**Microbial Bioavailability of 2,2',4,4'-Tetrabromodiphenyl Ether (BDE-47)
in Natural Sediments from Major Rivers of China**

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Abstract

Microbial degradation plays a crucial role in eliminating polybrominated diphenyl ethers (PBDEs) in environments. However, the microbial bioavailability of PBDEs in aquatic sediments is not well understood. In this work, the bioavailability of 2,2',4,4'-tetrabromodiphenyl ether (BDE-47), a typical PBDE congener, to PBDE-degrading microorganisms in natural sediments from six Chinese rivers under anaerobic conditions was investigated. The contents of black carbon (BC) and total organic carbon (TOC) in the six sediment samples were in the range of 0.025% to 0.30% and 0.03% to 3.38%, respectively. BDE-47 desorption from various sediments was fitted well with the first-order three-compartment desorption model. The desorbing fraction of sediment-associated BDE-47 at each desorption time interval exhibited a significant negative correlation with the BC content ($p < 0.01$). In the sediments, the anaerobic debromination of BDE-47 by microorganisms underwent a stepwise debromination pathway generating mainly three lower brominated congeners (BDE-28, -17 and -4). The microbial debromination ratio of BDE-47 ranged from 4.21% to 7.89% in various sediments after 120 d incubation anaerobically, and it negatively correlated with the content of sediment BC significantly ($p < 0.01$). However, the desorbing fraction and microbial debromination ratio of BDE-47 only showed weak correlations with the TOC content in sediments ($p > 0.05$). Furthermore, there was a significant positive correlation of desorbing fraction of BDE-47 from sediments with its microbial debromination ratio ($p < 0.01$) as well as with the level of its three lower brominated products ($p < 0.05$) after the first 20 d incubation. This study suggests that the BDE-47 bioavailability to microorganisms in anaerobic river sediments is mainly influenced by the content of sediment BC which controls the desorbing fraction of sediment-associated BDE-47.

Keywords: Polybrominated diphenyl ethers; Desorption; Microbial debromination; Black carbon; Bioavailability; Riverine sediments

1. Introduction

Polybrominated diphenyl ethers (PBDEs), a typical kind of hydrophobic organic compound (HOC), are widely used as flame retardants in various industrial and commercial fields (Alaee et al., 2003; Schreder and La Guardia, 2014). Over the past few decades, the global market demand and consumption of PBDEs have increased dramatically, reaching 67,000 tons in 2001 (Ma et al., 2012). Because of their extensive application, PBDEs will be inevitably released into environments during their production, usage, and disposal (Tang et al., 2014). PBDEs are worldwide distributed in almost all kinds of abiotic environmental components and biological tissues (Shaw et al., 2008; Yuan et al., 2012; Ge et al., 2014). More notably, PBDEs would also potentially pose toxic effects on higher animals and humans, which is of great interest and concern to researchers (Hakk and Letcher, 2003; Lee and Kim, 2015).

In aquatic environments, the sediment is usually considered as a major sink for PBDEs due to their hydrophobicity (Wei et al., 2012; Lee et al., 2015; Wang et al., 2015). It is well known that the environmental behavior and fate of HOCs are largely governed by the characteristics of aquatic sediments, especially by the sediment organic matter (SOM) (Cornelissen et al., 2008). The composition and content of SOM play a crucial role in determining the sorption and desorption of sediment-bound HOCs, thus subsequently affecting their mobility and bioavailability (Huang et al., 2003; Sormunen et al., 2009a; Wang et al., 2011). Cornelissen et al. (2005) reported that the “carbonaceous geosorbents” (CG), such as black carbon (BC) and kerogen, would significantly enhance the HOC sorption on sediment relative to amorphous organic matter. Also, the bioaccumulation of HOCs would be lowered correspondingly with the increase of the CG/TOC (total organic matter) ratio in sediments (Cornelissen et al., 2005). Moreover, Jia and Gan (2014) recently found that the free dissolved concentration (C_{free}) of PBDEs declined remarkably in sediments when the BC content increased, indicating the potential reduction in PBDE bioavailability by the sediment BC.

Microbial degradation is a crucial pathway to eliminate organic contaminants in environments (Zhang et al., 2013; Kolvenbach et al., 2014). HOC bioavailability to microorganisms is of great importance since it is closely related to microbial degradability of HOCs in soils and sediments (Kickham et al., 2012; Liang et al., 2014). However, although there are a few studies reporting the PBDE biotransformation by microbial isolates in artificial aqueous media (Robrock et al., 2008), research on microbial-mediated debromination of PBDEs in natural sediments is still very scarce. Furthermore, most of previous research on the bioavailability of PBDEs mainly focus on their bioaccumulation potentials in aquatic or soil organisms (Gaylor et al., 2014; Li et al., 2014), while our understanding is still very limited about the PBDE bioavailability to microorganisms in riverine sediments and the effect of SOM on the microbial bioavailability of PBDEs.

Therefore, the objective of the present study is to investigate the PBDE bioavailability to PBDE-degrading microorganisms in natural sediments with various contents of BC and TOC and attempt to elucidate the role of BC and TOC in influencing the microbial bioavailability and debromination of PBDEs. Different sediments from six typical rivers in China, including the Yangtze River (YZR), Yellow River (YER), Haihe River (HHR), Pearl River (PR), Hanjiang River (HJR), and Yongding River (YDR), were used in this work. The 2,2',4,4'-tetrabromodiphenyl ether (BDE-47) was selected as a representative of PBDE congeners due to its prevalent distribution in aquatic environments (Ma et al., 2012). Desorption processes of sediment-bound BDE-47 were measured by Tenax extraction technique. The anaerobic debromination of BDE-47 by PBDE-degrading microorganisms in different sediments was determined. Meanwhile, the relationship of the BC and TOC content with the desorbing fraction as well as with the debromination ratio of BDE-47 was analyzed to explore the influencing mechanism of the BDE-47 bioavailability to microorganisms in natural riverine sediments.

2. Materials and Methods

2.1 Chemicals

Individual PBDE congener standards dissolved in isooctane, including 2,2'-dibromodiphenyl ether (BDE-4), 2,2',4-tribromodiphenyl ether (BDE-17), 2,4,4'-tribromodiphenyl ether (BDE-28), BDE-47, and 3,3',4,4'-tetrabromodiphenyl ether (BDE-77) and the standard solution of decachlorobiphenyl (PCB-209) were obtained from AccuStandard (New Haven, CT). Acetone, dichloromethane, and hexane were of HPLC grade and purchased from J. T. Baker (Phillipsburg, U.S.A.). Trichloroethylene, ethanol, resazurin, sodium propionate, and all other inorganic reagents used in this study were of analytical grade or higher and purchased from Sinopharm Chemical Reagent Co., Ltd. (Beijing, China). Vitamins (purity > 99%), neutral alumina (100-200 mesh), and silica gel (100-200 mesh) were supplied by J&K Scientific Ltd. (Beijing, China). Diatomite was purchased from Celite China Inc. (Beijing, China). Tenax-TA resin (60-80 mesh) was obtained from Kanglin Science and Technology Ltd. (Beijing, China).

2.2 Sediment Sample Collection

Surface sediments (0-5 cm) were collected using a pre-cleaned stainless steel shovel from six typical rivers in China, including the YZR, YER, HHR, PR, HJR, and YDR. [Figure S1](#) in the [Supporting Information \(SI\)](#) shows the sampling sites which were chosen far away from polluted sources to avoid potential sediment contamination. The collected sediment samples were kept in clean polyethylene bags and then transported back to the laboratory at 4 °C. The stones, peddles, and plant debris in sediments were removed, and the sediments were air-dried in a clean fume hood. The dried sediments were sieved through a 1-mm sieve and irradiated by γ -rays at a dosage of 25 kGy to sterilize prior to use. The background PBDE concentrations of the YZR and PR sediments were 0.15 and 1.10 ng g⁻¹, respectively, and the concentrations of PBDEs were below the detection limit in the other four river sediment samples.

2.3 Sediment TOC and BC Determination

The BC content in sediments was measured by the thermochemical oxidation method with slight modification described by [Liu et al. \(2011b\)](#). Briefly, 0.5 g of dried and sieved sediment was put into a ceramic crucible and then heated in a muffle furnace at 375 °C for 24 h under air flow for oxidation of organic matter. After cooling, to remove the inorganic carbonates, the samples were acidified by addition of 0.1 M HCl until no bubbles were observed. Afterwards, the sediment samples were oven-dried at 60 °C overnight. The residue carbon was analyzed by a Vario EI elemental analyzer (Elementar Inc., Germany). TOC in sediment could be determined through similar procedures only omitting the step of removing organic matters by thermal oxidation at 375 °C.

2.4 Aging and Desorption of BDE-47 in Sediments

2.4.1 Aging

Two grams of sediment was weighted into a 50 mL Teflon centrifuge tube and spiked with 1 mL of BDE-47 stock solution to get the final BDE-47 concentration of 10 µg g⁻¹ in each sediment sample. After evaporation of the solvent in the fume hood for approximately 2-3 h, 2.5 mL ultrapure water and 30 µL of NaN₃ solution (20 g L⁻¹) which acted as a microbial inhibitor were added into each tube. The spiked sediments were then aged at room temperature in the dark for 100 d, which was long enough to simulate the sequestration of BDE-47 in natural environments ([Liang et al., 2010](#); [Xia et al., 2010](#)).

2.4.2 Desorption

After 100 d aging, each centrifuge tube was added with 20 mL of ultrapure water and 200 µL of NaN₃ solution. The Tenax-TA beads were ultrasonic-cleaned for three times with ultrapure water, acetone, and hexane (10 mL g⁻¹ Tenax) successively and oven-dried at 75 °C overnight before use. A weight of 0.2 g Tenax-TA beads was enclosed in a 200-mesh string bag and added into each tube. Then the tubes were placed in an incubator and shaken at the speed of 150 rpm at 30 °C. Desorption of BDE-47 from

sediments is a physicochemical process without any transformation or conversion of BDE-47, in which oxygen may not be an important factor. Therefore, the experiment on desorption of sediment-sorbed BDE-47 was not performed under anaerobic conditions, similar to other research (Sun et al., 2014). At predetermined time intervals (i.e. 1, 4, 9, 12, 24, 48, 72, 180, 336, 480 and 600 h), the string bags were removed out of the tubes for subsequent BDE-47 extraction and replaced with bags enclosing new Tenax-TA beads. Desorption of BDE-47 from sediments was described using the first-order kinetics model with three compartments (Harwood et al., 2015):

$$S_t/S_0 = F_r \cdot e^{-k_r \cdot t} + F_s \cdot e^{-k_s \cdot t} + F_{vs} \cdot e^{-k_{vs} \cdot t} \quad (1)$$

$$F_r + F_s + F_{vs} = 1 \quad (2)$$

where, S_0 and S_t represent the quantity of sediment-bound BDE-47 at the beginning and time t (h) during the desorption period, respectively; F_r , F_s , and F_{vs} are the rapidly, slowly, and very slowly desorbing fraction, respectively; k_r , k_s , and k_{vs} (h^{-1}) are the desorption rate constants for respective desorbing compartments. Due to the strong hydrophobicity of BDE-47 ($\log K_{ow} = 6.81$) and great sorption capacity of Tenax TA resin, the aqueous concentration of BDE-47 is assumed to be negligible during the desorption process. We also assumed that BDE-47 desorption from the three compartments of sediments was independent on each other.

2.5 Anaerobic Debromination of BDE-47 by Microorganisms in Sediments

One mL of BDE-47 stock solution ($500 \mu\text{g mL}^{-1}$) was spiked into 50 g of sediment in a 125 mL headspace bottle, yielding a final BDE-47 concentration of $10 \mu\text{g g}^{-1}$ in all sets of microcosms. After volatilization of the solvent, the bottle was added with 50 mL of sterile deionized water and then sealed using a Teflon-silicon cap. The spiked sediments were aged aseptically and anaerobically under room temperature in the dark for 100 d. After aging, each bottle was added with 30 mL of basal medium and sealed tightly. Then, syringe needles were inserted through the Teflon-silicon caps to discharge the air inside

the bottles during the process of autoclaving. After autoclaving at 121 °C for 30 min, the syringe needles were removed immediately, resulting in an anaerobic condition inside the bottles. Afterwards, each bottle was added sequentially with 1 mL of phosphate buffer solution, 0.3 mL of NaHCO₃ solution (0.5 mol L⁻¹), 0.3 mL of sodium propionate solution (0.5 mol L⁻¹), 0.3 mL of Na₂S solution (1.0 mol L⁻¹), 50 µL of vitamin solution, and 0.5 mL of trichloroethylene solution (200 mg L⁻¹). The microbial culture used herein was originally isolated from the groundwater sample polluted by chlorinated organics. The details about the culture and basal medium preparation are provided in [SI](#). Finally, 1 mL of microbial culture suspension was inoculated into the BDE-47 debromination experiment sets, and 0.5 mL of NaN₃ solution was added to the control sets. All bottles were incubated anaerobically at 30 °C with the shaking speed of 150 rpm. At predetermined time intervals (i.e. 10, 20, 30, 45, 60, 80, 100 and 120 d), three replicate bottles for each sediment sample were taken out for determining the lower brominated products of BDE-47.

2.6 Extraction, Cleanup and Analysis of PBDE Congeners

Accelerated solvent extraction (ASE 300, Dionex, Sunnyvale, CA, U.S.A.) was used to extract the PBDE congeners in sediments as reported elsewhere ([de la Cal et al., 2003](#)). Cleanup of the concentrated extracts was carried out by chromatography columns filled with silica gel and alumina ([Mai et al., 2005](#)). The PBDE congeners were determined by an Agilent 7890A gas chromatography (GC) coupled to an Agilent 5975C mass spectrometer (MS). Detailed procedures of PBDE extraction and cleanup from sediments and Tenax-TA resin as well as the operation conditions of GC-MS are described in [SI](#).

2.7 Quality Assurance and Quality Control

The ratios of signal to noise for PBDE congeners greater than 3 were considered as valid data. The detection limit of BDE-47 was 1 µg L⁻¹, and those of BDE-4, -17, and -28 were 0.5 µg L⁻¹. Correlation coefficients of the standard curves for all PBDE congeners were

greater than 0.99. The recoveries of the PBDEs were monitored using BDE-77 as the surrogate standard and ranged from 78% to 97% in all experiment sets. All experiment sets were carried out in dark to avoid photochemical degradation of BDE-47. Each experiment was performed in triplicate and the relative standard deviations were less than 10%. In the desorption experiment, the aqueous level of BDE-47 detected in method blank without Tenax-TA beads was far below ($< 0.1\%$) the levels on the Tenax-TA beads in the experiment sets. Meanwhile, the debromination ratio of BDE-47 after 120 d in the sterilized control samples was below 0.1%.

3. Results and Discussion

3.1 TOC and BC Contents in Different Sediments

The surface sediment samples were collected from six typical rivers in different areas of China. As shown in [Table 1](#), contents of BC and TOC varied remarkably among these samples. The quantity of TOC ranged from 0.03% for the YER to 3.38% for the YDR, and the BC contents was in the range of 0.025% to 0.30%. Except for the YDR and HJR, the TOC in sediments from other four rivers were all below 1%, indicating their relatively low contents of TOC. It is noted that the sediment from the YER showed both the smallest amount of TOC and BC. This is ascribed to the fact that the YER sediment is primarily derived from the soil erosion of the Loess Plateau which contains low level of organic matter ([Dong et al., 2015](#)).

3.2 Desorption Kinetics of Sediment-associated BDE-47

Tenax extraction method is an effective desorption-based way to estimate the bioavailability of HOCs in soils and sediments ([Harwood et al., 2015](#)). The mass balance was determined by comparing the sum of the quantity of BDE-47 sorbed on Tenax-TA beads and that remaining on the sediment with the original quantity on the sediment. It ranged from 88% to 115% for all the sediment samples. As shown in [Figure 1](#), the desorption process generally underwent three phasic patterns: an initial relatively fast

release of BDE-47, followed by a subsequent slow desorption phase and a final very slow phase of BDE-47 desorption from sediments. The first-order kinetics desorption model with three compartments could sufficiently describe the desorption behavior of sediment-bound BDE-47 as the correlation coefficients (r^2) of the fitting model for all sediment samples were higher than 0.95 (Table 2).

The F_r , F_s , and F_{vs} for all sediments were in the range of 18.5%-44.8%, 15.7%-57.1%, and 18.5%-53.2%, respectively (Table 2). The relatively larger F_r and F_s for the sediments from YER (44.8%) and YDR (57.1%) indicated that desorption of BDE-47 from these two sediment samples mainly occurred at the rapid and slow desorbing stage, respectively. In contrast, the F_{vs} for the sediments from the YZR, HJR, HHR, and PR were 53.1%, 53.2%, 45.2%, and 36.4%, respectively, which were relatively higher than their F_r and F_s . This suggested the dominant role of very slow desorption controlling the release of BDE-47 associated with these sediments. de la Cal et al. (2008) investigated desorption of sediment-bound BDE-47 using the Tenax-assisted method and found that the F_{vs} accounted for 35.1%, which was less than the F_{vs} herein expected for the YDR sediment (Table 2). Compared with their experiment conditions (4 °C and 18 d), the higher aging temperature (room temperature) and longer aging time (100 d) herein might be responsible for the larger F_{vs} in the present work. It is known that the movement of HOC molecules into sediment particles would be enhanced by higher temperature (Sormunen et al., 2009b), and the extended aging time would lead to the stronger association between HOCs and sediments, impeding the release of sediment-bound HOCs (Liang et al., 2010). Additionally, in terms of desorbing rate constants, the rapidly desorbing rate constant (k_r) of BDE-47 for all six sediment samples ranged from $4.02 \times 10^{-2} \text{ h}^{-1}$ to $12.3 \times 10^{-2} \text{ h}^{-1}$, in agreement with other HOC desorption from sediments reported previously (Cornelissen et al., 1997; Greenberg et al., 2005). However, the slowly and very slowly desorbing rate constants herein were much lower in comparison with the results reported by Liu et al. (2011a). This was probably due to the much longer aging time

herein (100 d) relative to their 14 d aging time, making it more difficult for the BDE-47 to desorb from sediments in this research (Rhodes et al., 2012).

3.3 Effects of BC and TOC on BDE-47 Desorption

As shown in Table 1, the contents of BC in six sediment samples were in the order of YZR > YDR > PR > HJR > HHR > YER, and meanwhile Figure 1 showed that the desorbing fractions of BDE-47 from these sediments followed an opposite sequence: YER > HHR > HJR > PR > YDR > YZR. This revealed a trend that the desorbing fraction of sediment-associated BDE-47 decreased with the increasing BC content, suggesting an important role of BC in governing the desorption process of sediment-associated BDE-47. This was further confirmed by analyzing the correlation of the desorbing fraction of BDE-47 from various sediments at different time intervals with the BC and TOC contents. As shown in Table 3, the desorbing fraction of BDE-47 at each time interval was negatively correlated with the BC content in sediments significantly ($p < 0.01$). This finding was consistent with other studies (Liu et al., 2011a; Ogbonnaya et al., 2014). However, although a negative correlation existed between the desorbing fraction of BDE-47 and the TOC content, the correlation was not statistically significant at the 95% confidence level ($p > 0.05$) (Table 3). These results suggested that in comparison with TOC, BC could play a more important role in dominating desorption of sediment-bound BDE-47. It is generally believed that the desorption process of HOCs from soils or sediments is related not only with the quantity of organic matter but with its quality as well (Huang et al., 2003). The SOM is heterogeneous and usually comprised of an amorphous or “soft” carbon phase such as humic matter and a condensed or “hard” carbon phase such as BC and kerogen (Weber et al., 1992; Huang et al., 2003). Many studies reported that BC had a much greater affinity to HOCs than the “soft” organic matter, leading to the HOC desorption hysteresis from soils and sediments (Liu et al., 2012; Yang et al., 2012). Consequently, desorption of sediment-associated BDE-47 was greatly impeded with the increase of BC amount. This induced the reduction in the

275 BDE-47 desorbing fraction, which could explain its significant negative relationship with
276 the sediment BC content (Table 3).

277 Additionally, after 600 h desorption experiments, we observed a significant positive
278 correlation between the residual amounts of BDE-47 associated with sediments and the
279 BC content ($p < 0.01$) (Figure S2). Interestingly, when the residual amounts of
280 sediment-bound BDE-47 was normalized by the BC content, there were still
281 discrepancies among the residual BDE-47 associated with different sediment samples,
282 especially for the YER and HHR as shown in Figure S3. Thus, our results implied that,
283 besides the content of sediment BC, the BC source in these different sediments would
284 also affect the desorption behavior of BDE-47. The sources of BC in sediments and soils
285 mainly include burning of biomass (e.g. wood, cornstalk, grass, etc.) and combustion of
286 fossil fuels (e.g. coal, petroleum, gas, etc.) (Liu et al., 2011b), and the sources are related
287 to the local economic, agricultural and industrial conditions. Therefore, BC in these river
288 sediments collected from different regions of China was probably derived from different
289 sources, thus influencing the BDE-47 desorption.

290 Table 2 also showed that there was a significantly negative correlation between rapidly
291 desorbing fraction (F_r) and the sediment BC content ($p < 0.05$), indicating the BC content
292 would substantially affect the F_r of BDE-47 in sediments. This might attribute to the
293 association of more BDE-47 with BC as the BC content increased, which brought about
294 the declined amount of BDE-47 releasing from sediments during the rapid desorption
295 phase. Besides, the slowly desorbing fraction (F_s) positively correlated with the TOC
296 content significantly ($p < 0.01$) as shown in Table 2. This means that the increased
297 sediment TOC content would result in larger amount of BDE-47 releasing from
298 sediments during the slow desorption phase. Previous research indicated that TOC
299 played a significant role in the process of slow desorption of polychlorinated biphenyl
300 from sediments (Cornelissen et al., 1998). Rhodes et al. (2010) studied the desorption
301 behavior of phenanthrene from different soils, and they also found that the soils with

higher content of TOC would exhibit larger F_s than soils containing lower TOC. However, no significant correlation between the very slowly desorbing fraction (F_{vs}) and the TOC and BC was observed ($p > 0.05$), suggesting neither TOC nor BC could strongly influence the very slow desorption phase of sediment-associated BDE-47. This might be due to the heterogeneity of sediment matrixes so that many variables are responsible for the association of BDE-47 with sediments, and influences of other variables could attenuate the effect of BC and TOC (Luo et al., 2012).

3.4 Anaerobic Microbial Debromination of BDE-47 in Sediments

After the microbial debromination of BDE-47 in six sediment samples for 120 d under anaerobic conditions, mainly three kinds of newly borne lower brominated PBDE congeners were detected in sediments including two tribromo BDEs (BDE-17 and -28) and one dibromo BDE (BDE-4) (Figure 2). In the sterilized control set, no lower brominated PBDE products were detected, indicating that the debromination of BDE-47 was ascribed to the microbial activity. Figure 2 also showed that BDE-17 was the predominant lower brominated congener for all riverine sediments, and its concentrations in these sediments ranged from 291.75 to 555.87 ng g⁻¹ after microbial debromination for 120 d. The relative higher level of BDE-17 suggested that the BDE-47 molecule underwent the removal of bromines at the *para* position by the PBDE-degrading microbes in sediments anaerobically, which agreed with previous research (Robrock et al., 2008). In addition, we observed a stepwise debromination pathway for the microbial debromination of BDE-47 in anaerobic sediments. That is to say, the relative proportion of tribromo products decreased, while the relative level of dibromo products increased accordingly during the 120 d experiment. For example, from the 10th d to 120th d of BDE-47 debromination in the anaerobic YER sediment, the relative proportion of BDE-4 increased from 0.5% to 8.6%, while the relative proportion of BDE-17 and -28 decreased from 91.2% to 87.7%, and from 8.3% to 3.7%, respectively (Figure S4). Similar stepwise debromination pathways were also reported by other

329 researchers (Tokarz et al., 2008; Song et al., 2015).

330 Also, the debromination ratio of BDE-47 in various sediments was determined by
331 dividing the sum of the molar quantity of three lower brominated PBDE products by the
332 molar quantity of initially spiked BDE-47. As shown in Figure 2, for all sediments, the
333 debromination ratio of BDE-47 increased with the incubation time, in the range from
334 4.21% for the YZR to 7.89% for the YER after 120 d anaerobic incubation. This is
335 comparable to other research (Tokarz et al., 2008). Nevertheless, Zhu et al. (2014)
336 recently reported that the amount of BDE-47 debrominated by microbes reached more
337 than 90% in anaerobic sediments, which was much higher than the results herein. This
338 difference might be due to the much longer incubation time (seven months) in their
339 study which led to more complete PBDE removal from sediments.

340 Furthermore, we analyzed the relationship of the microbial debromination ratio of
341 BDE-47 with the BC and TOC content in various riverine sediments. The results are
342 displayed in Table 3. At different time intervals, the debromination ratio of BDE-47
343 negatively correlated with the sediment BC content significantly ($p < 0.01$) with the
344 exception of the sediment samples incubated for 10 d, while it exhibited a weak negative
345 correlation with the TOC content ($p > 0.05$). This suggested that BC was an overriding
346 factor which influenced BDE-47 debromination in sediments. Research has shown that
347 the BC has stronger binding to HOCs relative to other SOM so that the bioavailability of
348 HOCs to organisms would be affected by the presence and quantity of sediment BC
349 (Wang et al., 2011; Jia and Gan, 2014). For example, Sundelin et al. (2004) found that the
350 log value of biota-sediment accumulation factors (BSAFs) was negatively related with
351 the log of BC content during the bioaccumulation of polycyclic aromatic hydrocarbons
352 (PAHs) in amphipod in sediments, indicating the reduced bioavailability of PAHs by the
353 strong BC sorption. Also, the microbial mineralization of phenanthrene would
354 substantially decrease when the BC content increased in soils (Rhodes et al., 2008).

For the sediment-associated HOCs, the bioavailable fraction is generally recognized as the proportion which could desorb from the sediment reversibly (Xia and Wang, 2008; Lydy et al., 2015). Correlation between the debromination ratio of BDE-47 and its desorbing fraction after the first 20 d was further explored using the linear regression. As shown in Figure 3, the desorbing fraction of BDE-47 from sediments displayed significant positive correlation with the debromination ratio ($p < 0.01$) as well as with the three lower brominated PBDE products ($p < 0.05$). This suggests that the bioavailability of BDE-47 to microorganisms was mainly controlled by its desorption from sediments. BDE-47 underwent desorption from sediments followed by the debromination by PBDE-degrading microorganisms. Therefore, based on the discussion above, the sediment samples with relatively higher content of BC would sorb BDE-47 more strongly and thus lead to the reduction in desorbable proportion to a less extent as discussed in Section 3.3, which caused lower BDE-47 bioavailability to PBDE-degrading microbes and eventually its lower microbial debromination ratio. Our results revealed that the content of BC could act as an important factor in influencing the microbial bioavailability of BDE-47 in sediments.

4. Conclusions

This study investigated the PBDE bioavailability to PBDE-degrading microorganisms in various natural sediments from several major Chinese rivers. Our results indicated that in comparison with the TOC, the sediment BC, which had a greater affinity to HOCs, played a controlling role in affecting the desorption process of sediment-associated BDE-47. Similarly, we also found that the BC content was a more influential factor that governed the microbial debromination of BDE-47 in sediment anaerobically. Based on these findings and combined with the fact that the BDE-47 debromination by microbes positively correlated with its desorbing fraction from sediments, we postulated that the strong sorption ability of sediment BC would lower the desorbing parts of sediment-bound BDE-47, thus reducing the BDE-47 bioavailability to the degrading

microorganisms. The results of this work implied the quantity of sediment BC should be considered as a key factor when evaluating the environmental behaviors and risks of PBDEs in natural riverine sediments.

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520

521 Table 1 The BC and TOC contents in various river sediments

	Rivers ^a					
	YDR	HHR	YER	HJR	YZR	PR
BC (%)	0.29	0.09	0.025	0.19	0.30	0.28
TOC (%)	3.38	0.68	0.031	1.02	0.52	0.89

522 ^a YDR, HHR, YER, HJR, YZR and PR denote the Yongding River, the Haihe River, the
523 Yellow River, the Hanjiang River, the Yangtze River and the Pearl River, respectively.

524

525 Table 2 Fitting parameters of desorption kinetics for BDE-47 from various sediments

	F_r (%)	k_r (h ⁻¹)	F_s (%)	k_s (h ⁻¹)	F_{vs} (%)	k_{vs} (h ⁻¹)	r^2
YZR	18.5%	5.66E-02	28.4%	7.33E-07	51.4%	1.16E-125	0.966
YER	44.8%	1.23E-01	15.7%	5.67E-03	39.7%	5.35E-129	0.992
HJR	25.0%	1.04E-01	21.8%	4.53E-03	52.1%	3.29E-101	0.994
HHR	31.8%	9.18E-02	23.0%	2.63E-03	45.1%	1.76E-117	0.997
YDR	24.4%	4.95E-02	57.1%	1.72E-07	17.4%	5.84E-128	0.984
PR	29.9%	4.02E-02	33.7%	3.87E-123	33.7%	1.78E-125	0.952

526

527

Table 3 Correlation of the desorbing fraction and debromination ratio of BDE-47 with the BC and TOC content

		Desorbing fraction ^a							
		$F_{24\text{ h}}$	$F_{72\text{ h}}$	$F_{336\text{ h}}$	$F_{480\text{ h}}$	$F_{600\text{ h}}$	F_{r}	F_{s}	F_{vs}
BC	r	-0.955**	-0.946**	-0.958**	-0.960**	-0.959**	-0.847*	0.699	-0.200
	p	0.003	0.004	0.003	0.002	0.002	0.033	0.123	0.704
TOC	r	-0.499	-0.472	-0.500	-0.494	-0.494	-0.422	0.940**	-0.767
	p	0.314	0.344	0.313	0.319	0.319	0.405	0.005	0.075
		Debromination ratio ^b							
		$R_{10\text{ d}}$	$R_{20\text{ d}}$	$R_{30\text{ d}}$	$R_{45\text{ d}}$	$R_{60\text{ d}}$	$R_{80\text{ d}}$	$R_{100\text{ d}}$	$R_{120\text{ d}}$
BC	r	-0.753	-0.938**	-0.978**	-0.947**	-0.984**	-0.983**	-0.975**	-0.973**
	p	0.084	0.006	0.001	0.004	0.000	0.000	0.001	0.001
TOC	r	-0.168	-0.537	-0.531	-0.480	-0.533	-0.500	0.478	-0.502
	p	0.751	0.272	0.278	0.335	0.276	0.312	0.337	0.310

^a $F_{24\text{ h}}$, $F_{72\text{ h}}$, $F_{336\text{ h}}$, $F_{480\text{ h}}$, and $F_{600\text{ h}}$ denote desorbing fraction at 24, 72, 336, 480 and 600 h, respectively.

^b $R_{10\text{ d}}$, $R_{20\text{ d}}$, $R_{30\text{ d}}$, $R_{45\text{ d}}$, $R_{60\text{ d}}$, $R_{80\text{ d}}$, $R_{100\text{ d}}$, and $R_{120\text{ d}}$ denote the debromination ratio of BDE-47 in sediment at 10, 20, 30, 45, 60, 80, 100, and 120 d, respectively.

* Significant at the 95% confidence level.

** Significant at the 99% confidence level.

537 **Figure Captions**

538 Figure 1 Desorption kinetics curves of BDE-47 from various sediments. Data represent
539 the mean value $\pm$ standard deviation (n=3).

540 Figure 2 Debromination ratio of BDE-47 and its lower debrominated PBDE products in
541 sediments. Solid symbols represent samples with PBDE-degrading microorganisms and
542 hollow symbols represent the sterilized controls. Data represent the mean value $\pm$
543 standard deviation (n=3).

544 Figure 3 Correlation of the desorbing fraction of sediment-associated BDE-47 after 20 d
545 of desorption with the debromination ratio and with the concentration of three lower
546 brominated products after 20 d of anaerobic incubation for the six sampling sites. Data
547 represent the mean value $\pm$ standard deviation (n=3)

548

Figure 1 Desorption kinetics curves of BDE-47 from various sediments. Data represent the mean value $\pm$ standard deviation (n=3).

Figure 2 Debromination ratio of BDE-47 and its lower debrominated PBDE products in sediments. Solid symbols represent samples with PBDE-degrading microorganisms and hollow symbols represent the sterilized controls. Data represent the mean value $\pm$ standard deviation (n=3).

559

560 Figure 3 Correlation of the desorbing fraction of sediment-associated BDE-47 after 20 d

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564