

Delineation of PCB uptake pathways in a benthic sea star using a radiolabelled congener

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ABSTRACT

| 2 | Asterias rubens, a common sea star in North Sea waters, was selected to study the |
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| 3 | bioaccumulation of an important polychlorinated biphenyl congener, ¹⁴ C-labelled PCB#153 |
| 4 | from two contrasted sources: sea water and sediments. After 4 weeks of acclimation to |
| 5 | laboratory conditions, sea stars were exposed for 34 days to realistic concentrations (30 ng l ⁻¹ ir |
| 6 | sea water and 9.5 ng g-1 DW in sediments) of the contaminant during which time |
| 7 | bioaccumulation of PCB#153 was followed in 6 body compartments. Results showed that (1) |
| 8 | for each body compartment, PCB uptake kinetics were generally asymptotic and |
| 9 | bioaccumulation was far greater when A. rubens was exposed via sea water than via sediments |
| 10 | (2) body wall and podia were the body compartments showing the greatest affinity for the PCB |
| 11 | congener making them ideal tissues for biomonitoring purposes, and (3) the concentrations |
| 12 | reached in body compartments were in the range of values reported in several field studies |
| 13 | Because radioisotopic techniques are extremely sensitive, they allow taking into account key |
| 14 | organs which are sometimes too small for standard analysis of PCBs. |

KEYWORDS

Polychlorinated biphenyls; PCB#153; bioaccumulation; kinetics; Asterias rubens; echinoderm

RUNNING HEADING

21 PCB bioaccumulation in sea stars

INTRODUCTION

1

2 Polychlorinated biphenyls (PCBs) are strictly anthropogenic chemicals that constitute one of the most problematic and widespread group of contaminants. These xenobiotics, represented by 3 4 209 congeners, are extremely resistant to degradation (physico-chemical or biological), are 5 bioconcentrated by living organisms, and can cause various adverse effects depending on their 6 pattern and degree of chlorine substitution (Metcalfe 1994). For PCBs entering the marine 7 environment, bottom sediments are the ultimate repository where they may become a source 8 for uptake by marine organisms through direct or indirect contact or, for filter feeders, by 9 ingestion; however, information about their impact on benthic species is relatively scarce 10 (Chapman 1995, Carr et al. 1996, Wood et al. 1997). According to different authors, the asteroid Asterias rubens qualifies as an excellent 11 bioindicator organism for monitoring heavy metal contamination in the North Sea and NE 12 Atlantic benthic ecosystems (Knickmeyer et al. 1992, den Besten et al. 1993, Everaarts et al. 13 14 1998, Temara et al. 1998, Warnau et al. 1999). It is indeed a widely distributed and abundant key species (sensu Lewis 1978) that is easy to collect, identify and maintain in the laboratory. 15 16 In addition, A. rubens is a top predator feeding mainly on mussels and living on or in the 17 proximity to bottom sediments which are the main reservoir of many contaminants, including 18 PCBs. The biological and ecological characteristics of A. rubens as well as its potential 19 economic impact (as a predator of commercially important mussels) have lead some authors to 20 use this species as a tool to assess the degree of PCB contamination in the North Sea (den Besten et al. 1989, 1993, Everaarts et al. 1998). However, to the best of our knowledge, no 21 22 study has investigated PCB bioaccumulation processes in A. rubens. The only two experimental studies investigating PCB bioaccumulation in echinoderms concern sea urchins 23 exposed to contaminated sediments (Weisberg et al. 1996, Schweitzer et al. 2000), and only 24 Weisberg et al. (1996) examined the kinetic aspects of PCB uptake. 25

Such data are however needed to further assess the value of Asterias rubens as a bioindicator of 1 2 PCB contamination. Therefore, in the present study, we have investigated the kinetics of PCB uptake in A. rubens exposed either to the contaminant in sea water or associated with 3 sediments, i.e. the two extreme pathways of contamination from the viewpoint of absolute PCB 4 concentrations. Indeed, the high hydrophobicity of PCBs result in a characteristic partitioning 5 with concentrations in sea water typically in the range of pg to ng l⁻¹ while sediment б concentrations are in the range of µg to mg kg⁻¹ (see Table 1). The PCB congener IUPAC #153 7 (2,2',4,4',5,5' hexachlorobiphenyl) was selected because it is the most abundant in marine 8 biota (Stebbing et al. 1992) and has been shown to be an excellent indicator of total PCB 9 10 contamination (Atuma et al. 1996).

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MATERIALS AND METHODS

- Sampling. The sea stars Asterias rubens (Linnaeus 1758) were collected in April 1999 in the
- 14 intertidal zone at Audresselles (Pas-de-Calais, France). Prior to experimentation, specimens
- were acclimated to laboratory conditions for 1 month in constantly aerated closed circuit
- aguaria (salinity: 36 %, T: 16 ± 0.5 °C, 12/12 h dark/light cycle).
- In order to follow PCB#153 bioaccumulation under realistically simulated conditions, a ¹⁴C-
- labelled congener was used and measured using highly sensitive β spectrometry.
- Radiotracer. The 14 C-labelled 2,2',4,4',5,5' hexachlorobiphenyl (purity \geq 95%) was purchased
- from Sigma Chemicals, USA. Specific activity was 925 MBq mmol⁻¹. Stock solutions were
- 21 prepared in acetone at a concentration of 1 µg ml⁻¹.
- 22 Sample treatment and liquid scintillation counting. Water samples (2 ml) were directly
- transfered to 20 ml glass scintillation vials (Packard, USA) and 10 ml of Ultima Gold XR®
- 24 (Packard Instruments) scintillation liquid were added. Samples of sediment and sea star tissue
- 25 (previously crushed) were placed in a vial containing 2 ml of Acetonitrile[®] in an ultrasonic bath
- for 10 min. Acetonitrile® was then collected and replaced by another 2 ml of Acetonitrile® and

- the ultrasonic operation was repeated a second time. This treatment gave 4 ml of liquid phase
- 2 (viz. the extraction) and a residue. The residue was digested overnight at 70°C with 2 ml of
- 3 Soluene[®], and 10 ml of Hionic Fluor[®] scintillation liquid were then added. The liquid phase (4
- 4 ml) was added to 16 ml of filtered seawater and extracted twice using 2 ml of n-Hexane
- 5 (Sigma, USA) under constant agitation. The organic phase (4 ml) and the aqueous phase (20
- 6 ml) were treated separately. The entire organic phase and 2 ml of the aqueous phase were each
- 7 added separately to 10 ml of Ultima Gold XR® scintillation liquid.
- 8 ¹⁴C-radioactivity was then measured using a 1600 TR Liquid Scintillation Analyzer (Packard),
- 9 compared to standards of known activities, and corrected for quenching, background and
- physical decay of the radiotracer. Counting times were adjusted to obtain counting rates with
- relative propagated errors less than 5 %. PCB concentrations were expressed on a total lipid
- content basis where lipids were determined according to the method of Barnes & Blackstock
- 13 (1973). A schematic diagram of the sample treatment is shown in Figure 1.

Experimental procedures

- 15 Uptake from sea water: Asteroids (n = 24) were placed for 34 d in a 70 l glass aquarium
- 16 (constantly aerated closed circuit aquaria; salinity 36 %; 16 ± 0.5 °C; 12/12 h dark/light cycle)
- 17 containing natural sea water spiked with ¹⁴C-labelled PCB#153. One day prior to the
- experiments, four 5 l glass beakers were filled with filtered sea water (36 \%; 16 ± 0.5 °C),
- spiked with the radiolabelled PCB stock solution, and constantly stirred using an orbital
- agitation plate. Contaminated water then was poured into the glass aquaria and uncontaminated
- sea water was added to obtain a final volume of 70 l. Sea water and radiotracer were renewed
- every second day during the entire experiment. Activity was checked before and after each
- renewal to assess the stability of the labelled PCB concentration in sea water (Table 1). The sea
- stars were fed unlabelled mussels (*Mytilus edulis*) every second day just before the seawater
- 25 renewal. After 2 hours uningested mussels were removed to limit as much as possible PCB
- incorporation via the food. Periodically (after 2, 4, 7, 11, 14, 21 and 34 days), sea stars (n = 3)

were removed, dissected into seven body compartments (oral and aboral body walls, pyloric

2 caeca, gonads, rectal caeca, central digestive system, and podia), and radioanalyzed to

determine uptake kinetics and body distribution of the incorporated PCB.

4 Uptake from sediments: Sediments (2.5 kg dry wt) from the North Sea (Audresselles, Pas-de-

Calais, France) were contaminated for 4 days with the ¹⁴C-labelled PCB using the rolling jar

method (Murdoch et al. 1997). Sea stars (n = 24) were placed in a 70 l glass aquarium

(constantly aerated open circuit aquarium; salinity 36 %; 16 ± 0.5 °C; 12/12 h dark/light cycle)

containing a 10 cm layer of sea water running over a 2 cm layer of spiked sediments. A

seperate group of 5 sea stars were placed in the same aquaria, but in another compartment (not

in contact with the sediments), to serve as a control for possible cross-contamination through

sea water. The sea stars were fed every second day with mussels (Mytilus edulis). Uningested

food was removed after 2 hours. The radioactivity of the labelled PCB was measured weekly in

the sediments to check for possible leaching (Table 1). Periodically (after 2, 4, 7, 11, 14, 21,

and 34 days), 3 individuals were removed, dissected as described above, and their tissues

counted for radioactivity.

Data analyses. Uptake of the PCB congener from sea water and sediments was expressed as

change in PCB concentration (ng g-1 total lipids) over time. Uptake kinetics were described

either by using a saturation exponential model (eq 1), a single component exponential model

(eq 2), or a combined model (logistic and single component exponential) (eq 3):

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$$C(t) = Css (1-e^{-k.t})$$
 (eq 1)

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$$C(t) = C(0) e^{k.t}$$
 (eq 2)

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$$Ct = Css (1-e^{-k.t}) / 1+e^{-k.(t-1)}$$
 (eq 3)

where C(t), C(0), and Css are the PCB concentrations (ng g⁻¹ total lipids), respectively, at time t

2 (d), at time 0 and at steady state, k is the rate constant (d⁻¹), and I is the time (d) at the inflexion

point. The model showing the best fitting accuracy (based on the calculation of the

determination coefficient, R^2 , and examination of the residuals) was used.

5 Constants of the different models and their statistics were estimated by iterative adjustment of

the models and Hessian matrix computation, respectively, using the nonlinear curve-fitting

routines in the Systat® 5.2.1 software (Wilkinson 1988). Differences between PCB

concentrations in the different sea star body compartments were tested by 1-way ANOVA and

the multiple comparison test of Tukey (Zar 1996). Changes in PCB body distribution were

tested for significance using the G-test (adapted from the log-likelihood ratio test) for 2xk

contingency tables (Zar 1996). Prior to the latter test, data were arcsin-transformed using the

correction of Freeman-Tukey (1950) described by Zar (1996). The level of significance for

statistical tests was always set at $\alpha = 0.05$.

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RESULTS

The uptake of PCB#153 by Asterias rubens was investigated through separate exposures to

contaminated sea water or sediments. As differences between accumulation kinetics in aboral

and oral body walls were never found in any experiment (p always > 0.1), these two

compartments were pooled and are presented as a single compartment (body wall) throughout

the text. The uptake kinetics of PCB congener #153 in 6 different body compartments (body

wall, pyloric caeca, gonads, rectal caeca, central digestive system, podia) are shown in Figures

2 and 3 for the sea water and sediment exposures, respectively.

Contamination via sea water. Depending on the body compartment, accumulation from sea

water was best described by a combined (logistic and exponential) model (viz. uptake in body

wall, pyloric caeca, gonads and podia) or a single component exponential model (viz. uptake in

central digestive system and rectal caeca) (Fig. 2, Table 2A).

- Body wall was the compartment that concentrated ¹⁴C-PCB#153 to the greatest degree, up to
- 2 two orders of magnitude higher than the rectal caeca ($p_{Tukey test} \le 0.0001$; Table 3).
- 3 Body distribution of incorporated ¹⁴C-PCB#153 varied significantly along the timecourse of
- 4 the experiment ($p_{G-test} < 0.05$). Initially, the contaminant was mostly present in the podia (74 \pm
- 5 % of total body load after 2 days of exposure) and secondarily in the body wall (26 ± 5 %).
- Progressively, the proportion of the PCB associated with body wall increased, reaching 69 ± 5
- 7 % of the total body burden after 34 days of exposure, while during the same time the podia
- 8 proportion had decreased to $7 \pm 2 \%$ (Table 4).
- 9 Contamination via sediments. Frequent radioanalysis of the contaminated sediments indicated
- that the maximum difference between measured ¹⁴C-PCB#153 activities was 13.1% and that no
- significant decreasing trends occurred; therefore, concentrations in labelled PCB remained
- relatively stable throughout the 34 d-long experiment (9.5 \pm 1.1 ng g⁻¹ dry wt; see Table 1).
- Similarly, radioactivity in the sea water and in control sea stars remained below the detection
- limit, indicating that no significant ¹⁴C-PCB was incorporated from suspended sediments
- possibly ingested by the mussels on which they fed nor from sea water due to cross
- 16 contamination.
- Accumulation from contaminated sediments was best described either by a single component
- exponential model (gonads), a saturation exponential model (podia), or a combined model
- 19 (body wall, rectal caeca, pyloric caeca and central digestive system) (Fig. 3, Table 2B). As
- 20 noted during the seawater exposure, body wall and podia were the body compartments that
- accumulated ¹⁴C-PCB#153 to the highest levels when exposed to labelled sediments (Table 5).
- The distribution of ¹⁴C-PCB in sea star tissues was determined at different times during the
- 23 timecourse of the experiment. Relative transfers among body compartments appeared quite
- 24 different from those observed during the seawater uptake experiment. Indeed, the proportion of
- 25 contaminant in the body wall and podia remained relatively constant throughout the

experiment. Body wall and podia contained the major part (ca. 60%) of the total body burden 1

of 14 C-PCB, while the lowest percentage was found in the rectal caeca ($\leq 0.3\%$) (Table 4).

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DISCUSSION

The present study reports the first experimental data on the bioaccumulation kinetics of a key PCB congener in the sea star Asterias rubens, a common species widely distributed in the North Sea and NE Atlantic. The fact that organisms were also exposed to very low background concentrations of stable PCB#153 (Table 1), showed that they were actually exposed to a global concentration of PCB#153 that did not differ significantly from the ¹⁴C-PCB concentrations added experimentally to sea water or sediments (Table 1). Experimental concentrations in sea water were higher than those usually reported for PCB#153 in the natural North Sea waters. However, the latter concentrations most generally concern the dissolved 12 fraction whereas our measurements involve both dissolved and particulate fractions. Although available PCB data on bulk seawater samples mostly concern the sum of congeners or PCB mixture equivalents, it is noteworthy that the experimental concentrations used here are quite close (even much lower if considering extreme hot spots) to values reported for moderate to highly contaminated marine locations (Table 1). In addition, the ratio between seawater and sediment PCB concentrations added is similar to the ratio between the background PCB concentrations measured in sea water and sediments used in the experiments (Table 1). Therefore, the experimental exposures may be considered as acceptable simulations of field exposure situations that may actually occur in the field. Data on PCB concentrations in Asterias rubens in the field are scarce, and even less are available when looking for congener-specific data (e.g. Everaarts et al. 1998, den Besten et al. 2001). It is noteworthy that the total PCB#153 concentrations (background + incorporated) reached in the pyloric caeca at the end of the experiments matched the concentrations reported in the same organs of sea stars from moderate to highly contaminated North Sea locations

(Table 6). No field data were found concerning PCB concentrations in the body wall. 1 2 Considering the whole body, PCB concentrations reached in experimentally-exposed sea stars were 2 to 10 times higher than the few data available from the literature (Everaarts & Fischer 3 4 1989; Table 6). However, these comparisons should be made with caution, since the latter 5 field values are derived from sea stars collected during the spawning period. Indeed, it has been 6 shown that the whole-body content of extractable lipids is strongly dependent on the sexual 7 state of individuals, and may fluctuate by a factor of 2 to 3, particularly during the spawning 8 period. This may result in a similar range of variations of PCB concentrations occurring within 9 a few weeks (Knickmeyer 1992; Everaarts et al. 1998; Table 6). 10 Whether sea water or sediments were considered as a contamination source, a steady-state was reached or tended to be reached in most body compartments during the course of the 11 experiments. This suggests either that target sites are rapidly saturated, or that a metabolization 12 mechanism is induced quite rapidly following PCB exposure. Although a MFO-like system has 13 14 been described in pyloric caeca of Asterias rubens by den Besten et al. (1990, 1993, 1998), it is well documented that PCB#153 is quite resistant to biological degradation (Sipes & 15 16 Schnellmann 1987, Letcher et al 2000) due to its specific structure lacking hydrogen atoms on 17 the biphenyl molecule (Borlakoglu & Wilkins 1993). Therefore, the hypothesis regarding target site saturation is considered to be the most plausible explanation. 18 19 It is also noteworthy that when a steady-state in uptake was observed, equilibrium 20 concentrations of PCB#153 were generally reached quite rapidly (around day 20), indicating that the sea star could be used as a bioindicator to pinpoint a PCB contamination event soon 21 after its occurrence. 22 Concentrations of incorporated ¹⁴C-PCB#153 at steady-state were much higher (up to 300 23 24 times) in body wall and podia than in any other compartment. Being easily dissected and constituting 70-80% of the total body weight, body wall is of particular interest with respect to 25 26 field surveys, and it should be recommended as a body compartment to monitor

- 1 complementarily to pyloric caeca which are the only body compartment that has been used in
- previous studies (e.g., Everaarts et al. 1998, den Besten et al. 1993, 2001).
- 3 Concentrations incorporated into the rectal caeca were always low, between one and two orders
- 4 of magnitude lower than all the other compartments. This is somewhat surprising but could be
- 5 related to the functions of the rectal caeca which are well known to play an essential role in sea
- 6 star digestion and excretion processes (Jangoux 1982, Warnau & Jangoux 1999).
- 7 Our results have shown that PCB uptake is far more efficient in sea stars exposed to spiked sea
- 8 water than to labelled sediments when compared to exposure concentrations. For a given body
- 9 compartment, calculated concentration factors (CFs) based on sea water were between 2 and 3
- orders of magnitude higher than transfer factors (TFs) from sediments (Tables 3 and 5).
- Therefore, over the long term, despite the fact that sediments constitute the main reservoir of
- PCBs in the marine environment and that seawater PCB concentrations are comparatively
- extremely low, sea water would be an important route for PCB bioaccumulation in this sea star
- as it has been suggested for certain benthic infauna (e.g. Fowler et al. 1978). However, this
- does not imply that sea water would be the predominant pathway for PCB uptake, since our
- results showed that final concentrations reached in the different body compartiments following
- the two types of exposure were generally of the same order of magnitude. In addition, direct
- trophic transfer was not addressed here and could also contribute significantly to PCB
- 19 bioaccumulation in the sea star.
- While this work constitutes the first report on PCB bioaccumulation kinetics in a sea star,
- 21 several previous studies have used radiolabelled ¹⁴C-PCB to examine bioaccumulation kinetics
- in other aquatic organisms (e.g., Goerke et al. 1973, Gooch & Hamdy 1982, Schweitzer et al.
- 23 1997). However, surprisingly, these studies mostly concern PCBs as Aroclor equivalents (see
- e.g. Butcher et al. 1997). The main advantage of the ¹⁴C approach to measure PCB fluxes and
- 25 transfers in aquatic biota is obviously the high sensitivity and the rapidity of the detection,
- 26 compared to analytical techniques using gas chromatography. It therefore constitutes an

- 1 interesting tool, since current research on the behaviour of PCBs in the environment tends to
- 2 focus on congener-specific information (Safe 1990, Metcalfe 1994, Letcher et al. 2000).
- 3 Furthermore, it allows working with low (realistic) PCB concentrations, and assessing uptake
- 4 in organs which are often too small to be analyzed by classical chemical methodologies.

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REFERENCES

- Atuma SS, Linder CE, Andersson Ö, Bergh A, Hansson L, Wicklund-Glynn A (1996) CB153
- as indicator for congener specific determination of PCBs in diverse fish species from
- swedish waters. Chemosphere 33:1459-1464
- Barnes H, Blackstock J (1973) Estimation of lipids in marine animals and tissues: detailed
- investigation of the sulfophosphovanillin for «total lipids». J Exp Mar Biol Ecol 12:103-
- 20 118
- 21 Bergen BJ, Nelson WG, Pruell RJ (1996) Comparison of nonplanar and coplanar PCB
- congener partitioning in seawater and bioaccumulation in blue mussels (Mytilus edulis).
- 23 Environ Toxicol Chem 15:1517-1523
- Boon JP, Van Zantvoort MB, Govaert MJMA (1985) Organochlorines in benthic polychaetes
- 25 (Nephtys spp) and sediments from the southern North Sea. Identification of individual
- PCB components. Neth J Sea Res 19:93-105

- 1 Borlakoglu JT, Wilking JPG (1993) Correlations between the molecular structures of
- 2 polyhalogenated biphenyls and their metabolism by hepatic microsomal monooxygenases.
- 3 Comp Biochem Physiol 105(C):113-117
- 4 Butcher JB, Gauthier TD, Garvey EA (1997) Use of historical PCB aroclors measurement:
- 5 Hudson river fish data. Environ Toxicol Chem 16:1618-1623
- 6 Chapman PM (1995) Ecotoxicology and Pollution: key issues. Mar Pollut Bull
- 7 31:167-177
- 8 Carr RS, Long ER, Windom HL, Chapman DC, Thursby G (1996) Sediment quality
- 9 assessment studies of Tampa Bay, Florida. Environ Toxicol Chem 15:1218-1231
- den Besten PJ (1998) Cytochrome P450 monooxygenase system in echinoderms. Comp
- 11 Biochem Physiol 121(C):139-146
- den Besten PJ, Herwig HJ, Zandee DI, Voogt PA (1989) Effect of cadmium and PCBs on
- reproduction of the sea star *Asterias rubens*: aberration in the early developpement.
- Ecotoxicol Environ Saf 18:173-180
- den Besten PJ, Herwig HJ, Van Donselaar EG, Livingstone DR (1990) Cytochrome P450
- monooxygenase system and benzo(a)pyrene metabolism in echinoderms. Mar Biol
- 17 107:171-177
- den Besten PJ, Lemaire P, Livingstone DR, Woodin B, Stegeman JJ, Herwig HJ, Seinen W
- 19 (1993) Time-course and dose-response of the apparent induction of the cytochrome P450
- 20 monooxygenase system of pyloric caeca microsomes of the female sea star Asterias rubens
- L. by benzo(a)pyrene and polychlorinated biphenyls. Aquat Toxicol 26:23-40
- den Besten PJ, Valk S, van Weerlee E, Nolting RF, Postma JF, Everaarts JM (2001)
- Bioaccumulation and biomarkers in the sea star Asterias rubens (Echinodermata:
- Asteroidea): a North Sea field study. Mar Environ Res 51:365-387
- Elder D (1976) PCBs in NW Mediterranean coastal waters. Mar Pollut Bull 7:63-64

- 1 Everaarts JM, Fischer CV (1989) Microcontaminants in surface sediments and macrobenthic
- 2 invertebrates of teh North sea. NIOZ Report 1989-6, 44 pp
- 3 Everaarts JM, Otter E, Fischer CV (1990) Cadmium and polychlorinated biphenyls: different
- distribution patterns in North Sea benthic biota. Neth J Sea Res 26:75-82
- 5 Everaarts JM, den Besten PJ, Hillebrand MTJ, Halbrook RS, Shugart LR (1998) DNA strand
- 6 breaks, cytochrome P450-dependant monooxygenase system activity and levels of
- 7 chlorinated biphenyl congeners in the pyloric caeca of the seastar (Asterias rubens) from
- 8 the North Sea. Ecotoxicology 7:69-79
- 9 Fowler SW, Polikarpov GG, Elder DL, Parsi P & Villeneuve JP (1978) Polychlorinated
- biphenyls: accumulation from contaminated sediments and water by the polychaete *Nereis*
- 11 diversicolor. Mar Biol 48:303-309
- Goerke H, Ernst W (1977) Fate of ¹⁴C-labelled di-, tri-, and pentachlorobiphenyl in the marine
- annelid *Nereis virens*. Chemosphere 9:551-558
- Gooch JA, Hamdy MK (1982) Depuration and biological half-life of ¹⁴C-PCB in aquatic
- organisms. Bull Environ Contam Toxicol 28:305-312
- Harvey GR, Steinhauer WG (1976) Biogeochemistry of PCB and DDT in the North Atlantic.
- In: Nriagu JO (ed) Environmental Biogeochemistry, vol. 1. Ann Arbor Science Publ, Ann
- 18 Arbor, p. 203-221
- Jangoux M (1982) Digestive systems: Asteroidea. In: Jangoux M, Lawrence JM (eds)
- Echinoderm Nutrition. Balkema, Rotterdam, p 235-272
- 21 Knickmeyer R, Landgraff O, Steinhart H (1992) Cyclic organochlorines in the seastar Asterias
- *rubens* from the German Bight, December 1988-May 1989. Mar Environ Res 33:127-143
- Laane RWPM, Sonneveldt HLA, Vander Weyden AJ, Loch JPG, Groenveld G (1999) Trend in
- 24 the spatial and temporal distribution of metals (Cd, Cu, Zn and Pb) and organic

- compounds (PCBs and PAHs) in the dutch coastal zone sediments from 1981 to 1996: a
- 2 model case study for Cd and PCBs. J Sea Res 41:1-17
- 3 Letcher RJ, Klasson-Wehler E, Bergman A (2000) Methyl sulfone and hydroxylated
- 4 metabolites of polychlorinated biphenyls. In: Paasivirta J (ed) The handbook of
- 5 environmental chemistry Vol 3, Part K: New types of persistent halogenated compounds.
- 6 Springer-Verlag, Berlin, Heidelberg, p 315-359
- 7 Lewis JR (1978) The implications of community structure of benthic monitoring studies. Mar
- 8 Pollut Bull 9:64-67
- 9 Marchand M, Abarnou A, Marcaillou-Lebaut C (1990) Les polychlorobyphényles (PCB) en
- milieu marin : biogéochimie et écotoxicologie. Rapp Scient Techn IFREMER 18, 162 pp
- 11 Metcalfe CD (1994) Polychlorinated biphenyls. In: Kiceniuk JW, Ray S (eds) Analysis of
- contaminants in edible aquatic resources. VCH Press, Berlin, p. 305-338
- Murdoch MH, Chapman PM, Norman DM, Quintino VM (1997) Spiking sediment with
- organochlorines for toxicity testing. Environ Toxicol Chem 16:1504-1509
- Safe S (1990) Polychlorinated biphenyls (PCBs), dibenzo-p-dioxins (PCDDs), dibenzofurans
- 16 (PCDFs) and related compounds: environmental and mechanistic considerations which
- support the development of toxic equivalency factors (TEFs). CRC Crit Rev Toxicol
- 18 3:293-303
- 19 Schultz-Bull DE, Petrick G, Kannan N, Duinker JC (1995) Distribution of individual
- 20 chlorobiphenyls (PCB) in solution and suspension in the Baltic Sea. Mar Chem 48:245-
- 21 270
- Schweitzer LE, Bay SM, Suffet IH (2000) Dietary assimilation of a polychlorinated biphenyl in
- adult sea urchins (*Lytechinus pictus*) and maternal transfer to their offspring. Environ
- 24 Toxicol Chem 19:1919-1924

- 1 Schweitzer LE, Hose JE, Suffet IH, Bay SM (1997) Differential toxicity of three
- 2 polychlorinated biphenyl congeners in developing sea urchin embryos. Environ Toxicol
- 3 Chem 16:1510-1514
- 4 Sipes IG, Schnellmann RG (1987) Biotransformation of PCBs: metabolic pathways and
- 5 mechanisms. In: Safe S (ed) Polychlorinated biphenyls: mammalian and environmental
- 6 toxicology, Springer Verlag, Berlin, p. 97-110
- 7 Stebbing ARD, Dethlefsen V, Carr M (1992) Biological effects of contaminants in the North
- 8 Sea. Mar Ecol Prog Ser 91 (1-3), 361 pp
- 9 Telli-Karakoç F, Tolun L, Henkelmann B, Klimm C, Okay O, Schramm KW (2002) Polycyclic
- hydrocarbons (PAHs) and polychlorinated biphenyls (PCBs) distributions in the Bay of
- Marmara sea : Izmit Bay. Environ Pollut 119:383-397
- Temara A, Skei JM, Gillan D, Warnau M, Jangoux M, Dubois Ph (1998) Validation of the
- asteroid Asterias rubens (Echinodermata) as a bioindicator of spatial and temporal trends
- of Pb, Cd, and Zn contamination in the field. Mar Environ Res 45:341-356
- Warnau M, Jangoux M (1999) *In vitro* and *in vivo* investigations of the excretory function of
- the rectal caeca in the asteroid *Asterias rubens* (Echinodermata). Comp Biochem Physiol
- 17 123(A):263-267
- Warnau M, Teyssié JL, Fowler SW (1996) Biokinetics of selected heavy metals and
- radionuclides in the common Mediterranean echinoid *Paracentrotus lividus*: sea water and
- food exposures. Mar Ecol Prog Ser 141:83-94
- Warnau M, Fowler SW, Teyssié JL (1999) Biokinetics of radiocobalt in the asteroid Asterias
- *rubens* (Echinodermata): seawater and food exposures. Mar Pollut Bull 39:159-164
- Weisberg S, Francisco C, Hallock D (1996) Bioaccumulation and toxicity of PCB in sea
- urchins exposed to contaminated sediments. Southern California Coastal Water Res.
- 25 Project Annual Report 1996, Westminster, CA, USA
- Wilkinson L (1988) Systat: the system to statistics. Systat Inc, Evanston, IL

- Wood L, O'Keefe P, Bush B (1997) Similarity analysis of PAH and PCB bioaccumulation
- 2 patterns in sediment-exposed *Chironomus tentans* larvae. Environ Toxicol Chem 16:283-
- 3 292
- 4 Zar JH (1996) Biostatistical analysis, 3rd edition. Simon & Schuster/A Viacom company,
- 5 Upper Saddle River, NJ, 850 pp

CAPTIONS TO FIGURES: Figure 1. Schematic representation of sample processing before β-spectrometry analysis Figure 2. Asterias rubens-Seawater experiment. Uptake of ¹⁴C-PCB#153 from seawater in different body compartments of the sea star (mean concentration in ng g^{-1} total lipids \pm SD, n=3) Figure 3. Asterias rubens-Sediment experiment. Uptake of ¹⁴C-PCB#153 from sediments in different body compartments of the sea star (mean concentration in ng g^{-1} total lipids \pm SD, n=3).

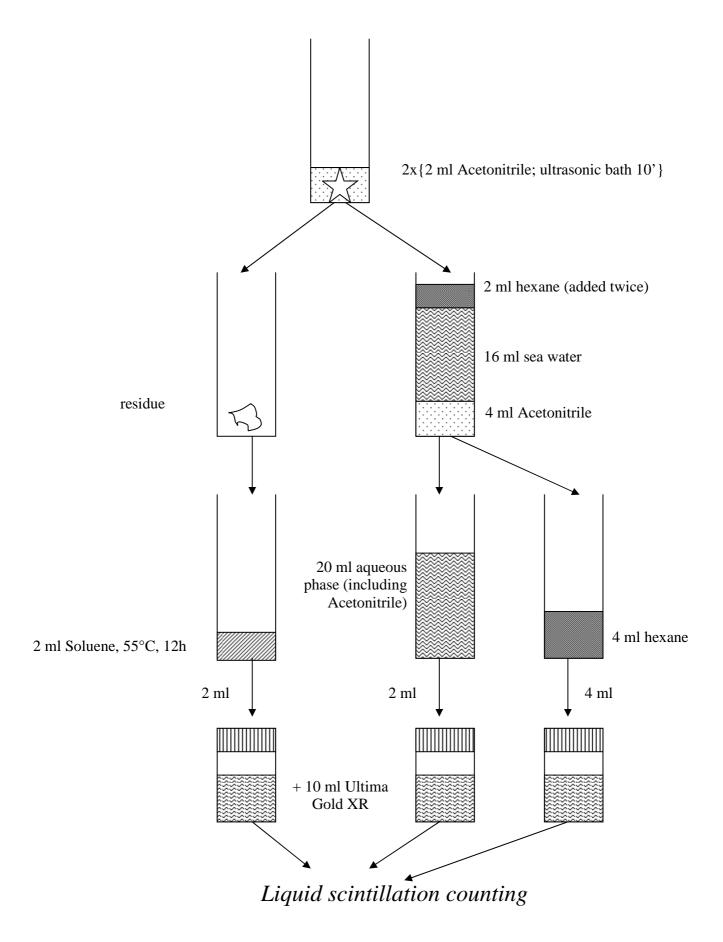


Figure 1

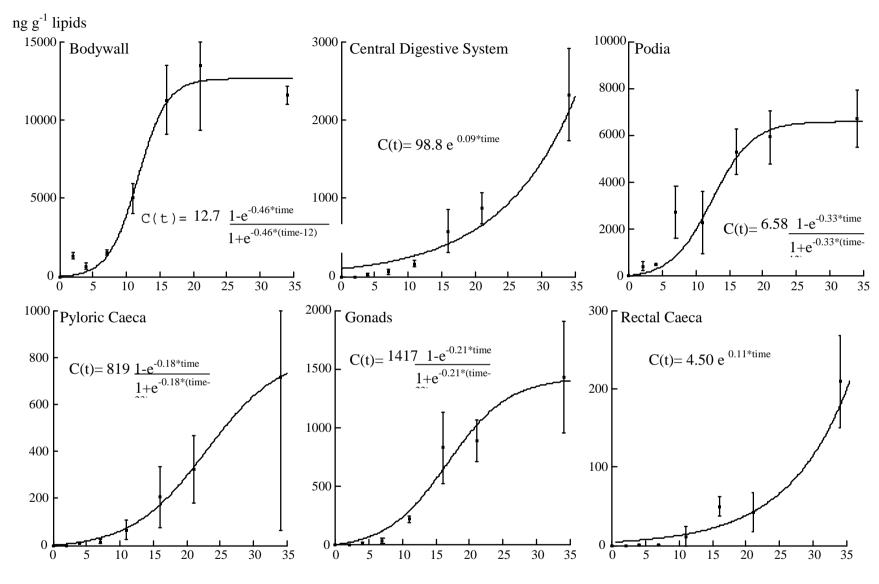


Fig. 2

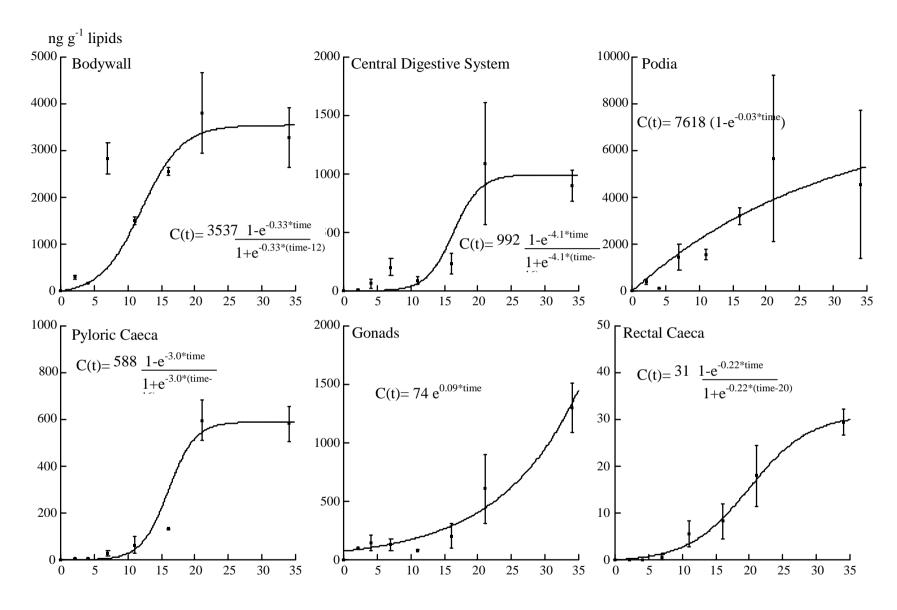


Fig. 3

Table 1. Characteristics of the background and added concentrations of PCB#153.

Background concentrations were measured in sediments, sea water and sea stars (body wall and pyloric caeca) the day before starting the experiment; added concentrations were measured in subsamples of sediments and sea water regularly collected in the experimental microcosms throughout the experiment. Ranges of values of PCB#153 (unless specified) reported for sediments and sea water in the field are given for comparison.

| | Compartment | PCB concentration | Location | References |
|--------------|--|---|---------------------------------|----------------------------------|
| Background | Sediments | $0.017 \text{ ng g}^{-1} \text{ dry wt } (n = 6)$ | | |
| | Sea water | $0.026 \text{ ng } 1^{-1} \text{ (n = 6)}$ | | |
| | Pyloric caeca | $522 \pm 167 \text{ ng g}^{-1} \text{ lipids (n = 6)}$ | | |
| | Body wall | $559 \pm 17 \text{ ng g}^{-1} \text{ lipids } (n = 6)$ | | |
| Added | Sediments | $9.49 \pm 1.14 \text{ ng g}^{-1} \text{ dry wt (n = 12)}$ | | |
| | Sea water (dissolved + particulate) | $31.4 \pm 15.6 \text{ ng l}^{-1} \text{ (n} = 36)$ | | |
| Field Values | Sediments | 22 - 4,060 ng g ⁻¹ dry wt | North Sea, German Bight | Stebbing et al 1992 |
| | | 0.27 - 47 ng g ⁻¹ dry wt (sum ₇ PCB) | North Sea, Dutch coastal zone | Boon et al. 1985 |
| | | $2.2 - 32 \text{ ng g}^{-1} \text{ dry wt (sum}_7 \text{ PCB)}$ | North Sea, Dutch coastal zone | Laane et al. 1999 |
| | Sea water (dissolved) | 0.1 - 67.2 pg l ⁻¹ | Baltic Sea | Shultz-Bull et al. 1995 |
| | Sea water | $0.8 - 8.7 \text{ ng } 1^{-1} \text{ (Aroclor 1260)}$ | Atlantic | Harvey & Steinhauer 1976 |
| | (dissolved + particulate) | 1.5 – 38.0 ng l ⁻¹ (Phenoclor DP-5) | Mediterranean French coasts | Elder 1976 |
| | | $0.2 - 370 \text{ ng } 1^{-1}$ (Phenoclor DP-5/DP-6) | Med. and Atlantic French coasts | Marchand et al. 1990 |
| | | $0.34 - 4.93 \text{ ng } 1^{-1} \text{ (sum hexa-CB)}$ | Marmara Sea | Telli-Karakoç <i>et al.</i> 2002 |
| | Sea water (extreme hot spot) | dissolved: $1.8 \pm 0.3 \mu g l^{-1}$ | New Bedford Harbor, USA | Bergen et al. 1996 |
| | | particulate: $14 \pm 3.9 \mu g \Gamma^1$ | | |

Table 2. Asterias rubens. Parameters and statistics of the equations describing the uptake of ¹⁴C-PCB #153 from sea water and sediments in the body compartments of the sea star.

E (exponential model): $C(t) = C_0 e^{k.t}$; S (saturation model): $C(t) = Css.(1-e^{-k.t})$; C (combined model): $C(t) = Css.(1-e^{-k.t})/(1+e^{-k.(t-I)})$;

where C_0 , C(t), $Css: {}^{14}C\text{-PCB}$ #153 concentrations (ng g^{-1} lipids) respectively at time 0, at time t (d) and at steady-state; k: rate constant (d^{-1}); I: time (d) at the inflexion point; ASE: asymptotic standard error; R^2 : corrected determination coefficient.

2A. Sea water

| Body compartment | Model | C_0 (ASE) | Css (ASE) | k (ASE) | I (ASE) | R^2 |
|--------------------------|-------|-------------|--------------|---------------|-------------|-------|
| Body wall | C | | 12,665 (691) | 0.46 (0.13) | 11.7 (0.67) | 0.92 |
| Central digestive system | E | 98.8 (27.7) | | 0.093 (0.009) | | 0.77 |
| Rectal caeca | E | 4.5 (2.0) | | 0.11 (0.01) | | 0.91 |
| Pyloric caeca | C | | 819 (398) | 0.18 (0.17) | 22.9 (8.2) | 0.80 |
| Gonads | C | | 1,417 (231) | 0.21 (0.11) | 23 (2.8) | 0.90 |
| Podia | С | | 6,584 (449) | 0.33 (0.12) | 12.4 (0.87) | 0.93 |

2B. Sediments

| Body compartment | Model | C_0 (ASE) | Css (ASE) | k (ASE) | I (ASE) | \mathbb{R}^2 |
|--------------------------|-------|-------------|--------------|---------------|-----------|----------------|
| Body wall | C | | 3,537 (206) | 0.33 (0.08) | 12 (0.92) | 0.93 |
| Central digestive system | C | | 992 (81) | 4.1 (29) | 16 (2.0) | 0.81 |
| Rectal caeca | C | | 31 (2.3) | 0.22(0.05) | 20 (1.1) | 0.94 |
| Pyloric caeca | C | | 588 (18) | 3.0 (11) | 16 (1.5) | 0.97 |
| Gonads | E | 74 (19) | | 0.085 (0.008) | | 0.89 |
| Podia | S | | 7,618 (4266) | 0.034 (0.029) | | 0.57 |

Table 3. Asterias rubens. Concentration factors (CF; maximum, minimum and mean values) in the body compartments of the sea star after 34 days of exposure via sea water.

CFs are calculated as the ratio between PCB#153 concentration in the sea star body compartments (ng g⁻¹ total lipids) and its concentration in sea water (ng g⁻¹).

| | Body wall | Central dig. syst. | Gonads | Rectal caeca | Pyloric caeca | Podia |
|---------|--------------|--------------------|--------------|--------------|-------------------|---------------|
| Max. CF | $3.91\ 10^5$ | $9.16\ 10^4$ | $6.01\ 10^4$ | $7.90\ 10^3$ | $4.75 \cdot 10^4$ | $2.43 \ 10^5$ |
| Min. CF | $3.52\ 10^5$ | $5.44\ 10^4$ | $2.96\ 10^4$ | $4.58\ 10^3$ | $1.05 10^4$ | $1.72 \ 10^5$ |
| Mean CF | $3.74\ 10^5$ | $7.50 \ 10^4$ | $4.62\ 10^4$ | $6.76\ 10^3$ | $2.31\ 10^4$ | $2.17 \ 10^5$ |

Table 4. Asterias rubens. PCB distribution (mean $\% \pm SD$, n = 3) in the different body compartments of the sea star after 34 days of exposure via sea water or sediments.

| Body compartments | ¹⁴ C-PCB-153 distribution (%) | | | |
|--------------------------|--|-------------------|--|--|
| | Seawater exposure | Sediment exposure | | |
| Body wall | 68.8 ± 1.4 | 20.7 ± 4.4 | | |
| Central digestive System | 13.9 ± 4.1 | 8.9 ± 2.3 | | |
| Gonads | 8.4 ± 2.5 | 12.7 ± 2.5 | | |
| Rectal caeca | 0.2 ± 0.1 | 0.3 ± 0.1 | | |
| Pyloric caeca | 1.8 ± 0.7 | 5.7 ± 1.4 | | |
| Podia | 6.8 ± 1.9 | 39.9 ± 14.6 | | |

Table 5. Asterias rubens. Transfer factors (TF; maximum, minimum, and mean values) in the body compartments of the sea star after 34 days of exposure via sediments.

TFs are calculated as the ratio between PCB#153 concentration in the sea star body compartments (ng g⁻¹ total lipids) and its concentration in sediments (ng g⁻¹ dry wt).

| | Body wall | Central dig. syst. | Gonads | Rectal caeca | Pyloric caeca | Podia |
|---------|-----------|--------------------|--------|--------------|---------------|-------|
| Max. TF | 417 | 109 | 150 | 3.43 | 70 | 863 |
| Min. TF | 286 | 81 | 111 | 2.91 | 55 | 258 |
| Mean TF | 343 | 94 | 137 | 3.10 | 61 | 479 |

Table 6. *Asterias rubens*. Comparisons among PCB #153 concentrations obtained in the present study (background + incorporated concentrations) and previous field studies in the North Sea.

| Body compartment | PCB#153 concentration (ng g ⁻¹ lipids) | Specifications | References |
|------------------|---|---|--------------------------|
| Pyloric caeca | 608 - 1,111 | Experimental conditions; seawater uptake | Present study |
| | 920 - 1,377 | Experimental conditions; sediment uptake | Present study |
| | 41 - 1,054 | Pre-spawning period 1995; southern North Sea | den Besten et al. 2001 |
| | 450 - 1,050 | Pre-spawning period 1995; Dutch coastal zone | Everaarts et al. 1998 |
| | 40 - 125 | Pre-spawning period 1995; southern North Sea | Everaarts et al. 1998 |
| Bodywall | 728 - 4,360 | Experimental conditions; seawater uptake | Present study |
| | 1,215 - 14,068 | Experimental conditions; sediment uptake | Present study |
| Whole body | 8,190 - 9,300 | Experimental conditions; seawater uptake | Present study |
| | 2,330 - 2,810 | Experimental conditions; sediment uptake | Present study |
| | 550 - 940 | Spawning period 1986; Dutch coastal zone | Everaarts & Fischer 1989 |
| | 100 - 235 | Spawning period 1986; southern North Sea | Everaarts & Fischer 1989 |
| | 4,300 (sum ₃₅ PCB) | Spawning period 1989 ; German Bight | Knickmeyer et al. 1992 |
| | 2,400 (sum ₃₅ PCB) | Post-spawning period 1989; German Bight | Knickmeyer et al. 1992 |