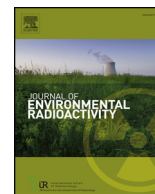




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Radioecological aftermath: Maternal transfer of anthropogenic radionuclides to shark progeny is sustained and enhanced well beyond maternal exposure

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ABSTRACT

Cartilaginous dogfish *Scyliorhinus canicula* continued to transfer four anthropogenic radionuclides (⁶⁵Zn, ⁶⁰Co, ¹³⁴Cs and ²⁴¹Am) to their eggs for over six months, after two months of continued maternal exposure to radio-labelled food. Unexpectedly, rates of radionuclide transfers to eggs and their yolk & embryo during maternal depuration were equivalent for ⁶⁰Co and ²⁴¹Am, or even enhanced for ⁶⁵Zn and ¹³⁴Cs by factors of c.200–350%, over two-three months, compared to their maximal transfer rates at the end of the maternal uptake phase. These rates of maternal transfer of radionuclides to yolk & embryo were positively associated with their previously determined efficiencies of assimilation (AE) from ingested radio-labelled food. Thus progeny may be more exposed via maternal transfer to those radionuclides which have greater rates of maternal assimilation from food. As maternal depuration continued beyond 60–80 up to 180–200 days the transfers of all four radionuclides to eggs did diminish but were still substantial at mean values of 18% for ²⁴¹Am, 17% for ¹³⁴Cs and 9 and 8% for ⁶⁰Co and ⁶⁵Zn, respectively. In the yolk & embryo the mean rates of transfer over this period were further reduced for ²⁴¹Am (13.5%), ⁶⁰Co (2.5%) and ⁶⁵Zn (5.8%), but were still appreciable for ¹³⁴Cs at 56%. These results for *S. canicula* have demonstrated a potential enhanced radiological risk of extended duration due to the particular biokinetics of maternal transfer in this species. This study draws further attention to the current paucity of knowledge about the maternal: progeny transfer pathway, particularly in the context of the known heightened radio-sensitivity of early life stages in fish and other vertebrates, compared to later life stages.

1. Introduction

There is a current paradox in marine radioecology and radio-protection. Radiobiological studies among a range of vertebrate species including teleost fish and humans have repeatedly demonstrated that early life stages are more radio-sensitive than the adult life stages, for equivalent degrees of radiation exposure (International Commission for Radiological Protection, 1990; Freeman et al., 2014; Geiger et al., 2006; Hu et al., 2016; Jaafar et al., 2013; Walker and Streisinger, 1983). Yet there is currently little information on the capacities of early life stages to bioaccumulate radionuclides compared to their adult life stages, as expressed in their comparative biota: water concentration factors (CFs) (Beresford, 2010; Howard et al., 2013). These data are important for radio-protection because any increased bioaccumulation capacities in the early life stage relative to the better-studied adult life stage would further exacerbate its greater vulnerability to radiation impact, relative to the adult phase. There is also a paucity of experimental information

which adequately compares the degrees of exposure of early and adult marine life stages, given that their exposure pathways can be qualitatively different. This concern led to the first study (Jeffree and Johansen, 2017) which particularly investigated this question, based on previous experimental exposures of the encased embryo and free-swimming juvenile life stages of the cartilaginous dogfish *Scyliorhinus canicula*, for their respective normalised exposure pathways for ⁶⁵Zn, ⁶⁰Co, ¹³⁴Cs and ²⁴¹Am (Jeffree et al., 2006a & b; 2007, 2008; 2010, 2015). This study showed that encased embryos can be more exposed to these radionuclides by up to two orders of magnitude compared to the juvenile of the adult life stage, even after short-term exposure periods (14 days), and that the maternal transfer pathway is particularly important in radionuclide transfers to the egg's yolk & embryo. Yolk which is laden with radionuclides via maternal transfer also has potential longer-term consequences for the embryo's absorption of radionuclides during its long developmental period prior to hatching (Lechenaute et al., 1993).

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Another radioecological lag phase was suggested by the linear relationships between cumulative radio-activities which had been maternally ingested and levels in oviposited eggs, which showed no indications of any equilibration being achieved over a period of 61 days of maternal exposure to radiolabelled food (Jeffree et al., 2015). Given this indicated enhanced exposure of the egg and yolk & embryo via the maternal pathway (Jeffree and Johansen, 2017), and the non-equilibrium status of the maternal transfers to eggs, even after 61 days of maternal exposure to radio-labelled food (Jeffree et al., 2015), it was also important to investigate the duration and magnitude of the exposure of progeny to this maternal radio-contaminant legacy, once maternal exposure to radionuclides has actually ceased.

This experimental study provides further and complementary bio-kinetic data on maternal transfers of ^{65}Zn , ^{60}Co , ^{134}Cs and ^{241}Am to eggs and their yolk & embryo during a maternal depuration period of more than 6 months. These patterns of maternal: egg transfer of radionuclides during maternal depuration were also interpreted in relation to previous results on their differential rates of assimilation from food (Mathews et al., 2008) by dogfish and also their rates of transfer to eggs during maternal uptake (Jeffree et al., 2015).

2. Materials and methods

2.1. Maternal histories of exposure to radionuclides prior to maternal depuration

This depuration experiment followed on directly from a period of maternal uptake of radionuclides from radio-labelled mussel tissues (*Mytilus edulis*), which were consumed within a period of up to 61 days of food provision, as detailed in Jeffree et al. (2015). *M. edulis* had previously been chosen as an experimental food because of the prominence of bivalve molluscs in the natural diet of *S. canicula* with increasing frequency in larger individuals (Lyle, 1983).

The average daily weight of ingested food for each of three maternal dogfish (70–80 cms total length range) were 1.5, 2.8 and 0.03% food ingested/body weight/day, of which the two higher values occurred within the range which has been determined for various other Chondrichthyans (Wetherbee and Cortes, 2004; Holmgren and Nilsson, 1999). The four radionuclides used in this previous study, zinc-65, cobalt-60, americium-241 and cesium-134, are typically associated with effluents entering the marine environment from coastal nuclear facilities and are thus available for subsequent bioaccumulation (e.g. Alexander and Rowland, 1966; Whicker and Schultz, 1982).

2.2. Experimental conditions of maternal dogfish during their depuration of radionuclides

These three female dogfish that produced eggs during the uptake phase of the experiment were maintained during their depuration phase in a 3000 L tank under experimental conditions that were the same as those used for their experimental exposure to radiolabelled food over 61 days. They continued to be partitioned into corrals of 750 L volume under separate confinement so that their individual rates of egg production and patterns of ingestion of radionuclide-free food could be determined and which was offered to them for up to 233 days, during five days (Monday-Friday) per week. Following the delivery of food portions of measured weight to each individual dogfish, the residual food that was not consumed within 15 min was then retrieved by pipette in order to determine, by subtraction, the food weight that was actually ingested by each fish, where rates of consumption were similar to those observed during the maternal uptake phase (Section 2.1).

2.3. Radio-analysis of eggs and their yolk & embryo

A high-resolution gamma spectrometry system was used for all radiometric analyses of dogfish reproductive organs, total egg and yolk

& embryo samples, consisting of four coaxial Germanium (N- or P-type) detectors (EGNC 33–195-R, Intertechnique; 40–70% efficiency). The detectors were connected to a multi-channel analyzer and a personal computer employing spectral analysis software (Interwinner 6, Intertechnique). The radioactivity levels of samples were determined by comparison with known standards of appropriate geometry, including a phantom dogfish egg, and were corrected for background and physical decay of a given radioisotope. During counting the specimens were situated consistently relative to the counter. Counting times were adapted to obtain count rates with relative propagated errors of less than 5%, viz. typically 10–30 min for whole egg and yolk & embryo radio-analyses. The radio-analytical techniques used in this study were identical to those used in previous experiments on shark eggs and their components (Jeffree et al., 2006b, 2015).

2.4. Data calculations and statistical analyses

In a previous study on juvenile *S. canicula* that had also absorbed these radionuclides from food, the whole body biological half-lives in the long-lived compartment of *S. canicula* were (in days): ^{57}Co -32, ^{134}Cs -87 and ^{65}Zn -58, with no significant efflux rate constant being measured for ^{241}Am over 20 days (Mathews et al., 2008). Similarly, maternal depuration of the radionuclides (previously accumulated from radio-labelled food) to the external milieu would be expected to result in diminishing maternal body burdens, as previously determined experimentally for *S. canicula* juveniles over 29 days of exposure to radio-tracer free seawater (Jeffree et al., 2006a), and consequently their reduced maternal transfer to eggs.

The previous study (Jeffree et al., 2015) of the maternal transfer of these radionuclides demonstrated significant ($p < 0.05$) linear relationships (constrained through the origin) between the total Bq maternally ingested in radio-labelled food and those in a) eggs and b) yolk & embryo, for each radionuclide. These linear regression equations which varied considerably in slope among radionuclides (Table 1) were used to predict the total Bq in eggs and yolk & embryo at the last recorded day of maternal ingestion of radio-labelled food, which was then used as the benchmark against which to assess the rates and patterns of maternal transfers of each radionuclide to eggs and their yolk & embryo during maternal depuration. Although the consumption of radio-labelled food was terminated, the maternal transfer to eggs from ingested radionuclides could be expected to continue for a period which represented a lag between maternal ingestion and transfer to the egg. Factors which had been identified as contributors to this lag period included food digestion and absorption into the blood, transfer to those organs responsible for egg production, and the time between when the egg construction was complete and its oviposition. This lag period was

Table 1
Radionuclide assimilation efficiencies (AE) and maternal-to-egg/yolk & embryo transfer rates during maternal uptake (mTF) and depuration.

Radionuclide	Percentage Assimilation Efficiency (AE) ^a	mTF slope ^b ($\times e3$)	Averaged percentage transfer during the heightened phase of maternal depuration
<i>Egg</i>			
^{65}Zn	17	0.7	252
^{134}Cs	73	1.0	89
^{60}Co	11	0.3	63
^{241}Am	6	0.17	80
<i>Yolk & Embryo</i>			
^{65}Zn	17	0.7	194
^{134}Cs	73	0.2	253
^{60}Co	11	0.2	82
^{241}Am	6	0.1	55

^a From Mathews et al. (2008).

^b From Jeffree et al. (2015).

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