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Trophic transfer of ^{110m}Ag in the turbot *Scophthalmus maximus* through natural prey and compounded feed items

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Abstract

Industrial incidents can result in radionuclide release in the environment, among which ^{110m}Ag . Indeed, under particular circumstances, non-negligible amounts of ^{110m}Ag have been measured in the marine environment (as observed in Fukushima Dai-ichi incident). This element can be accumulated by aquatic organisms through different pathways including the trophic transfer. The present study aimed at examining the variation of ^{110m}Ag assimilation efficiency (AE) by turbot, *Scophthalmus maximus*, when exposed through different feeds. Pulse-chase feeding experiments were carried out in mesocosms, using radiolabelled feeds (natural prey and commercial pellets). Depuration kinetics of ^{110m}Ag over 21 days were generally fitted by a two-component exponential model; the ingested radioelement was poorly assimilated by turbot regardless of the food item that was used (AE always <3%). Concentration and subcellular distribution of ^{110m}Ag in prey did not seem to influence its assimilation by turbot. These results suggest that physiological mechanisms could occur in fish that would prevent the transfer of ^{110m}Ag from gut lumen to internal organs (e.g. ^{110m}Ag neutralization in the lumen of the stomach, detoxification mechanisms occurring in the gut).

Keywords: Silver; Accumulation; Aquaculture; Feeds; Radionuclides

1. Introduction

The impact of artificial radionuclides on aquatic ecosystems, in which they act as micro-pollutants, is a topic of particular interest, both in terms of radiation protection and of environmental assessment. Under normal operational conditions of nuclear facilities, ^{110m}Ag is one of the radionuclides that can be emitted in small quantities (Adam et al., 2001). Under particular circumstances, non-negligible amounts of ^{110m}Ag have been measured in the marine environment, such as after the Chernobyl and Fukushima Dai-ichi Nuclear Power Plant accidents (Buessler et al., 2012; Aono et al., 2014).

Information on ^{110m}Ag bioaccumulation in fish is until now essentially confined to freshwater organisms (e.g. Baudin and Garnier-Laplace, 1994; Ausseil et al., 2002; Bertram and Playle, 2002). Although no biological role has been identified, these studies showed that the trophic transfer is an important pathway of accumulation in freshwater fish. In the few studies carried out on marine fish, the trophic transfer seemed to be a less important accumulation pathway of ^{110m}Ag (Pentreath, 1977; Long and Wang, 2005). However, only one type of food per species was investigated in these studies. There is thus currently a lack of knowledge on the possible influence of food quality on ^{110m}Ag assimilation by marine fish.

Dissolved or particulate contaminations occur and contribute simultaneously in natural environment to the global metal bio-accumulation in organisms. The delineation of the relative contribution of the different exposure pathways has to be assessed using models (see Thomann, 1981, Landrum et al., 1992; or Metian et al., 2008a), to really identify a main uptake route of contamination. These models that are using the kinetic parameters experimentally obtained are more and more considered in the literature but this is not systematic, and some papers argue the importance of one exposure pathway on another without mathematical validation. In the same time,

it is key to determine within these models the variability of parameters that can affect the final determination of the main uptake pathway, such as for example the effect of food quality on metal transfer.

In this study, the turbot *Scophthalmus maximus* was chosen to examine the transfer of ^{110m}Ag from four different types of feeds: three different natural prey (seabream *Sparus aurata*, shrimp *Palaemon serratus*, and ragworm *Hediste diversicolor*) and manufactured pellets; this latter feed was selected since the turbot is a common species produced in aquaculture. In order to better understand assimilation processes, two levels of biological organization were considered: (1) the whole individual (determination of the depuration kinetics and of assimilation efficiencies) and (2) the organs and tissues (dissections and determination of body distribution).

2. Materials and Methods

2.1. Origin and acclimation of organisms

In January 2014, one hundred juvenile turbot *S. maximus* were purchased from a fish farm (France Turbot, France) and shipped to the International Atomic Energy Agency premises in the Principality of Monaco. Fish were acclimated to laboratory conditions for 21 days (open circuit, 500-L aquarium; water renewal: 100 L h^{-1} ; $0.45 \mu\text{m}$ filtered seawater; salinity: 38 p.s.u.; temperature: $15 \pm 0.5^\circ\text{C}$; pH: 8.0 ± 0.1 ; light/dark: 12h/12h). During the acclimation period, the fish were fed to a daily ration of 2% of their estimated biomass with 1.1-mm pellets (Le Gouessant, France).

In order to investigate the influence of the diet on ^{110m}Ag assimilation by *S. maximus*, 1.1-mm manufactured pellets (Le Gouessant, France) and three different natural prey of turbot were used (e.g. Sparrevohn et al., 2008; Florin and Lavados, 2010): juvenile fish (*S. aurata*; 60-day-old

hatchlings, weight: $0.060 \pm 0.013\text{g}$), shrimp (*P. serratus*, weight : $0.58 \pm 0.11\text{g}$) and worms (*H. diversicolor*, weight: $0.82 \pm 0.14\text{g}$). Juvenile fish were obtained from a hatchery (Poissons du Soleil, France), shrimp were purchased from a fisherman (Poissons Vivants, France), and worms were purchased from fishing bait seller (Normandie Appats, France). All living feeds were acclimated to the same laboratory conditions than predator fish for a minimum of two weeks prior to experiments. Shrimp and worms were fed a mix of fish feed and crushed mussels whereas juvenile fish were fed 300- μm pellets (Biomar, France). Since body size is known to affect metal bioaccumulation in marine organisms (Boyden, 1974; Warnau et al., 1995), only individuals with homogeneous size of each prey species were used in the experiments.

2.2. Radiotracers and Counting

Experiments were carried out using a high-specific $^{110\text{m}}\text{Ag}$ radiotracer purchased from POLATOM, Poland ($^{110\text{m}}\text{Ag}$ as AgNO_3 in 0.1 M HNO_3 ; $[T_{1/2}] = 250$ days; specific activity: 520 MBq mg^{-1}). The radioactivity of the tracer was measured using a high-resolution γ -spectrometer system composed of 5 Germanium - N or P type - detectors (EGNC 33-195-R, Canberra[®] and Eurysis[®]) connected to a multi-channel analyser and a computer equipped with a spectra analysis software (Interwinner 6, Intertechnique[®]). The radioactivity was determined by comparison with standards of known activity and of appropriate geometry (calibration and counting). Measurements were corrected for background and physical radioactive decay. Organisms were placed in counting tubes filled with clean seawater during the counting period. The counting time was adjusted to obtain a propagated counting error less than 5% (e.g. Metian et al., 2008b), typically 25-60 min for whole organism radioanalysis. This counting duration did not affect organism welfare, as shown by their inter-counting activity and feeding behavior.

2.3. Experimental Procedure

2.3.1. Radiolabelling of feeds

Twelve grams of pellets were dipped for 1 h in 16.5 mL of seawater spiked with 1.8 kBq $^{110\text{m}}\text{Ag}$ mL⁻¹. Pellets were then dried for 48 hours at 50°C and kept in a dry environment in order to prevent mold growth. Preliminary tests were performed to determine the possible leakage into the water of radioisotopes from the pellets during the feeding. During the feeding, acclimated fish were consuming pellets in less than 1 min. Therefore, preliminary tests consisted in pouring radiolabelled dry pellets (100 mg per treatment) for 1 and 10 min in 50 mL seawater and to measure any radioactivity in the seawater. The leakage of pellet-radioactivity in water did not exceed 0.8% and 2% of the initial activity after 1 min and 10 min, respectively. Although these tests confirmed the single-pathway contamination (viz. food) of the fish, one turbot was used in each treatment, as a control to take into account the possibility of $^{110\text{m}}\text{Ag}$ recycling through water (see Section 2.3.2).

Radiolabeling of the natural prey was carried out by exposing them for 7 to 21 days in seawater spiked with the radiotracer (nominal activity of 0.5 kBq $^{110\text{m}}\text{Ag}$ L⁻¹, corresponding to 27 nmol L⁻¹ equivalent stable Ag). No change in pH was detectable in the aquarium (close circuit) after tracer addition. The spiked seawater was renewed daily in order to keep the radiotracer activity as constant as possible. Activity of the metal tracer in seawater was checked daily, before and after each seawater renewal, to determine time-integrated activities (Rodriguez y Baena et al., 2006). Prey were fed after each seawater renewal and just prior to radioisotope addition. Exposures of prey were made in aerated, 20-L aquaria. For shrimp exposure, each organism was kept individually in a cylindrical PVC container (drilled to allow for free water circulation) during the whole duration of the experiment in order to avoid cannibalism during moulting and to facilitate

individual identification. For the worms, the walls of the aquarium were obscured and plastic tubes were added as artificial burrows.

2.3.2. Exposure of turbot via radiolabelled feeds

Four sets of experiments were realized for each type of feed considered. Each time, 8 to 12 juvenile turbot (11.9 ± 5.5 g, wet weight) were randomly picked and transferred in an aerated, open circuit, 70-L aquarium. One week before the exposure to radiolabelled feed, fish were daily fed the selected type of feed and individually identified by slits cut in fins.

Each experiment consisted in a single exposure to radiolabelled feed (single feeding method also called pulse chase feeding; see e.g. Hédouin et al., 2010; Metian et al., 2010). For each experiment, turbot were fed *ad libitum* for 30 min; the uneaten food was then removed. Ragworms and shrimp were cut into pieces prior to the feeding in order to facilitate ingestion. Two hours after the initiation of the feeding, each fish was whole-body γ -counted alive and then replaced in clean, flowing seawater conditions (parameters as previously described). No regurgitation of the radiolabelled feeds was observed. Fish were then fed daily non-labeled pellets (2% of their biomass). In parallel, after the feeding period, an additional turbot was placed in each aquarium within in a net to control any possible radiotracer recycling from seawater due to radiotracer leaching from the contaminated food or from fish depuration. Fish were then whole-body γ -counted alive daily over 21 days (including control individuals). They were moved to another, clean 70-L aquarium after each counting to avoid contamination from ^{110m}Ag contained in faeces.

After the depuration period, 4 individuals were dissected in 7 compartments: (1) the 4 fillets (skinned muscles), (2) the kidney, (3) the liver, (4) the gall bladder, (5) the digestive tract, (6) the head (including gills) and (7) the remaining parts (including skin, skeleton, fins, heart and muscle

residues) and were separated, weighed (wet wt) and radioanalysed to determine the radiotracer body distribution. During experiments, no mortality was recorded.

2.3.3. ^{110m}Ag compartmentalization in feeds

For the natural prey, the distribution of ^{110m}Ag between the soluble and insoluble fractions was determined according to a method adapted from Bustamante and Miramand (2005). Briefly, an aliquot of contaminated feed stored at -80°C was crushed and tissues homogenized using a T25 Ultra-Turrax Basic (IKA[®]) in approximately 10 volumes (w:v) of TRIS-HCl buffer 0.02 M sucrose 0.25 M with 1 mM protease inhibitor (PMSF, phenylmethylsulfonylfluoride) and 5 mM reducing agent (DTT, dithiothreitol), at pH 8.06. The homogenates were centrifuged at 45 000 G for 2 h at 4°C (Sorvall Evolution RC Superspeed Centrifuge, Sorvall instruments[®]) to separate particle-free supernatant from the debris. Aliquots of the homogenates and trophically available metal (TAM) obtained were radioanalyzed in order to determine the radiotracers compartmentalization. The same procedure was repeated over time for each prey.

2.4. Data treatment and statistical analyses

Depuration of radiotracers was expressed as the percentage of remaining radioactivity (radioactivity at time t divided by the initial radioactivity measured in the organism at the beginning of the depuration period; Warnau et al., 1996). Depuration kinetics were fitted using non-linear model. They were best fitted using the following two-component model that includes an exponential component and a constant (Eq. 1) (decision based on F test and ANOVA tables):

$$A_t = A_{0s} \cdot e^{-k_{es}t} + AE \quad (\text{Eq. 1})$$

where A_t and A_{0s} are the remaining activities (%) at time t (d) and 0, respectively; k_{es} is the depuration rate constant (d^{-1}) and AE is the assimilation efficiency (%). The first component

$(A_{0s} \cdot e^{-k_{es}t})$ represents the depuration kinetics of the radiotracer fraction that is weakly associated to the organisms and rapidly eliminated, whereas the second component (AE) describes the radiotracer fraction that is retained in the body (Warnau et al., 1996). A short-term biological half-life can be calculated ($T_{b1/2}$) from the depuration rate constant according to the relation $T_{b1/2s} = \ln 2/k_{es}$. By definition, biological half-life of the “AE” component is infinite. Model constants were estimated by iterative adjustment of the model using the nonlinear curve-fitting routines in the Statistica® software 7.0. Distribution patterns of ^{110m}Ag among organs and tissues were compared by a G-test using R software 3.0.1 (R Development Core Team, 2014). The level of significance for statistical analyses was always set at $\alpha = 0.05$.

3. Results

In order to evaluate the influence of food quality on ^{110m}Ag assimilation in the turbot *S. maximus*, 4 food items were first radiolabelled through seawater. The seabream *S. aurata*, the shrimp *P. serratus*, the ragworm *H. diversicolor* and the manufactured compounded feed concentrated ^{110m}Ag differently: the average activities measured in these feeds just before the feeding of the turbot were respectively 7 Bq g⁻¹, 37 Bq g⁻¹, 117 Bq g⁻¹ and 1072 Bq g⁻¹ (Table 1).

When turbot were fed seabream or shrimp, activities could be measured accurately in turbot after feeding. However, after 48 h of depuration, uncertainties of counts became very high (> 20%). After two weeks, the activities were below detection limits. Therefore, depuration kinetics could not be modeled for these two types of food.

When turbot were fed with pellets, whole-body depuration kinetics were best fitted by a two-component model (an exponential phase and a constant; Figure 1; R^2 : 0.99). ^{110m}Ag was poorly

assimilated by the fish ($AE = 0.32 \pm 0.75\%$, $p > 0.05$). Virtually all ingested ^{110m}Ag was very rapidly lost ($T_{b/2s} = 0.77 \pm 0.02$ d; mean \pm SD; Fig. 1).

When ragworms were used for feeding turbot, depuration kinetics were best fitted by the same two-component model as with pellets (Fig. 1; R^2 : 0.99). The short-term depuration of ^{110m}Ag was hereto fast ($T_{b/2s} = 0.66 \pm 0.03$ d; mean \pm SD; Fig. 1) but, conversely to the other feeds, ^{110m}Ag assimilation was high enough to be accurately estimated ($AE: 3 \pm 0.7\%$; mean \pm SD; Table 1, $p < 0.05$).

Post-feeding distribution of ^{110m}Ag in turbot at the end of the 21-d depuration period is shown in Table 2, for the different types of food. Similar patterns of ^{110m}Ag distribution among compartments were observed in turbot fed with the different feeds ($p > 0.05$): liver and digestive tract accounting for until 65% of the total ^{110m}Ag load (Table 2). These body compartments represented, respectively, less than 10% and 1% of the body weight. Most of the remaining activity was distributed in the head and in remaining tissues.

Regarding subcellular partitioning of ^{110m}Ag in natural prey, the majority was found in the soluble fraction: the average proportion ranged from 50 to 72%. Significant difference was recorded for the two extreme values ($p < 0.01$): the highest proportion of ^{110m}Ag in the soluble fraction ($72 \pm 5\%$) was measured in shrimp whereas this proportion was $50 \pm 4\%$ in ragworms (Fig. 2).

4. Discussion

^{110m}Ag can contribute up to 20% of the low level gamma radioactivity in liquid effluents from some pressurized reactors under normal operating conditions (Baudin and Garnier-Laplace, 1994). For several years, radioecological field studies have detected ^{110m}Ag in the main components of aquatic ecosystems downstream from nuclear power stations (e.g. Eyrolle et al., 2008; Aono et al., 2014).

Moreover, the recent Fukushima Dai-ichi Nuclear Power Plant accident has led to the release of a massive amount of this radionuclide in the environment (Buessler et al., 2012; Aono et al., 2014). In this context, it is essential to understand the transfer of this radionuclide in aquatic organisms to better assess the risk for the ecosystem (food chain) and for Human.

Despite the potential importance of trophic transfer, the contribution of the food pathway for ^{110m}Ag global bioaccumulation in marine fish has been poorly investigated (e.g. Pentreath, 1977; Rouleau et al., 2000; Wood, 2011). Experimental approaches appear to be the most appropriate to better understand, estimate and model radionuclide transfer from prey to predators (Adam et al., 2002).

To undertake this study, different natural prey and compound pellet were exposed to waterborne ^{110m}Ag . Concentration Factors (CFs) were calculated at the end of the exposure period; they were 76 ± 9 in shrimp, 119 ± 77 in seabream and 203 ± 101 in ragworm. Although the exposure time varied among prey (7-21 days), results suggested that ragworms, exposed for 7 days in this study, have a higher ^{110m}Ag accumulation capacity than the other feeds.

To get a better understanding of ^{110m}Ag biological storage mechanisms at intracellular level in natural prey, ultracentrifugation was used for determining the subcellular fractioning of this element. The method used allows separating insoluble fraction (i.e., intracellular metal-rich granules and membrane fragments) from soluble fraction (i.e., cytosol and soluble metallothioneins and heat-sensitive proteins; Vijver et al., 2004). Most studies using such a technique were focusing on target organs and showed that ^{110m}Ag was mainly stored in an insoluble form, for example in the gills of the trout *Oncorhynchus mykiss* (80-95%; Galvez et al., 2002; Wood et al., 2002) or in the digestive gland of the scallop *Pecten maximus* ($93 \pm 2\%$; Metian et al., 2008a). Subcellular storage of ^{110m}Ag (and thus partitioning) may vary among organs for a given species, as shown in the bivalve species, *Gafrarium tumidum*; where 65% of ^{110m}Ag was found in the insoluble fraction

of the visceral mass whereas 90% was found in this fraction of the gills (Metian et al 2005). Therefore, in the present study, we decided to centrifuge the whole organisms (prey) to get a better idea of the integrated subcellular partitioning. Results revealed that a large proportion of the ^{110m}Ag was included in the soluble fraction of prey (50 -72%).

Differences of ^{110m}Ag accumulation in food items (in terms of concentration or subcellular distribution) did not have any influence on assimilation of the radionuclide by turbot. Indeed, our results indicated clearly that ^{110m}Ag was poorly assimilated and retained by the turbot whatever the feed considered. The assimilation efficiency of the radioelement ingested with ragworms and pellets was <3%. AEs could not even be calculated in the case where turbot were fed with seabream and shrimp (activities measured in turbot were rapidly below the detection limits; Table 1). These results are consistent with the literature, although a wide range of food items was never looked at. Estimated AEs in individuals fed ragworms and pellets ($3 \pm 0.7\%$ and $0.3 \pm 0.8\%$, respectively; mean \pm SD) are similar to the AE of $4.2 \pm 2.8\%$ estimated in plaice *Pleuronectes platessa*, fed with radiolabelled ragworms (46 days of depuration; Pentreath, 1977). To the best of our knowledge, no study has been using pellets as food for studies on ^{110m}Ag AE. However, Rouleau et al. (2000) followed Ag depuration in individuals of the American plaice, *Hippoglossoides platessoides*, fed with a ^{110m}Ag -labelled wet paste (composition close to manufactured pellets; Provencher, 1995); authors found a very low retention of ^{110m}Ag with this food, with an estimated AE of $8.6 \pm 4.8\%$. For this latter study, authors have used gravid females (adults) and thus direct comparison is not possible as we used juvenile fish and sexual maturity may affect the retention of trace elements due to the energy allocation for the oocyte production (Bang et al., 2008).

Some authors are linking AE of elements in predators with the subcellular partitioning of these elements in their prey. The concept of Trophically Available Metal (TAM), as defined by Wallace

and Luoma (2003), states that the fraction of elements found in the soluble fraction of prey should reflect the element fraction that is bioavailable to predators ingesting that prey. In our study, results revealed that although ^{110}mAg was preferentially found in the soluble fraction of prey (50e72%), AEs were always low. Therefore, the positive relationship between TAM and AE stated by Wallace and Luoma (2003) could be true for simple organisms (as shown for bivalves fed phytoplankton; Wang and Fisher, 1996; Reinfelder et al., 1997) but was not verified in the case of our more complex prey (teleost, crustacean and polychaete). Even considering the lower value of soluble fraction of ^{110}mAg (23.7e26.8%) previously found in the whole body *H. diversicolor* by Rainbow et al. (2006), the AE of ^{110}mAg is still too low compared to what would be expected by TAM concept. Possible explanations for this low AE in turbot despite a theoretically important source of bioavailable ^{110}mAg are: (1) the occurrence of neutralization processes such as speciation or complexation of the element due to acidic conditions within the stomach of the fish, that would limit the element accessibility in the digestive tract or (2) the occurrence of detoxification mechanisms in the wall of the digestive tract, preventing ^{110}mAg to penetrate into the internal compartments of the fish. The first hypothesis is backed up with our pellet experiment for which ^{110}mAg was not biological bound but chemically adsorbed. Indeed, although the radionuclide was virtually 100% under “soluble” form in the latter experiment, ^{110}mAg was not assimilated either by the fish. Regarding the second hypothesis, our body distribution results show that most of the assimilated ^{110}mAg is located in the liver and digestive tract and not eliminated (AE component), suggesting storage of the element in these organs and tissues of the turbot. This can be seen in relation to previous observations made, for example, in several bivalves (freshwater and seawater species; Berthet et al., 1992), the sea urchin *Paracentrotus lividus* (Warnau et al., 1996) and the fish *Oryzias latipes* (Chae et al., 2009), where Ag is stored within the intestinal wall as amorphous

Ag (Ag₂S) generally found precipitated in the basement membranes and underlying connective tissues: Berthet et al., 1992).

The occurrence of mechanism(s) impeding the transfer of the element across the fish digestive barrier was also mentioned in several studies, which suggested that intestinal tissues help buffer diet-borne metal uptake (Cu and Zn), by acting as a barrier (Maage and Julshamn, 1993; Clearwater et al., 2000; Handy et al., 2000).

Results from the transfer of ^{110m}Ag to the turbot via manufactured pellets highlighted a low risk of contamination of fish produced in aquaculture in the case of an environmental contamination by ^{110m}Ag. The potential for pellet to accumulate ^{110m}Ag by adsorption exists in the environment but the related AE in fish would be extremely low (Table 1). However, the bioaccumulation of ^{110m}Ag in fish through seawater was not investigated in our study and should be considered to establish a comprehensive risk to fish and consumers.

As radioactive isotopes have nearly identical physical and chemical properties to equivalent stable isotopes, the findings of the present study can also be extended to the stable element (*viz.* Ag) and its bioaccumulation mechanisms in fish (e.g. Metian et al., 2008a; Metian et al., 2010). Here, we demonstrated the low silver retention regardless the 4 type of feeds used (natural prey and manufactured pellet) in a marine fish. This information is also of interest in the context of the occurrence of silver pollution in the marine environment, e.g. in relation to the rapid increase in the use of silver nanoparticles (Savery et al., 2013) or to the urban poor sewage treatment (Sañudo-Wilhelmy and Flegal, 1992).

5. Conclusion

This present study confirmed the low assimilation efficiency of ^{110m}Ag from food in a marine fish, the turbot *S. maximus*, with the use of a range of food items that are the main components of the diet of the fish in natural conditions or in aquaculture production. Results showed a poor assimilation and retention of ^{110m}Ag by the predator fish regardless of the type of food.

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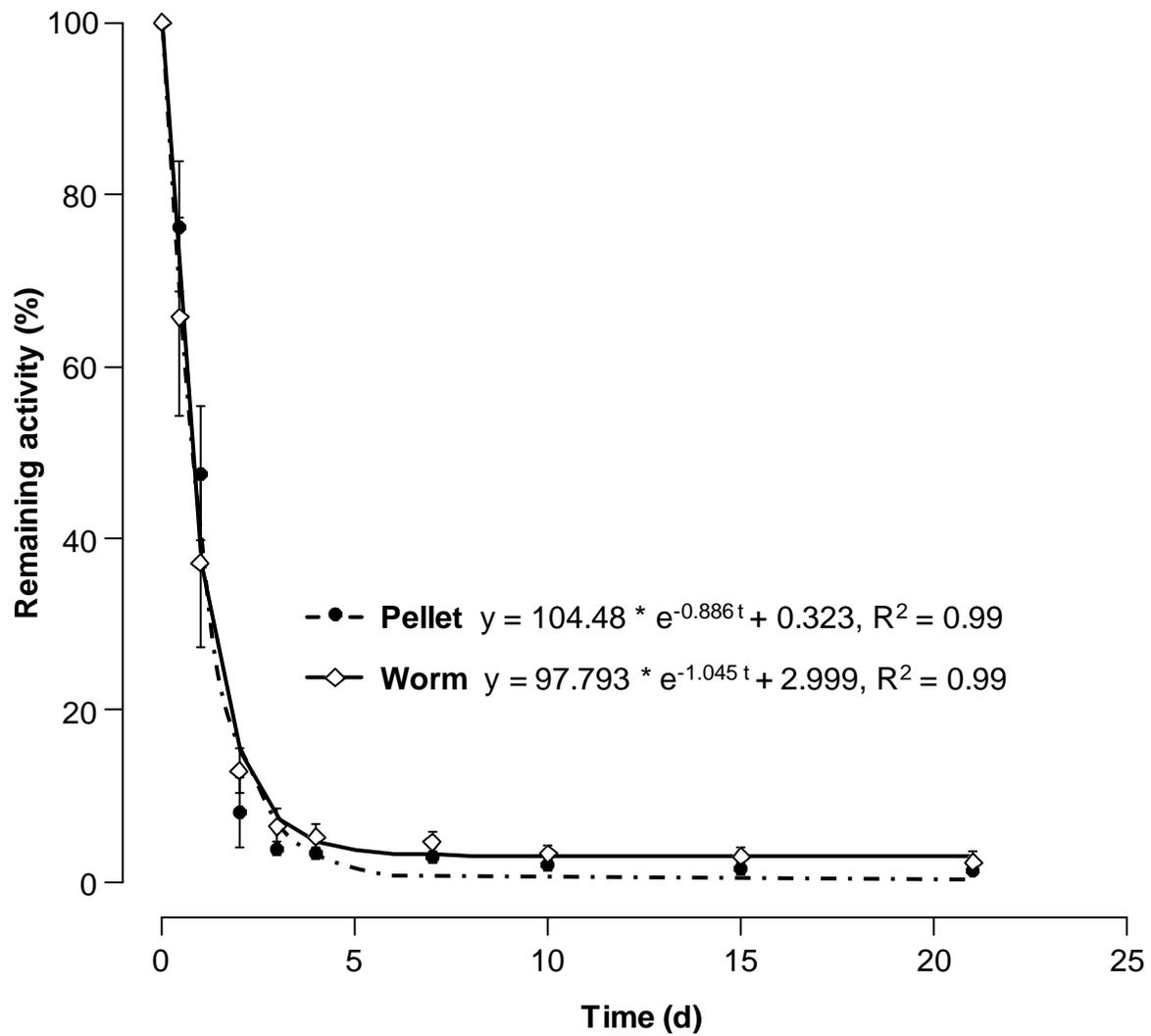


Figure 1. Influence of type of food on whole-body depuration of ^{110m}Ag in turbot (% remaining activities, mean \pm SD).

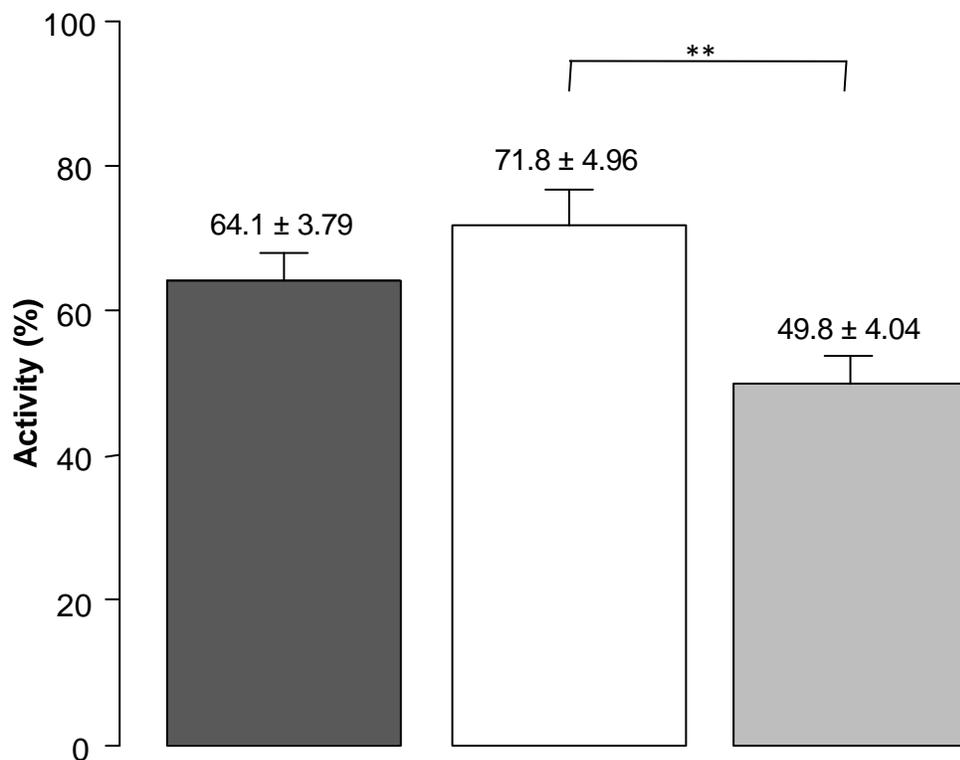


Figure 2. Percentage of ^{110m}Ag in soluble fraction of the three food items (seabream in black, shrimp in white and ragworm in grey, $n = 4$). * $p < 0.05$. ** $p < 0.01$.

Table 1. Activities (Bq g⁻¹ wwt, mean ± SD) in the different feeds (n=4) and in the turbot (n = 8-12) after the single-feeding and at the end of depuration period. Assimilation efficiencies (%) related to the different feeds are also indicated.

Feed	Activity in feed (Bq g ⁻¹)	Activity in turbot		Remaining activity (%)
		(2h after the single-feeding, Bq g ⁻¹)	(end of depuration, Bq g ⁻¹)	
Seabream	7	0.67 ± 0.39	<0.03 ± 0.009 ^a	<6.5 ± 5.2 ^b
Shrimp	37	0.98 ± 0.45	<0.02 ± 0.007 ^a	<2.1 ± 0.6 ^b
Pellet	1072 ^c	15.44 ± 7.17	0.19 ± 0.09	1.3 ± 0.65
Worm	117	11.65 ± 5.61	0.29 ± 0.10	2.3 ± 1.2

^a Values indicated are the detection limits.

^b Values calculated using detection limits.

^c Concentration (Bq g⁻¹) dry weight

*Different from 0 (p< 0.0001)

Table 2. Body distribution (% , mean \pm SD, n = 4) of ^{110m}Ag in the turbot fed with radiolabelled feeds and then maintained for 21d in uncontaminated seawater. Values with high uncertainty (>5%) are shown in italic. Weights were expressed as a percentage of total mass.

Compartments	Seabream ^a		Shrimp ^a		Pellet ^b		Worm	
	Weight (%)	Distribution (%)	Weight (%)	Distribution (%)	Weight (%)	Distribution (%)	Weight (%)	Distribution (%)
Digestive tract	9.7 \pm 2.4	16.1 \pm 14.5	5.9 \pm 0.6	10.7 \pm 2.9	6.0 \pm 0.9	14.0 \pm 2.8	5.4 \pm 0.78	15.5 \pm 4.5
Filets	18.6 \pm 1.5	16.5 \pm 3.0	20.8 \pm 0.6	13.8 \pm 11.2	22.1 \pm 1.6	4.69 \pm 1.4	22.9 \pm 1.8	2.8 \pm 1.2
Gall bladder	<1	5.1 \pm 1.2	<1	7.0 \pm 1.5	<1	2.4 \pm 1.6	<1	2.2 \pm 1.1
Head	23.3 \pm 3.0	21.4 \pm 8.1	20.1 \pm 1.1	15.5 \pm 4.3	20.2 \pm 4.38	18.0 \pm 6.68	26.2 \pm 1.7	12.9 \pm 4.8
Kidneys	<1	13.7 \pm 9.9	<1	5.4 \pm 1.6	<1	1.4 \pm 0.7	<1	<1
Liver	1.2 \pm 0.4	9.0 \pm 3.3	<1	17.8 \pm 15.1	<1	29.8 \pm 13.9	<1	49.7 \pm 17.3
Remaining parts	46.6 \pm 2.0	18.3 \pm 7.6	51.6 \pm 0.7	29.7 \pm 9.7	50.3 \pm 4.8	29.7 \pm 5.1	44.5 \pm 1.18	16.1 \pm 9.1

^aFor these experiments, activities are closed to the detection limits.

^bFor this experiment, counting errors exceed 5% due to the low activities measured.