	0.7	(*38*)
$n(C_{org})/n$(N)	8	8.8	8.5	8.2	8	4–10	(*38*)
w(S)/%	0.1	0.3	0.05	0.16	0.14	0.06	(*29*)
k_{meth}/(%/day)	0.2	0.6	4.8	2.5	3.2	0.7*	(*29*)
k_{demeth}/(%/day)	3	17	10	3	4	–	(*29*)
k_{meth}/k_{demeth}	0.07	0.04	0.48	0.83	0.8	–	
J_{Hg}/(ng/(m^2·day))	–	52	–20	–21 to 41	–	70	(*39,40*)
J_{MeHg}/(ng/(m^2·day))	–	6	2	–0.6 to 5.2	–	8	(*39,40*)

using ^{203}Hg (*29*), *^{197}Hg (*41*) which is not readily bioavailable (*46*). Hg transformations include the variation of Hg mobility due to interaction with organic and inorganic phases and bacterial transformations, including Hg methylation and MeHg demethylation. Hg methylation is, hence, influenced by sedimentary geochemical (redox) conditions, including the quantity and quality of organic matter, bioturbation and local hydrodynamics. Microbial processes, including sulphate and iron reduction as key activities, produce large amounts of MeHg under optimal conditions encompassing the presence of substrate (dissolved inorganic Hg in pore waters), organic matter and sulphate- (SRB) and iron-reducing (IRB) bacteria (*34,47*). The fine-grained sediments in the central part of the Gulf of Trieste with degradable marine (autochthonous) organic matter and a shallow redoxcline (*38,45*) are examples of areas favouring methylation primarily by SRB (*29*). The methylation rate constant (k_{meth}) increases from the river outflow towards the south (Table 1), except in the southern 'pristine' sediments (lower k_{meth} at F_1; Table 1) where Hg is of prevalently autochthonous marine (less fluvial) origin (*15*). This increase appears mostly related to the benthic sulphur cycling, *i.e.* microbial sulphate reduction and mobilization of bioavailable inorganic Hg in cinnabar by sulphide production, despite the decrease of sulphate reduction rates (*29*). High sulphide concentration can imobilize Hg as HgS that is not readily methylated (*46*), while low sulphide concentration (μM range), like that normally present in pore waters of the Gulf of Trieste (*45*), promotes formation of HgS^0, which is considered to be the predominant chemical species controlling the bioavailability of inorganic Hg for methylation by SRB (*48*). In regions with intermediate sulphide concentrations, charged Hg-S complexes dominate (*3*), which are less available for MeHg production. Amendments of a sulphate reduction inhibitor, *i.e.* molybdate (*49*), confirmed that SRB are important methylators in the sediments of the gulf (and the adjacent Grado and Marano Lagoon) and they are most active in summer and autumn. In winter, IRB can prevail (*49*). Organic matter can also influence the methylation rate (*3*). However, the pore water DOM (*39*) seems to be of lesser importance since in the environment with low sulphide concentration the charged Hg-S complexes, less available for MeHg production, are not expected to out-compete sulphide for Hg (*3*).

Less is known about MeHg demethylation and its role in controlling net MeHg production (*50*). Demethylation rate constants (k_{demeth}, Table 1) decrease from the Isonzo/Soča river mouth towards the central part of the Gulf of Trieste (*8,29*). Comparison between winter and summer MeHg demethylation rates (*8,29*) showed that the oxidized surface sediment layer in winter promotes a higher proportion of the reductive demethylation pathway. This process is catalyzed by the bacterial detoxification system *mer* (*34*), transforming the pore water MeHg to Hg^0 and methane. In other periods, demethylation is mostly oxidative, producing Hg(II), which is again transformed to MeHg, and CO_2 (*8,29*). The amendment of inhibitors, *i.e.* molybdate (*49*), proposed the primary role of SRB as active demethylators in warmer months, but not in winter, suggesting that other bacteria, including methanogens, denitrifiers and Fe-reducers, can act as active demethylators (*51*).

The importance of redox conditions in controlling Hg methylation and Hg and MeHg fluxes at the sediment-water interface was revealed during: (*i*) periods of bottom water anoxia when MeHg fluxes increased as a result of their release during the reduction of iron oxides, especially goethite, and (*ii*) oxic periods when reoxidation lowers MeHg benthic fluxes due to enhanced demethylation and adsorption of Hg and MeHg onto iron oxides (*39,44*). Sediment bioturbation by benthic infauna introduces oxidants (O_2, nitrate, sulphate) into anoxic sediment (*45*) and influences methylation by preventing the formation of high sulphide levels (*52*). The products of benthic Hg transformations, *i.e.* the difference between methylation and demethylation, are subsequently released into the water column by benthic diffusive and advective fluxes (*40*) and resuspension (*3*). They are successively incorporated into the benthic food webs or released to the water column and incorporated into the pelagic food webs (*53*). The experimentally determined Hg methylation and MeHg demethylation rates are largely unknown, due to a lack of knowledge regarding levels of 'bioavailable' Hg species (*29*). However, using Hg pore water data for the calculation of Hg methylation rates and correcting for MeHg demethylation yielded net methylation rates that were of the same order as MeHg benthic fluxes (*29*).

The role of pelagic and benthic phototrophs is mostly in the production of organic compounds (macromolecules), bonding Hg, while bacterial enzymes, mostly of heterotrophs, split into substrates for subsequent microbial and photochemical transformations (*54*). The colloidally immobilized Hg is less available to microbial transformations (*55*), for example Hg methylation in the water column. The produced MeHg may be an important source for bioaccumulation in the phytoplankton and consequently in the whole pelagic food web, but to date is unknown, except in the Mediterranean French Thau lagoon (*56*). Moreover, the pelagic MeHg demethylation, which may control the level of bioavailable MeHg, is also completely unknown. Recently, using a new highly sensitive ^{197}Hg method (*57*), it has been shown that Hg reduction is active in the water column of the Gulf of Trieste, while methylation was not detected (*41*). Seasonally, the reduction seems to be affected more by biological events rather than by the physical structure of the water column. Bacterial activity due to Alpha- and Gammaproteobacteria and Bacteroidetes in the microbial loop (*41,58*) may be involved in this process probably linked to the production and degradation of 'fresh' labile (mostly phytoplanktonic) organic matter limited by the low phosphorus availability in northern Adriatic waters (*32, 59*).

Bioaccumulation in Marine Biota

In the water column, MeHg is transferred to phytoplankton and then to the coastal and, by bioadvection, to the shelf food webs (*5*). It is well documented that the percentage of Hg as MeHg increases along the aquatic food web, averaging about 10 % in the particulate matter, 15 % in phyto- and 30 % in zooplankton, and about 95 % in the muscle of fish on top of the food web (*60*), due to the trophic variability and differences in ecology and metabolism. However, relatively little is known about Hg and MeHg accumulation in marine, as well as in the Mediterranean food webs (*4*). Bernhard (*61*) reported that in a Mediterranean food web composed of seawater→ plankton→pilchard→tuna the percentage of Hg as MeHg increases from 2 % in seawater through 60–90 % in pilchard to 100 % in tuna. Since the Gulf of Trieste is one of the areas most severely polluted by Hg in the Mediterranean and worldwide (*3*), there should be increased bioaccumulation of toxic MeHg in local species. We tried to decode the MeHg bioaccumulation in food webs of the Gulf of Trieste using data from ray species and their prey (*62*), because they are apex predators, like their relatives sharks, and they are also consumed by humans. We also used data on mean MeHg contents in seawater and zooplankton (Table 2; *10,17,52,62–64*). Considering Hg and MeHg in various trophic levels of the Gulf of Trieste, the percentage of MeHg increases along the benthic and pelagic food webs (Table 2). The accumulation of MeHg from unfiltered seawater to particulate matter, including the accumulation in phytoplankton, is low since the majority of total seawater MeHg in the Gulf of Trieste is associated with organic and inorganic particles. The MeHg bioaccumulation between particulate matter and zooplankton is high, amounting to about 10^5.

There is little information on MeHg in marine phytoplankton because of experimental difficulties, especially in coastal waters, associated with discrimination between inorganic particles, detritus and microalgal cells. In phytoplankton, the MeHg content in total Hg exceeds 10 % (*65*) and the bioaccumulation between water and phytoplankton is the greatest (>10^5) of the whole marine food web with rather small interspecies variations (*66–68*). The factors controlling temporal and spatial variations of MeHg bioconcentration in marine phytoplankton are still not completely understood (*3*) and may comprise the chemical speciation of MeHg in relation to seawater chloride and sulphur concentrations and inorganic and organic complexes (*69*) including extracellular polymers (mainly polysaccharides). Recent data indicate the presence of active and passive (and facilitated) Hg and MeHg uptake with bonding onto cell wall in the latter mechanism (*70–72*). The active uptake remains less clear and may include Hg and MeHg bonding onto organic compounds, actively taken by the cell, or they can represent a surrogate for the metal actively absorbed by the cell. Hg and MeHg, bonded especially through thiol groups (*25,26*), in organic complexes control the active and passive uptake mechanisms into microalgal cells (*71*). The complexed metals seem less available to phytoplankton compared to free ions (*73*). Since the quality and quantity of organic compounds, forming complexes with Hg and MeHg, vary temporally and spatially, the availabilty of Hg species for phytoplankton uptake tends to vary (*74*). Once absorbed, MeHg is more penetrating into the cell cytoplasm and complexed with phytochelatins, metallothioneins and other chelators (*75,76*), resulting in a higher concentration (biomagnification) in the food web compared to Hg. Hg and MeHg complexed by dissolved organic matter (DOM), including microbial exopolymers, lower their availability for phytoplankton uptake by imobilization (*24*). Since the quality and quantity of DOM is temporally changing by degradation, the biological availability for phytoplankton use also changes. This transfer also occurs in the benthos where benthic (micro)algae absorb MeHg from sediment pore waters and solid phase, which is then available to the benthic fauna and benthic food web. Little is known regarding the Hg and MeHg bioaccumulation in benthic microalgae, where sampling and experimental approach are more difficult (*77*). The role of benthic micro-, mostly diatoms, and to lesser extent macroalgae and macrophytes in the Gulf of Trieste can be important (*77*) because the euphotic zone extends to the seabed (*36*).

The Hg and MeHg bioconcentrated in the first trophic level are subsequently transferred into the pelagic and benthic food webs (*4*). MeHg biomagnification between seawater and higher trophic levels of the Gulf of Trieste (*62*) amounted to 10^6 in small pelagic fish (anchovy) and molluscs (squid), and to 10^7 in the muscles of larger pelagic predators (pelagic stingray) and is at the higher limit of reported biomagnification between water and the muscle of predatory fishes (*3*). MeHg bioaccumulation between surface sediment, including benthic microalgae, and benthic invertebrates (murex) was 10^2, while that between small fish (red bandfish) and the muscles of benthic apex predators (bull ray, eagle ray, common stingray) was lower, ranging from 0.1 to 60 (*62*). The trophic transfer ultimately resulted in a 10^3 biomagnification between the sediment and the muscles of benthic rays. MeHg contents were positively correlated with length, mass and age of grey mullet and all ray species (*53,62*), but in the bull ray the slower MeHg increase was independent of Hg increase probably due to demethylation (*62*) involved in detoxification mechanism. The relative importance of these two bioaccumulation pathways suggests greater accumulation of MeHg by pelagic feeding species, suggesting that MeHg benthic fluxes and diffusion in the water column could be more important than biotransfer mechanisms (*5*).

Human Exposure

The limit of Hg content in wet mass of seafood in the EU is 0.5 μg/g, while that in the fast accumulating species listed in the Directive, encompassing those located at the highest trophic levels (predators) and in the benthos is 1 μg/g (*78*). It is to note that high Hg and MeHg levels and high Hg methylation rate constants in sediments of the gulf are not generally reflected in fish and other biota living in this area (Table 2), compared with those from the open Adriatic Sea (*79*). The reasons may include high MeHg demethylation (*29*) and Hg reduction (*57*) potentials observed in the gulf. On the other hand, many specimens of bull ray and pelagic stingray exceed the EU guideline values for commercial use and human health (*62*). If we consider that all studied ray species are fast accumulating species, the number of specimens exceeding the safe Hg and MeHg levels for benthic fish and predators might be somewhat lower. Considering the weekly intake tolerance for humans of 1.3 μg of MeHg per kg of body mass (*80*), a portion of 100 g per week for many specimens of apex predators would be exceeded.

Table 2. Biomagnification of methylmercury (MeHg) in pelagic and benthic food webs in the Gulf of Trieste

	w(Hg)*/(μg/kg)	(m(MeHg)/m(THg))/%	Ref.
Pelagos			
Seawater (dissolved)	BDL-0.005	<0.02	(*17*)
Seawater (particulate)	BDL-0.011	0.6	(*17*)
Zooplankton (>200 μm)	130–370	6–17	(*17*)
Grey mullet (*Liza aurata*)	114	57	(*52*)
Anchovy (*Engraulis encrosicholus*)	159	38–71	(*62*)
Pelagic stingray (muscle) (*Dasyatis violacea*)	499–1285	100	(*62*)
Squid (*Loligo vulgaris*)	372	100	(*63*)
Benthos			
Sediment	6.7–80	0.3–1	(*10*)
Posidonia (*Posidonia oceanica*)		29	(*64*)
Gastropod (*Murex tranculus*)	276	52	(*62*)
Mussel (*Mytilus galloprovincialis)*	18–51		(*62*)
Red bandfish (*Cepola rubescens*)	39	57–77	(*62*)
Common pandora (*Pagellus erythrinus*)	297	77	(*52*)
European conger (*Conger conger*)	671	75	(*52*)
Bull ray (muscle) (*Pteronylaeus bovinus*)	61–1740	90–100	(*62*)
Common stingray (muscle) (*Dasyatis pastinaca*)	400	90–100	(*62*)
Eagle ray (muscle) (*Myliobatis aquila*)	50–183	90–100	(*62*)

*values are expressed on wet mass basis
THg=total Hg; BDL=below detection limit

Conclusions and Future Research Directions

Available data show that the processes controlling microbial production and degradation of toxic MeHg in the historically Hg-contaminated Gulf of Trieste are localized in the surface anoxic sediments and strongly influenced by biogeochemical conditions, especially those linked to S and Fe(Mn) cycling. Organic matter seems of lesser importance except for its role in fuelling the microbial metabolism involved in Hg cycling. The MeHg sedimentary content, decreasing from the riverine Hg pollution source, is a function of benthic flux and both Hg methylation and MeHg demethylation, the latter representing a natural bioremediation process. The Hg sedimentary transformation pathways are linked to the composition of microbial communities: SRB predominate over IRB in Hg methylation and MeHg demethylation, except in winter when IRB can dominate methylation and a variety of bacterial species can be involved in demethylation including aerobes, SRB, IRB and nitrate reducers. Conversely, in the pelagic system (water column), Hg cycling is poorly understood. There it appears that Hg(II) reduction (microbial and photochemical) is the main process making the Gulf of Trieste a net source of dissolved gaseous Hg (DGM) in the atmosphere. Subsequent evasion of Hg^0, also understudied, reduces the potentially bioavailable pool for MeHg production. Pelagic Hg(II) reduction seems connected with the microbial loop and controlled by labile organic matter. In this context, MeHg demethylation and Hg reduction studies especially should be pursued in the near-shore water column.

All sources and forms of Hg contribute to its accumulation in benthic and pelagic food webs with greater accumulation of MeHg in pelagic feeding species, suggesting that its benthic flux and water column advection should be more important than the biotransfer mechanism. However, it seems likely that efficient sedimentary MeHg demethylation and water column Hg(II) reduction are important factors preventing extensive contamination of marine biota although some MeHg fish levels exceed the limits. There is surprisingly little information on MeHg in edible marine species from the gulf and in this context more future studies should address this issue. Comprehensive and systematic investigations of MeHg bioaccumulation and biomagnification in lower food web organisms, *i.e* phytoplankton and fractionated zooplankton, should be performed in the future since the initial bioconcentration of MeHg by phytoplankton is thought to represent the greatest single contribution to bioaccumulation in the marine food web. The role of dissolved organic compounds, including their formation and degradation, on Hg and MeHg uptake in phytoplankton is of primary importance. Finally, all outcomes should be included in the annotated Hg biogeochemical (mass balance) models of coastal waters.

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