Ecotoxicology
October 2023, Volume 32 Issue 8 Pages 994-1009
https://doi.org/10.1007/s10646-023-02679-y
https://archimer.ifremer.fr/doc/00842/95422/

Are tunas relevant bioindicators of mercury concentrations in the global ocean?

Médieu Anaïs [1, *], Lorrain Anne [1], Point David [2]

[1] IRD, Univ Brest, CNRS, Ifremer, UMR 6539, LEMAR, Plouzané, France
[2] Observatoire Midi-Pyrénées, GET, UMR CNRS 5563/IRD 234, Université Paul Sabatier Toulouse 3, Toulouse, France

* Corresponding author : Anaïs Médieu, email address : anais.medieu@gmail.com

Abstract :

Humans are exposed to toxic methylmercury mainly by consuming marine fish. The Minamata Convention aims at reducing anthropogenic mercury releases to protect human and ecosystem health, employing monitoring programs to meet its objectives. Tunas are suspected to be sentinels of mercury exposure in the ocean, though not evidenced yet. Here, we conducted a literature review of mercury concentrations in tropical tunas (bigeye, yellowfin, and skipjack) and albacore, the four most exploited tunas worldwide. Strong spatial patterns of tuna mercury concentrations were shown, mainly explained by fish size, and methylmercury bioavailability in marine food web, suggesting that tunas reflect spatial trends of mercury exposure in their ecosystem. The few mercury long-term trends in tunas were contrasted and sometimes disconnected to estimated regional changes in atmospheric emissions and deposition, highlighting potential confounding effects of legacy mercury, and complex reactions governing the fate of mercury in the ocean. Inter-species differences of tuna mercury concentrations associated with their distinct ecology suggest that tropical tunas and albacore could be used complementarily to assess the vertical and horizontal variability of methylmercury in the ocean. Overall, this review elevates tunas as relevant bioindicators for the Minamata Convention, and calls for large-scale and continuous mercury measurements within the international community. We provide guidelines for tuna sample collection, preparation, analyses and data standardization with recommended transdisciplinary approaches to explore tuna mercury content in parallel with observation abiotic data, and biogeochemical model outputs. Such global and transdisciplinary biomonitoring is essential to explore the complex mechanisms of the marine methylmercury cycle.

Keywords : Methylmercury, Minamata Convention, Biomonitoring, Spatial gradients, Temporal trends, Standardized monitoring guidelines

1. Introduction

Mercury (Hg) is a potent neurotoxin that poses health risks to the global population. It is emitted into the environment from natural sources, but also originates from multiple anthropogenic sources including atmospheric emissions and direct releases to land and water (Outridge et al., 2018). Once released, Hg transport and fate is complex and influenced by chemical, physical, and biological processes. Once deposited or taken up in the ocean, an expected substantial but yet not well defined fraction of Hg is naturally transformed into the more toxic and bioavailable methylmercury (MeHg) form (Bowman et al., 2020; Mason and Fitzgerald, 1990), which is easily absorbed by organisms and can biomagnify along marine food webs. This compound also bioaccumulates within individuals over time, resulting in higher MeHg concentrations in bigger fish. Humans are exposed to MeHg mainly by their consumption of seafood (Sunderland et al., 2018), especially top predators like some tunas that exhibit high MeHg concentrations relative to the rest of the food web (e.g., Bodin et al., 2017; Choy et al., 2009; Houssard et al., 2019). The adverse effects of MeHg exposure on human and wildlife health are well documented (Axelrad et al., 2007; Genchi et al., 2017), and MeHg health and socioeconomic costs are estimated to be a billion dollars per year worldwide (Bellanger et al., 2013; Zhang et al., 2021).

Mercury is easily transported and deposited across national boundaries (Horowitz et al., 2017), and thus requires international cooperation for its control. The United Nation Minamata Convention on Mercury is a legally binding international agreement (ratified by 141 countries in June 2023, www.mercuryconvention.org) with the objective of protecting human health and the environment from anthropogenic Hg emissions and releases. This implies a mix of measures to control, reduce, and eliminate major sources of Hg, but also an assessment of the resulting changes in emissions and releases, and ultimately in human and ecosystem exposures. Given the complexity of the global Hg cycle, such assessment of Hg changes in the environment requires a collection of metrics that complement each other and capture different spatial and temporal dimensions of effectiveness.

In the atmosphere, a few long-term monitoring networks have been measuring air Hg concentrations and deposition in contrasted regions of the world since the 1990s (Slemr et al., 2020, 2015; Zhang et al., 2016). Anthropogenic Hg emissions are thought to have tripled total Hg concentrations in thermocline waters (100 – 1,000 m) of the global ocean relative to deeper older waters (Lamborg et al., 2014), yet still unclear is how these anthropogenic Hg inputs are converted into MeHg in oceans. Further developments of clean sampling protocols, and lower detection limits, coupled to the establishment of global-scale oceanographic survey programs (i.e., CLIVAR and GEOTRACES) in the past two decades, have led to more than 200 high-resolution and full depth profiles of Hg speciation covering the Arctic, Atlantic, Pacific, and Southern Ocean (Bowman et al., 2020). Although these measurements in the open ocean have improved our understanding of the biogeochemical Hg cycle in the global ocean, they remain too scarce and limited in space and time to evaluate the effectiveness of the Minamata Convention. To complement this abiotic monitoring, bioindicators are commonly used for assessing environmental loads and associated ecological and human health impacts resulting from controls on point sources.

An exemplary long-term biotic monitoring effort is being conducted by the Arctic Council's Arctic Monitoring and Assessment Program (AMAP), and involves Hg measurements in fishes, birds, marine mammals, and people to complement Hg observations in air and seawater (AMAP, 2021). Atmospheric Hg levels in the Arctic are generally decreasing, while both increasing and decreasing trends of Hg in Arctic biota have been observed over the last decades. These contrasted trends likely

result from concomitant reducing Hg emissions from North America and Europe, and/or climate-induced confounding factors (e.g., thawing permafrost, melting glaciers, and changes to vegetation cover) altering Hg uptake and re-emission (Jiskra et al., 2020), and MeHg production/degradation and biomagnification in upper predators (Masbou et al., 2015; Point et al., 2011; Wang et al., 2019). In the meantime, there are very few long-term Hg biomonitoring efforts in tropical regions, while populations from these regions are often considered as vulnerable regarding Hg exposure given that seafood represents their primary source of animal proteins (Bell et al., 2009; Béné et al., 2015). In tropical marine ecosystems, tunas have been identified as potential human and ecological health bioindicators (UN Environment, 2019). There is indeed a consensus that tunas can pose concern for human exposure to MeHg as they are one of the most important global sources of seafood (FAO, 2018), while MeHg represents the major chemical form of total Hg in muscle (> 90 %) (Houssard et al., 2019; Mergler et al., 2007). There is yet no evidence that tunas are direct relevant bioindicators species of Hg concentrations in the ocean, i.e., that they are able to reflect Hg exposure in their ecosystem, integrating local and global Hg source mixing and oceanic transformation into MeHg over different spatial and temporal scales.

This article reviews information from the literature to investigate the relevance of tunas as Hg bioindicators in the ocean for the Minamata Convention, focusing on muscle, the recommended matrice to biomonitor MeHg exposure in fishes with a high percent MeHg like tunas (UN Environment, 2019). While the term "tunas" refers to 15 different ocean fish species, including seven of commercial importance, this review focuses only on the four worldwide-distributed species to ensure comparability of Hg exposure and accumulation at both regional and global scales. This includes the three tropical tunas, i.e., bigeye (*Thunnus obesus*), yellowfin (*T. albacares*), and skipjack (*Katsuwonus pelamis*), and albacore (*T. alalunga*, a temperate and tropical species). Although classified as highly migratory, we hypothesize that tropical tunas, and to a lesser extent albacore, are suitable to explore high resolution subregional patterns of marine MeHg bioavailability and accumulation as these species are suspected to display relative site fidelity (Fonteneau and Hallier, 2015; Houssard et al., 2017) contrary to Pacific (*T. orientalis*), Atlantic (*T. thynnus*) and Southern (*T. maccoyii*) bluefin tunas that undergo large transoceanic migrations (Block et al., 2005; Hobday et al., 2015; Madigan et al., 2014). Tropical tunas and albacore are also widely exploited (representing 99 % of the global tuna catches) (ISSF, 2021), and monitored by regional fishery management organizations through several sampling programs. Given this context, these four species represent a possible gold mine of biological samples to generate robust and comparable datasets, a mandatory requirement to get accurate and sensitive temporal and spatial Hg biomonitoring studies in the global ocean. While focussing on these four tuna species, some results on bluefin tunas are also presented as they bring valuable information, not available for tropical species nor albacore, to discuss the relevance of tunas as bioindicators of Hg in the ocean.

2. State of mercury data in tunas

Since the 1970s, more than 50 studies have documented Hg concentrations in tropical tunas and albacore. These studies were conducted mainly in the Pacific (n = 26) and in the Atlantic Oceans (n = 20), and to a lesser extent in the Indian Ocean (n = 10) (Figure 1A; SI Appendix Table S1). Only one study has documented Hg concentrations for the global ocean for yellowfin (Nicklisch et al., 2017). Yellowfin is indeed the most studied tuna species regarding its Hg concentrations (number of

individuals with published Hg concentration, *n* = 3,423), followed by bigeye (*n* = 1,674), albacore (*n* = 1,435), and skipjack (*n* = 1,420) (Figure 1B). Mercury levels were found to vary among tuna species, but also in space and time, as discussed in the three following sections, respectively, regarding the expected objectives of Hg biomonitoring in the open ocean.

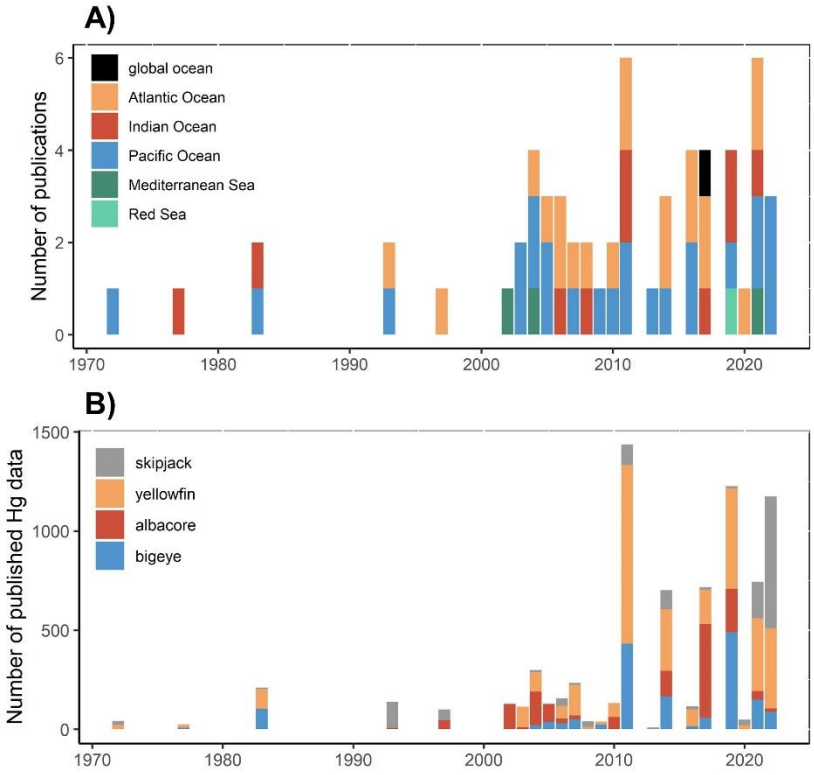

Figure 1. Review of published mercury data in tunas through time. A) Number of publications per year and ocean documenting mercury (Hg) concentrations in muscle tissues of tropical tunas and albacore. **B)** Number of Hg concentration data reported in publications per year and tuna species. References and detailed information used to produce these histograms are reported in SI Appendix Table S1.

3. Inter-species differences in tuna mercury concentrations

Mercury concentrations have been shown to differ among tuna species (Figure 2). While bigeye, albacore, and yellowfin can display Hg concentrations exceeding the food safety guidelines of 1 µg.g^{-1} wet weight (FAO Codex Alimentarius Commission, 2021), skipjack samples always show Hg levels below the limit (Figure 2; SI Appendix Table S1). Samples with Hg concentrations exceeding the food safety guidelines are generally associated to bigger individuals, illustrating the natural bioaccumulation of Hg in organisms through time, and the consequent need to consider both tuna species and fish size when addressing recommendations in terms of food safety regarding Hg content.

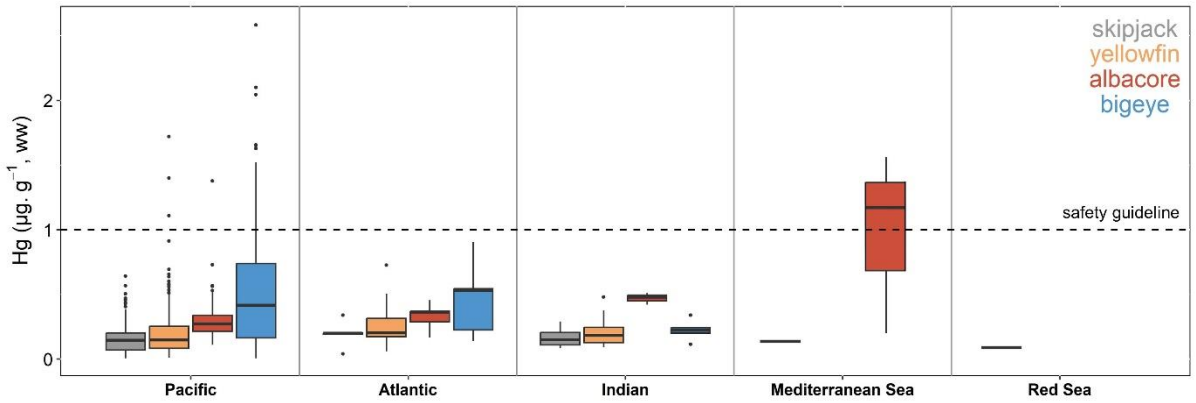

Figure 2. Gradient of mercury concentrations in tropical tunas and albacore. Boxplots illustrate the inter-species variability of mean total mercury (Hg) concentrations (µg.g^{-1}, wet weight) measured in white muscle tissue of skipjack (grey), yellowfin (orange), albacore (red), and bigeye (blue) caught in the Pacific, Atlantic, and Indian Oceans, and in the Mediterranean Sea and the Red Sea. The dashed line represents the food safety guideline of 1 µg.g^{-1} wet weight. Thick bars are the median values, points are outliers, and the boxes contain 50% of the data. References and detailed information used to produce these boxplots are reported in SI Appendix Table S1.

Relative differences of Hg concentrations among tuna species are consistent among study regions of the global ocean, following this general pattern in Hg concentrations: bigeye > albacore > yellowfin and skipjack (Blum et al., 2013; Bodin et al., 2017; Choy et al., 2009; Garcia et al., 2007; Houssard et al., 2019; Médieu et al., 2021a, 2021b; Yamashita et al., 2005). The highest Hg concentrations in bigeye compared to the three other species is presumably the result of three confounding factors: i) a higher trophic position of this species, ii) a deeper vertical habitat facilitating its access to mesopelagic prey with enhanced Hg concentrations, and iii) a longer lifespan (Choy et al., 2009; Ferriss and Essington, 2011; Houssard et al., 2019). Tropical tunas and albacore indeed display distinct growth rates (i.e., skipjack > yellowfin > albacore > bigeye), lifespan (i.e., skipjack < yellowfin < albacore < bigeye), vertical habitats (bigeye generally dive deeper than the epipelagic skipjack and yellowfin), and diet preferences, which may also vary among ocean basins (Murua et al., 2017; Olson et al., 2016; Pethybridge et al., 2018). Further investigation at the global scale remains needed to disentangle the relative importance of these ecological and biological processes, alongside the natural marine biogeochemical functioning, to explain the inter-species differences of tuna Hg concentrations. Yet, these preliminary results shed light on the potential power of combining these four tuna species to investigate both horizontal and vertical variability of MeHg bioavailability in the water column, and MeHg biomagnification pathways along pelagic food webs.

4. Spatial variability of mercury concentrations in tunas

Within a same tuna species, Hg concentrations also vary among ocean regions (Figure 3). Despite the scientific interest and the social and economic needs to document Hg variability in the global ocean, the vast majority of studies available to date have been conducted on a small spatial scale, relying most of the time on imprecise tuna catch locations (Figure 4A). Since the 2010s, regional collaborations and exchange of tuna Hg data associated to precise tuna catches enabled highlighting regional differences in Hg concentrations in yellowfin and bigeye tunas from the eastern Pacific Ocean (Figure 4C) (Ferriss and Essington, 2011), and in albacore from the western Indian Ocean (Chouvelon et al., 2017). In the global ocean, an eightfold difference in Hg concentrations was documented in yellowfin samples among 12 different locations (Nicklisch et al., 2017). Here, our compilation of all Hg data in tropical tunas and albacore from the global ocean reveals that mean observed Hg concentrations vary across sites by a factor 64 (0.01 – 0.64 $\mu g.g^{-1}$, wet weight, min-max), 172 (0.01 – 1.72 $\mu g.g^{-1}$), 14 (0.11 – 1.56 $\mu g.g^{-1}$), and 258 (0.01 – 2.58 $\mu g.g^{-1}$) in skipjack, yellowfin, albacore, and bigeye, respectively (Figure 3). These species-specific and global-scale compilations confirm that Hg concentrations are highly variable among species and sites. They also highlight that the vast majority of the studies documenting Hg concentrations in tunas rely on spot samples acquired at low spatial resolution, which do not allow exploring regional spatial patterns of Hg exposure and accumulation in pelagic biota. Recently, international collaborations and access to tuna samples from extensive tuna tissue banks have enabled to produce broad-scale and high-resolution maps of Hg concentrations in bigeye, yellowfin, and albacore from the western central Pacific region (Houssard et al., 2019) (Figure 4E), and in skipjack for the entire Pacific Ocean (Médieu et al., 2022). Such maps allow refining the assessments of risk exposure to Hg associated with human consumption according to tuna species and tuna catch areas.

In most of the low resolution spatial studies, fish body size or weight has been identified as the best predictor of intra-species variability of tuna Hg concentrations, illustrating the natural bioaccumulation property of Hg in individuals over time (Figure 4B) (e.g., Besada, 2006; Boush and Thieleke, 1983; Cai et al., 2007; Chen et al., 2014, 2011; Kojadinovic et al., 2007; Peterson et al., 1973; Rivers et al., 1972; Sompongchaiyakul et al., 2008; Teffer et al., 2014). Yet, this body size/weight effect is not often considered when comparing tuna Hg concentrations at regional scales. For instance, between Hawaii and the North East Pacific Ocean, Nicklisch et al. (2017) reported a 4-fold increase of Hg concentrations in yellowfin (mean total Hg = 0.602, and 0.154 $\mu g.g^{-1}$, ww, respectively), but a two-fold difference of size (mean standard length = 117 and 56 cm, respectively). The use of quantitative methods (e.g., mixed effects in multiple regression models) can help account for the body size effect and quantify its relative importance compared to regional effects when exploring the regional variability of tuna Hg concentrations. In the eastern Pacific Ocean, such an approach revealed higher Hg concentrations in bigeye in the eastern equatorial region than in Hawaii, central- and mid-equatorial Pacific, not explained by fish size differences (Figure 4D) (Ferriss and Essington, 2011). Recently, length-standardization methods of tuna Hg concentrations have been developed to remove the bias associated to fish size differences among individuals, and to characterize the natural bioaccumulative properties of Hg in tunas when exploring Hg spatial patterns (see methodological details in section 6.2.) (Houssard et al., 2019; Médieu et al., 2022). This method revealed that differences in tuna sizes explained high observed Hg concentrations in bigeye in the equatorial western Pacific, but not in the southwestern region where standardized Hg concentrations remained highest than in any other region (Figure 4E & F) (Houssard et al., 2019). In the literature, individual fish length

is rarely reported alongside individual tuna Hg concentration, which prevents us from standardizing most of the published Hg concentrations with this method, and producing compilation maps of standardized Hg concentrations in the four tuna species. Contrary to a simple comparison of observed Hg concentrations measured in different locations (Figure 3), mapping standardized Hg concentrations at a high resolution and broad spatial scale allows exploring the spatial distribution of Hg concentrations anomalies not explained by Hg bioaccumulation and fish sizes among individuals, but likely due instead to other processes such as tuna trophic ecology, ocean biogeochemistry or anthropogenic Hg emissions.

Identifying and evaluating the relative importance of these underlying processes, other that body size effect, to explain spatial patterns of tuna Hg levels can be complicated given the difficulty with quantifying these processes, and disentangling possible interplay among them. Tuna foraging depth is considered as a key driver to explain higher Hg concentrations in mesopelagic species like bigeye (Choy et al., 2009); yet still unclear is the relative importance of both food chain length and/or baseline Hg variations as possible causal effects (Ferriss and Essington, 2011). The recent exploration of standardized Hg concentrations in tunas alongside ecological data (e.g., tuna trophic position estimates), biogeochemical (e.g., dissolved O_2 concentrations), physical (e.g., thermocline depth), and anthropogenic (e.g., atmospheric elemental Hg concentrations) model outputs through quantitative models (e.g., generalized additive models) offers new perspectives to disentangle the relative importance of ecological, biogeochemical and anthropogenic processes. In the western central Pacific Ocean, this transdisciplinary approach revealed that spatial variability of standardized Hg concentrations in bigeye, yellowfin, and albacore were mainly related to tuna foraging depth and local ocean biogeochemistry, and that spatial trophic changes were of minor importance (Houssard et al., 2019). Similarly, large spatial patterns of standardized Hg concentrations in the epipelagic skipjack were mainly explained by the natural functioning of the Pacific Ocean, likely via variations in the MeHg concentrations and the depth at which MeHg peaks within the water column (Médieu et al., 2022). In this latter study, authors also revealed the local influence of anthropogenic emissions in Asia, enhancing skipjack Hg concentrations in the northwestern Pacific Ocean. Overall, these two broad-scale and transdisciplinary studies lead to the hypothesis that the spatial variability in tuna Hg concentrations is mainly explained by both tuna foraging depth and the vertical and horizontal variability of MeHg bioavailability in the ocean. These results shed light on the potential power of combining tropical tunas and albacore to investigate both horizontal and vertical variability of MeHg bioavailability in the water column, and MeHg biomagnification pathways along pelagic food webs. These findings call for deeper global comparisons with observed Hg data in the ocean.

Such comparisons are rarely achievable because of the mismatch in space and time between available tuna Hg data and scarce observations of Hg concentrations and speciation in the ocean. In the southwestern Pacific Ocean, Hg concentrations in tropical tunas and albacore were recently shown to mirror ambient seawater MeHg concentrations, with higher Hg concentrations in the mesopelagic bigeye reflecting higher dissolved MeHg concentrations in subsurface waters (Barbosa et al., 2022). At the global scale, Hg concentrations in bluefin tunas were also found to reflect global patterns of Hg bioavailability and pollution in each ocean basins (Tseng et al., 2021). Together, these results exploring Hg concentrations in tunas in parallel with global and high-resolution model outputs, or with dissolved MeHg observations, confirm that tunas tend to reflect global and regional changes of Hg exposure in their ecosystem. In the framework of the Minamata Convention, such coupled approaches could benefit the general understanding of Hg accumulation in tunas in the global ocean, and could therefore

offer perspective in modelling responses of Hg concentrations in marine top predators to changes in oceanic MeHg concentrations in the context of different emissions scenarios.

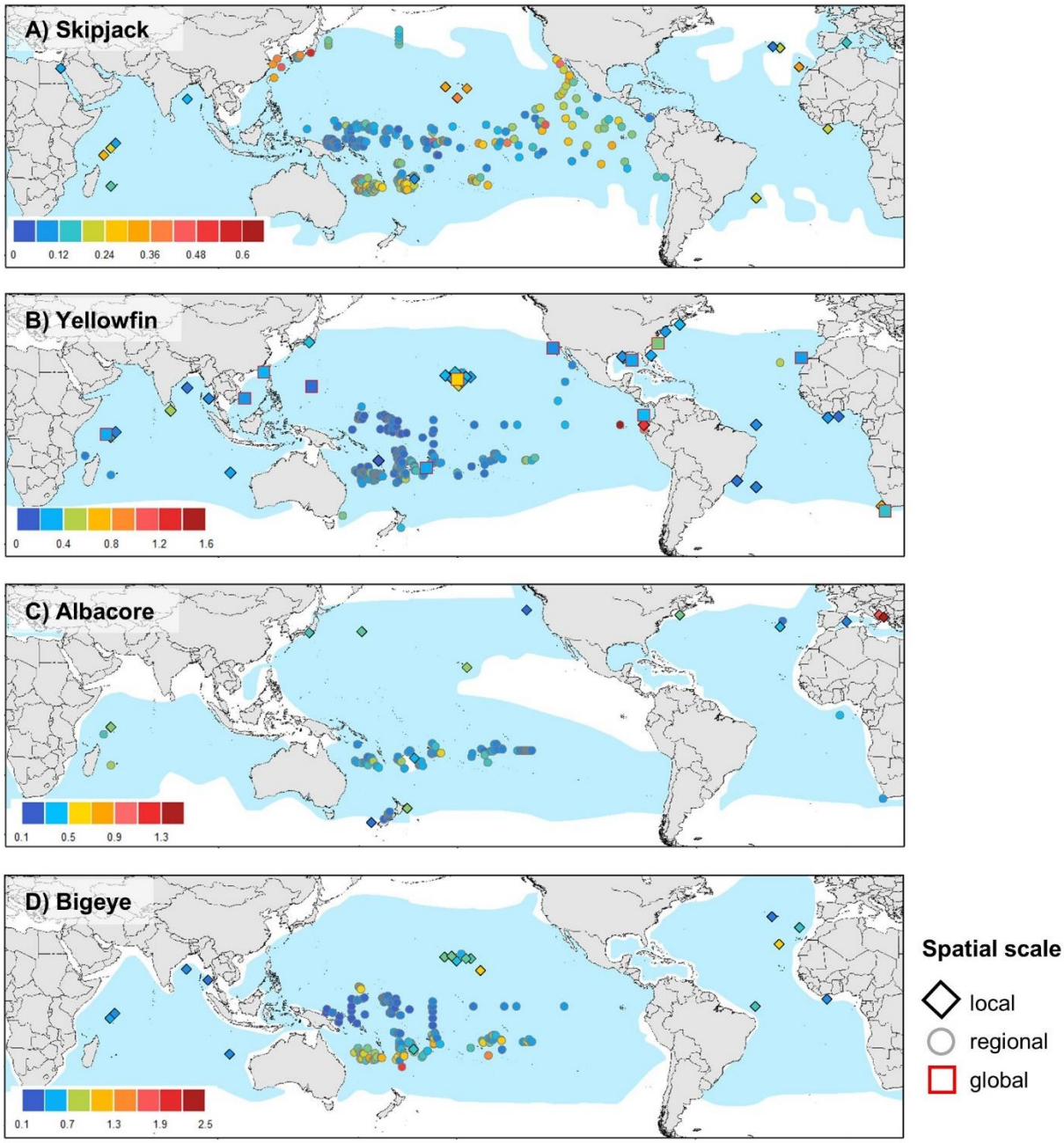

Figure 3. Review of mercury concentrations in tropical tunas and albacore in the global ocean. Spatial variability of total mercury (Hg) concentrations (µg.g^{-1}, wet weight) in white muscle tissue (coloured symbols) and potential habitat distribution (light blue) of **A)** skipjack, **B)** yellowfin, **C)** albacore, and **D)** bigeye at the global scale. The colour of symbols refers to total Hg concentrations measured in tuna muscle samples. The shape of symbols refer to the spatial scale of the studies from which tuna Hg data originate: the 12 squares represent the 12 samples locations used in the unique global study (Nicklisch et al., 2017), the circles are the locations used in regional studies (Chen et al., 2011; Chouvelon et al., 2017; Endo et al., 2016; Ferriss and Essington, 2011; Garcia-Vazquez et al., 2021; Houssard et al., 2019; Kojadinovic et al., 2006; Médieu et al., 2022; Ordiano-Flores et al., 2011; Yamashita et al., 2005), and the diamonds represent locations where tuna Hg concentrations are reported in local studies. Potential habitat distributions per species were obtained from the IUCN Red List Spatial Data of tunas (IUCN, 2017). References and detailed information used to produce these maps are reported in SI Appendix Table S1.

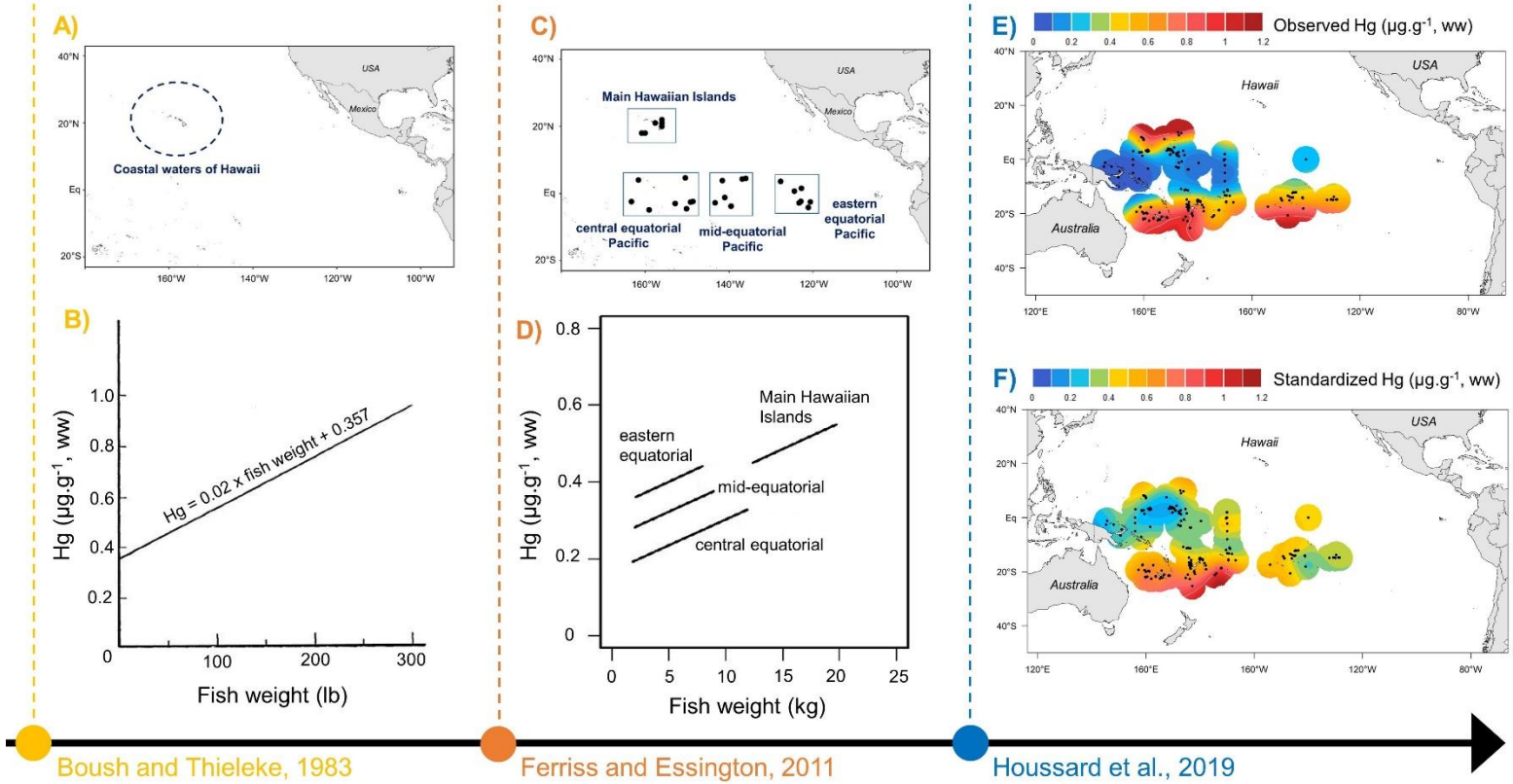

Figure 4. Temporal evolution of the research effort to document mercury accumulation in tunas. Different sampling strategies and standardization methods to investigate mercury (Hg) concentrations in bigeye in the Pacific Ocean. In the 1980s, **A)** bigeye samples were reported at a rough spatial resolution (i.e., coastal waters of Hawaii symbolised by the dashed circle), and **B)** Hg concentrations (µg.g^{-1}, wet weight) in bigeye were found to increase linearly with individual fish weight illustrating the natural bioaccumulation of Hg through time, adapted from Boush and Thieleke (1983). In the beginning of 2010s, **C)** precise catch locations were provided for each bigeye samples (black dots), allowing to investigate **D)** differences of Hg bioaccumulation patterns (black lines) in bigeye among four regions of the eastern Pacific Ocean, adapted from Ferriss and Essington (2011). In the late 2010s, a large and high-resolution compilation of Hg data in bigeye samples (black dots) allowed to map **E)** large spatial differences of bigeye Hg concentrations (µg.g^{-1}, wet weight) in the western central Pacific Ocean. Moreover, robust standardization methods of Hg concentrations to individual fish length helped removing bias associated to different individual fish sizes and revealing **F)** large spatial differences of standardized Hg concentrations (at 100 cm fork length, µg.g^{-1}, wet weight) in bigeye in the western central Pacific Ocean, not explained by natural Hg bioaccumulation through time, but rather by ecological and/or biogeochemical processes, adapted from Houssard et al., (2019).

5. Temporal variability of mercury concentrations in tunas

Another key aspect when evaluating the relevance of tunas as bioindicators for the Minamata Convention is their ability to reflect temporal changes of Hg trends and inputs to the environment at both local and global scales. Only four temporal studies of Hg concentrations are available to date in tropical tunas, plus one in Atlantic bluefin, all showing distinct results regarding Hg long-term trends. In the central north Pacific Ocean (i.e., Hawaii), stable Hg concentrations were found in yellowfin and bigeye between the 1970s and the 1990s (Kraepiel et al., 2003). A re-evaluation of these stable trends in the light of new data showed increasing Hg concentrations in yellowfin (+ 5.5 ± 1.6 %/year during 1998-2008), and in bigeye (+ 3.9 ± 2.1 %/year during 2002-2008), which were suggested to result from an increasing transport and deposition of anthropogenic Hg, as well as the lateral flow of water masses from the north west Pacific (Drevnick et al., 2015; Drevnick and Brooks, 2017). Conversely, Lee et al. (2016) reported an annual decrease of Hg content of 0.018 ± 0.003 % between 2004 and 2012 in Atlantic bluefin from the northwestern Atlantic Ocean, which was assumed to reflect a reduction in North American Hg emissions. Unfortunately, no complementary trophic ecology data were available in these studies to discuss if confounding temporal changes in tuna diet could contribute to the contrasted temporal trends of Hg content, and potentially biased the response of tuna Hg burden to changes of anthropogenic releases to the environment. In the southwestern Pacific (i.e., New Caledonia and Fiji), the evaluation of Hg concentrations in the three tropical tunas between 2001 and 2018, alongside ecological data (i.e., nitrogen and carbon stable isotopes), revealed inter-annual variability but no significant long-term trends of both tuna diet and tuna Hg concentrations (Médieu et al., 2021). The absence of temporal trends in tuna Hg concentrations in the southwestern Pacific contrasts with the other available temporal studies in tunas from the northern hemisphere (Drevnick et al., 2015; Drevnick and Brooks, 2017; Lee et al., 2016), and the increasing trend of Hg emissions reported for the southern hemisphere since the 1980s (Streets et al., 2017). Conversely, it is in accordance with low and stable anthropogenic Hg emissions into the atmosphere from Oceania (Streets et al., 2019). Such disconnect between estimated regional variations in emissions and deposition and stable Hg concentrations in biota has been documented in other marine and freshwater ecosystems (Wang et al., 2019), and likely result from two main confounding factors: i) the large quantities of legacy Hg that remain available for bioaccumulation, and ii) changes occurring in multi-causal, local, and regional processes (e.g., climate) that control the speciation, bioavailability, and bioaccumulation of both current and legacy emitted Hg.

Differences of temporal trends in tuna Hg concentrations could also result from the operational difficulty of getting robust and continuous datasets over long periods of time to determine sensitive and accurate Hg time series. In the framework of AMAP, several metrics of performance have been developed to describe and discuss the statistical power of Hg time series, and ensure statistically powerful Hg biomonitoring through time in the Arctic (AMAP, 2021). Among them, the “adequacy” has been developed to compare the time series ability to detect trends and at the same time indicate whether trend detection is justified for the time series (Bignert et al., 2004). It is defined as the number of actual monitoring years in a time series divided by the number of years of sampling required to detect a 5 % annual change in Hg concentrations. A ratio ≥ 1 implies that the time series are adequate. Here we calculated the adequacy for each of the published Hg time series in tropical tunas and reveal that only two of the five were statistically powerful (Figure 5). This result highlights the urgent need for long and continuous monitoring programs of Hg concentrations in tunas at these two locations, as

well as in other regions of the global ocean to explore the potential time lags between potential changes in Hg emissions to the atmosphere and MeHg levels in marine food webs.

Overall, global Hg temporal data in tunas remain too limited in time (i.e., short and/or discontinuous time series) and space (i.e., only two sites) to be able to state whether or not tunas are capable of reflecting the temporal variability of Hg exposure in their ecosystem, at both regional and global scales. Nevertheless, preliminary results, which contrast among ocean regions, and in particular between hemispheres, highlight distinct regional ocean patterns of Hg anthropogenic inputs and deposition, and suggest different responses to changes in emissions given the complex biogeochemical processes governing the speciation, bioavailability, and biomagnification of Hg in the ocean.

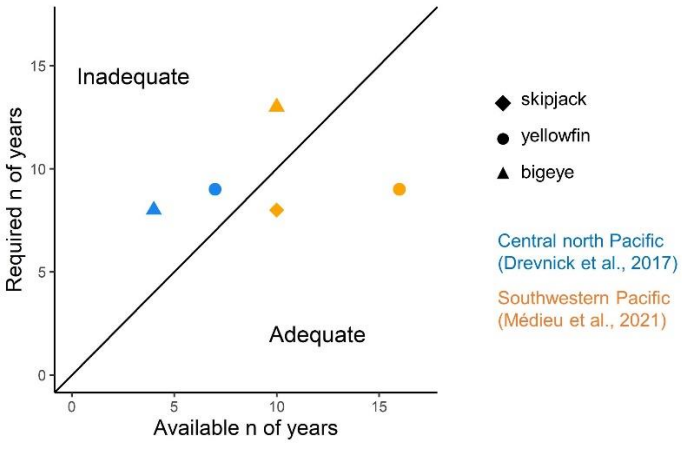

Figure 5. Statistical power of published time series of mercury concentrations in tunas in the global ocean. Adequacy values of mercury (Hg) time series data in skipjack (diamonds), yellowfin (dots), and bigeye (triangles) in the central north (blue, from Drevnick and Brooks, 2017) and the southwestern (orange, from Médieu et al., 2021a) Pacific Ocean. Adequacy was calculated following the methods of Bignert et al. (2004), considering a significance level of $p < 0.05$ and 80 % statistical power. Datasets falling within the lower right portion of the graph are more than adequate to detect a 5 % annual change in Hg concentrations, while those in the upper left portion are inadequate.

6. Guidelines for collecting and reporting relevant monitoring information on mercury in tunas

6.1. Data collection, reporting and organization

The use of tuna Hg data to evaluate the effectiveness of the Minamata Convention at reducing Hg emissions requires worldwide collaboration and coordination, framed around a scientifically sound strategy, based on transparent processes, harmonized methodologies, with reliable and comparable datasets. Among all studies documenting Hg concentrations in tunas, less than 13 % (7 over 56) provide all the necessary information for a relevant Hg biomonitoring. Below, we therefore provide recommendations and guidelines for collecting and reporting relevant information on tunas (Figure 6) to monitor tuna MeHg concentrations within an interdisciplinary framework, at both regional and global spatio-temporal patterns, while taking into account possible biological and ecological confounding effects.

Precise coordinates and date of capture: To be able to investigate spatial and temporal variability of Hg concentrations in tunas at a high resolution, it is crucial to collect the precise date and location of the tuna fishing operation. Collecting such information is quite easy onboard scientific vessels, as well as on commercial fishing vessels thanks to onboard observers involved in the several sampling programs deployed in the global ocean by regional fishery management organizations. Conversely, fishing information of tuna captured by the artisanal fishery and sampled at the landing can be difficult to obtain. In this case, rather than attributing tuna samples to the port where they were collected, we suggest estimating fishing date and area based on the expertise of the fishermen, or if available, on geo-localised data of the fishing trip (e.g., vessel monitoring system, VMS). Tuna sampling at the market is not adequate for an accurate spatio-temporal Hg biomonitoring as the origin of tuna (i.e., fishing date and coordinates) is difficult to obtain.

Tuna individual straight fork length: Given the natural bioaccumulative property of MeHg in organisms through time, it is necessary to have biological information associated to tuna samples when exploring Hg patterns in tunas. Individual tuna age is the most appropriate biological data to account for bioaccumulation through time; yet it is rarely used because of the inherent uncertainty in tuna ageing methods (e.g., growth models, tagging, interpretation of calcified structures, or analysis of length-frequency data) (Murua et al., 2017). Tuna length and weights therefore represent a good alternative to account for Hg bioaccumulation in individuals. While individual length can be easily measured either onboard or at the lab, precise fish weight cannot be obtained onboard because boats are not stable enough. For a homogeneous and global Hg biomonitoring program with tunas, we therefore recommend measuring individual straight fork length, i.e. from the tip of tuna snout to the fork of the tail (Figure 6), which is the most common and best fitted biometric for fish species like tunas that have forked caudal fins (avoid curved fork length, total length, and standard length).

Muscle tissue collection: There are many possible matrices for biomonitoring and tissue choice depends on monitoring objectives, interests, and outcomes. In high trophic level fish species like tunas, it is recommended to monitor MeHg exposure in muscle tissues as i) MeHg is the dominant chemical form (> 90 %, SI Appendix Table S1) in tuna muscle, and ii) muscle is the most edible part of tunas (UN Environment, 2019). Given red muscle exhibits significantly higher total Hg concentrations than white muscle (Bosch et al., 2016), it is important to collect only white muscle tissue during tuna sampling. For homogeneity, we recommend collecting white muscle samples in dorsal position (Figure 6) to account for possible variable lipid contents in different muscle parts among tuna species and ocean regions. If not possible (e.g., no permission to collect samples in dorsal position from fishermen), muscle samples can be alternatively collected in anal position. No difference in total Hg, MeHg and inorganic Hg concentrations were found between dorsal and anal white muscles of yellowfin caught off south Africa (Bosch et al., 2016), yet we suggest first checking for Hg variability between anal and dorsal positions when compiling tuna Hg data obtained in both positions. Tuna muscle samples can be stored in labelled individual cryotubes, plastic zip lock bags, or glass vials. When possible, we advise collecting sufficient amount of muscle tissue without skin (~ 2 g wet weight) in order to anticipate future analyses (e.g., Hg concentrations, stable isotopes, fatty acids, major and trace elements), while taking into account the weight loss after freeze-drying (~ 70 %, SI Appendix Table S1).

Other matrices might be interesting to investigate pelagic top predators MeHg exposure, in addition to white muscle tissues. Blood in particular represents a possible non-lethal marker of Hg distribution in tunas, similar to bird and mammal species in other marine ecosystems. While Hg kinetics in tunas

remain poorly understood, blood circulation is suspected to play an important role in MeHg distribution across tissues (Leaner and Mason, 2004), and blood is expected to be representative of a short exposure time (~ weeks) compared to muscle tissues (~ months) (Bearhop et al., 2000). In the western central Pacific Ocean, consistency in the spatial distribution patterns of total Hg concentrations in blood and white muscle tissues has been evidenced in yellowfin and bigeye, highlighting the pertinence of these two tissues for large-scale Hg monitoring studies (Barbosa et al., 2022). Such biomonitoring coupling blood and muscle tissues is promising to better understand MeHg accumulation pathway in tunas, and ultimately refine Hg predictions in top predators to address the main challenges of the Minamata Convention.

Tissue conservation and preparation prior to analyses: To avoid elements (i.e., Hg, carbon and nitrogen) degradation, white muscle samples must be stored frozen immediately after collection, at minimum -20°C. Also, to avoid concern about variable moisture content, we recommend freeze-drying muscle samples during 48-72h (depending on the amount of tissue collected), instead of drying samples at 60°C, to obtain and compare Hg concentrations on a dry weight basis, as recommended by the Global Mercury Assessment (UN Environment, 2019). At this stage and when possible, we advise estimating gravimetrically the percentage of moisture (i.e., water content), calculating the difference between wet and dry masses of samples after freeze-drying. This allows precise conversion of concentrations from dry weight to wet weight in order to compare tuna Hg concentrations with safety guideline values that are currently expressed on a wet weight basis (FAO Codex Alimentarius Commission, 2021). Otherwise, a mean value of 70 % can be used (SI Appendix Table S1) (Bodin et al., 2017; Houssard et al., 2019; Kojadinovic et al., 2006; Teffer et al., 2014; Vlieg et al., 1993). Finally, muscle samples must be ground into a fine and homogeneous powder to guarantee repeatability and accuracy in lab analyses.

Mercury concentrations and ecological tracers: Methylmercury exposure in tuna muscle is recommended to be assessed with total Hg concentrations, as i) MeHg is the dominant chemical form (> 90 %, SI Appendix Table S1) in tuna muscle, and ii) analysis of total Hg concentrations is more cost effective and accessible to those without advanced and costly laboratory facilities (UN Environment, 2019). Total Hg concentrations are measured on powdered and homogenized freeze-dried muscle samples by either thermal decomposition, gold amalgamation, and atomic absorption spectrometry, or acid digestion followed by cold vapor atomic fluorescence spectroscopy. Both methods are relevant as long as blanks and biological standard reference materials (e.g., BCR-464, tuna muscle, total Hg = 5240 ± 100 $ng.g^{-1}$; TORT-3, lobster hepatropancreas, total Hg = 292 ± 22 $ng.g^{-1}$) are routinely used in each analytical batch and reported by authors with the corresponding recoveries to check Hg measurements accuracy and traceability. Total Hg concentrations are expressed on a dry weight basis, and can be converted on a wet weight basis to be compared to safety guidelines values (FAO Codex Alimentarius Commission, 2021).

To explore the potential confounding effects of changes in tuna trophic position, habitat, and feeding pathway, we recommend systematically analysing bulk muscle carbon and nitrogen stable isotope values ($\delta^{13}C$ and $\delta^{15}N$) in addition to total Hg concentrations. Carbon and nitrogen stable isotopes are measured on powdered and homogenized freeze-dried muscle samples packed in tin cups and analysed with an elemental analyser coupled to an isotope ratio mass spectrometer. Results are reported in the δ unit notation and expressed as part of thousand (‰) relative to international standards. Similar to Hg analyses, a combination of certified and/or in-house reference materials must

be used to ensure replication, accuracy, and precision within typical acceptable ranges. Lipid extraction on a subaliquot of the bulk muscle powder (Hg being measured on the bulk fraction) can be performed with neutral solvents (e.g., dichloromethane, cyclohexane) prior to $\delta^{13}C$ analysis to overcome bias related to lipid content (Bodin et al., 2009; Ménard et al., 2007). On preserved lipid-free samples initially prepared for tuna foraging ecology (e.g., Bodin et al., 2020) that represent a gold mine of already collected samples for Hg biomonitoring, it is yet possible to analyse total Hg concentrations as lipid removal has been shown to have no effect on total Hg concentrations in tropical tunas (Médieu et al., 2021b). Another alternative to avoid chemical lipid removal is to measure both total Hg concentrations and $\delta^{13}C$ and $\delta^{15}N$ values on bulk muscle powders, and to correct $\delta^{13}C$ values in samples with elevated lipid content (C:N > 3.5), using a mass balance equation mathematical derived from Atlantic bluefin tuna muscle (Logan et al., 2008).

When using tuna $\delta^{15}N$ ratios to explore possible confounding trophic effects on Hg concentrations, it is important to keep in mind that tuna $\delta^{15}N$ values reflect both trophic dynamics along food webs, and biogeochemical processes at the base of marine food webs (e.g., demand for the nitrogenous nutrients by primary producers, or variability in the dominant dissolved nitrogen species present; Lorrain et al., 2015). While this baseline effect may be negligible at a local spatial or temporal scale, it may significantly bias tuna trophic position estimates when working at larger scales given the large spatial and temporal variations of $\delta^{15}N$ baseline signature in the global ocean (McMahon et al., 2013). In section 8.1, we provide alternatives to better account for baseline variability when exploring the relative importance of trophic processes on Hg accumulation in tunas.

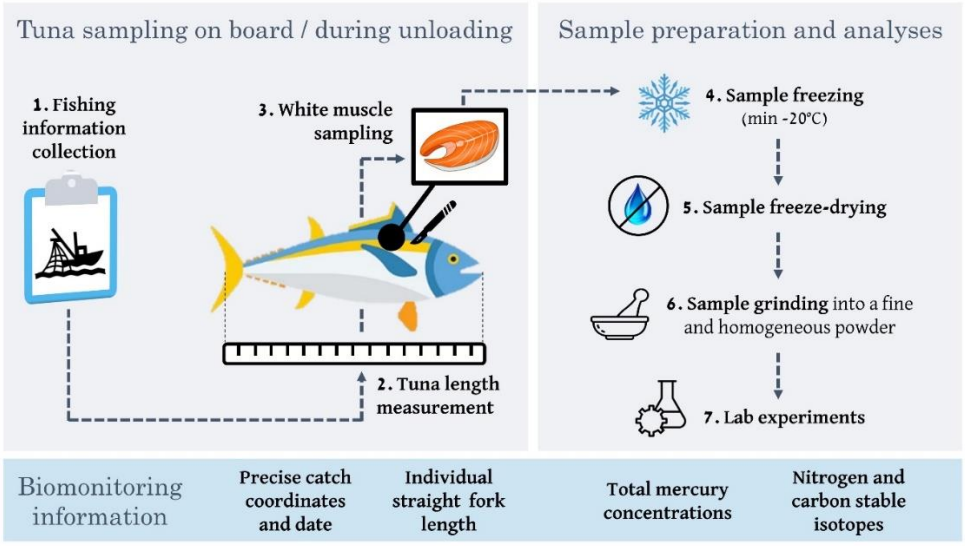

Figure 6. Conceptual framework of the standardized guidelines for collecting and reporting relevant monitoring information on tunas.

6.2. Data standardization

When exploring Hg patterns at both spatial and temporal scales, we recommend standardizing Hg concentrations to remove the bias associated to fish size differences among individuals, and to characterize the extent of the natural bioaccumulative processes of Hg in tunas, following the

methodology used in three recent studies in the Pacific Ocean (Houssard et al., 2019; Médieu et al., 2022, 2021a). Briefly, this consists in fitting power-law relationships ($\log(Hg) = a \times (FL - b)^c - d$) between log-transformed Hg concentrations and tuna fork length (FL). Residuals from these length-based models (i.e., observed values – predicted values) are extracted and used to calculate length-standardized Hg concentrations at mean species lengths (e.g., 100 cm FL for bigeye, yellowfin and albacore, and 60 cm FL for skipjack), thereafter defined as "standardized Hg concentrations", in contrast to "observed Hg concentrations". Such standardization at fish length allows the exploration of spatial and temporal Hg patterns within a same tuna species, as evidenced in sections 4 and 5 respectively, but also inter-species differences to explore different Hg accumulation pathways in relation to tuna foraging depths (section 2).

For an accurate global comparison of Hg concentrations in tunas, age-standardization, rather that length-standardization, might be more relevant given that tuna growth varies among ocean basins (Murua et al., 2017; Tseng et al., 2021). To do so, power-law relationship can be fit between logHg and tuna age, with tuna age estimated from tuna fork length using species- and ocean- specific growth models, as recommended by fisheries scientists for tuna stock assessments. Nevertheless, given the uncertainties related to tuna growth models and the resulting potential bias on age estimations, we assume that length-standardization of Hg concentrations remains relevant to explore spatio-temporal variability of tuna Hg content when working at small or large regional scales.

7. Caveats and limitations

Being highly exploited worldwide and monitored by extensive regional fishery management organizations, tropical tunas and albacore represent a possible gold mine of biological samples to get robust and comparable Hg datasets, as required for an effective Hg biomonitoring in the context of the Minamata Convention. This sampling strategy provides a considerable gain of time, effort, and cost for scientists in charge of sample collection. Yet, this implies that the spatial and temporal coverage, as well as the fishing gear and technique rely solely on the fishermen and fishing companies, leading possibly to poorly documented ocean regions, and/or discontinuous Hg time series. This sampling strategy can also induce variability in tuna length resulting for different fishing gear selecting preferentially certain tuna fish sizes. In the eastern Pacific Ocean for instance, fishing companies principally target surface tunas with purse seine, i.e., mainly skipjack, yellowfin and juvenile bigeye (Inter-American Tropical Tuna Commission, 2021), while in the southwestern Pacific Ocean, fishermen preferentially target adult yellowfin and bigeye in deeper waters with longlines (ISSF, 2021). Although the length- (or age-) standardization approach recommended above allows taking into account this spatial variability of fishing gear and tuna length among samples, it is important to keep in mind the biological and ecological consequences of this sampling bias when exploring Hg accumulation in tunas.

In addition, while relatively easy to collect, tuna samples remain available for the last few years/decades only, mostly when regional fishery management organizations started to build specimen tissue banks, and the oldest published Hg data are from the 1970s. Contrary to seabirds, it is not possible to analyse historical samples (e.g., feathers) stored in natural history museum collections to obtain historical Hg data. Such biological archives are yet valuable to discuss Hg temporal trends in marine predators in parallel to Hg emissions and deposition trends over a long period of time, and to better assess the impacts of anthropogenic Hg releases on marine ecosystems on multidecadal

timescales (e.g., Bond et al., 2015; Carravieri et al., 2016; Monteiro and Furness, 1997). The combined use of body feathers from museum specimens and free-living birds of sooty tern recently highlighted an increase of 58.9 % of mean Hg concentrations between the 1920s and the 2020s, likely reflecting ecosystem-wide changes in the tropical South Atlantic Ocean and/or anthropogenic Hg emissions to the environment (Cusset et al., 2023). Overall, this highlights the need to combine several biomonitoring species to capture large spatial and temporal scales of Hg exposure in different ecosystems.

8. Future directions

8.1. Data integration with oceanic and ecological variables

In addition to the provision of information listed above, other metadata are relevant to interpret tuna Hg concentrations regarding possible confounding changes in tuna trophic ecology, and ultimately to predict Hg concentrations in tunas according to changes in anthropogenic releases.

The recent development of global physical and biogeochemical models now allow better quantifying the variability of ocean (thermo-)dynamics and baseline processes, and tracking nutrient flow along marine food webs (e.g., NEMO-PISCES, Buchanan et al., 2021; and MOBI, Somes et al., 2017). Such models, providing baseline $\delta^{13}C$ and in particular $\delta^{15}N$ estimates in phytoplankton for each tuna Hg data location, offer new perspectives to take into account natural baseline processes while exploring the relative importance of trophic factors on Hg variability in tunas. In the Pacific Ocean for instance, both estimated $\delta^{15}N$ values in particulate organic matter, and baseline-corrected tuna $\delta^{15}N$ values (i.e., difference of $\delta^{15}N$ values in tunas and particulate organic matter) helped disentangling the relative importance of baseline and trophic processes, respectively when explaining the large spatial patterns of standardized Hg concentrations in skipjack (Médieu et al., 2022).

Moreover, developing simulations of MeHg concentrations in seawater and phyto/zooplankton by global 3-D models (e.g., MITgcm, Zhang et al., 2020) can offer new alternatives to the scarce observations of Hg concentrations and speciation in the ocean to explore global patterns of tuna Hg concentrations alongside regional changes in MeHg production, bioavailability, and biomagnification. Physical and biogeochemical models have been shown to be other relevant tools to investigate the mechanisms governing MeHg bioavailability in seawater. The complementary use of standardized tuna Hg concentrations alongside biogeochemical (e.g., subsurface dissolved O_2 concentrations) and physical (e.g., depth of oxycline) model outputs already evidenced the importance of marine biogeochemical processes, in particular variable vertical profiles of dissolved MeHg concentrations among ocean regions, to explain large spatial patterns of tuna Hg concentrations (section 4) (Houssard et al., 2019; Médieu et al., 2022).

Finally, atmospheric Hg models represent a good alternative to missing high-resolution and worldwide observed Hg data in the atmosphere to explore tuna Hg concentrations in parallel to changes in anthropogenic Hg emissions and Hg concentrations in the atmosphere. Recently, the complementary use of tuna standardized Hg concentrations alongside atmospheric elemental Hg concentrations predicted by the GEOS-Chem model (Angot et al., 2018; Travnikov et al., 2017) at each tuna location revealed the relationship between actual Asian Hg emissions on elevated skipjack Hg concentrations in the northwestern Pacific Ocean (Médieu et al., 2022).

While the use of biological, physical, and anthropogenic model outputs in parallel to standardized Hg concentrations in tunas is promising to improve our understanding of the main processes driving the spatio-temporal variability of Hg content in pelagic top predators, it remains important to keep collecting measurements of Hg concentrations and speciation in the atmosphere and the ocean at large spatial and temporal scales in contrasted tropical regions. The comparison of these observations data with standardized Hg concentrations in tunas, as done in recent studies (Barbosa et al., 2022; Tseng et al., 2021), is indeed crucial to validate the main hypotheses formulated with model outputs, and to refine predictions of Hg responses in pelagic biota to the different emissions and climate change scenarios of the Minamata Convention.

8.2. Mercury stable isotopes

Among the elemental and molecular analytical tools available to explore the Hg cycle, Hg stable isotope signatures measured in biotic tissues has improved significantly our understanding of the biogeochemical, ecological, and metabolic factors driving MeHg exposure. Thanks to the unique photochemical mass-independent fractionation of Hg isotopes (reported as Δ^{199}Hg), it is now possible to trace trophic resources and the vertical foraging habitat of pelagic species (Le Croizier et al., 2020; Madigan et al., 2018). Following light attenuation with depth, Δ^{199}Hg values decrease from the surface to the aphotic waters (Blum et al., 2013), but are conserved during trophic transfer between a prey and its predator (Kwon et al., 2016; Laffont et al., 2011), leading to lower Δ^{199}Hg values in deeper marine predators. Mercury stable isotopes also offer new perspectives to investigate how climate change can affect MeHg concentrations in marine food webs. In the Arctic, Δ^{199}Hg values in marine biological samples showed the influence of sea ice on Hg photodegradation (Point et al., 2011), and documented the effect of climate change of Hg cycle (Masbou et al., 2015). In tropical ecosystems, the combined use of Hg concentrations, and carbon, nitrogen and Hg stable isotopes, alongside biogeochemical model outputs in seabirds in the northern Humboldt current system off Peru revealed that stable Hg concentrations over time were masking concomitant reduced MeHg formation (due to the deepening of the oxycline) and degradation (due to reduced productivity, carbon export, and remineralization) (Renedo et al., 2021). Similar studies using the global tuna tissue bank are therefore of interest to investigate anthropogenic- and climate-induced changes of MeHg formation, degradation, and accumulation in pelagic marine food webs of tropical ecosystems, at both regional and global scales.

Acknowledgements: We are grateful to research communities working on tuna trophic ecology, mercury cycle and biogeochemical modeling who indirectly contributed to this work. We also thank Anders Bignert from the Swedish Museum of Natural History for the adequacy calculation. This study benefited from thoughtful comments from two anonymous referees.

Funding: This study was conducted in the framework of ANR-17-CE34-0010 MERTOX (unravelling the origin of methylMERcury TOXin in marine ecosystems, 2017 – 2021, PI David Point) from the French Agence Nationale de la Recherche. It also benefited from financial support of the Région Bretagne and Université de Bretagne Occidentale (UBO).

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