ASSESSING METAL BIOMAGNIFICATION AND UPTAKE PATHWAYS IN MEDITERRANEAN FISH

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Abstract
Metal biomagnification in marine food chains is an important yet contentious issue, and one that has significant regulatory and public health consequences. Nevertheless, few estimates of metal assimilation and retention are available for upper trophic level organisms. Using radiotracers in a laboratory study we measured the trophic transfer of 8 metals (Am, Cd, Co, Cr, Cs, Mn, Se, Zn) and assessed the relative importance of exposure route in metal accumulation in 3 representative species of carnivorous fish that are commonly harvested for human consumption in the Mediterranean. Results indicate that: (1) inter-metal differences in fish assimilation far exceed interspecies differences; (2) the relative potential for biomagnification of metals decreases such that: Cs >Se >Zn >Mn >Cd >Co >Am; (3) for all metals except Am and Cs, dietary exposure accounts for >90% of tissue metal concentrations in marine fish.
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Fish are known to accumulate trace metals, sometimes to levels that are toxic to themselves or human consumers. Numerous studies of metal concentrations in fish have found great variability, both intra- and interspecifically, but mechanistic studies have been largely lacking, and consequently the processes governing such variability remain largely unknown [1]. Mercury is the only metal recognized to biomagnify in top level predators, but recent evidence indicates the potential of other metals to biomagnify in some food chains [2]. The kinetic parameters determining biomagnification are not fully quantified at upper trophic levels despite the fact that biomagnification has important regulatory and public health concerns. In general there are relatively few studies that provide quantitative measurements of the kinetics of metal uptake and depuration or that delineate the relative importance of diet or the aqueous phase as sources in marine fish . In addition, while early studies considered metal uptake from solution and indeed many environmental regulatory agencies still consider only aqueous exposure in risk assessment and wildlife protection, there is increasing recognition of the importance of metal accumulation from dietary exposure in marine food webs [1, 3].

In order to assess the importance of dietary metal uptake in marine fish and the extent to which it may vary between fish, we measured metal uptake from dietary exposure in three fish species (two teleosts and one elasmobranch) with different phylogenies and life histories. Included in the study were the spotted dogfish, *Scyliorhinus canicula*, the turbot, *Psetta maxima*, and the sea bream, *Sparus auratus*, three species that are abundant in the Mediterranean and that comprise important and valuable commercial fisheries.

Biokinetic models can provide a quantitative description of the processes governing metal bioaccumulation in aquatic organisms. Under steady state conditions, metal accumulation in organisms from both dietary and aqueous exposure can be calculated from the following equation:

$C_{ss} = [(k_u \text{ x } C_w)/(k_{ew} + g)] + [(AE \text{ x } IR \text{ x } C_f)/(k_{ef} + g)]$

where C_{ss} is the metal concentration in fish at steady state, k_u is the dissolved uptake rate constant, C_w is the dissolved metal concentration, AE is the assimilation efficiency from food, IR is the ingestion rate, C_f is the metal concentration in food, k_{ew}, k_{ef} is the efflux rate constant from water and food respectively, and g is the growth rate constant [4]. The trophic transfer factor (TTF) of a metal through the food chain is then: $TTF = C_n / C_{n-1} = (AE \text{ x } IR) / (k_e + g)$, where C_n is the concentration of metal in an organism at trophic level n and C_{n-1} is the concentration of metal in an organism from the trophic level below. A value of TTF >1 indicates the possibility of biomagnification of the metal in question; a TTF <1 indicates biodiminution [5]. Currently, TTFs remain largely unknown in marine food chains [2].

Pulse-chase radiotracer methods are commonly used to quantify the parameters used in biokinetic models. The use of gamma-emitting radioisotopes enables the simultaneous measurement of multiple elements at environmentally realistic concentrations via non-destructive sampling of live organisms. In our study, we labeled prey fish (juvenile *Sparus auratus*) via the aqueous phase with ^{241}Am, ^{109}Cd, ^{60}Co, ^{51}Cr, ^{134}Cs, ^{54}Mn, ^{75}Se and ^{65}Zn. 12 individuals each of *Sparus auratus* and *Psetta maxima* and 6 individuals of *Scyliorhinus canicula* were then fed with a pulse of radiolabeled food and radioactivity was measured in each fish over a 3-wk period to assess the uptake rate, assimilation efficiency, retention, and tissue distribution of these metals in the three fish species.

Here we compare the relative ranking of TTF of the 6 metals; values were averaged for the 3 fish species as no significant differences were evident among the fish. We report relative TTF values for each metal (AE/k_e). Of the metals examined the greatest potential for biomagnification was shown by Cs, Se, and Zn, with Am and Co showing the lowest potential (Table 1). We combined the kinetic parameters obtained from dietary exposure with those determined from aqueous exposure in the laboratory for the same fish species to determine the relative importance of exposure route of metals to fish. Our modeling indicates that, with the exception of Am and Cs, dietary exposure is the dominant exposure route, accounting for $\geq$90% of total metal in fish (Table 1). Regulatory bodies that determine appropriate water quality criteria need to consider dietary exposures in protecting aquatic animals from toxic metal contamination.

Tab. 1. Relative potential for biomagnification, expressed as AE/k_e (see text), and percent of metal from dietary exposure in carnivorous Mediterranean fish. Percent from diet was calculated using data from this study and from Jeffree et al. [6]. Values represent means + SD of values obtained for all fish species.

Metal	AE/k_e	Percent from diet
Am	14 ±5	4 ±2
Cd	27 ±6	99 ±0.2
Co	23 ±10	93 ±9
Cs	56 ±30	35 ±26
Mn	30 ±6	96 ±5
Zn	34 ±17	99 ±0.1

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