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Microbial reductive dechlorination of weathered and exogenous co-planar polychlorinated biphenyls (PCBs) in an anaerobic sediment of Venice Lagoon

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Abstract

The occurrence of reductive dechlorination processes towards pre-existing PCBs and five exogenous coplanar PCBs were investigated in a contaminated sediment of Porto Marghera (Venice Lagoon, Italy) suspended, under strictly anaerobic conditions, in water collected from the same site. PCB dechlorination started after five months of incubation, when sulfate initially occurring in the microcosms was completely depleted and methanogenesis was in progress. It was ascribed to sulfate-reducing bacteria. Several pre-existing hexa-, penta- and tetra-chlorinated biphenyls were slowly bioconverted into tri- and di-, *ortho*-substituted PCBs from the 5th to the 16th month of experiment. Spiked coplanar PCBs, i.e., 3,3′,4,4′-tetrachlorobiphenyl, 3,3′,4,4′,5- and 2,3′,4,4′,5-pentachlorobiphenyls, 3,3′,4,4′,5,5′- and 2,3,3′,4,4′,5-hexachlorobiphenyls, were extensively transformed (by about 90%) into lower chlorinated congeners, such as 3,3′,5,5′-/2,3′,4,4′-tetrachlorobiphenyl, 3,3′,5-, 2,4,4′-, 2,3′,4- and 2,3′,5-trichlorobiphenyl, 3,4-/3,4′- and 3,3′-dichlorobiphenyl and 2-chlorobiphenyl. The reductive dechlorination of spiked PCBs did not influence significantly the biotransformation rate and extent of pre-existing PCBs.

Introduction

PCBs are contaminants of great environmental concern that, due to their poor biodegradability and high hydrophobicity, tend to strongly accumulate in anoxic sediments rich in reduced organic matter (Brown & Wagner 1990).

PCBs occurring in freshwater sediments have been proven to be reductively dechlorinated in anaerobic primary microcosms developed with defined mineral media (Bedard & Quensen 1995; Wiegel & Wu 2000). With few exceptions, the dechlorination activity was generally directed to the *meta*- and *para*-chlorines of the biphenyl molecule, resulting in the bioconversion of highly chlorinated PCBs into low-chlorinated *ortho*-substituted congeners (Bedard & Quensen 1995; Wiegel & Wu 2000). Such an activity has been often ascribed to methanogenic bacteria that fortuitously use PCBs as terminal electron acceptors (Bedard & Quensen 1995; Wiegel & Wu 2000). Several studies showed that supplementation of contaminated freshwater sediments with PCBs or polybrominated biphenyls (Bedard et al. 1996, 1997; Van Dort et al. 1997), brominated and iodinated benzoic acids (Bedard et al. 1998; Wu et al. 1999) can stimulate dehalogenating microorganisms, thus increasing rate and yield of weathered PCB dechlorination.

Much less is still known about the occurrence of PCB reductive dechlorination in marine sediments (Lake et al. 1992; Alder et al. 1993; Øfjord et al. 1994; Berkaw et al. 1996; Palekar et al. 2003).

Furthermore, the majority of studies available on marine sediments have been done on uncontaminated sediments spiked with mixtures of PCBs and suspended in synthetic media, and this markedly limits the practical relevance of the information provided. To our knowledge, reductive dechlorination of weathered PCBs has been only documented in a marine sediment of the New Bedford Harbor suspended in synthetic mineral medium under methanogenic conditions (Alder et al. 1993). A similar study was done on marine sediments of LCP Chemical Superfund site (Georgia) suspended in filtered water from the same contaminated site, but no dechlorination of pre-existing PCBs was observed (Palekar et al. 2003). Very recently, dechlorination of weathered PCBs was documented in three contaminated marine sediments of the Brentella Canal (Porto Marghera, Venice Lagoon) when suspended either in a synthetic marine medium or in water coming from the site (i.e., under geochemical conditions very close to those occurring *in situ*). The dechlorination was more extensive in the presence of the site water than with the mineral medium (Fava et al. 2003a), and it was apparently mediated by sulfate-reducing spore forming indigenous bacteria, selective towards *meta* and *para* positions of PCB molecules, and not 'primed' by the addition of exogenous 2,3,4,5,6-pentachlorobiphenyl (Fava et al. 2003b).

Therefore, more information on the biological fate of weathered PCBs in marine sediments is desirable. It is also desirable that such information is obtained through laboratory experiments that closely mimic geochemical conditions occurring at the site, as this might permit to estimate the actual potential of *in situ* microbial processes in the final site decontamination/detoxification. On the basis of these considerations, we found of relevance to extend our survey on the occurrence of PCB microbial dechlorination processes to other sediment samples of the Brentella Canal, which has been closed to navigation due to its high chemical pollution. Therefore, in this work we looked at microbial transformation processes of weathered PCB in a contaminated sediment collected from a new district of that Canal. We also investigated the possible fate of five highly toxic coplanar PCBs in the same sediment, and the effects of their addition on the microbial transformation of pre-existing PCBs.

Materials and methods

Sediment and water samples

A sample of 2 kg of anaerobic sediment was obtained from the top 20 cm of the sediment layer in the Brentella canal of the first industrial area of Porto Marghera, Venice Lagoon (see Moret et al. 2001 for a map of the area), where a 4 l sample of site water was also collected. Both sediment and water samples were stored at 4 °C in plastic jars filled to the top until used. The sediment was black, silty mud and contained approximately 1.6 mg kg^{-1} (on dry wt basis) of a mixture of PCBs, which could be partially ascribed to PCBs of Aroclor 1242 and Aroclor 1254. The density, pH and dry weight of the sediment were 1663.4 ± 24.3 g l^{-1}, 7.50 ± 0.10 and 59.1 ± 1.8 % (w/w), respectively. Lagoon water collected from the same marine environment contained 20.45 ± 0.49 g l^{-1} Cl^-, 43.87 ± 0.05 mg l^{-1} NO_2^-, 2.05 ± 0.09 g l^{-1} SO_4^{2-} and had a pH of 7.40 ± 0.05. No PCBs were detected in that lagoon water.

Microcosms preparation and sampling

All microcosms were prepared from a preliminary sediment slurry as described by Fava et al. (2003b). In brief, 0.45 l of the marine water sampled from the lagoon were vigorously mixed through a magnetic stirrer and purged with 0.22-μm filter-sterilized O_2 free N_2: CO_2 (70:30) with a Hungate-similar apparatus in a 1-l Erlenmeyer flask (previously deoxygenated with filter-sterilized O_2 free N_2: CO_2 70:30) at room temperature for 2 h. Anaerobic sediment (~0.15 l) was then added and the resulting slurry (25 % v/v) was mixed and purged for 2 h. While stirring and purging, 50 ml aliquots of sediment slurry where transferred into eight 50-ml sterile serum bottles equipped with a sterile magnetic bar and purged with filter-sterilized O_2 free N_2:CO_2 (70:30). The bottles were then sealed with sterile Teflon-coated butyl stoppers and aluminum crimp sealers. Four of them were temporarily stored at 4 °C in the dark, while the others were autoclaved at 121 °C for 1 h on three consecutive days with incubation at 28 °C between each autoclaving treatment to prepare sterile controls. All microcosms were then opened under sterile conditions, stirred with a magnetic bar and kept under filter-sterilized O_2 free N_2:CO_2 (70:30)

flushing. While mixing and flushing, each exogenous PCB was individually added (123 μl of a 10,000 mg l^{-1} solution prepared in acetone) to four of the microcosms (two biologically active and two autoclave-sterilized) to a final concentration of 100 mg kg^{-1} of dry sediment, whereas a volume of pure acetone identical to that employed to supply PCBs in the spiked microcosms was added to the parallel non-spiked microcosms. All microcosms were then immediately re-capped with sterile Teflon-coated butyl stoppers, further stirred for 5 min and incubated stationary at 25 ± 1 °C in the dark for 16 months, during which they were periodically sampled and analyzed to determine the volume and the composition of the head-space gas and the concentration of SO_4^{2-}, Cl^-, NO_3^-, NO_2^- and PCBs.

The microcosms sampling was performed as described by Fava et al. (2003b). In brief, each sealed microcosm was first subjected to stirring for 5 min followed by sampling of the head-space gas. Each microcosm was then opened under sterile conditions, stirring and purging with filter-sterilized O_2 free N_2: CO_2 (70:30) were applied, followed by sampling of sediment slurry for PCB and anion analysis. The head space was then flushed with filter-sterilized O_2 free N_2: CO_2 (70:30) and the microcosm recapped with a new sterile Teflon-coated butyl stopper and aluminum crimp sealer.

PCBs extraction and analytical procedures

PCBs were extracted from duplicate 0.3 ml aliquots of sediment slurry placed in 1.5-ml vials for gas chromatography (GC) equipped with Teflon-coated screw caps (Hewlett-Packard Co., Palo Alto, CA, USA) by using anhydrous diethyl ether (900 μl) in the presence of octachloronaphthalene (OCN; 10 μl of a 40 mg l^{-1} stock solution in hexane) and elemental mercury (150 μl), as reported in Fava et al. (2003b). The extraction protocol used yielded more than 90% of the initial amount of the five exogenous congeners employed from all active spiked microcosms (at the beginning of the experiment) and sterile spiked microcosms (throughout the whole experiment). The qualitative and quantitative analysis of the extracted PCBs was performed with a gas chromatograph (5890 series II) equipped with a HP–5 capillary column (30 m by 0.25 mm), a ^{63}Ni electron capture detector and a 6890 series II automatic sampler (Hewlett-Packard Co., Palo Alto, CA, USA) under the analytical conditions described by Fava et al. (2003b).

Qualitative analysis of the sediment-carried PCBs and of the freshly spiked PCBs and their possible dechlorination products was performed by comparing the retention time (relative to OCN) of the CG peaks obtained from the analysis of the sediment organic extracts with those of pure congeners and of PCBs occurring in standard Aroclor 1242 and Aroclor 1254 mixtures analyzed under identical conditions. Aroclor PCBs, injected in the presence of OCN, were identified as described by Fava et al. (2003b). Quantitative analysis of PCBs was performed by using the GC–ECD response factor of each target PCB obtained through linear calibration curves of Aroclors and pure congeners as previously reported (Fava et al. 2003b). Linear 5-point calibration curves (0.5–10.0 mg l^{-1} range) were also used to determine the GC–ECD response factor for the freshly spiked 3,3′,4,4′-tetrachlorobiphenyl, 3,3′,4,4′,5- and 2,3′,4,4′,5-pentachlorobiphenyl, 3,3′,4,4′,5,5′- and 2,3,3′,4,4′,5-hexachlorobiphenyl, as well as for their dechlorination products not occurring in the standard Aroclor mixtures and for lower chlorinated PCBs, such as 2,4-/2,5-, 2,4′-/2,3-3,4-, 3,4′- and 3,3′-dichlorobiphenyl and 2-, 3- and 4-chlorobiphenyl. Response factors were verified at each sampling/analysis. For data calculations, co-eluting congeners and homologues were assumed to be present in equal proportions. The concentrations were expressed as μmol of PCB kg^{-1} of dry sediment.

Gas production was measured with an air-tight syringe while its composition in CH_4, CO_2, N_2 and O_2 was analyzed with a Varian TCD 3300 gas chromatograph equipped with a Carbosieve S-II stainless steel column (3 m by 1/8 inch internal diameter) (Supelco, Inc., Bellefonte, PA, USA) and thermal conductivity detector (Varian Inc., Palo Alto, CA, USA) as described by Fava et al. (2003b).

The concentration of SO_4^{2-}, Cl^-, NO_3^- and NO_2^- in the sediment slurry was determined by using a Dionex DX-120 ion chromatograph equipped with an IonPac AS14 4 × 250 mm column, a conductivity detector combined to an ASRS-II Ultra conductivity suppressor system (Dionex, Sunnyvale, CA, USA) as described elsewhere (Fava et al. 2003b). Linear 4-point calibration curves (2.5–50.0 mg l^{-1} range) for SO_4^{2-}, Cl^-, NO_3^- and NO_2^- were obtained by using mixtures of these compounds.

Chemicals

3,3′,4,4′- Tetrachlorobiphenyl, 3,3′,4,4′,5- and 2,3′,4,4′,5- pentachlorobiphenyls, 3,3′,4,4′,5,5′- and 2,3,3′,4,4′,5- hexachlorobiphenyls, OCN, Aroclor 1242, Aroclor 1254 and all pure PCBs used as analytical standards were provided by Ultra-Scientific (North Kingstown, RI, USA). Anhydrous diethyl ether, acetone and hexane (both for pesticide analysis in capillary column GC systems) as well as the ultra-resi analyzed water for ion chromatography were supplied by Mallinckrodt-Baker (Phillipsburg, NJ, USA). Inorganic ions for IC analysis were provided by Aldrich (Strenheim, Germany).

Results and discussion

Biotransformation of weathered PCBs in the non-spiked microcosms

The initial concentration of total PCBs pre-existing in the sediment was 1.60 ± 0.13 mg kg^{-1} of dry sediment. No transformation of such PCBs was observed in the sterile microcosms throughout the experiment. On the contrary, a significant change in PCB profile was observed in the corresponding biologically active microcosms starting from the 5th month of incubation. At the end of the experiment (i.e., after 16 months), a marked depletion of several hexa-, penta- and tetra-chlorinated congeners, such as 2,2′,3,4,4′,5′-hexachlorobiphenyl, 3,3′,4,4′-tetrachlorobiphenyl/2,3,3′,4′,6–pentachlorobiphenyl, 2,3′,4,4′-tetrachlorobiphenyl/2,2′,3,5′,6-pentachlorobiphenyl, 2,3′,4′,5-, 2,2′,3,4-/2,2′,4,5-/2,4,4′,6-, 2,2′,5,5′-, 2,2′,3,6′- and 2,2′,3,6-tetrachlorobiphenyl, along with the accumulation of less chlorinated PCBs, such as 2,2′,5-/2,2′,4-/4,4′-chlorobiphenyl, 2,4'-/2,3-, 2,3'- and 2,4-/2,5-dichlorobiphenyl, were observed (Figure 1). 4-monochlorobiphenyl also accumulated (2.32 ± 0.27 μmol kg^{-1} of dry sediment) as compared to the sterile ones, where it occurred at 1.66 ± 0.54 μmol kg^{-1} of dry sediment. Conversely, 2-monochlorobiphenyl that occurred in the sterile microcosms at 1.34 ± 0.07 μmol kg^{-1} of dry sediment was depleted to 0.93 ± 0.34 μmol kg^{-1} of dry sediment. Remarkably, the total amount of PCBs, on a molar basis, did not change in the active microcosms throughout the 16-month experiment.

The accumulation of 2,3′-, 2,4′-, 2,2′,4- and 2,2′,5- chlorobiphenyls is a typical feature of dechlorination process H′ described by Bedard and Quensen (1995). In fact, the dechlorination activity was apparently directed mainly towards flanked *para* chlorines and *meta* chlorines of 2,3- and 2,3,4-chlorophenyl rings. However, accumulation of 2,4-/2,5-dichlorobiphenyl indicates that additional *meta* and *para* dechlorination patterns also occurred. Therefore, pre-existing PCBs were dechlorinated through both pattern H′ and pattern M, as previously observed, according to Bedard and Quensen (1995), in slurries of autoclaved sediment from Owasco Lake (NY) inoculated with microorganisms eluted from Hudson River sediments, where dechlorination process H′ and *meta* dechlorination typical of process M were

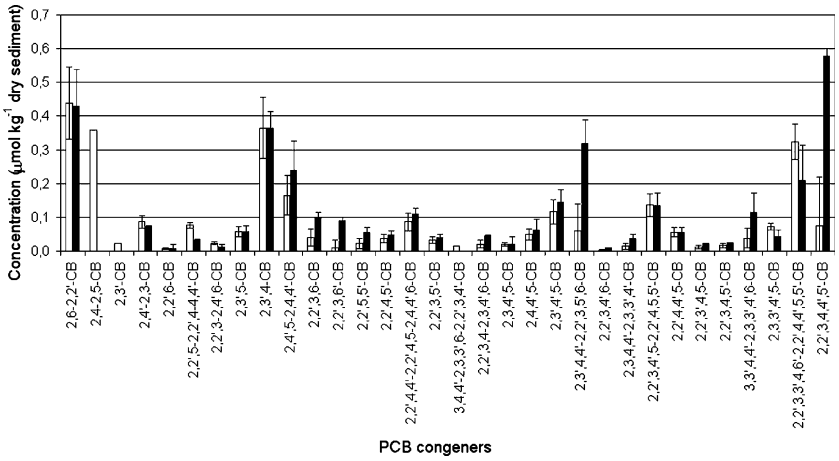

Figure 1. Average concentration of each sediment-carried PCB congener (± standard deviation as error bar) in the non-spiked sterile (black column) and biologically active (white column) microcosms after 16 months of incubation.

consecutively detected (Rhee et al. 1993), and in pyruvate and Aroclor 1242-supplemented enrichments of microorganisms from Hudson River sediments (Morris et al. 1992). Similar processes were found to describe the reductive dechlorination of weathered PCBs in other few marine sediments studied so far (Alder et al. 1993; Fava et al. 2003a, b). Alder et al. (1993) reported the occurrence of dechlorination pattern H′ towards pre-existing Aroclor 1242 PCBs in a sediment from New Bedford Harbor, whereas a prevalent *meta* dechlorination activity, tentatively ascribed to dechlorination process M, was detected in a sediment of the Brentella Canal (Venice Lagoon, Italy) contaminated by Aroclors 1242 and 1254 PCBs (Fava et al. 2003b).

Neither sulfate consumption nor biogas and methane evolution was detected in the sterile microcosms throughout the whole experiment (Figure 2). On the contrary, a little sulfate consumption was observed in the active microcosms since the first month of incubation. Sulfate was then quickly depleted, becoming 8.4% of its initial concentration (1.95 ± 0.03 g l^{-1}) at the end of the 2nd month of incubation and totally depleted at the end of the 3rd month of experiment (Figure 2). No significant biogas production was observed in the active microcosms until total sulfate depletion (Figure 2). A large amount of biogas (35.6 ± 9.2 ml), consisting of more than 47% (v/v) of methane, was detected in the same microcosms between the 3rd and the 5th month of incubation (Figure 2). PCB reductive dechlorination became detectable concomitantly to the end of sulfate reduction and the beginning of methanogenesis (data not shown). Similar results were obtained by Zwiernik et al. (1998) on a aged PCB-contaminated sediment of Hudson River amended with $FeSO_4$ and they seem to suggest, as proposed by Zwiernik et al. (1998), that sulfate-reducing bacteria capable of dechlorinating PCBs after sulfate depletion were responsible for PCB biodegradation. This hypothesis is also supported by the results of our recent work performed on another sediment of the same area of the Venice Lagoon (Fava et al. 2003b). However, it cannot be totally excluded that methanogenic bacteria were involved in the process, as it has been reported for other marine sediments (Alder et al. 1993), or that it was mediated by either sulfate-reducing and methanogenic bacteria along with fermentative bacteria, as it has been observed in other circumstances (Kim & Rhee 1999; Wiegel & Wu 2000).

Biotransformation of exogenous coplanar PCBs and of pre-existing PCBs in the spiked microcosms

3,3′,4,4′,5,5′- and 2,3,3′,4,4′,5-Hexachlorobiphenyl, 3,3′,4,4′,5- and 2,3′,4,4′,5-pentachlorobiphenyl

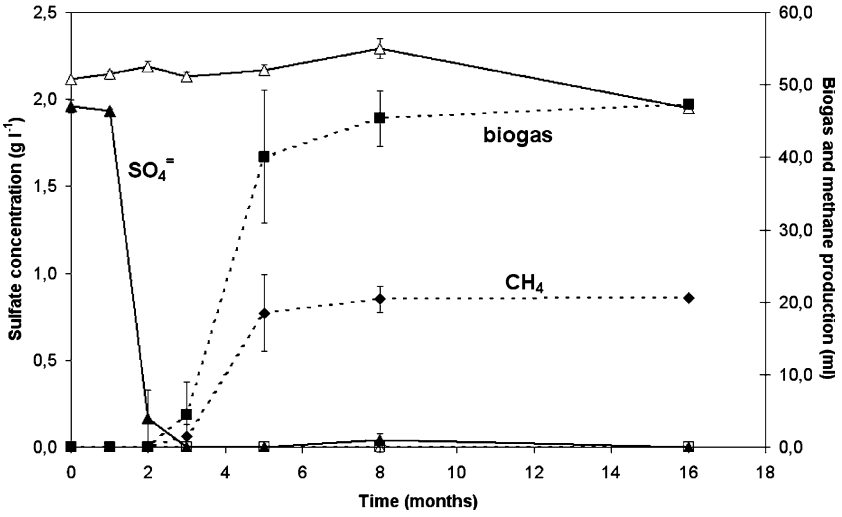

Figure 2. Sulfate concentration (triangles, solid line), biogas production (squares, dashed line) and methane production (diamonds, dashed line) detected in the non-spiked active microcosms (solid symbols) and sterile microcosms (empty symbols) throughout the 16-month experiment.

and 3,3′,4,4′-tetrachlorobiphenyl occurred in the spiked microcosms at the initial concentration of 534.94 ± 47.92, 327.53 ± 20.67, 296.63 ± 24.73, 333.70 ± 10.17 and 424.89 ± 13.91 μmol kg^{-1} of dry sediment, respectively. Their concentration did not significantly change in the sterile microcosms throughout the whole experiment. On the contrary, all exogenous PCBs were markedly bioconverted into less chlorinated congeners, such as 3,3′,5,5′-/2,3′,4,4′-, 3,3′,4′,5-, 2,3′,4′,5- and 2,4,4′,5-tetrachlorobiphenyl, 3,3′,5-, 2,4,4′-, 2,3′,4- and 2,3′,5-trichlorobiphenyl and 3,4-/3,4′- and 3,3′-dichlorobiphenyl, in the biologically active microcosms starting from the 5th month of incubation. The rapid accumulation of 2,3,3′,4,4′-pentachlorobiphenyl and 3,3′,4,5′-tetrachlorobiphenyl (Figure 3) indicates that 2,3,3′,4,4′,5-hexachlorobiphenyl and 3,3′,4,4′,5-pentachlorobiphenyl were the first two spiked PCBs to undergo dechlorination. Such dechlorination products, along with other products, such as 2,3′,4′,5- and 2,4,4′,5-tetrachlorobiphenyl, were found to be further transformed after the 8th month of incubation. Conversely, the metabolites 3,3′,5,5′-/2,3′,4,4′-tetrachlorobiphenyl, 3,3′,5-, 2,4,4′-, 2,3′,4- and 2,3′,5-trichlorobiphenyl and 3,4-/3,4′- and 3,3′-dichlorobiphenyl, along with 2-chlorobiphenyl, progressively accumulated throughout the whole 16-month experiment (Figure 3). At the end of the experiment, all the exogenous PCBs were found to be depleted by more than 90%. These findings are of great relevance, as they indicate that the microorganisms of the contaminated sediment employed in this study were able to rapidly and extensively dechlorinate five of the most toxic coplanar PCBs reported in the literature (Kimbrough 1995). Their activity was probably assisted by the high bioavailability of pollutants, which were surely much more bioavailable than aged, preexisting PCBs (Wiegel and Wu, 2000; Cornelissen et al. 2004).

The dechlorination of 3,3′,4,4′-tetrachlorobiphenyl, 3,3′,4,4′,5-pentachlorobiphenyl and 3,3′,4,4′,5,5′-hexachlorobiphenyl was also studied in two estuarine sediments of Tansui River and Erjen River by Kuo et al. (1999). In those sediments, 3,3′,4,4′-tetrachlorobiphenyl and 3,3′,4,4′,5-pentachlorobiphenyl were dechlorinated through rates, extents and lag periods that markedly depended on the sediment employed. Tansui River sediment exhibited only *para* dechlorination, whereas in the Erjen River sediment *meta* dechlorination of 2,5- and 3,5-chlorophenyl rings occurred only after complete *para* dechlorination. On the contrary, 3,3′,4,4′,5,5′-hexachlorobiphenyl was not transformed at all in microcosms of both sediments (Kuo et al. 1999). Therefore, the microbial population occurring in the Tansui River and Erjen River estuarine sediments exhibited a narrower and lower dechlorination activity towards coplanar congeners with respect to that displayed by microbes occurring in the marine sediment of Brentella Canal.

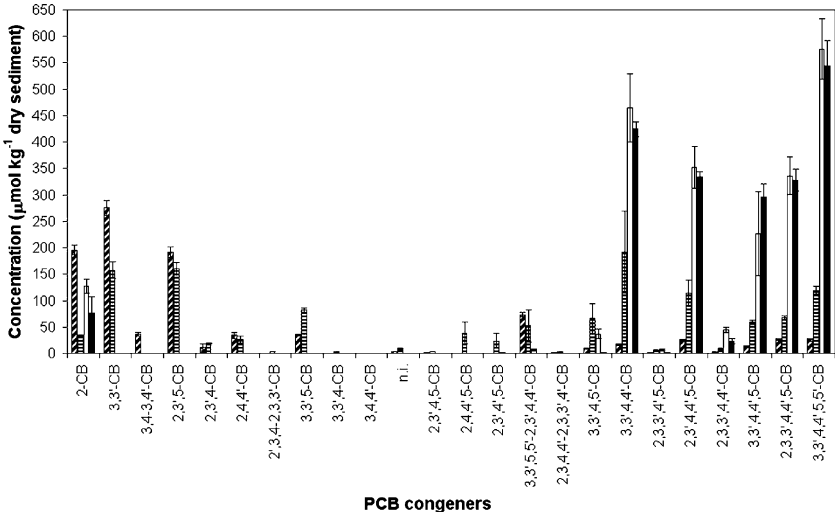

Figure 3. Average concentration of coplanar PCBs and their potential dechlorination products (±standard deviation as error bar) in the spiked sterile microcosms after 16 months of incubation (■) and in the parallel biologically active microcosms after 5 (□), 8 (▤) and 16 (▨) months of incubation.

Weathered PCBs were also significantly dechlorinated in the spiked microcosms. Some of the products of dehalogenation of exogenous PCBs had GC retention times comparable to those of some pre-existing PCBs. This obligated us to study the fate of only those sediment-carried PCBs whose GC peak area was not affected by this phenomenon (Figure 4). A significant transformation of such selected PCBs was detected after the 5th month of incubation. At the end of the experiment, some of the penta- and tetra-chlorinated congeners were found to be more extensively dechlorinated than in the parallel non-spiked microcosms (Figure 4). Furthermore, 2,2′,5-/2,2′,4-/4,4′-chlorobiphenyls and 2,2′,3-/2,4′,6-trichlorobiphenyl, that were slightly accumulated in the non-spiked microcosms, were here found to be depleted (Figure 4). On the contrary, 2,4′-/2,3-dichlorobiphenyl was apparently consumed in the non-spiked microcosms and accumulated in the spiked microcosms (Figure 4). Therefore, the dechlorination of exogenous PCBs only slightly influenced the bioconversion extent and pattern of sediment pre-existing PCBs (Figure 4), probably because the poor bioavailability of the latter controlled the process (Wiegel & Wu, 2000).

The lack of marked priming effects by exogenous PCBs has been already observed in another sediment of the Brentella Canal (Fava et al. 2003b), in the estuarine sediment of the LCP Chemicals Superfund site spiked with 2,6-dibromobiphenyl and 2,3,4,5,6-pentachlorobiphenyl (Palekar et al. 2003) and in a sediment of the New Bedford Harbor spiked with Aroclor 1242 (Alder et al. 1993).

Sulfate depletion and methane production profiles similar to those reported in Figure 2 for the non-spiked microcosms were observed in the biologically active spiked microcosms. Sulfate was completely consumed after 3 months of incubation, when methanogenesis started to occur concomitantly to PCB dechlorination (Figure 5). However, the overall amount of produced methane (13.6 ± 0.4 ml) was about 60% of that observed in the non-spiked ones (20.5 ± 8.6 ml). This finding suggests that the occurrence of exogenous PCBs adversely affected methanogenesis, probably because of their ability to sustain the growth of sediment microorganisms competing with methanogenic bacteria. This evidence further supports the hypothesis according to which sulfate reducing bacteria using PCBs as final electron acceptors once sulfate is completely depleted are responsible for PCB dechlorination in the microcosms developed in this study.

Conclusions

The occurrence of microbial-mediated reductive dechlorination processes towards weathered PCBs and five spiked coplanar PCBs have been shown in a contaminated sediment of the Brentella Canal of Porto Marghera area (Venice lagoon, Italy). Analogously to previous studies (Fava et al. 2003a,

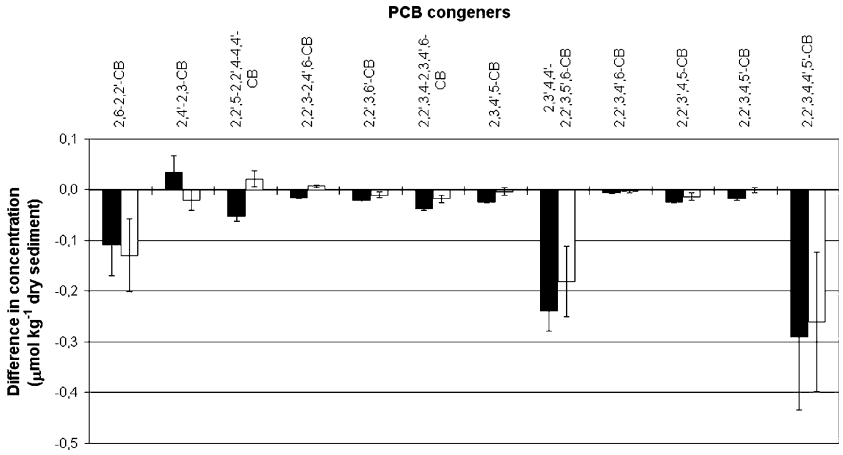

Figure 4. Changes in the native PCB concentration in the biologically active non spiked (white bar) and spiked (black bar) microcosms (all estimated vs. the sterile ones) at the end of the 16th month of incubation.

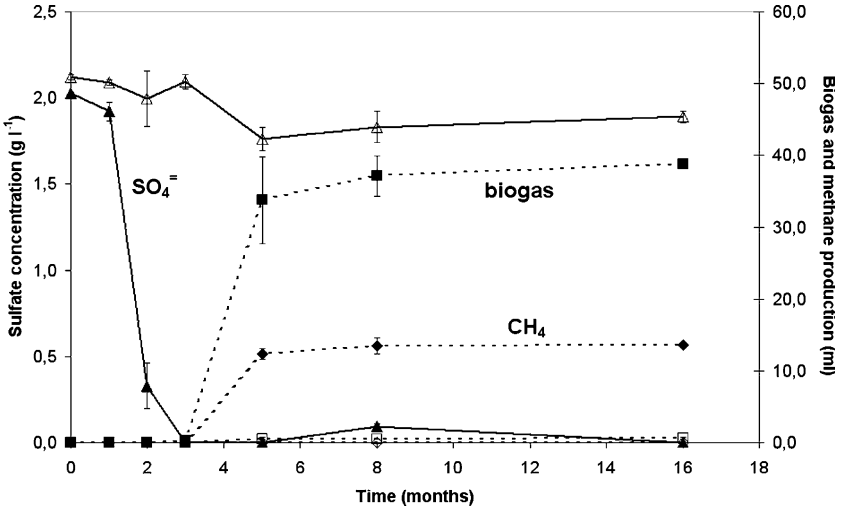

Figure 5. Sulfate concentration (triangles, solid line), biogas production (squares, dashed line) and methane production (diamonds, dashed line) detected in the spiked active microcosms (solid symbols) and sterile microcosms (empty symbols) throughout the 16-month experiment.

b), the dechlorination of pre-existing PCBs was slow, partial, not significantly primed by the addition of exogenous PCBs and probably mediated by sulfate-reducing bacteria; differently from them, it was here ascribed to the concomitant occurrence of dechlorination pattern H′ and M. This difference in dechlorination patterns let us speculate, according to Wiegel and Wu (2000), that different microbial populations with different dechlorinating specificities are involved in PCB dechlorination in the Brentella Canal. Further, the results of this and of the other studies quoted above support the hypothesis that reductive dechlorination of PCBs is a widely spread process in the sediments of the Brentella Canal.

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