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Temperature Effects on Uptake and Retention of Contaminant Radionuclides and Trace Metals by the Brittle Star *Ophiothrix fragilis*

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ABSTRACT

Recent revelations of extensive dumping of radioactive wastes in Arctic seas emphasize the need to understand the processes affecting accumulation of contaminants in polar marine food webs. Little is known, however, about the effects of low temperatures on bioaccumulation of radionuclides and toxic metals. To address this question, we examined the effects of temperature on uptake and retention of 11 dissolved radioisotopes (including both nuclear waste components and required, and toxic metals) by the brittle star Ophiothrix fragilis, *typical of species which dominate Arctic benthic communities. Lower temperatures significantly reduced uptake rates of all elements examined, but had little effect on loss rates. These results raise questions about the validity of extrapolating previous work on biological dynamics of dissolved contaminants, largely carried out at temperate zone temperatures, to polar ecosystems. This work suggests that the effects of low Arctic temperatures may need to be taken into consideration in order to understand the potential for food chain accumulation of nuclear wastes and toxic metals in high-latitude seas. Copyright © 1996 Elsevier Science Ltd*

INTRODUCTION

Industrialized northern countries release large quantities of heavy metals and radionuclide contamination (from nuclear accidents, fallout from weapons testing and nuclear wastes) into high-latitude temperate or Arctic seas (Presley, 1994; Mount *et al.*, 1994). The Kara Sea, in particular, is a site of considerable interest due to the release of almost 100,000 TBq of radioactive waste in this region since the 1950s (White Book 3, 1993). Most of the radionuclides present are fission products (Mount *et al.*, 1994). Despite the elevated contaminant inputs to northern and polar oceans, little attention has been given in the radioecological trace element literature to the effects of low temperatures on the bioaccumulation of radioactive wastes and other potentially toxic trace elements in marine food chains. Since metabolic effects of cold temperatures in ectothermic marine organisms could influence uptake and mobilization of contaminants by the biological community, the results of experiments conducted at temperate zone temperatures may not always be applicable to cold-water marine habitats (Boisson *et al.*, in press; Hutchins *et al.*, 1996).

To address this question, we examined the uptake and retention of a wide variety of radionuclides from seawater by the ophiuroid *Ophiothrix fragilis* (Echinodermata, Ophiuroidea) at two temperatures, one typical of polar regions and one more representative of temperate waters. We examined this organism because ophiuroids and other benthic echinoderms have been shown to effectively concentrate a number of heavy metals, radioactive fission and activation products, and transuranic isotopes from seawater (Grillo *et al.*, 1981; Galey *et al.*, 1983; Fowler & Carvalho, 1985; Chassard-Bouchaud *et al.*, 1988; Fowler & Teyssié, 1995; Hutchins *et al.*, 1996). The ability of ophiuroids to retain pollutants, along with their high relative abundance in northern and polar benthic communities (Miquel, in press), has led to suggestions that they may be useful as bioindicators of contaminant exposure (Chassard-Bouchaud *et al.*, 1988).

We examined the biological interactions of several gamma-emitting radioisotopes of anthropogenic contaminant elements with contrasting geochemical characteristics. These included components of the disposed nuclear wastes in the Kara Sea (^{241}Am, ^{152}Eu, ^{106}Ru, ^{60}Co, ^{134}Cs, ^{133}Ba) as well as radioisotopes of required or toxic trace metals (Cd, Ag, Zn, Mn, Co-cobalamine). The chemical behavior of this group of elements ranges from highly particle-reactive (Am, Eu, Ru) to relatively non-particle-reactive (Ba, Cs) in seawater. Biological characteristics of these elements also differ; some are not biologically required and can exert toxic effects (Am, Eu, Ru, Cd, Ag), others are required micronutrients in small amounts but can be toxic at higher concentrations (Zn, inorganic Co). Still others are micronutrients which are usually not toxic at environmentally realistic concentrations (Mn, Co-cobalamine).

Chemical speciation can also affect contaminant uptake by the biota. For instance, Co is accumulated more efficiently as Co-cobalamine (vitamin B12) than as inorganic Co by some marine organisms (Nolan *et al.*, 1992). To examine speciation effects on the uptake of this activation product, an important component of the Kara Sea wastes (Mount *et al.*, 1994), we employed a double-labeling technique (Nolan *et al.*, 1992) to compare ophiuroid bioaccumulation of inorganic Co-60 and cobalamine Co-57.

METHODS

Ophiuroids (*Ophiothrix fragilis*) were collected in February, 1995 by divers at a depth of 24 m from nearshore waters of the North Sea (Wermeldinge, Zealand, the Netherlands). Ambient temperature and salinity were 5–6°C and 33–34 ppt. In the laboratory, the ophiuroids were gradually acclimated to experimental conditions and temperatures (2°C or 12°C, 34 ppt salinity) over a period of several days.

Radioisotope uptake experiments were conducted using 15 animals held in 1500 ml of 34 ppt 0.2 μm-filtered Mediterranean surface water (2°C) or five animals in 500 ml of the same filtered seawater (12°C). Animals of similar size (0.9–1.5 g) were used to minimize the possibility of size-related differences in uptake and retention of the radioisotopes. Ophiuroids were fed just prior to the experiments and were then held without feeding during the uptake and depuration periods.

Exposure to gamma-emmiting isotopes was accomplished using two isotope mixtures at each temperature. Isotopes added to each mixture were chosen to minimize spillover of gamma emissions between detection windows. Mixture 1 contained ^{133}Ba (9.96 Bq/ml), ^{152}Eu (7.15 Bq/ml), ^{106}Ru (7.11 Bq/ml), and ^{54}Mn (6.75 Bq/ml). Mixture 2 consisted of ^{241}Am (6.84 Bq/ml), ^{109}Cd (10.30 Bq/ml), cobalamine ^{57}Co (6.05 Bq/ml), inorganic ^{60}Co (5.34 Bq/ml), ^{65}Zn (5.36 Bq/ml), ^{110}Ag (7.30 Bq/ml), and ^{134}Cs (8.95 Bq/ml). Isotopes were added as microliter quantities in 0.5N HNO_3 or HCl (except ^{57}Co-cobalamine, added as a benzyl alcohol solution), and acid additions were neutralized by appropriate quantities of 1N NaOH to yield a final pH of 7.8–8. Because many of the isotopes were rapidly accumulated by the animals and depleted from the water, isotope mixtures were renewed every 2–3 d during the uptake period.

Gamma activities of the samples were measured using a high-resolution germanium detector (Intertechnique Nucleaire, EGNC 33-195-R). Measurements of gamma energy peaks for isotopes in Mixture 1 were made at 356.0 keV (^{133}Ba), 511.9 keV (^{106}Ru), 834.8 keV (^{54}Mn), and 1408.0 keV (^{152}Eu); for Mixture 2, peak energies measured were 59.5 keV (^{241}Am), 88.0 keV (^{109}Cd), 122.1 keV (^{57}Co), 604.7 keV (^{134}Cs), 657.8 keV (^{110}Ag), 1115.6 keV (^{65}Zn), and 1332.5 keV (^{60}Co). Corrections for background, energy spillover between detection windows and radioactive decay were made using Intergamma software. For each type of sample (animal or water), the detector was calibrated using activity standards in the appropriate geometries. Counting times were adjusted to yield propagated counting errors of $< 6\%$ during uptake and $< 13\%$ during depuration.

Uptake and loss of all isotopes (except ^{106}Ru) were monitored non-destructively on live animals for 12–14 d periods. Because insufficient ^{106}Ru isotope was available to complete the uptake portion of the experiment, uptake for this isotope at 12°C was monitored for only 6 d, and the depuration period at this temperature was therefore extended to 24 d. Animals were thoroughly rinsed with unlabeled seawater before counting to minimize activity carryover from any adhering radioactive water.

Uptake data for all elements are reported as pmol/g live weight. Specific activities of each element were calculated as Bq/litre added, divided by the sum of the concentrations of added isotope carrier (if any) and ambient seawater levels (M). Ambient levels for Mediterranean surface seawater were obtained from literature values where available, and from other surface seawater values when Mediterranean data were not available. Ambient seawater concentrations used in these calculations are presented in Table 1 along with molar concentrations and calculated specific activities of the added isotopes. For some radionuclides or metals, the addition of the radioisotope represented a significant increase over ambient seawater levels (Am, Ag, Co-cobalamine, Eu, Ba and Ru). For the other elements (Cs, Zn, Cd, inorganic Co, and Mn), molar amounts added were substantially below ambient concentrations. Because of the possibility of inadvertant contamination of the trace metals Zn, Ag, Co, Cd and Mn, molar uptake values for these elements should be taken to represent minimum values. Uptake of the other elements probably represents true molar values, as contamination of these elements is likely to be negligible. At the end of the uptake period, 14 d concentration factors were calculated as Bq/g animal divided by the average activity in the water (Bq/ml). It should be noted that in some

TABLE 1

Radioisotopes Added to Mixtures 1 & 2, Including Concentrations of Activities Added (Bq/liter), and Total Molar Concentrations Added with Radioisotope (Including Radioisotope and Carrier if any, mols/liter). Specific Activities Added were Calculated as the Ratio of Radioactivity Added (Bq/liter) to Total Mols of Element (mols/liter Added With Isotope + Estimated mol/liter Ambient Concentration). Values for Estimated Ambient mols/liter were Obtained from Literature Values for Mediterranean Surface Seawater (Where Available) or from Values for Other Surface Seawater where Mediterranean Values were Not Available

Radioisotope	*Bq/liter added*	*Mols/liter added (radioisotope + carrier)*	*Estimated ambient mols/liter*	*Reference for ambient molar concentrations*	*Calculated specific activity (Bq/mol total)*
Mixture 1					
^{133}Ba	9.96×10^3	9.1×10^{-6}	3.2×10^{-8}	Bruland, 1983	1.09×10^9
^{152}Eu	7.15×10^3	1.2×10^{-10}	9×10^{-13}	Bruland, 1983	5.92×10^{13}
^{54}Mn	6.75×10^3	1.4×10^{-10}	1×10^{-9}	Statham *et al.*, 1985	5.92×10^{12}
^{106}Ru	7.11×10^3	3.7×10^{-12}	6.9×10^{-12}	IAEA, 1985	6.71×10^{14}
Mixture 2					
^{110}Ag	7.30×10^3	2.81×10^{-9}	1.7×10^{-12}	Flegal *et al.*, 1995	2.60×10^{12}
^{241}Am	6.84×10^3	2.4×10^{-10}	1×10^{-19}	Fisher *et al.*, 1983a	2.85×10^{13}
^{109}Cd	10.30×10^3	2.37×10^{-12}	7.5×10^{-14}	Spivack *et al.*, 1983	1.36×10^{12}
^{57}Co(cobalamine)	6.05×10^3	5.5×10^{-13}	7.4×10^{-14}	Nolan *et al.*, 1992	9.71×10^{15}
^{60}Co (inorganic)	5.34×10^3	1.4×10^{-13}	6.8×10^{-11}	Huynh-Ngoc *et al.*, 1989	7.87×10^{13}
^{134}Cs	8.95×10^3	1.8×10^{-10}	2.2×10^{-9}	Bruland, 1983	3.75×10^{12}
^{65}Zn	5.36×10^3	2.44×10^{-12}	4×10^{-9}	Sherrell & Boyle, 1988	1.34×10^{12}

cases these values may not necessarily be those that would be obtained at steady state (see below).

Depuration was monitored by placing the labeled ophiuroids into individual containers with 250 ml of unlabeled, 0.2-μm filtered seawater at the appropriate temperatures and measuring the loss of radioactivity in the animals periodically. Seawater in the depuration containers was changed daily to minimize recycling of isotopes.

At the end of the uptake and depuration periods in the 2°C experiment, animals were removed for simple dissections for initial characterization of isotope distributions between arms and central discs. Dissected samples were solubilized in 10 ml HCl before counting to assure even distribution of isotopes and simplify counting geometries.

RESULTS

Results of the uptake portion of the experiment for all 11 elements (pmol/g/d) are presented in Fig. 1. Molar amounts accumulated by the ophiuroids during the 13 d uptake period ranged from <0.1 pmol/g (Eu, Ru) to >14 nmol/g (Ba) at 12°C and from <0.06 pmol/g (Eu, Ru) to >8nmol/g (Ba) at 2°C. Despite this five-order-of-magnitude range in molar uptake, patterns of uptake were similar for all elements examined. Accumulation was more rapid at 12°C than at 2°C; at the higher temperature, uptake was biphasic with an initial rapid uptake rate during the first 2 d being followed by a slower rate for the rest of the uptake period. Accumulation at 2°C could be modeled by a single linear rate for the entire uptake period.

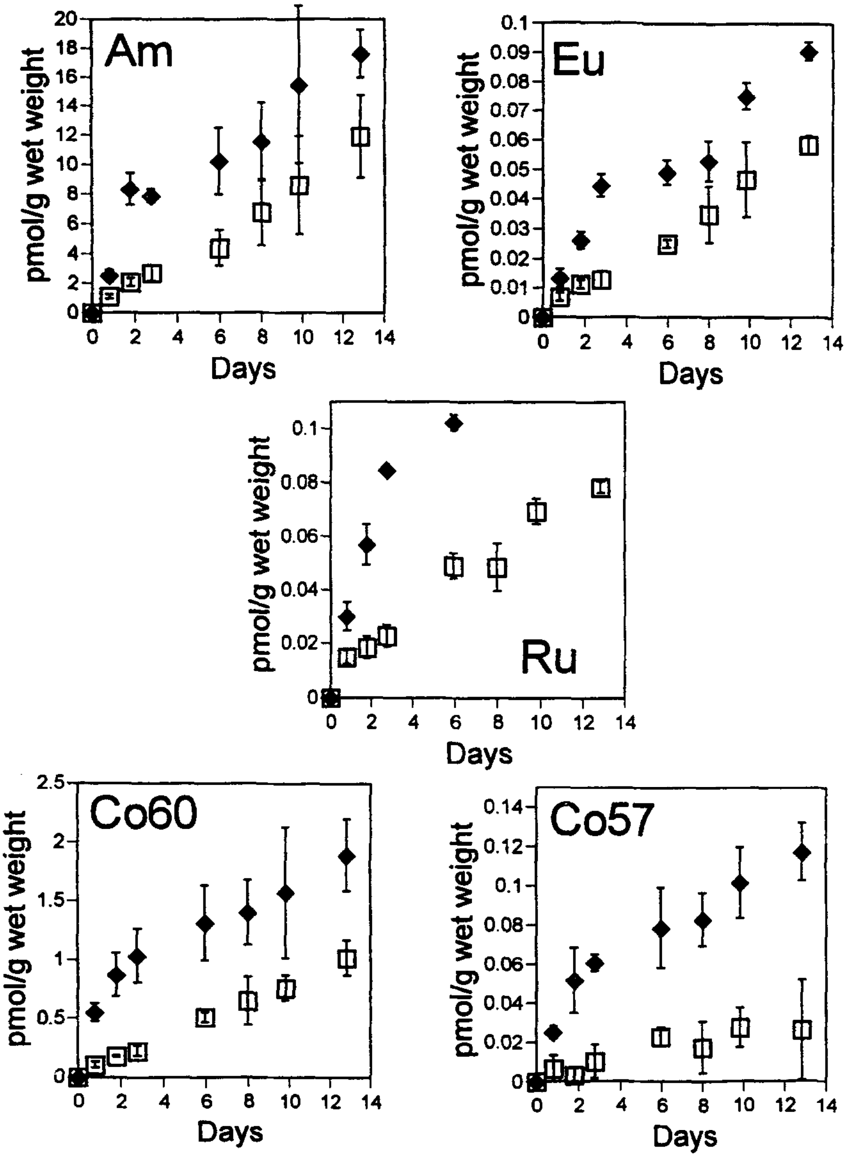

Fig. 1. Uptake of 11 elements (pmol/g) at 2°C (open squares) and 12°C (closed diamonds). Error bars represent the standard deviation of three or four individual animals.

Initial uptake rates (first 2 d) at both temperatures are compared in Table 2. Q_{10} values were calculated with the initial uptake rates at 2°C and at 12°C. Most elements exhibited uptake Q_{10} values between 3.2 and 4.6 (Ag, Am, inorganic Co, Cs, Eu, Mn, Ru and Zn). The Q_{10} for Ba uptake (1.5) was significantly below this range, and values for Co-cobalamine (7.7) and Cd (12.2) were substantially higher.

Table 2 also presents 14 d concentration factors (CF_{14d} = mol/g animal divided by mol/ml water in the dissolved phase) at the end of the uptake period, as well as CF_{14d} Q_{10} values (the ratio of the CF_{14d} at 12°C to the CF_{14d} at 2°C). In all cases, concentration

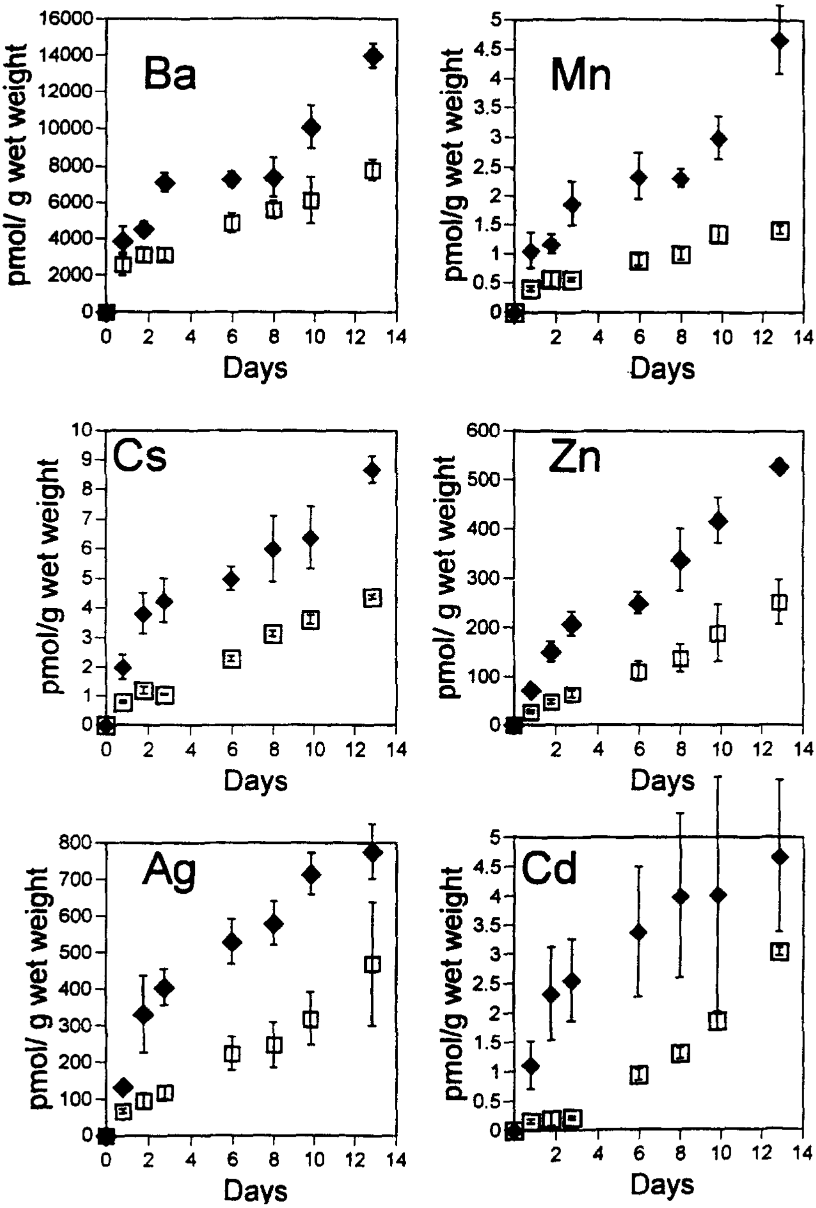

Fig. 1. Contd.

factors were calculated based on a running average of measured activities in the water. All elements were concentrated more at the higher temperature, with Co-cobalamine showing the greatest response to temperature and Ru the smallest of all the elements tested. Ag was concentrated most effectively at both temperatures; other elements with relatively large CF_{14d} values (> 30) included Am, Cd, Co-cobalamine, Eu, and Zn. Ba and Mn were not concentrated at 2°C (CF_{14d} = 1), and along with Cs exhibited the lowest CF_{14d} values at both temperatures. The high Q_{10} values derived from initial uptake rates may

TABLE 2

Initial Uptake Rates (pmol/g/d) and Resulting Q_{10} Values for Each Element at 2°C and 12°C. Also Shown are 14 d Concentration Factors (CF_{14d}) for Each Element at the End of the Uptake Period at Both Temperatures and Final CF_{14d} Q_{10} Values

	Ag	Am	Ba	Cd	Co (cobalamine)	Co (inorganic)	Cs	Eu	Mn	Ru	Zn
Initial uptake rate (pmol/g/d)											
2°C	4.10×10^{1}	9.76×10^{-1}	1.60×10^{3}	7.45×10^{-2}	2.93×10^{-3}	7.97×10^{-2}	3.77×10^{-1}	4.54×10^{-3}	1.94×10^{-1}	7.64×10^{-3}	2.27×10^{1}
12°C	1.52×10^{2}	3.18×10^{0}	2.37×10^{3}	9.56×10^{-1}	2.25×10^{-2}	3.67×10^{-1}	1.56×10^{0}	1.58×10^{-2}	6.12×10^{-1}	3.04×10^{-2}	7.56×10^{1}
Q_{10}	3.7	3.3	1.5	12.2	7.7	4.6	4.1	3.5	3.2	4.0	3.3
14 d Concentration Factor (CF_{14d})											
2°C	271	48	1	34	69	16	3	62	1	9	78
12°C	480	53	2	46	360	29	4	102	5	10	166
CF_{14d} Q_{10}	1.8	1.1	2	1.4	5.2	1.8	1.3	1.6	5	1.1	2.1

overemphasize temperature effects in the long term when compared to CF_{14d} Q_{10} values derived from final 14 d concentration factors (Table 2). CF_{14d} Q_{10} values ranged from 1.1 (Am, Ru) to 5.2 (Co-cobalamine), substantially lower than uptake Q_{10} values, but nevertheless clearly demonstrating temperature enhancement of elemental accumulation during the two week exposure period. The effect of temperature on CF Q_{10} values at steady state (not reached in these experiments — see discussion) is unknown.

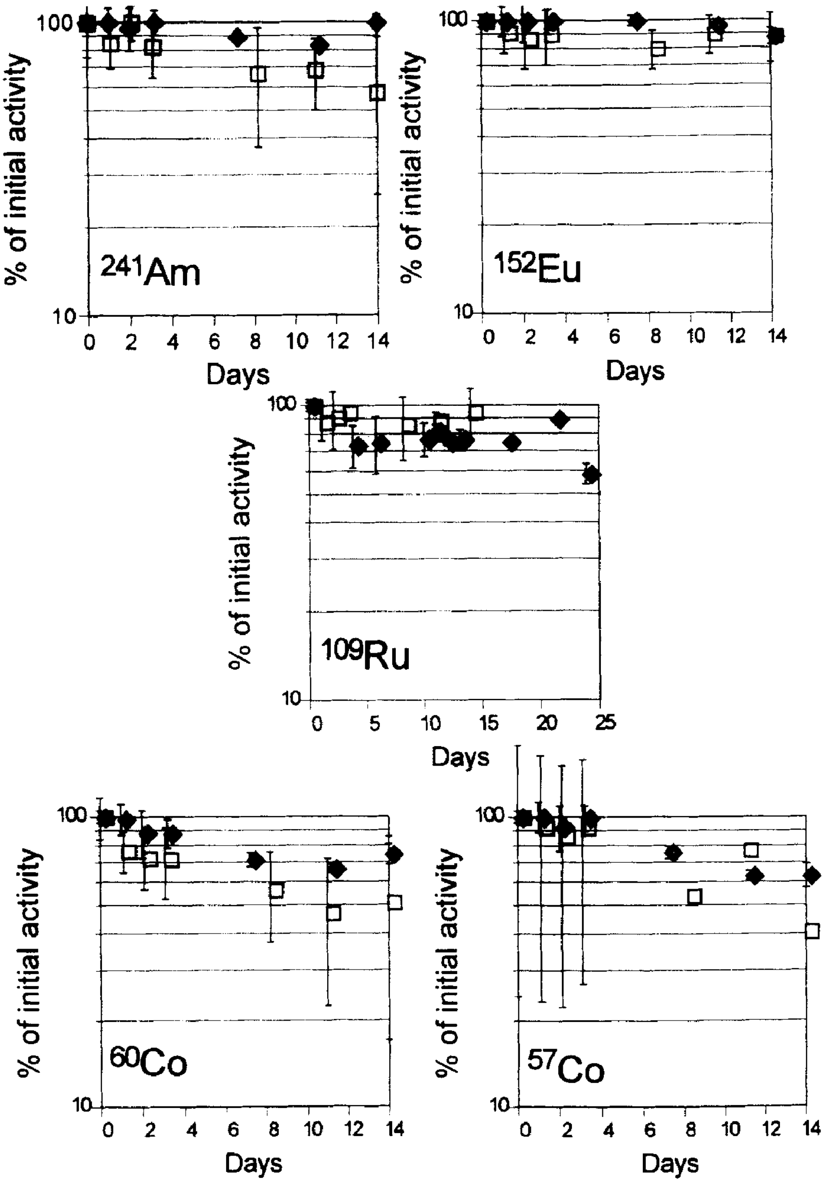

Fig. 2. Retention of 11 elements over a 14 d depuration period, expressed as the percent remaining of the element present in the animals at the end of the uptake period (time 0 of depuration). Symbols and error bars as in Fig. 1.

Retention of the 11 radioisotopes in the ophiuroids, at both temperatures, is illustrated in Fig. 2. Variability in loss rates between replicate individuals during the 14 d depuration period was greater at 2°C than at 12°C, and was especially large for Co-cobalamine and Cd (note error bars, Fig. 2). However, some of the individual variability, especially for Cd, was likely to be due to lower total activities and therefore larger propagated counting errors during the depuration period than during uptake.

Loss during the two week depuration period could be best described by a one compartment model at both temperatures for most of the isotopes. However, loss of Ba, Mn

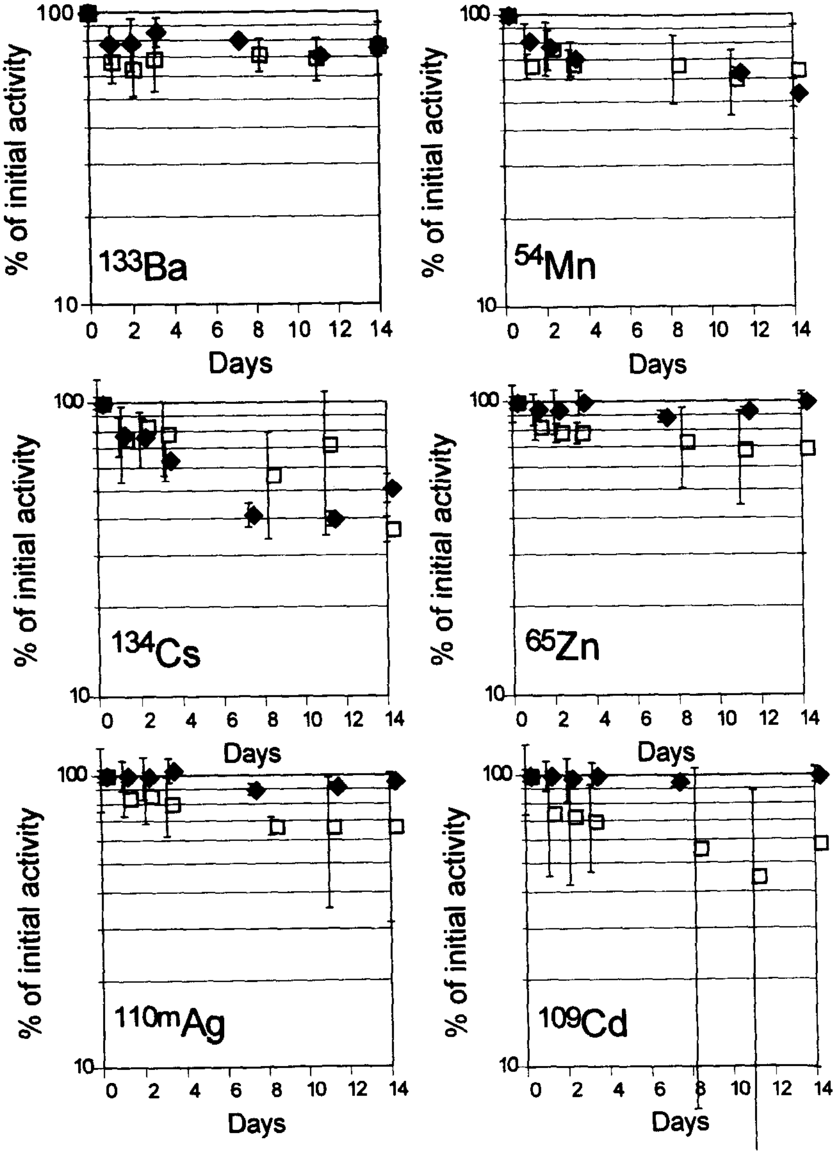

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TABLE 3

Loss Rates (% d^{-1}) and Biological Half-Lives for the Long-Term Pool (Days 2–14 of Depuration) for Each Isotope at 2°C and 12°C. 0^* Denotes Slopes Which are Not Significantly Different from Zero at $P < 0.05$

	Ag	Am	Ba	Cd	Co (cobalamine)	Co (inorganic)	Cs	Eu	Mn	Ru	Zn
2°C											
loss rate (% d^{-1})	0*	3.00	0*	0*	0*	2.16	0*	0*	0*	0*	0.93
$tb_{1/2}$ (days)	∞	16.2	∞	∞	∞	14.3	∞	∞	∞	∞	55
12°C											
loss rate (% d^{-1})	0*	0*	0*	0*	0*	1.55	2.23	0*	1.70	0*	0*
$tb_{1/2}$ (days)	∞	∞	∞	∞	∞	35.4	18.0	∞	27.9	∞	∞

and Cs appeared to conform with a two compartment model, with a rapid loss in the first 2 d followed by a slower loss for the remainder of the experiment. For this reason, loss rates for all isotopes were calculated between days 2 and 14 (Table 3). Loss rates were calculated using the equation:

$$A_t = A_0(e^{-kt}),$$

where A_0 and A_t are the activities at the beginning and end of depuration, respectively and t = 12 d. Loss rates (% d^{-1}) were calculated based on linearization of ln-transformed data, and in all cases $r^2 \geq 0.6$.

For most of the elements examined, there was no significant difference in long-term loss rates at 2°C and 12°C (Student's *t*-test, $P < 0.05$). Although visual examination of Fig. 2 suggests loss of the slowly exchanging pool for many of the elements examined, at the $P < 0.05$ confidence level, loss rates significantly different than zero for Ag, Ba, Cd, Co-cobalamine, Eu, and Ru at either temperature were not detectable (Table 3). However, loss rates were more rapid at 2°C than at 12°C for Am, inorganic Co, and Zn, and more rapid at 12°C than at 2°C for Cs and Mn. No consistent trend for the effects of temperature on loss rates was observed during the 14 d depuration period, and in most cases rates were very similar at both temperatures (Fig. 2, Table 3). These data suggest that temperature variations within the range examined here do not exert a major influence on excretion of elements obtained from the dissolved phase by ophiuroids, at least in relatively short-term loss experiments such as those presented here.

Concentrations of these elements in the discs and arms are compared at the end of the uptake and depuration periods in Table 4. After 13 d of uptake, there was no significant difference ($P < 0.05$) in arm and disc concentrations for any element except Eu, which was more enriched in arms. After depuration, the only significant differences were for Ba and cobalamine-Co, which were more enriched in discs. For all other elements examined, arm and disc concentrations were not significantly different after either uptake or depuration.

TABLE 4

Concentrations of Each Element (pmol/g ± std dev.) Accumulated in Ophiuroid Discs and Arms at the End of the 2°C Uptake Period and Remaining at the End of the 2°C Depuration Period. Bold Values are Significantly Different for Arms and Discs ($P \leqslant 0.05$)

	End of uptake (n = 6)		End of depuration (n = 4)	
	Discs	Arms	Discs	Arms
Ag	340 ± 93	410 ± 100	350 ± 79	290 ± 14
Am	10 ± 2.7	15 ± 3.9	8.6 ± 0.9	9.6 ± 1.7
Ba	10500 ± 1220	10600 ± 917	**6600 ± 96**	**5200 ± 545**
Cd	1.4 ± 0.6	1.9 ± 0.5	1.96 ± 1.3	1.93 ± 0.08
Co cobalamine	0.033 ± 0.021	0.025 ± 0.018	**0.026 ± 0.0016**	**0.013 ± 0.0018**
Co inorganic	0.7 ± 0.079	0.93 ± 0.12	0.51 ± 0.16	0.53 ± 0.025
Cs	5.2 ± 1.5	4.1 ± 0.86	2.5 ± 0.48	1.8 ± 0.31
Eu	**6.6 ± 1.3**	**7.8 ± 1.3**	5.4 ± 0.61	5.2 ± 0.11
Mn	2.1 ± 0.21	1.7 ± 0.13	0.84 ± 0.38	0.65 ± 0.2
Ru	0.121 ± 0.012	0.101 ± 0.012	0.075 ± 0.019	0.061 ± 0.034
Zn	230 ± 70	240 ± 32	250 ± 65	140 ± 20

DISCUSSION

The results of these experiments demonstrate temperature-enhanced initial uptake rates from water of all elements examined. In contrast, depuration was relatively unaffected by temperature over the 14 d time frame of the loss experiments. Increases in accumulation rates at higher temperatures may be partially due to increased metabolic activity. Animals were observed to be much more active at 12°C than at 2°C, and increased internal circulation of water through the respiratory and digestive systems may result in increases in the amounts of all elements bound to internal body surfaces. Additionally, production of metal-binding proteins or external mucus may be greater at elevated temperatures. Increases in proteins rich in cysteine or other sulfhydryl-containing residues, such as metallothioneins, would especially affect elements such as Ag, Cd, and Zn, which have very high affinities for sulfur (Nieboer & Richardson, 1980).

These results suggest that initial accumulation of most contaminant elements from the dissolved phase by ophiuroids should be reduced in polar environments in comparison to exposure to the same concentrations in temperate marine waters. Animals from temperate waters could also show a seasonal cycle in contaminant uptake, with accumulation rates being higher during warmer months and reduced during the winter. Seasonal changes in uptake or retention are suggested by the observations of Chassard-Bouchaud *et al.* (1988), who observed elevated elemental concentrations in *O. fragilis* individuals collected in spring in comparison to those collected in autumn.

However, once accumulated from water, contaminants may be lost at nearly identical rates in both high and low temperature regimes. The very long retention times for most of the elements examined here at both temperatures ($tb_{1/2}$ values of the slowly exchanging pool for most elements were not significantly different from infinity) support the view that ophiuroids would be excellent bioindicators of ambient dissolved radionuclides and trace metals, efficiently retaining a record of their exposure to soluble contaminants. Long retention times should also increase the potential for trophic transfer of contaminants to predators (such as flatfish) which consume ophiuroids. However, exposure from food sources, not investigated in these experiments, should also be considered since assimilation by this route could substantially alter total depuration rates.

Concentration factors varied widely between elements, but indicate that ophiuroids would be especially appropriate bioindicators for exposure to Class B or borderline metals such as Ag, Zn, and Cd, as well as Co in cobalamine form. In particular, Ag is concentrated very effectively by this species, and ophiuroids might be ideal sentinel organisms for exposure to this contaminant, which is commonly enriched in sewage outflows (Sanudo-Wilhelmy & Flegal, 1992). Our results also suggest that ophiuroids might not be as effective as bioindicators for components of nuclear wastes such as those in the Kara Sea material, including ^{241}Am, ^{152}Eu, and ^{106}Ru. Concentration factors for these particle-reactive isotopes were lower than those reported for similar exposure periods using other potential bioindicator organisms, such as sea stars or bivalves (Bjerregaard *et al.*, 1985; Fowler & Teyssié, 1995; Guary *et al.*, 1995; Hutchins *et al.*, 1996). Concentration factors of the less particle-reactive elements such as Cs, Ba, and Mn were very low, suggesting little or no accumulation from water at environmentally realistic levels.

Accumulation of cobalamine-Co in these experiments was much higher than that of inorganic Co, similar to results obtained in experiments on uptake and retention from water by fish (Nolan *et al.*, 1992). Chemical speciation could have important effects on contaminant uptake and retention by marine invertebrates, especially since many other metals (including Zn, Cd, and Cu) are known to be dominated by organic complexation in seawater (Bruland *et al.*, 1991).

The results of these experiments, involving relatively short-term exposure to radioisotopes, suggest the concentration factors that ophiuroids might display under field conditions following a pulsed release from radioactive waste dumpsites such as those in the Kara Sea. None of the elements examined appeared to exhibit uptake saturation kinetics in the time frame of these experiments (about 2 weeks). The relatively linear uptake rates at both temperatures (after the initial rapid uptake at 12°C; Fig. 1, Table 2) suggest that an equilibrium had not been reached for radioisotope partitioning to the animals; hence the elemental concentration factors presented here are not based on steady-state conditions. Animals exposed to chronic low environmental levels of contaminants for months, years or a lifetime can be expected to concentrate these materials to a greater extent (i.e. higher concentration factors) than would animals acutely exposed for days or weeks.

For instance, the concentration factors obtained in this study after two weeks of exposure to ^{241}Am were about 50, consistent with the value of 60 measured for another ophiuroid species after a three week exposure (Grillo *et al.*, 1981). Extrapolation of the ^{241}Am uptake rate at 2°C in our study (9.76×10^{-1} pmol/g/d) to a four month exposure period yields a much higher calculated concentration factor of 472 at the water concentration (2.48×10^{-1} pmol/ml) used here. Although tissue concentrations may attain equilibrium with water concentrations over shorter periods of time than this, these calculations illustrate the potential for order-of-magnitude discrepancies between laboratory concentration factors obtained from short-term uptake studies with animals and field concentration factors in chronically contaminated ecosystems, where exposure is likely to occur over extended periods of time. Short-term experiments can, however, effectively simulate the type of pulsed contaminant exposure likely to occur during events such as leakage from or rupture of nuclear waste containers.

This discrepancy between laboratory and field concentration factors is less evident with small, short-lived organisms (e.g. phytoplankton and some zooplankton), since their entire life span, from birth through death, can occur in the time frame of the laboratory experiments (Fisher *et al.*, 1983a,b; Fisher, 1985). However, animals such as ophiuroids and other benthic macroinvertebrates may take longer to reach equilibrium due to longer lifespans, low surface-to-volume ratios, and differential uptake and exchange among tissues. It is evident that there is a need for long-term investigations so these organisms can equilibrate with the dissolved contaminants.

For most of the elements examined here, there was little or no difference between ophiuroid arms and discs in the amount accumulated after either uptake or depuration. Brugmann & Lange (1988) reported enrichment of Zn and other elements in arms of field-collected asteroid echinoderms, and enrichment of Cd and Mn in central discs. The results of our study may suggest a difference in body distribution patterns between asteroids and ophiuroids, or may simply reflect differences in the amount of time required for various tissues to reach equilibrium with ambient levels. In more detailed dissections, Grillo *et al.* (1981) demonstrated substantial enrichment of ^{241}Am in the body wall of the ophiuroid *Ophiura texturata* relative to the digestive tract and gonads (97% and 3% of total radioactivity, respectively). Further investigations into the kinetics of uptake into various tissues and subsequent exchange between organ systems (Galey *et al.*, 1983) would help clarify the factors affecting body distributions of contaminants in potential bioindicator species such as ophiuroids.

Repeated handling of fragile ophiuroids in extended experiments inevitably results in partial loss of arms in some individuals. There is evidence that Cd accumulation in ophiuroids differs between undamaged and regenerating individuals, and may impair recovery from sublethal injuries (D'Andrea *et al.*, 1996). We cannot rule out the possiblity that minor damage to some individuals in our experiments may have affected the extent to which some elements were accumulated or retained.

Accumulation of radioisotopes and heavy metals from food, not investigated in these experiments, represents another potential source of contaminant exposure to marine organisms. *Ophiothrix fragilis* is primarily a suspension feeder (Chassard-Bouchaud *et al.*, 1988), while many other ophiuroid species are deposit feeders and most asteroids are predators. Differing trophic modes will result in exposure from different sources, possibily leading to variability in assimilation efficiencies and retention times. Temperature variations may especially impact dynamics of contaminants obtained from food, due to thermal effects on metabolic activity and turnover of physiologically assimilated pools (Hutchins *et al.*, 1996). Trophic transfer experiments to determine the relative importance of food and water sources in various marine organisms are a necessary prerequisite to their use as monitors of environmental quality.

These experiments underscore the necessity for realistic simulations of environmental variables such as temperature in investigations into contaminant interactions with marine biota. Evidence is accumulating that biological mobilization of anthropogenic contaminants may differ considerably among contrasting regimes, and characteristics of each area should be carefully considered when modeling contaminant accumulation and selecting bioindicator species.

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