

# Sea water uptake, sediment transfer and histo- autoradiographic study of plutonium ( $^{239}\text{Pu}$ ) and americium ( $^{241}\text{Am}$ ) in the edible cockle *Cerastoderma edule*

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**ABSTRACT:** In laboratory experiments we investigated the uptake of americium 241 and plutonium 239 from labelled sea water or sediment by the suspension feeding cockle *Cerastoderma edule* (Linné) as well as the localisation of these elements in its tissue. Kinetics of  $^{239}\text{Pu}$  and  $^{241}\text{Am}$  accumulation by the cockle were identical for sea water contaminated at either  $\text{Bq ml}^{-1}$  or  $\text{mBq l}^{-1}$ . In both cases, the cockles accumulated americium preferentially to plutonium. For the flesh, the concentration factor after a 60 d experiment was thus about 8 times higher for  $^{241}\text{Am}$  ( $\text{CF} \approx 1200$ ) than for  $^{239}\text{Pu}$  ( $\text{CF} \approx 160$ ). In the case of plutonium, the cockle tissues (flesh, pallial fluid, shell) and the whole animal quickly reached equilibrium (28 d) with the element contained in the sea water. In the case of americium, however, equilibrium was not reached even after 88 d of accumulation. Cockle shells fixed 80 % of the americium but only 36 % of the americium was accumulated by the whole animal. In the flesh, 96 % of the americium was fixed by the combined viscera and digestive tract. In these organs, americium and plutonium were located (histo-autoradiography) in the basal cells of the gut wall and the cells of the digestive tubule wall close to the gut. Transfer factors for americium and plutonium from a contaminated sediment were low at around 0.01. When cockles from the sediment only were labelled, the distribution of americium in their tissues was identical to that recorded for transfer from the water only. Measurement of the  $^{241}\text{Am}$  in the interstitial water extracted from the experimental sediment enabled the calculation of the interstitial water/species concentration factor. These data suggest the hypothesis that, in the case of this suspension-feeding bivalve, the americium is transferred from the sediment via the interstitial water. Direct transfer from sedimentary particles can be regarded to be nil for cockles under the conditions of experiment.

## INTRODUCTION

A limited number of studies have investigated the transfer of americium and plutonium from labelled sediments to certain sediment-dwelling marine invertebrates (Beasley & Fowler 1976, Murray & Renfro 1976, Miramand et al. 1982, Vangenechten et al. 1983, Germain et al. 1983, Miramand 1983). The purpose of these experiments was to quantify transfers of transuranic elements strongly fixed on sediments to benthic marine species. Interest in this type of study has been heightened by the 'sea bed' programme, with its plan

for burying containers of long-life radioactive waste, and in particular transuranic waste, in the sedimentary strata of the ocean deeps (Sousselier 1977, Talbert 1977).

With a view to adding to these data, this paper describes experiments on the transfer of americium ( $^{241}\text{Am}$ ) and plutonium ( $^{239}\text{Pu}$ ) to the suspension-feeding mollusc *Cerastoderma edule*, the common cockle. This mollusc constitutes a substantial biomass in certain sediments and is, moreover, eaten by human beings. In order to determine and quantify the transfer paths to this species, we studied the uptake of americium and plutonium from sea water and sediment, and measured the amount of americium contained in the interstitial water extracted from the experimental sediment. In addition, the localisation of

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these elements in the cockle tissues and cells was identified by histo-autoradiography.

## MATERIALS AND METHODS

*Cerastoderma edule* (L.) and the sediment in which they live were collected at St. Vaast la Hougue (east coast of the Cotentin peninsula).  $^{241}\text{Am}$  (half-life 434 y) and  $^{239}\text{Pu}$  (half-life 24386 y), conditioned in 1N  $\text{HNO}_3$ , were obtained from the C.E.A. (France). Before use, the  $^{239}\text{Pu}$  was put into a (VI) oxidation state according to Duursma & Parsi method (1974). The  $^{241}\text{Am}$  is assumed to be in a (III) state.

*Uptake from sea water.* Two experiments were performed: one with a large volume, using  $^{241}\text{Am}$  and  $^{239}\text{Pu}$  concentrations likely to be found in theory in parts of the sea where industrial waste is discharged; the other using concentrations 20000 to 90000 times greater, i.e. similar to those often used in experimental studies. These experiments were carried out in order to see if transfers differ for weak and strong concentrations of radionuclides in sea water.

For the first experiment, 180 individuals (average wet weight  $10 \pm 2$  g) were put into an aquarium containing 600 l of sea water filtered through a millipore filter ( $0.45 \mu\text{m}$ ) at  $14 \text{ }^\circ\text{C} \pm 1 \text{ }^\circ\text{C}$ . The  $^{239}\text{Pu}$  ( $37 \text{ mBq l}^{-1}$ , or  $1 \text{ pCi l}^{-1}$ ) and the  $^{241}\text{Am}$  ( $18.5 \text{ mBq l}^{-1}$ , or  $0.5 \text{ pCi l}^{-1}$ ) were then added. Over the 60 d of the experiment, the sea water was changed weekly using water of the same quality containing the same concentrations of radionuclides. Each time the sea water was changed, 10 l of old and new solution were drawn off for analysis.

Every 10 d, 30 individuals were collected, weighed and dissected. The tissue (flesh, shell, pallial fluid) were oven-dried at  $90 \text{ }^\circ\text{C}$  to constant weight and then ground. After internal tracers ( $^{236}\text{Pu}$  and  $^{243}\text{Am}$ ) had been introduced, the transuranic elements were extracted and separated by laboratory radiochemical methods (Grenaut et al. 1983).

For the second experiment, 100 cockles similar in weight to those used previously, were placed into aquaria filled with 20 l ( $^{241}\text{Am}$ ) and 15 l ( $^{239}\text{Pu}$ ) of filtered sea water ( $0.45 \mu\text{m}$ ) at  $14 \text{ }^\circ\text{C} \pm 1 \text{ }^\circ\text{C}$ .  $888 \text{ mBq ml}^{-1}$  ( $24 \text{ pCi ml}^{-1}$ )  $^{239}\text{Pu}$  and  $1665 \text{ mBq ml}^{-1}$  ( $45 \text{ pCi ml}^{-1}$ )  $^{241}\text{Am}$  were then added. Every 48 h the sea water was changed using water of the same quality containing the same concentration of the radioisotopes. Each time the water was changed, aliquots of the new and old solutions were drawn off for analysis. Five individuals from each aquarium were removed and dissected (flesh, shell, pallial fluid) and their radioactivity was measured. Counting techniques were the same as those described earlier (Miramand et al. 1982).  $^{241}\text{Am}$

gamma ray (60 keV) was measured with 25 % efficiency with a gamma spectrometer coupled to a Na I (Tl) well crystal.  $^{239}\text{Pu}$  alpha emission was counted by liquid scintillation spectrometry, with over 95 % efficiency in all cases.

The results of both experiments are expressed as concentration factor, defined as  $\text{cpm g}^{-1}$  individual wet weight/ $\text{cpm ml}^{-1}$  sea water.

*Histo-autoradiography.* The cockles from sea water were labelled by the method described for the second experiment. Experimental waters contained about  $18.5 \text{ Bq ml}^{-1}$  ( $500 \text{ pCi ml}^{-1}$ ) of  $^{241}\text{Am}$  and about  $7.4 \text{ Bq ml}^{-1}$  ( $200 \text{ pCi ml}^{-1}$ ) of  $^{239}\text{Pu}$ . Cockles were taken from each tank after 17 d of labelling for  $^{239}\text{Pu}$  and after 88 d of labelling for  $^{241}\text{Am}$ . Their flesh was immediately separated from the shell, rinsed with clean sea water and immersed in Bouin's fluid for 48 h. In both cases, only 5 % activity was lost in the Bouin's fluid.

Tissues were prepared for histo-autoradiography by the method described earlier (Miramand & Guary 1981). After fixing, the tissues were dehydrated and impregnated with paraffin wax. Sections  $5 \mu\text{m}$  thick were placed on glass slides, dried and dewaxed. Autoradiographs were prepared with Ilford K2 nuclear emulsion (diluted 50 % with 1 % glycerin solution) at  $50 \text{ }^\circ\text{C}$  using the dipping technique (Rogers 1973). The slides were then dried, placed in hermetically-sealed boxes and stored in the dark at  $4 \text{ }^\circ\text{C}$ . Each week, autoradiographs were developed, fixed, washed and coloured with Masson trichrome. The slides were dehydrated and mounted for microscopic examination.

*Accumulation from sediment.* Muddy sand sediment was collected at Pont de Saire near St. Vaast La Hougue (Manche, France) where the cockles were collected. It contained 1.2 % organic matter and 20 % carbonates. Two kg of dry sediment (2.84 kg wet weight) were contaminated at the level of  $37.10^4 \text{ Bq kg}^{-1}$  dry weight ( $10 \mu\text{Ci kg}^{-1}$  dry weight) as previously described by Miramand et al. (1982). Following labelling, the sediment was placed in a tank. After settling, the depth of sediment in the tank was about 10 cm. The supernatant sea water was siphoned off and a flow of uncontaminated sea water was set up; 15 cockles were then placed in the sediment. The experiment continued for 14 d at  $14 \text{ }^\circ\text{C} \pm 1 \text{ }^\circ\text{C}$ . The cockles were then taken out, left in running sea water for 48 h to clear their digestive tract before analysis and then thoroughly rinsed. Measuring their radioactivity and that of the sediment enabled the calculation of the transfer factor (T.F.):  $\text{cpm g}^{-1}$  individual wet weight/ $\text{cpm g}^{-1}$  wet weight. After counting, the cockles were dissolved in hot nitric acid; no sedimentary particles were found in the tissues of mineralised cockles.

At the end of the experiments, the sediment was analysed to determine the degree of  $^{241}\text{Am}$  fixation to

it.  $^{241}\text{Am}$  was extracted from sediment aliquots with acetic acid at pH 2, then with a 50:50 mixture of concentrated  $\text{HCl-HNO}_3$  at  $80^\circ\text{C}$ . The interstitial water was extracted from 600 g of sediment with a press similar to that described by Presley et al. (1967). The concentration of  $^{241}\text{Am}$  in the water extracted was then measured. As the specific radioactivity of both sediment (As) and interstitial water (A Isw) were known, the distribution coefficient of  $^{241}\text{Am}$  between sediment and interstitial water ( $K_d = \text{As}/\text{A Isw}$ ) could be calculated.

## RESULTS

### Uptake of $^{239}\text{Pu}$ and $^{241}\text{Am}$ from sea water

The kinetics of uptake of these 2 transuranic elements by whole cockles (Fig. 1) or by their tissues (flesh, shell, pallial fluid) (Fig. 2) were identical for the 2 types of experiment: accumulation in a few litres of sea water, heavily labelled with americium or plutonium ( $\text{Bq ml}^{-1}$ ) or accumulation in a large volume of much less heavily labelled sea water ( $\approx \text{mBq l}^{-1}$ ).

The cockles preferentially accumulated americium

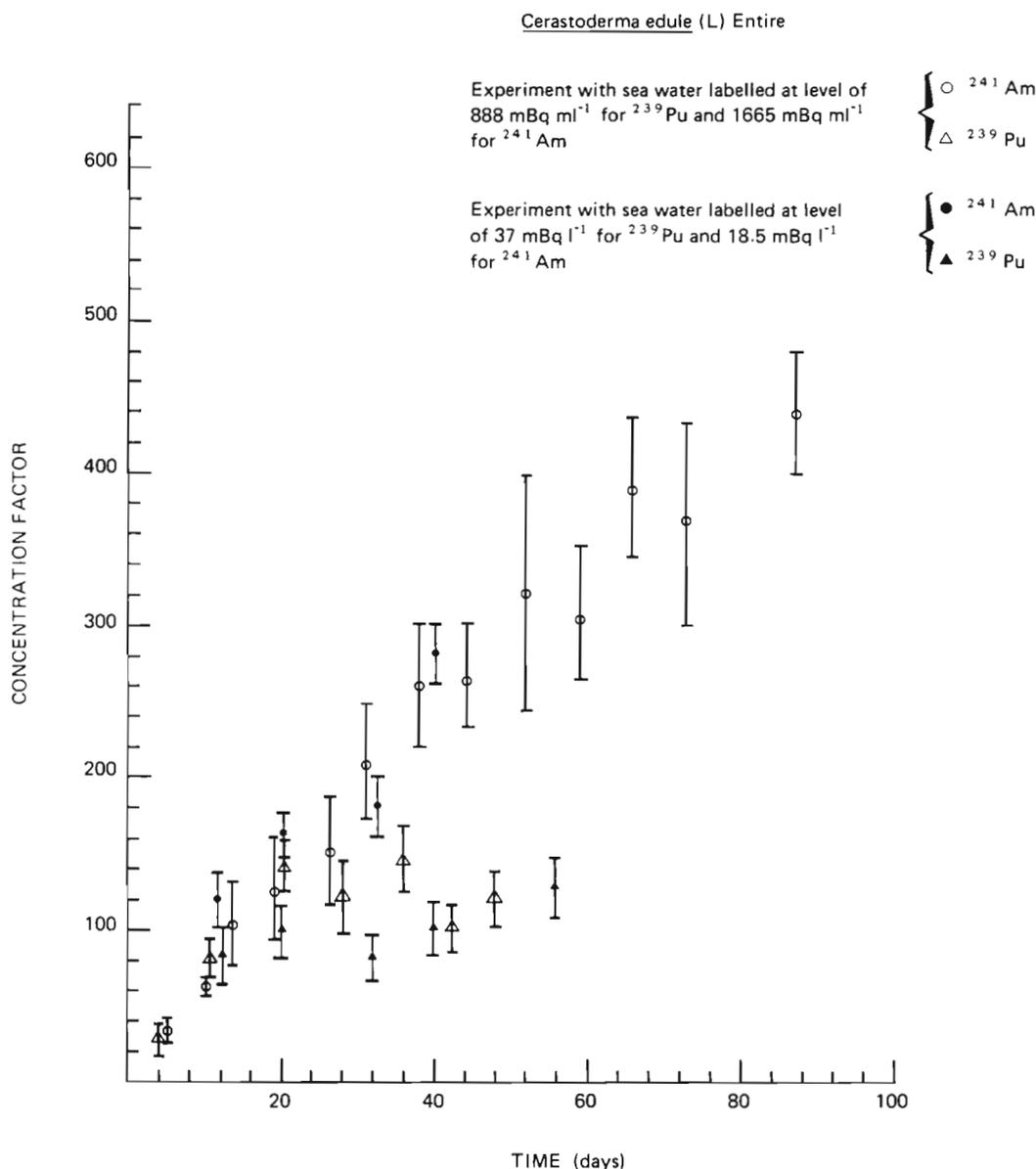


Fig. 1. *Cerastoderma edule*. Whole body accumulation of  $^{241}\text{Am}$  and  $^{239}\text{Pu}$  from labelled sea water ( $\bar{x} \pm \text{SD}$ ).  $T = 14^\circ\text{C} \pm 1^\circ\text{C}$  ( $n = 30$  per group for experiment with sea water, labelled at  $37 \text{ mBq l}^{-1}$   $^{239}\text{Pu}$  and  $18.5 \text{ mBq l}^{-1}$   $^{241}\text{Am}$ ;  $n = 5$  per group for experiment with sea water, labelled at  $888 \text{ mBq ml}^{-1}$   $^{239}\text{Pu}$  and  $1665 \text{ mBq ml}^{-1}$   $^{241}\text{Am}$ ). Mean wet weight of individuals =  $10 \pm 2 \text{ g}$  ( $\bar{x} \pm \text{SD}$ )

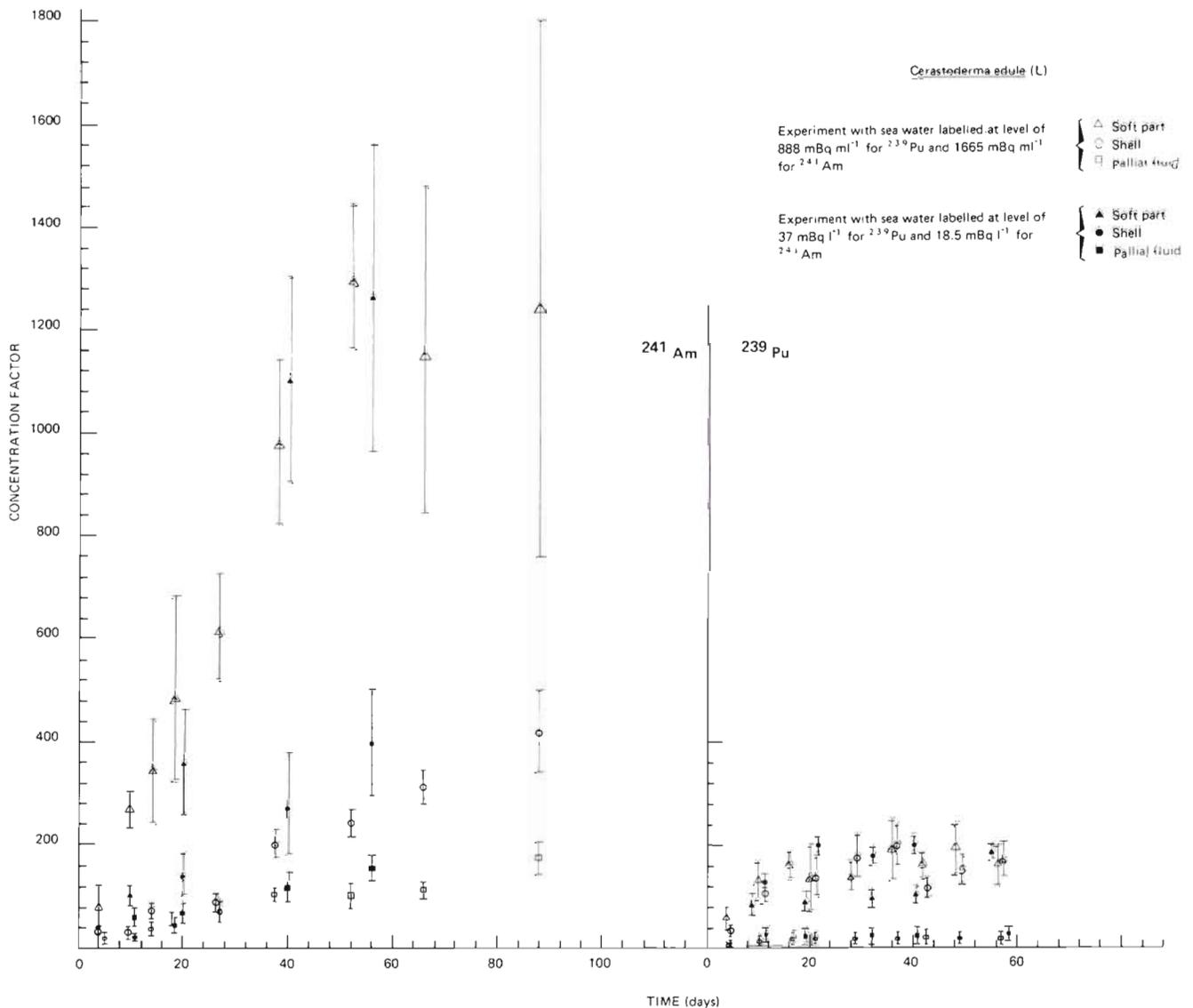


Fig. 2. *Cerastoderma edule*. Accumulation of <sup>241</sup>Am and <sup>239</sup>Pu from labelled sea water in shell, soft parts and pallial fluid ( $\bar{x} \pm SD$ ). T = 14 °C ± 1 °C (n = 30 per group for experiment with sea water, labelled at 37 mBq l<sup>-1</sup> <sup>239</sup>Pu and 18.5 mBq l<sup>-1</sup> <sup>241</sup>Am; n = 5 per group for experiment with sea water, labelled at 888 mBq ml<sup>-1</sup> <sup>239</sup>Pu and 1665 mBq ml<sup>-1</sup> <sup>241</sup>Am). Mean wet weight of individuals = 10 ± 2 g ( $\bar{x} \pm SD$ )

rather than plutonium (Fig. 1). Thus after the same period of exposure (56 d), the concentration factors for whole individuals were ca. 340 for <sup>241</sup>Am and 140 for <sup>239</sup>Pu. The difference was even more marked for flesh, to a lesser extent for the pallial fluid. For the flesh, after 56 d, the concentration factors were about 8 times higher for <sup>241</sup>Am (C.F. ≈ 1200) than for <sup>239</sup>Pu (C.F. ≈ 160). The shells, on the other hand, appeared to fix both elements more or less identically (C.F. ≈ 200). From Day 28 of exposure onwards, the cockles (flesh, pallial fluid, shell) quickly reached equilibrium with the plutonium in the sea water; but equilibrium with americium was not established even after 88 d of

labelling, despite a dip in the uptake curve after Day 40.

Major differences were also observed in the distribution of plutonium and americium in the tissues (flesh, pallial fluid, shell) of the cockles (Table 1). After a comparable period of accumulation (52, 56 d), cockle shells contained almost 80 % of the plutonium accumulated, compared to only 36 % for americium. The flesh, on the other hand, contained mostly americium, at about 57 % of the total, as against about 16 % for plutonium. The pallial fluid contained only 4 to 7 % of the 2 elements. Table 2 shows the distribution of americium in the organs of the soft parts of the

Table 1. *Cerastoderma edule*. Percentage tissue distribution of americium and plutonium following exposure to labelled sea water (52 d for  $^{241}\text{Am}$ ; 56 d for  $^{239}\text{Pu}$ ).

T = 14 °C ± 1 C° [ $\bar{x}$  ± SD (n = 5)]

Body part	% total $^{241}\text{Am}$ content	% total $^{239}\text{Pu}$ content	% total wet weight
Soft parts	57 ± 5	16 ± 3	17 ± 3
Shell	36 ± 5	80 ± 3	53 ± 4
Pallial fluid	7 ± 1	4 ± 1	30 ± 5

### Histo-autoradiographic location of $^{241}\text{Am}$ and $^{239}\text{Pu}$ in the flesh

Fig. 3 gives a general view of a section of cockle flesh from the digestive tract labelled with americium showing that americium is mainly fixed in the basal cells of the digestive tract wall. A further enlargement (Fig. 4) clearly shows that the black colouring of the wall observed in Fig. 3 is due to the very large quantity of the interlaced alpha traces. Fig. 3 also reveals a second major localisation of americium in the digestive tubules of the digestive gland. There are very few

Table 2. *Cerastoderma edule*. Concentration factors and tissue distribution of  $^{241}\text{Am}$  after 38 d exposure to labelled sea water. T = 14 °C ± 1 C° [ $\bar{x}$  ± SD (n = 5)]

Body part	C.F.	% total $^{241}\text{Am}$ content	Soft part: % $^{241}\text{Am}$ content	% total wet weight
Shell	200 ± 30	38 ± 3	–	54 ± 1
Pallial fluid	100 ± 10	9 ± 1	–	32 ± 5
Soft parts	1000 ± 200	53 ± 5	100	14 ± 3
– gill	70 ± 30	1.0 ± 0.5	1.3 ± 0.9	2.1 ± 0.4
– mantle	70 ± 20	1.0 ± 0.5	1.7 ± 1.2	3.7 ± 0.8
– muscle	40 ± 4	1.0 ± 0.2	1.1 ± 0.3	3.4 ± 0.9
– viscera	2600 ± 500	50 ± 6	96 ± 3	4.8 ± 0.9

cockles after 38 d of accumulation. The combined digestive tract/viscera contained about 50 % of all the americium accumulated by the cockles, this being 96 % of the americium fixed in the flesh. The capacity of these organs to accumulate americium is also extremely high (C.F. ≈ 2600). The muscles had the lowest concentration factor (about 40) and contained only 1 % of total americium.

or no alpha traces in the muscular, connective or gonad tissues. It can also be seen (Fig. 5) that the digestive tubules near the gut wall contain more alpha traces due to americium. The tubules furthest from the gut wall are less labelled. Fig. 5 also shows that the alpha traces are distributed mainly at the periphery of the tubules.

Distribution of plutonium in cockles tissues is identi-

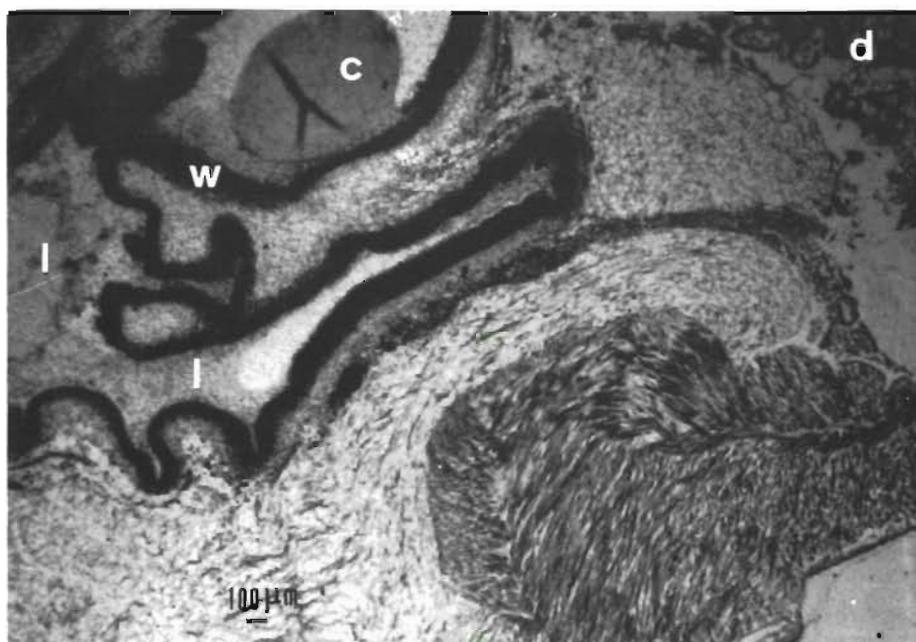


Fig. 3. *Cerastoderma edule*.  $^{241}\text{Am}$  autoradiography. General view of tissues slightly enlarged. Exposure time: 28 d. L: Lumen of digestive tract; W: wall of digestive tract; C: crystalline style; D: digestive tubules

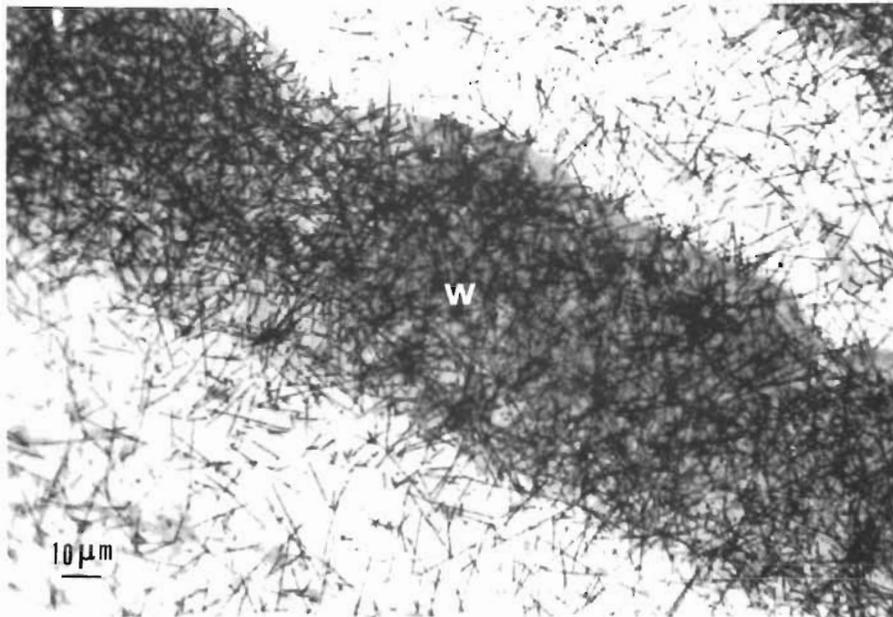


Fig. 4. *Cerastoderma edule*.  $^{241}\text{Am}$  autoradiography. Further enlargement reveals details of digestive-tract wall. Exposure time 28 d. w: wall of digestive tract

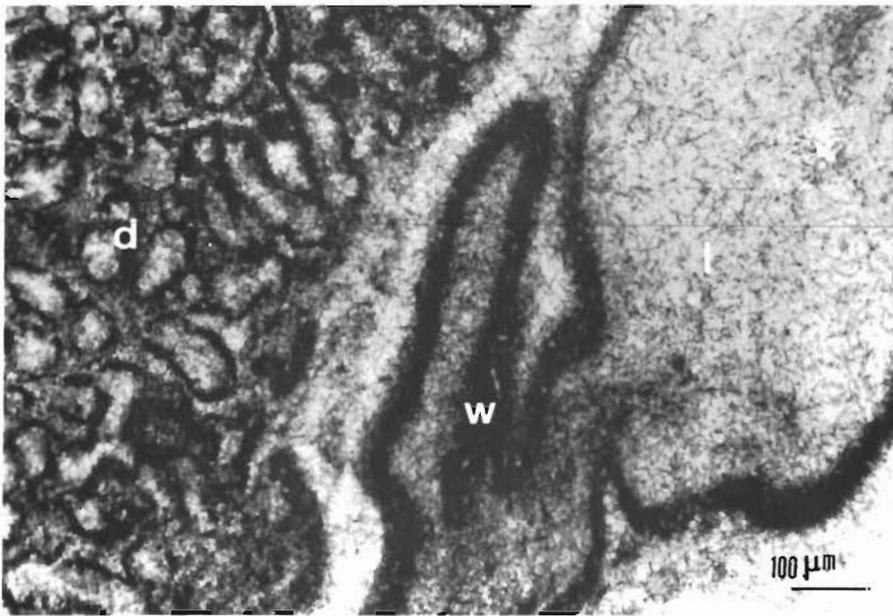


Fig. 5. *Cerastoderma edule*.  $^{241}\text{Am}$  autoradiography of digestive tubules. Exposure time: 28 d. l: lumen of the digestive tract; w: wall of the digestive tract; d: digestive tubules

cal. Plutonium is fixed in the cells of the digestive-tract wall and in the digestive tubules of the digestive gland (Fig. 6 and 7).

#### Accumulation of $^{239}\text{Pu}$ and $^{241}\text{Am}$ from contaminated sediment

$^{241}\text{Am}$  activity of the interstitial water extracted from the experimental sediment was about  $12 \text{ mBq ml}^{-1}$  ( $0.32 \text{ pCi ml}^{-1}$ ), giving an interstitial water/sediment  $K_d$  of  $3.10^4$ . Seventy-three % of the americium in the sediment was attached to sedimentary particles less

than  $50 \mu\text{m}$  in diameter, accounting for only 25 % by weight of the sediment. Sixty-five % of the americium can easily be desorbed from the sediment by a weak acid (acetic acid) at pH 2 which may indicate a degree of bio-availability. However, transfer of this element to whole cockles was low ( $\text{T.F.} \approx 0.008$ ) (Table 3).

The transfer factors for plutonium and americium are closely comparable at less than, or close to, 0.01 (Table 3) for both whole cockles and their tissues (flesh, shell, pallial fluid). Table 3 also shows the distribution of the americium and plutonium in cockle tissues. A large fraction of the plutonium is fixed on the shell (85 %), compared to only 39 % for americium.

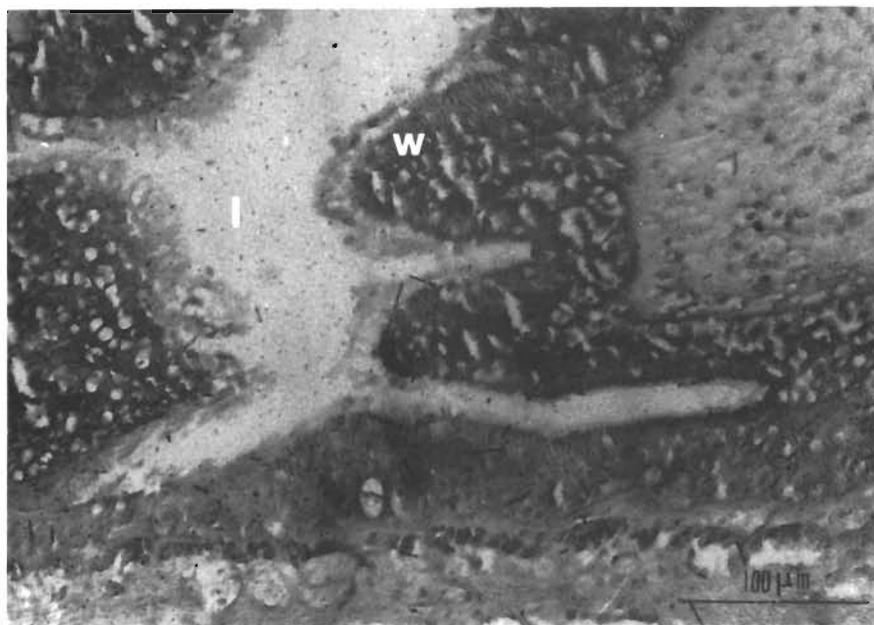


Fig. 6. *Cerastoderma edule*.  $^{239}\text{Pu}$  autoradiography of digestive-tract wall. Exposure time: 14 d. l: lumen of the digestive tract; w: wall of digestive tract

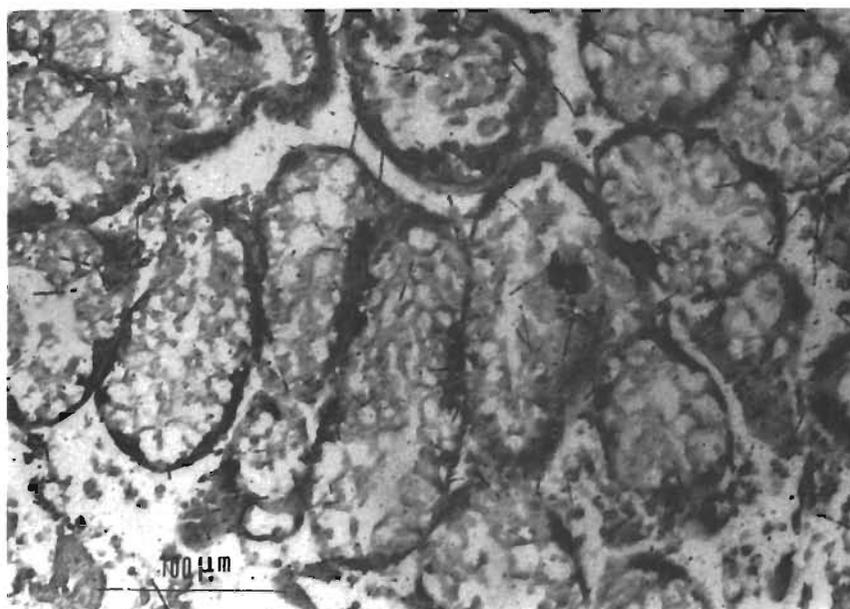


Fig. 7. *Cerastoderma edule*.  $^{239}\text{Pu}$  autoradiography of digestive tubules. Exposure time: 14 d

Table 3. *Cerastoderma edule*. Transfer factor of  $^{241}\text{Am}$  and  $^{239}\text{Pu}$  from contaminated sediment after 14 d.  $T = 14\text{ }^{\circ}\text{C} \pm 1\text{ }^{\circ}\text{C}$  [ $\bar{x} \pm \text{SD}$  ( $n = 15$ )].  $^{241}\text{Am}$  concentration in sediment:  $4209\text{ cpm g}^{-1}$  wet weight. Dry to wet ratio: sediment = 0.71; entire = 0.52; soft part = 0.07; shell = 0.91;  $^{239}\text{Pu}$  concentration in sediment:  $12355\text{ cpm g}^{-1}$  wet weight; dry to wet ratio: sediment = 0.77

Body part	Cpm $\text{g}^{-1}$ wet weight	$^{241}\text{Am}$		$^{239}\text{Pu}$		
		Transfer factor	% total $^{241}\text{Am}$ content	Cpm $\text{g}^{-1}$ wet weight	Transfer factor	% total $^{239}\text{Pu}$ content
Entire*	$35 \pm 8$	$0.008 \pm 0.002$	—	$169 \pm 49$	$0.014 \pm 0.004$	—
Soft parts	$58 \pm 32$	$0.014 \pm 0.008$	$53 \pm 19$	$56 \pm 28$	$0.005 \pm 0.002$	$9 \pm 4$
Shell	$32 \pm 13$	$0.008 \pm 0.003$	$39 \pm 25$	$198 \pm 49$	$0.016 \pm 0.004$	$85 \pm 5$
Pallial fluid	$13 \pm 4$	$0.003 \pm 0.001$	$7 \pm 3$	$38 \pm 20$	$0.003 \pm 0.002$	$6 \pm 4$

\* Reconstructed

## DISCUSSION

## Transfer from sea water

Concentrations of americium and plutonium in sea water are generally very low at the level of  $\mu\text{Bq l}^{-1}$  where fallout is exclusively atmospheric (Ballestra 1980) or at  $\text{mBq l}^{-1}$  where industrial wastes are discharged into the ocean (Hetherington et al. 1975, 1976, Germain & Miramand 1984, Pentreath et al. 1984). This raises the question of the validity of laboratory experiments using high concentrations around  $\text{Bq ml}^{-1}$  of americium and plutonium in sea water. In this study, the cockles accumulated the transuranic elements from sea water contaminated with either  $\text{mBq l}^{-1}$  or  $\text{Bq ml}^{-1}$  of these elements. The kinetics of accumulation of the americium and plutonium in the cockles were identical in both cases and therefore seem to be independent of the concentration of those elements in sea water, at least over the range tested. Experiments using a small volume of water and high concentrations of americium and plutonium are justified; such procedure considerably simplifies the experimental design (small volumes of water handled, easier counting because of the higher levels of radioactivity).

It is interesting to compare the accumulation of the transuranic elements by cockles and other lamelli-branchia. Table 4 gives the concentration factors and

3 to 7 times more plutonium in their flesh than did the 3 other bivalves tested. Furthermore, while plutonium is fixed in an identical manner on the shells of the 4 species, less americium appears to be retained on the shells of cockles. As a result, cockles behave differently than the other bivalves; their flesh retains most americium (66 % of total americium) whereas the shells of the other species retain 80 to 90 % of total americium. In terms of the return of transuranic elements to human beings *via* the digestive tract, the flesh of cockles would therefore appear to be a much more important vector than the flesh of mussels, tapes or scrobicularians.

## Transfer from sediment

As compared to the water/cockles transfers, transfers to cockles of americium and plutonium fixed on sediment were very small at ca. 0.01. This result confirms similar findings for other benthic species (Beasley & Fowler 1976, Murray & Renfro 1976, Miramand et al. 1982, Germain et al. 1983, Vangenechten et al. 1983).

The transfer factors of americium and plutonium from sediment to cockles are identical to those found for *Scrobicularia plana* (Miramand et al. 1982), but the paths by which americium is transferred from sediment to these 2 lamelli-branchia differ in some respects. In the case of *S. plana*, americium is transferred from the

Table 4. Comparison of concentration factors and percentages of distribution for americium and plutonium found in soft parts and shells of 4 species of bivalve mollusc after 21 d exposure to labelled sea water. T = 14 °C  $\pm$  1 C°

			<i>Mytilus galloprovincialis</i> (a)	<i>Tapes decussatus</i> (b)	<i>Scrobicularia plana</i> (c)	<i>Cerastoderma edule</i>
Soft part	Americium	C.F.	34	71	96	510
		% total Am content	9	19	11	66
	Plutonium	C.F.	28	18	38	130
		% total Pu content	9	8	10	18
Shell	Americium	C.F.	186	258	350	60
		% total Am content	91	81	85	24
	Plutonium	C.F.	148	165	200	175
		% total Pu content	91	92	86	75

(a) Guary (1980), (b) Grillo et al. (1981), (c) Miramand et al. (1982)

the percentages of plutonium and americium fixed in flesh and shells of *Mytilus galloprovincialis* (Guary 1980), *Tapes decussatus* (Grillo et al. 1981), *Scrobicularia plana* (Miramand et al. 1982) and *Cerastoderma edule* (this study); all data relate to the same period of accumulation of these elements from sea water (21 d) in the course of experiments conducted in an identical manner.

The cockles fixed 5 to 15 times more americium, and

sediment both indirectly *via* the americium in the interstitial water and also directly by desorption in the digestive tract of the americium fixed on ingested sediment particles. In the experiment with cockles, americium was measured in interstitial water extracted from the experimental sediment at a concentration of 12  $\text{mBq m l}^{-1}$ . With this value, it was possible to calculate the interstitial water/species concentration factor, CF Isw, and therefore to determine

the quotient C.F. Isw/C.F. sw., C.F. sw. was the concentration factor found in experiments on the accumulation of americium from sea water only after 14 d (Fig. 1). Provided the physicochemical state of americium in the interstitial water extracted from the sediment is identical with that of the americium present in the sea water from the experimental tanks, the quotient C.F. Isw/C.F. sw should be 1 if the sediment/species transfer of americium takes place through the interstitial water only. For soft parts of cockles, the quotient is  $0.9 \pm 0.8$ . In spite of the large relative error ( $\approx 90\%$ ) of the mean value (0.9), we can consider that the transfer of americium fixed on the sediment to this species is attributable to a straightforward interstitial water/species transfer. This hypothesis is reinforced by the fact that the distribution of americium in cockle tissue is exactly the same for accumulation of this element from both sediment and sea water. This finding is not surprising. In our experiment cockles, which are suspension feeding but burrowing species, did not ingest any sedimentary particles as the sediment was not suspended in the water. Consequently, in order to quantify all transfer paths for this species, it would be of interest to investigate whether americium is transferred from suspensions. It may be noted that transfer factors for this species obtained *in situ* are higher than those recorded in the laboratory (Germain & Miramand 1984) taking all transfer paths into account.

### Histo-autoradiographic localisation

We have located the fixation of americium and plutonium accurately in the flesh of the cockles examined. The artificial elements are principally localised in 2 types of cells: those of the digestive-tract wall and those of the wall of the digestive tubules. These cells, which have the main function of absorbing nutritive elements, are therefore also permeable to the transuranic elements which, although artificial, follow the digestive metabolism of these animals. This fact appears to show that digestive absorption is involved in the transfer of transuranic elements to cockles. Further study will, however, be necessary to establish details of the pattern and to determine the types of physiological mechanism involved, both active and passive. In this context, the role and extent of a possible transfer of these radionuclides through the branchial membrane and the haemolymph have still to be determined. However, the precise location of americium and plutonium in the cells of the digestive gland and tract poses the problem of calculating the radiation dose received by organs, because it is not distributed evenly over an organ but concentrated in certain types of cells.

Cockles accumulate americium preferentially from

sea water. Such preferential accumulation has already been demonstrated experimentally for many other aquatic organisms: the molluscs *Venerupis decussata* (Grillo et al. 1981), *Mytilus galloprovincialis* (Guary 1980), and *Scrobicularia plana* (Miramand et al. 1982); the crustaceans *Carcinus maenas* (Guary 1980), and *Corophium volutator* (Miramand et al. 1982); the echinoderms *Paracentrotus lividus* (Guary 1980), and *Stichopus regalis* (Grillo et al. 1981), and the annelids *Hermione hystrix* (Guary et al. 1981), and *Arenicola marina* (Miramand et al. 1982). However, some other species seem to accumulate plutonium rather than americium from water in experiments: this applies to the octopus *Octopus vulgaris* (Guary & Fowler 1982), the starfish *Coscinasterias tenuispina* (Guary 1980), the sand star *Ophiura texturata* (Guary et al. 1981) and the annelid *Nereis diversicolor* (Miramand 1983).

For all these species, in addition to their external parts in contact with the sea water, the organ retention sites are the same for americium and plutonium. In most cases, they are found in the digestive glands (Beasley & Cross 1980, Guary 1980). The problem is therefore to identify the reason for the different accumulation kinetics for americium and plutonium in benthic marine species. This histo-autoradiographic study has shown that in cockles, americium and plutonium are retained in the same types of cells. A similar finding was made with another bivalve, *Scrobicularia plana* (Miramand 1983). The explanation for the behavioural difference between americium and plutonium, demonstrated in these 2 species, may lie in biochemical or physicochemical differences between the 2 elements, associated in particular with their different valencies and specific activities.

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