Relationships between the bioavailability of polybrominated diphenyl ethers in soils measured with female C57BL/6 mice and the bioaccessibility determined using five in vitro methods

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ABSTRACT

Several in vitro methods for simulating human gastrointestinal digestion have been validated for predicting the bioavailability of heavy metals, but the methods for successfully predicting the bioavailability of organic pollutants are still limited. In this study, we used an adapted fasting in vitro digestion method (Fa-VDM) from the Simulator of the Human Intestinal Microbial Ecosystem and four other in vitro methods comprising In Vitro Gastrointestinal, a physiologically-based extraction test, the unified BARGE method, and Deutsches Institut für Normung e.V. in order to measure the bioaccessibility of polybrominated diphenyl ethers (PBDEs) in soils from an e-waste dismantling town, China, with a Standard Reference Material (SRM2585) as the control. Furthermore, the bioaccessibility data were compared with the bioavailability measured using female C57BL/6 mice. The bioavailability of PBDEs in the soils and SRM2585 were 1.7% to 38.1% and 3.9% to 48.8%, respectively, and the bioaccessibility determined using Fa-VDM were 1.6–55.4% and 6.7–32.1%. There were negative and parabolic correlations between octanol/water partition coefficient for PBDEs and the bioavailability and bioaccessibility, respectively, whereas the H/C ratios and organic matter contents of the soils did not correlate with them. The bioaccessibility data determined by Fa-VDM were generally higher than those obtained using the other four methods, mainly due to the higher bile concentration and larger liquid to solid ratio in the digestion solution in Fa-VDM. There was a significant linear relationship between the results according to the in vivo and in vitro method of Fa-VDM where the slopes varied from 0.83 to 1.16 ($R^2 > 0.73$) and intercepts from 0.3%–7.7% for BDE47, 99, 100, and 153 measured using Fa-VDM, thereby indicating that the bioaccessibility assessed by this method can potentially be used to predict the bioavailability of moderately brominated congeners in soils.

1. Introduction

Humans are generally exposed to environmental chemicals via three pathways: oral ingestion, inhalation, and dermal contact. Traditionally, human exposure to contaminants via oral ingestion is estimated as the product of the contaminant concentration and the mass of the ingested matrix. This approach generally assumes that 100% of the ingested matrix-borne contaminants are available for absorption in the human gastrointestinal tract. However, this assessment and its associated human health risk may be overestimated because the absorbed fraction of a contaminant is sometimes lower than 100% (Rostami and Juhasz, 2011).

Therefore, many researchers have suggested adding one of two parameters, i.e., oral bioavailability and bioaccessibility, for a more reasonable human exposure or health risk assessment. The oral bioavailability is defined as the fraction of an administered contaminant that reaches the central (blood) compartment from the gastrointestinal tract (Ruby et al., 1996). The oral bioaccessibility of a chemical refers to the fraction of the total accounted for by the chemical released from matrices. Thus, the bioaccessible chemical is potentially available for

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absorption in the gastrointestinal tract (Oomen et al., 2000). In vivo methods that employ animal models, such as mice and swine, have been used frequently to measure bioavailability in previous studies (Duan et al., 2014; Smith et al., 2012), but they are still expensive, time consuming, and ethically challenging (Smith et al., 2012; Li et al., 2016a, 2016b). Therefore, simple, rapid, and inexpensive in vitro methods should be developed to simulate the digestion of matrices, e.g., food and soil, in the human gastrointestinal tract in order to measure the bioaccessibility.

During the past few decades, several in vitro bioaccessibility methods have been developed, including the physiologically-based extraction test (PBET), unified BARGE method (UBM), Deutsches Institut für Normung e.V. (DIN), and In Vitro Gastrointestinal (IVG) (Li et al., 2015c, 2016c). Excluding DIN, the other three methods were originally developed to measure the bioaccessibility of heavy metals, although they can also be used to assess the bioaccessibility of some organic pollutants. However, poor relationships were generally observed between the bioavailability and bioaccessibility of the tested organic pollutants (Smith et al., 2012), with very few exceptions (Li et al., 2015c; Pu et al., 2004). Recently, we optimized a fasting in vitro digestion method (referred to as Fa-VDM in the following text) adapted from the Simulator of the Human Intestinal Microbial Ecosystem under worst-case scenario (considers the most severe possible bioaccessibility that can reasonably be obtained in a given digestion condition), which we optimized using the response surface methodology to measure the bioaccessibility of polybrominated diphenyl ethers (PBDEs, a typical class of persistent organic pollutants) in house dust (Yu et al., 2011). However, the Fa-VDM did not validated using in vivo assay, although it was used to determine the bioaccessibility of PBDEs in dust for assessing human exposure (Li et al., 2015a; Yu et al., 2012a, 2013). Subsequently, we also developed an absorption sink method by utilizing Tenax-TA (a porous polymer resin) as a sink to simulate the dynamic digestion and absorption processes in the gastrointestinal tract in order to study the bioaccessibility of PBDEs in air-conditioner filter dust (Yu et al., 2013). Moreover, Tenax-TA was employed as a sink to assess its performance with hydrophobic organic pollutants, such as PBDEs (Fang and Stapleton, 2014; Kademoglou et al., 2018) and polychlorinated biphenyls (Li et al., 2017). Furthermore, other materials such as silicone and cyclodextrins were used as sinks to simulate the dynamic digestion and absorption processes in the gastrointestinal tract (Zhang et al., 2015; Mayer et al., 2016; Gouliarmou et al., 2013).

However, the in vitro method developed with Fa-VDM and those with Tenax-TA as a sink still have some limitations. For example, the Tenax-TA assisted method is relatively complex. The Fa-VDM has not been validated using in vivo data as aforementioned, although the bioaccessibility data measured for PBDEs in dust using our method (Yu et al., 2012a, 2013) were similar to the bioavailability results reported in previous studies (Huwe et al., 2008; Ounnas et al., 2010). We also did not know whether there was positive in vivo–in vitro correlation (IVIVC) between our bioaccessibility and their bioavailability, because the data were determined from different samples. Thus, to confirm whether Ad-SNIME can be used to measure the bioaccessibility of PBDEs to predict the bioavailability and if it is not necessary assisted by using a sink such as Tenax-TA to simplify the in vitro method, the same samples should be used to validate the method of Fa-VDM and estimate the applicability.

Therefore, the main objectives of the present study were: (1) to determine the bioavailability of PBDEs in soils and a reference material using mice as model animals; (2) to compare the bioaccessibility data for PBDEs using five different in vitro methods comprising Fa-VDM, PBET, UBM, DIN, and IVG; (3) to determine the relationship between the bioavailability and bioaccessibility of PBDEs in soil; and (4) to estimate the suitability of these in vitro methods (especially the Fa-VDM) for predicting the bioavailability of PBDEs in soils.

2. Materials and methods

2.1. Soils and their properties

In total, 18 surface soils were collected from an e-waste-dismantling town in Southern China during 2013 (Wang, 2015). Only five of the soil samples were selected for the in vitro and in vivo experiments after considering the concentrations of PBDEs and the organic matter (OM) contents of the soils based on two criteria, i.e., the concentrations of PBDEs (total: 2.8×10^3–5.1×10^4 ng/g dw; dw: dry weight) and OM contents (7.2%–58.3%) with large ranges. The soils were lyophilized and sieved through a 65 mesh (250 μm) filter, and then storing at $-18\,^\circ$C until their use.

The OM contents of the five soils and a Standard Reference Material (organic contaminants in house dust, NIST SRM2585) were measured using the loss-on-ignition method, as described in our previous study (Yu et al., 2012a), and the elemental contents of C and H were analyzed with an elemental analyzer (EA3000, Leeman Co.).

2.2. Determination of PBDE concentrations in soils

The soils were treated using a method similar to that employed in our previous studies with some minor modifications (Yu et al., 2011, 2012a). Briefly, a soil sample (0.1 g) was Soxhlet extracted with 250 mL of a mixture of n-hexane and acetone (1:1, v:v) for 72 h. The extract was concentrated to approximately 1 mL, before diluting in 10 mL n-hexane for solvent exchange, and concentrating to approximately 1 mL again for purification by gel permeation chromatography and multilayer silica-alumina column chromatography, which were the same as our previous studies using (Yu et al., 2012a). The PBDE concentrations were determined using an Agilent 6890N gas chromatograph (GC) coupled to a 5975-mass spectrometer (MS). The same methodology was used to determine the concentrations of PBDEs in SRM2585. Each sample was measured five times on different dates and the average data were then used to assess the bioavailability and bioaccessibility.

2.3. Determination of PBDE bioavailability using an in vivo assay

Female C57BL/6 mice aged four to six weeks were used. All of the animal experiments were approved by the Animal Ethics Committee of Shanghai Jiao Tong University School of Medicine (Project Number 2014094) and conducted in compliance with the guidelines of the Care and Use of Laboratory Animals (certificated by Shanghai Committee of Science and Technology). All mice were raised in a climate-controlled room with a 12:12 h light:dark cycle for 1 week and given access to food ad libitum (SHOOBREE, SPF grade, from XIETONG · ORGANISM, Jiangsu Province, China) before the in vivo assay. The mice were then randomly divided into control (n = 3) and experimental groups (n = 3). To determine the bioavailability of PBDEs in soils and SRM2585, the samples were suspended in Milli-Q water with a soil concentration of 0.4 g/mL. Next, 0.5 mL of the suspension was administered by oral gavage to each female C57BL/6 mouse, i.e., 0.2 g soil or SRM2585 sample per mouse (approximately 10 g of soil/kg body weight). Only Milli-Q water was administered to the control group. All of the mice were placed in self-made stainless steel cages (Fig. S1) and were sacrificed at 24 h after administration. The gastrointestinal tract contents and remaining corpse including blood, skin, hair, and so on, were collected and weighed accurately. In addition, urine were collected using absorbent cotton and feces were collected using nipper, and the samples weighed, before treating them according to the procedure reported in our previous study (Feng et al., 2015). The tissue samples (gastrointestinal tract contents and remaining corpse) were also treated using a similar method to that described in our previous study (Yu et al., 2012b). The samples were lyophilized, Soxhlet extracted, and cleaned up by gel permeation chromatography, before separating on a multilayer silica-alumina column, and the samples were

Table 1
Concentrations (expressed as average $\pm$ standard deviation, ng/g dw) of PBDEs in the soils collected from the e-waste dismantling town and SRM2585.

	Soils (n = 5)					Reference dust (SRM2585) (n = 5)		
	# 2	# 3	# 5	# 9	# 18	Measured	Reference data	RSD
BDE17	58.7 ± 2.6	19.9 ± 1.8	4.0 ± 0.2	9.1 ± 0.6	3.6 ± 0.3	15.3 ± 1.3	11.5 ± 1.2	33.0%
BDE28	557 ± 48.1	111 ± 12.3	24.3 ± 1.0	52.7 ± 6.6	15.4 ± 0.6	50.3 ± 3.6	46.9 ± 4.4	7.2%
BDE47	1240 ± 171	677 ± 49.3	186 ± 4.3	273 ± 87.7	99.2 ± 3.1	413 ± 11.8	497 ± 46.0	16.9%
BDE66	379 ± 6.4	177 ± 34.3	42.0 ± 3.2	73.8 ± 11.0	24.2 ± 0.9	36.7 ± 2.8	29.5 ± 6.2	24.4%
BDE100	320 ± 32.7	49.3 ± 3.4	18.4 ± 0.9	24.9 ± 2.9	7.3 ± 0.4	117 ± 5.2	145 ± 11.0	19.3%
BDE99	1610 ± 232	765 ± 64.9	168 ± 11.3	295 ± 55.5	76.3 ± 3.5	746 ± 31.2	892 ± 53.0	16.4%
BDE85	131 ± 11.4	34.8 ± 4.6	8.9 ± 2.2	14.6 ± 1.6	3.1 ± 0.4	46.5 ± 3.4	43.8 ± 1.6	6.2%
BDE154	274 ± 3.7	49.5 ± 2.7	55.0 ± 3.4	20.3 ± 2.2	6.5 ± 0.4	81.6 ± 3.7	83.5 ± 2.0	2.3%
BDE153	958 ± 116	198 ± 17.9	59.7 ± 0.3	79.2 ± 10.5	17.5 ± 1.0	90.9 ± 8.7	119 ± 1.0	23.6%
BDE138	244 ± 15.6	16.0 ± 1.4	5.5 ± 0.4	9.1 ± 1.4	1.9 ± 0.3	13.2 ± 0.5	15.2 ± 2.0	13.2%
BDE183	4350 ± 446	118 ± 11.2	85.0 ± 12.1	16.4 ± 1.0	14.1 ± 0.7	43.2 ± 3.8	43.0 ± 3.5	0.5%
BDE190	843 ± 48.3	15.2 ± 3.8	7.5 ± 2.0	5.0 ± 0.7	1.9 ± 0.3	4.3 ± 0.7	5.1 ± 2.9	15.7%
BDE209	39,800 ± 6960	8980 ± 1050	5030 ± 302	2770 ± 497	2570 ± 278	2862 ± 486	2510 ± 190	14.0%

n: number of measurements; RSD: relative standard deviation.

finally tested by GC–MS according to the method mentioned above. In each batch of experiment, three mice used as control, three mice used as experiment group. The experiment was repeated two times. Finally, only five samples were used for the data analysis because there were some samples failures to test the concentrations of PBDEs.

2.4. Determination of PBDE bioaccessibility using five in vitro methods

To determine the bioaccessibility of PBDEs, five in vitro methods comprising Fa-VDM, PBET, UBM, DIN, and IVG were used with some modifications compared with the previously reported methods (Yu et al., 2011; Juhasz et al., 2009; Li et al., 2015b; Rodriguez and Basta, 1999; Wragg et al., 2007). Typically, 0.09 g of soil or SRM2585 (0.2 g samples were used for the other four methods) was digested in the artificially prepared gastric solution and then the intestinal solution at 37 °C. The components of the digestion solution and the associated parameters are all shown in Table S1.

The mixtures were incubated for a given time (Table S1) and then centrifuged at 3000 $\times g$ for 10 min at room temperature. The supernatants were filtered with polypropylene fiber membranes (0.45 μm). After filtration, the PBDEs in the filtrates were liquid–liquid extracted sequentially with acetone, n-hexane, and dichloromethane (2:1:3, v:v:v) similar to our previous study (Yu et al., 2009). The PBDEs in the pellets (soils) were extracted using acetone assisted by sonication. All of the extracts were treated as described in our previous studies and the final samples were stored at −18 °C until the analyses (Yu et al., 2013). Six in vitro experiments for five soil samples and SRM2585 and a procedural blank were carried out in each batch of in vitro test. The experiments were repeated at least three times.

2.5. Quality assurance and quality control

A procedural blank was run for each batch of samples to check for potential contamination in the laboratory. Calibration plots had satisfactory linear regression coefficients for all congeners ($R^2 > 0.998$). The limit of detection varied from 0.15 to 0.45 ng/g for tri- to hepta-BDE congeners and 1.8 ng/g for BDE209 in the soils. The congener of BDE71 was not quantified because of interfering peaks. The data were not corrected against the recovery rates of the surrogate standard ^{13}C-PCB141, and an average of 95.4 ± 10.3% was calculated based on all of the samples.

2.6. Calculations and statistical analysis

The bioavailability of a congener was calculated according to the following equation:

$$Bav(\%) = \frac{m_{wc}}{m_t + m_{gtc} + m_u + m_f} \times 100, \tag{1}$$

where m_{wc}, m_{gtc}, m_u, and m_f (pg) denote the mass of a PBDE congener in the whole corpse of a mouse, gastrointestinal tract contents, urine, and feces, respectively.

The bioaccessibility of each PBDE congener was calculated according to the following equation:

$$Ba(\%) = \frac{m_s}{m_s + m_p} \times 100, \tag{2}$$

where m_s and m_p (pg) denote the mass of a PBDE congener in the supernatant and pellet after digestion, respectively.

Statistical analysis was carried out using SPSS 13.0. The relationships among the variables were tested using linear or parabolic regression model. The level of statistical significance was set at $p < 0.05$, and a marginal significance was set at $p < 0.1$.

3. Results and discussion

3.1. Concentrations of PBDEs

The concentrations of PBDEs were first measured in the soils before testing the bioaccessibility and bioavailability. In addition, the PBDEs in SRM2585 were determined for quality assurance and quality control. We measured the concentrations of PBDEs five times similar to our previous study (Yu et al., 2006) because they would be used for the standard data for the calculations of bioaccessibility and bioavailability. The concentrations of the PBDEs including 13 congeners in the five soils and SRM2585 are presented in Table 1. The measured concentrations of PBDEs in SRM2585 generally agreed well with the reference data with the relative standard deviations < 25% with the exception of BDE17, thereby demonstrating the satisfactory quality assurance and quality control results in our laboratory. The total PBDEs in the five soils ranged from 2.84×10^3 to 5.08×10^3 ng/g dw. This is because PBDEs were released into the environment during the dismantling of electronic equipment and several studies have reported that environmental matrices such as air, soil, and water are heavily contaminated by PBDEs in this area (An et al., 2011; Gao et al., 2011; Labunsk et al., 2013; Leung et al., 2007; Xiong et al., 2015, 2016). Thus, the concentrations of PBDEs in these soils are much higher than those in other nonpoint source contaminated areas of China as well as in other regions, such as Shanghai in China (4.68–174.7 ng/g dw) (Wu et al., 2015) and Kocaeli in Turkey (0.70–203 ng/g dw) (Cetin, 2014).

The PBDE congener profiles in the five soils showed that BDE209 was the dominant congener in all five soil samples (Fig. S2) where it accounted for 76.1%–90.5% of the total PBDEs. In addition to BDE209,

Table 2
Bioavailability (%) of PBDEs in the soils collected from the e-waste dismantling town and the reference SRM2585 determined using female C57BL/6 mice.

Congeners	Soils (n = 5)					Reference dust (n = 5)
	# 2	# 3	# 5	# 9	# 18	SRM2585
BDE28	15.1 ± 5.8	12.1 ± 1.8	15.6 ± 3.7	31.5 ± 9.6	25.6 ± 10.2	45.0 ± 9.5
BDE47	21.3 ± 7.8	10.1 ± 3.4	24.3 ± 6.8	22.6 ± 8.7	33.8 ± 3.5	48.8 ± 5.7
BDE66	11.6 ± 3.7	5.5 ± 4.8	13.1 ± 3.2	15.4 ± 6.9	14.4 ± 4.7	30.2 ± 7.0
BDE100	31.7 ± 12.1	6.8 ± 6.3	38.1 ± 2.7	16.1 ± 8.3	37.0 ± 10.4	41.7 ± 8.6
BDE99	22.8 ± 5.3	5.7 ± 5.3	18.9 ± 3.6	13.1 ± 3.8	29.0 ± 0.4	42.8 ± 9.9
BDE154	20.9 ± 7.8	7.2 ± 4.1	23.4 ± 1.3	8.7 ± 6.4	31.0 ± 8.8	32.9 ± 9.9
BDE153	18.2 ± 4.3	6.5 ± 3.8	16.9 ± 2.8	7.8 ± 2.8	29.3 ± 2.0	37.8 ± 7.6
BDE183	9.3 ± 2.2	14.1 ± 4.2	9.2 ± 3.7	8.7 ± 6.9	18.3 ± 3.4	23.9 ± 6.3
BDE209	1.7 ± 1.4	3.5 ± 2.9	5.6 ± 2.6	5.6 ± 5.0	5.6 ± 4.7	3.9 ± 1.5

n: number of measurements.
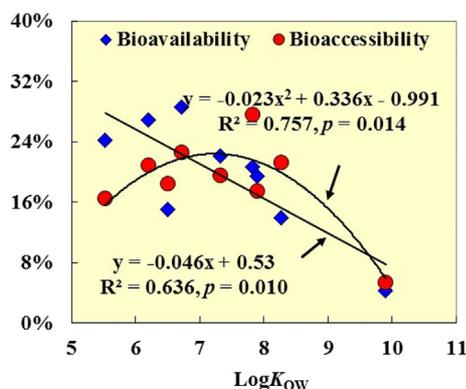
Fig. 1. Relationships between the average bioavailability or bioaccessibility of PBDEs in samples (the five soils and SRM2585) and the $LogK_{OW}$ values.

BDE47, and BDE99 were also important congeners where they accounted for 4.5% and 4.7% of the totals on average, respectively. Similar results were also reported from the same region and other cities in China (An et al., 2011; Yu et al., 2012a; Leung et al., 2007; Zhang et al., 2014). The high proportion of BDE209 in the soil samples implies that the main sources of PBDEs in the e-waste dismantling site were deca-BDE commercial products. Penta-BDE industrial products were also important sources of PBDE contamination in this region.

3.2. Bioavailability of PBDEs measured in soil using an in vivo assay

The bioavailability data for PBDEs in the soils and SRM2585 were determined using female C57BL/6 mice and the results are listed in Table 2. The bioavailability of four congeners could not be measured due to the low concentrations of BDE17, 85, 138, and 190 in three samples, and thus their bioavailability data were not reported. The bioavailability data for PBDEs in the soil samples collected from the e-waste dismantling town ranged from 1.7 ± 1.4% (BDE209) to 38.1 ± 2.7% (BDE100). For SRM2585, the bioavailability ranged from 3.9 ± 1.5% (BDE209) to 48.8 ± 5.7% (BDE47). In all of the samples, including the soil samples and SRM2585, the lowest bioavailability in each sample was observed for BDE209, whereas the highest varied among the other congeners for different soil samples, such as BDE28, BDE47, BDE100, and BDE183. Similar data were obtained with the lowest bioavailability of BDE209 (4.0% and 4.8%) in SRM2585 using Sprague-Dawley rats in a previous study (Huwe et al., 2008). Also, we believe that the low bioavailability of BDE209, as a decabrominated congener (Gerecke et al., 2005), was also can be attribute to transepithelial transport via intestinal cells because of the large molecular volume and bioaccessibility due to its status in the matrix as discussed in the followed text. For the other congeners, the bioavailabilities of the PBDEs in SRM2585 were generally lower in the present study compared

with the bioavailabilities of tri- to hexa-BDEs, which ranged from 25.1% to 76.1% according to Huwe et al. (2008) who added SRM2585 to corn oil and then mixed it with mouse chow before administration. This difference may have occurred because intestinal bile could emulsify with oil droplets to form small micelles that enhanced the bioaccessibility of PBDEs (Yu et al., 2010) and facilitated their intestinal absorption (Moser and Mclachlan, 2001), and thus the bioavailability was higher in the references. However, in our samples, the soils were only suspended in water and administered orally without the addition of oil. The results also implicated that higher lipid food would lead higher bioaccessibilities and bioavailabilities of PBDEs, and thus higher human health risks.

In the present study, the bioavailabilities of the different brominated congeners varied by an order of magnitude from 1.7% to 48.8%. In each sample, the trend in the variation was not obvious for different brominated congeners (Table 1). However, when we averaged the bioavailability of each congener in the soils and SRM2585, and plotted them against octanol/water partition coefficient ($LogK_{OW}$), a clear trend was observed where the average bioavailabilities of the PBDE congeners decreased from approximately 20% for low brominated congeners to 4.3% for BDE209 with an increase in $LogK_{OW}$ from 5.5 (BDE28) to 9.9 (BDE209) (Fig. 1). This trend can be explained based on intestinal absorption and diffusion through cellular membranes of the congeners with varied bromine substitution on the rings of PBDEs. As we well known, the unstirred water layer plays a very important role during intestinal absorption of drugs. The transport through unstirred water layer is the rate-limiting step for highly hydrophobic molecules as shown in Fig. S3 (Shanmukhan et al., 2018). In the present study, for the congeners with low $LogK_{OW}$ values, they are easier transport through unstirred water layer provided the higher intestinal absorption. However, when the $LogK_{OW}$ values increased, the transport of the high brominated congeners, especially for BDE209, increasingly limited and the absorption decreased. In addition, after the chemicals overcome the unstirred water layer, diffusive transport through the membranes of intestine is important, and the high brominated congeners with large $LogK_{OW}$ values and molecular volumes became increasingly limited and thus the absorption also decreased. A similar trend was found in our previous study where passive diffusion dominated the transepithelial transport of PBDEs in Caco-2 cells (derived from human colon carcinoma cells and widely used for investigating the absorption, transport, and metabolism of xenobiotics in the human intestine), and there was a negative linear relationship between transport and the $LogK_{OW}$ value where more brominated congeners were difficult to transport (Yu et al., 2017). Higher $LogK_{OW}$ values could limit the solubility of PBDEs from soil matrix to aqueous environment in the digestion fluids but can increase the intestinal absorption because of higher affinity to lipidic membranes. At the same time, the intestinal absorption of high brominated congeners would be limited because of large molecular volumes. Therefore, the influence of the $LogK_{OW}$ values of the congeners on the absorption and bioavailability of PBDEs is a combined result of

several factors.

To further understand the effects of different factors on the bioavailability of PBDEs, we determined the H/C ratios (a higher H/C ratio indicates the lower aromaticity of OM) and OM contents of the samples, as well as evaluating the correlations with the bioavailability of PBDEs (Figs. S4 and S5). The results showed that as the H/C ratios increased from 0.1 to 0.35, the average bioavailability of different brominated congeners in each sample (which varied between 7.94% and 34.1%) did not have positive or negative correlations with the ratios. Similar results were found for the OM contents, where the average bioavailability also varied as the OM contents increased from 7.2% to 58.3%. The results demonstrated that the bioavailability of PBDEs was not significantly correlated with the H/C ratios or OM contents. Thus, the H/C ratios and OM contents were not important factors that affected the bioavailability of PBDEs, although significant correlations were observed for bioaccessibility of PBDEs in house dust in our previous study (Yu et al., 2012a).

3.3. Bioaccessibility of PBDEs measured using five in vitro methods

The bioaccessibilities of PBDEs in the five soils and SRM2585 sample were measured using Fa-VDM, and the results are listed in Table 3. The results showed that the bioaccessibilities of the different congeners varied between 1.6% and 55.4%. In each soil sample, BDE209 was generally the congener with the lowest bioaccessibility (1.6%–9.4%). The bioaccessibilities of the PBDEs were also measured using the other four methods, i.e., PBET, UBM, DIN, and IVG (Fig. S6), and compared with those determined by Fa-VDM. The results showed that excluding sample No. 9 where the bioaccessibilities of tri- to hepta-congeners (9.8%–15.7%) measured using DIN were slightly higher than those determined by Fa-VDM (5.6%–12.6%), the bioaccessibilities of the PBDEs measured by PBET, UBM, DIN, and IVG in the other soils were generally lower than those measured with Fa-VDM. The bioaccessibilities of tri- to hepta-BDE congeners in the soils were generally higher than that of BDE209 in the same samples.

Similar results were obtained for SRM2585. The bioaccessibilities of tri- to hepta-BDE congeners in the reference material ranged from 23.8% to 32.1% and that in BDE209 was 6.7% according to Fa-VDM (Table 3). The bioaccessibilities of the PBDEs measured by PBET, UBM, DIN, and IVG varied between 6.6% and 19.6% (Fig. 2). Compared with the results obtained for the soil samples, the bioaccessibilities of the PBDEs in SRM2585 exhibited a similar trend where BDE209 was the congener with the lowest bioaccessibility. In the present study, the bioaccessibilities of the PBDEs in SRM2585 according to PBET ranged from 6.6% to 9.9%. Fang and Stapleton (2014) also obtained similar results (e.g., approximately 10% was obtained for BDE47) with PBET, but they combined the colon phase in the calculation. However, the bioaccessibilities of tri- to hepta-BDE congeners were approximately 40%–60% using a Tenax assisted colon-extended PBET method (Kademoglou et al., 2018), which were higher than our results and

those obtained by Fang and Stapleton (2014). In general, the bioaccessibility results obtained using the Fa-VDM method developed for organic compounds were more reliable than those measured by DIN, which was mainly developed for organic compounds (Adenugba et al., 2008), and the other three in vitro methods comprising IVG, UBM, and PBET, which were specifically developed for heavy metals (Li et al., 2016a; Cui et al., 2016).

As mentioned above, the bioaccessibilities of different brominated congeners varied in each sample and different results were obtained by the five in vitro methods. Internal and external factors were considered to further understand their effects on the different variations. The internal factors included the properties of PBDEs and the PBDE-bound matrices, whereas the external factors included the digestion conditions and the components of the digestion solutions used by the in vitro methods.

The bioaccessibilities of PBDE congeners in the soils were averaged and plotted against LogK_{OW} (Fig. 1). As LogK_{OW} increased for tri- to deca-BDEs from 5.53 to 9.9, a significant parabolic correlation (R^2 = 0.757) was observed between the bioaccessibilities of PBDEs measured using Fa-VDM and their LogK_{OW} values. In addition, compared with BDE209, the less brominated congeners of tri- to hepta-BDEs were more likely to be emitted from their commercial products, such as the polymer matrices, and then adsorbed onto the surfaces of soils. Therefore, their bioaccessibility increased as the LogK_{OW} value increased because the surfactant within the bile could increase the solubility of organic compounds in the digestion solution (Tang et al., 2006). BDE209 is not highly volatile and it is more difficult to release from matrices during digestion, thereby leading to lower bioaccessibility. Another study showed that the bioaccessibility of BDE209 was much higher in spiked soils than naturally contaminated soils, which were similar to the results for tri- to hepta-BDEs (Yu et al., 2013). Moreover, similar to the bioavailabilities, the bioaccessibilities of the PBDEs were not correlated with the H/C ratios or OM contents (Figs. S4 and S5). Therefore, the internal factor that mainly influenced the bioaccessibility of low brominated congeners was the LogK_{OW} value, and the original status had the main effect on BDE209. However, our previous research (Yu et al., 2012a) showed that the bioaccessibility of PBDEs, measured using a simulation system of human gastrointestinal tract showed significant negative correlations with OM content in dust. Additionally, the chemical composition of the soils (e.g., OM composition, or minerals), the soil microbiome, and so on, can also be a factor affecting the bioaccessibility or bioavailability. Moreover, in the human gastrointestinal tract, gut microbiome and food compositions might also can influence on the bioaccessibility or bioavailability. More studies are necessary to investigate the factors influence on the bioaccessibility and bioavailability.

The digestion conditions used in these methods were external factors that influenced the bioaccessibility. We found that the bioaccessibilities of PBDEs determined by Fa-VDM were generally higher than those obtained with the other four in vitro methods. This difference

Table 3
Bioaccessibility (%) of PBDEs in the soils collected from the e-waste dismantling town and the reference SRM2585 measured using Fa-VDM.

| Congeners	Soils (n = 3)					Reference dust (n = 3)
	# 2	# 3	# 5	# 9	# 18	SRM2585
BDE28	12.9 ± 2.1	8.2 ± 2.8	16.8 ± 2.2	16.2 ± 3.6	20.8 ± 3.0	23.8 ± 0.6
BDE47	12.2 ± 3.0	7.0 ± 2.9	28.5 ± 1.6	12.2 ± 1.6	33.0 ± 4.4	32.1 ± 4.2
BDE66	7.5 ± 1.5	7.2 ± 1.7	29.3 ± 3.0	12.0 ± 1.2	29.6 ± 3.1	25.1 ± 0.6
BDE100	17.2 ± 3.1	6.5 ± 3.5	32.8 ± 5.8	14.0 ± 2.6	36.4 ± 4.6	28.7 ± 1.1
BDE99	16.5 ± 3.5	4.7 ± 2.3	27.1 ± 5.7	8.1 ± 3.5	31.1 ± 2.9	29.7 ± 3.2
BDE154	18.8 ± 5.6	8.0 ± 3.0	42.7 ± 11.8	10.9 ± 2.6	55.4 ± 7.5	30.1 ± 3.0
BDE153	16.0 ± 4.4	6.8 ± 1.6	23.0 ± 10.7	5.6 ± 1.2	25.8 ± 5.0	27.9 ± 2.8
BDE183	13.3 ± 4.3	22.9 ± 1.7	27.9 ± 11.1	9.7 ± 2.0	28.1 ± 4.5	25.6 ± 3.7
BDE209	9.4 ± 4.3	4 ± 1.9	5.6 ± 2.9	1.6 ± 0.4	4.7 ± 1.4	6.7 ± 0.9

n: number of measurements.
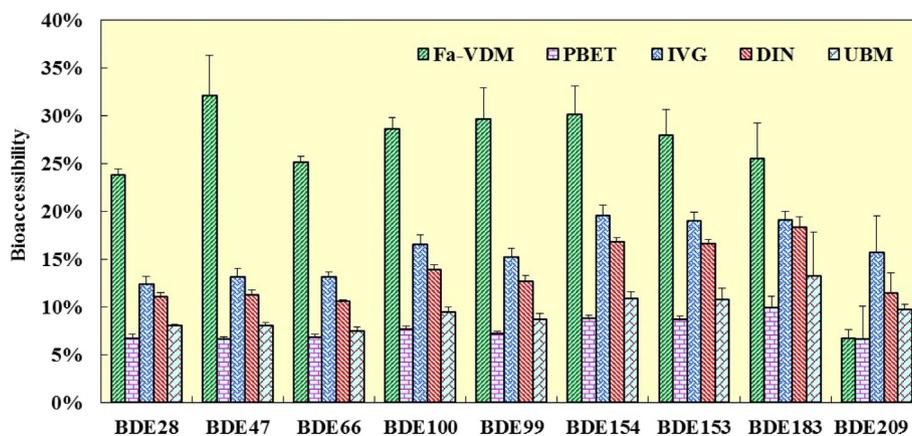
Fig. 2. Bioaccessibility of PBDEs in SRM2585 measured with five in vitro methods.

occurred because the bile concentration was around 5.5 g/L in the Fa-VDM digestion solution (Yu et al., 2011), which was higher than those in the PBET (1.75 g/L), IVG (3.5 g/L), UBM (3.7 g/L), and DIN (4.5 g/L) methods. The bile in the digestion solution could decrease the surface tension of the solution. It is well known that surface tension plays an important role in the mobilization of hydrophobic organic contaminants from a solid matrix due to the formation of bile salt micelles combined with the organic compounds, which affects their solubility (Oomen et al., 2004). In the present study, the formation of PBDEs with bile salt micelles increased the solubility of the chemicals in the intestinal solutions, and thus the bioaccessibility results obtained by Fa-VDM were higher than those with PBET, UBM, DIN, and IVG.

Furthermore, the ratio of the volume of the digestion solution relative to the mass of the digested solid matrix, i.e., the liquid to solid ratio (L/S), was selected as 200 in the Fa-VDM, which was also much higher than those in the PBET (100), IVG (150), UBM (97.5), and DIN (100) methods. This ratio was selected because the L/S ratio-dependent release of PBDEs was optimized with L/S ratios ranging from 50 to 200 in our previous study, and a relative steady state was obtained for ratios of 200 to 250 (Yu et al., 2011).

In addition, the incubation time during the intestinal phase was 6 h in Fa-VDM (Yu et al., 2011), which was longer than those in the other three methods (4 h for PBET, 1 h for IVG, and 4 h for UBM). The digestion time during the intestinal phase in DIN (6 h) was similar to that in Fa-VDM, but the lower concentration of bile (5.5 versus 4.5 g/L) and much lower L/S ratio (200 versus 100) could mainly account for the lower bioaccessibility measured by DIN. However, a previous study also showed that the bioaccessibility of spiked PBDEs did not increase in fish after digestion for 4 h (Yu et al., 2009). Therefore, the higher bioaccessibility of PBDEs tested using Fa-VDM was mainly due to the higher bile concentration and larger L/S ratio compared with those used in PBET, UBM, DIN, and IVG.

3.4. Correlations between the bioavailability and bioaccessibility of PBDEs in soils

We determined the correlations between the bioaccessibilities of PBDEs obtained in the soils using the five in vitro digestion models and the bioavailabilities measured using mice. When the bioaccessibilities of the PBDEs measured in the soils using Fa-VDM were correlated with the bioavailabilities, large IVIVC variations were observed for the different PBDE congeners (Table S2). The slopes for the PBDE congeners varied from -1.22 to 1.91 with R^2 values of 0.280–0.865 (p-values of 0.006–0.997), and the intercepts ranged from -7.72% to 10.4%. In general, no significant IVIVCs were obtained between the bioavailabilities and bioaccessibilities determined for the PBDEs by PBET, DIN, UBM, and IVG with the exception of BDE209 via PBET (Table S2). For the IVG method, the R^2 value determined for each PBDE congener

(0.265–0.613) was generally much higher than those obtained using the PBET, DIN, and UBM methods, except for BDE183 and 209. Their slopes were also generally higher than those obtained with PBET, DIN, and UBM. However, all of them were generally lower than those measured by Fa-VDM.

It is well known that the correlations between bioaccessibility and bioavailability data can be used to estimate the predictive capacity of the bioavailability based on the bioaccessibility tested with in vitro methods. In general, if the slope of the regression line ranges between 0.8 and 1.2 with an R^2 value higher than 0.6 and an intercept close to zero, the bioaccessibility of a chemical can be used to predict its bioavailability (Wragg et al., 2011). In the present study, the correlation slopes between the bioavailability and bioaccessibility for BDE47, 99, 100, and 153 in soils measured by Fa-VDM varied from 0.83 to 1.16 with R^2 values of 0.738–0.865, and intercepts of -7.42%, -2.44%, -0.25%, and 1.11%, respectively, which were in the general consensus ranges. Fig. 3 shows that there was a significant IVIVC with a slope of 1.06 ($R^2 = 0.849$) based on the average data for BDE47, 99, 100, and 153, although there are only marginal statistical significance ($p < 0.1$) for BDE47 and 99 (Table S2). However, no data were within the ranges using the other four in vitro methods with the exception of BDE209 via PBET (Table S2). These results indicate that the Fa-VDM method has the potential to predict the bioavailabilities of some PBDE congeners in soils, such as BDE47, 99, 100, and 153. The bioavailabilities can be used to more accurately calculate the estimate daily uptake of PBDEs for human exposure assessment. It should be noted that the Fa-VDM has not a good IVIVC for BDE209 (slope $= -1.22$; intercept $= 10.4$; $R = 0.549$), a dominant congener in the soil samples. As we well known, PBDEs are a kind of persistent organic pollutants with half-lives of the congeners varied from days to years in organisms, and much
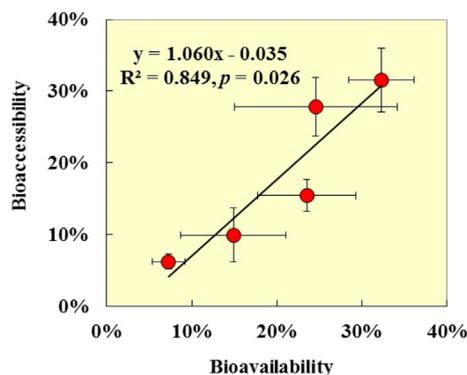

Fig. 3. Relationships between the average bioavailability and bioaccessibility of BDE47, 99, 100, and 153 in the five soils measured using female C57BL/6 mice and the Fa-VDM method.

shorter half-life of BDE209 was reported (Venier and Hites, 2011). The metabolism of BDE209 might also have certain effect on the bioavailability, although the present exposure time was only 24 h. More studies and suitable method are warranted.

However, it should be noted that it is difficult to use a single method to predict all PBDE congeners with large $LogK_{OW}$ ranges because this very important factor influences the bioaccessibility and bioavailability. However, its effects on the bioaccessibility and bioavailability were different, where the former was parabolic and the latter was negative linear. Thus, only the moderately brominated congeners exhibited good correlations between the bioaccessibility and bioavailability, thereby indicating the possible application of the bioaccessibility of moderately brominated congeners, such as BDE47, 99, 100, and 153, for predicting the bioavailability. To overcome the problem of the static in vitro model for measuring hydrophobic chemicals with large variations in $LogK_{OW}$, it is essential to devise an analogue to cells as a sink for simulating intestinal absorption. Several "absorption sinks" have been used, such as Tenax, silicone, and cyclodextrins (Fang and Stapleton, 2014; Kademoglou et al., 2018; Li et al., 2017; Zhang et al., 2015; Mayer et al., 2016; Gouliarmou et al., 2013), but they do not simulate the absorption process in organisms accurately. Thus, greater effort is needed to develop effective assays that accurately mimic the release and absorption of hydrophobic chemicals in the gastrointestinal tract.

4. Conclusion

In the present study, we measured the bioavailability and bioaccessibility of PBDEs in soils collected from an e-waste dismantling town and reference material SRM2585 using female C57BL/6 mice and five in vitro methods comprising Fa-VDM, PBET, DIN, UBM, and IVG. Higher brominated BDEs exhibited lower bioavailability and there was a negative relationship between the bioavailability and $LogK_{OW}$. However, there was a parabolic correlation between the bioaccessibility of PBDEs determined with Fa-VDM and $LogK_{OW}$. In general, the bioaccessibilities of PBDEs determined using Fa-VDM were higher than the values measured by the other four methods, mainly because of the bile concentrations and the L/S ratios. Significant IVIVCs with slopes that varied from 0.83 to 1.16 and intercepts of 0.3%–7.7% were obtained between the bioaccessibility and bioavailability results for BDE47, 99, 100, and 153, thereby suggesting that Fa-VDM can determine the bioaccessibility and it could potentially be used to predict the bioavailability of congeners in soils. Many challenges still affect bioaccessibility measurements for hydrophobic chemicals. Thus, greater effort is needed to develop effective in vitro methods in the future that accurately mimic digestion and absorption in the human gastrointestinal tract by using a better "absorption sink."

Notes

The authors declare no competing financial interest.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envint.2018.12.022.

References

Adenugba, A.A., McMartin, D.W., Beck, A.J., 2008. In vitro approaches to assess bioavailability and human gastrointestinal mobilization of food-borne polychlorinated biphenyls (PCBs). J. Environ. Sci. Health B 43 (5), 410–421.

An, T.C., Zhang, D.L., Li, G.Y., Mai, B.X., Fu, J.M., 2011. On-site and off-site atmospheric PBDEs in an electronic dismantling workshop in South China: gas-particle partitioning and human exposure assessment. Environ. Pollut. 159 (12), 3529–3535.

Cetin, B., 2014. Soil concentrations and source apportionment of polybrominated diphenyl ethers (PBDEs) and trace elements around a heavily industrialized area in Kocaeli, Turkey. Environ. Sci. Pollut. Res. 21 (13), 8284–8293.

Cui, X.Y., Xiang, P., He, R.W., Juhasz, A., Ma, L.Q., 2016. Advances in in vitro methods to evaluate oral bioaccessibility of PAHs and PBDEs in environmental matrices. Chemosphere 150, 378–389.

Duan, L.C., Palanisami, T., Liu, Y.J., Dong, Z.M., Mallavarapu, M., Kuchel, T., Semple, K.T., Naidu, R., 2014. Effects of ageing and soil properties on the oral bioavailability of benzo[a]pyrene using a swine model. Environ. Int. 70, 192–202.

Fang, M.L., Stapleton, H.M., 2014. Evaluating the bioaccessibility of flame retardants in house dust using an in vitro Tenax bead-assisted sorptive physiologically based method. Environ. Sci. Technol. 48 (22), 13323–13330.

Feng, Y., Hu, Q.L., Meng, G., Wu, X.M., Zeng, W.H., Zhang, X., Yu, Y.X., Wang, Y., 2015. Simulating long-term occupational exposure to decabrominated diphenyl ether using C57BL/6 mice: biodistribution and pathology. Chemosphere 128, 118–124.

Gao, S.T., Hong, J.W., Yu, Z.Q., Wang, J.Z., Yang, G.Y., Sheng, G.Y., Fu, J.M., 2011. Polybrominated diphenyl ethers in surface soils from E-Waste recycling areas and industrial areas in south China: concentration levels, congener profile, and inventory. Environ. Toxicol. Chem. 30 (12), 2688–2696.

Gerecke, A.C., Hartmann, P.C., Heeb, N.V., Kohler, H.P.E., Giger, W., Schmid, P., Zennegg, M., Kohler, M., 2005. Anaerobic degradation of decabromodiphenyl ether. Environ. Sci. Technol. 39 (4), 1078–1083.

Gouliarmou, V., Collins, C.D., Christiansen, E., Mayer, P., 2013. Sorptive physiologically based extraction of contaminated solid matrices: incorporating silicone rod as absorption sink for hydrophobic organic contaminants. Environ. Sci. Technol. 47 (2), 941–948.

Huwe, J.K., Hakk, H., Smith, D.J., Diliberto, J.J., Richardson, V., Stapleton, H.M., Birnbaum, L.S., 2008. Comparative absorption and bioaccumulation of polybrominated diphenyl ethers following ingestion via dust and oil in male rats. Environ. Sci. Technol. 42 (7), 2694–2700.

Juhasz, A.L., Weber, J., Smith, E., Naidu, R., Rees, M., Rofe, A., Sansom, L., 2009. Assessment of four commonly employed in vitro arsenic bioaccessibility assays for predicting in vivo relative arsenic bioavailability in contaminated soils. Environ. Sci. Technol. 43 (24), 9487–9494.

Kademoglou, K., Williams, A.C., Collins, C.D., 2018. Bioaccessibility of PBDEs present in indoor dust: a novel dialysis membrane method with a Tenax TA® absorption sink. Sci. Total Environ. 621, 1–8.

Labunsk, I., Harrad, S., Santillo, D., Johnston, P., Brigden, K., 2013. Levels and distribution of polybrominated diphenyl ethers in soil, sediment and dust samples collected from various electronic waste recycling sites within Guiyu town, southern China. Environ. Sci.: Processes Impacts 15 (2), 503–511.

Leung, A.O.W., Luksemburg, W.J., Wong, A.S., Wong, M.H., 2007. Spatial distribution of polybrominated diphenyl ethers and polychlorinated dibenzo-p-dioxins and dibenzofurans in soil and combusted residue at Guiyu, an electronic waste recycling site in southeast China. Environ. Sci. Technol. 41 (8), 2730–2737.

Li, C.L., Zhao, Z.H., Lei, B.L., An, J., Zhang, X.Y., Yu, Y.X., 2015a. Polybrominated diphenyl ethers in the air and comparison of the daily intake and uptake through inhalation by Shanghai residents with those through other matrices and routes. Environ. Sci. Pollut. Res. 22 (3), 1750–1759.

Li, J., Li, K., Cave, M., Li, H.B., Ma, L.Q., 2015b. Lead bioaccessibility in 12 contaminated soils from China: correlation to lead relative bioavailability and lead in different fractions. J. Hazard. Mater. 295, 55–62.

Li, K., Li, C., Yu, N., Juhasz, A.L., Cui, X.Y., Ma, L.N.Q., 2015c. In vivo bioavailability and in vitro bioaccessibility of perfluorooctanoic acid (PFOA) in food matrices: correlation analysis and method development. Environ. Sci. Technol. 49 (1), 150–158.

Li, C., Sun, H.J., Juhasz, A.L., Cui, X.Y., Ma, L.Q., 2016a. Predicting the relative bioavailability of DDT and its metabolites in historically contaminated soils using a Tenax-improved physiologically based extraction test (TI-PBET). Environ. Sci. Technol. 50 (3), 1118–1125.

Li, H.B., Zhao, D., Li, J., Li, S.W., Wang, N., Juhasz, A.L., Zhu, Y.G., Ma, L.Q., 2016b. Using the SBRC assay to predict lead relative bioavailability in urban soils: contaminant source and correlation model. Environ. Sci. Technol. 50 (10), 4989–4996.

Li, S.W., Sun, H.J., Li, H.B., Luo, J., Ma, L.Q., 2016c. Assessment of cadmium bioaccessibility to predict its bioavailability in contaminated soils. Environ. Int. 94, 600–606.

Li, C., Zhang, R.R., Li, Y.Z., Zhang, S.J., Gao, P., Cui, X.Y., Ma, L.Q., 2017. Relative bioavailability and bioaccessibility of PCBs in soils based on a mouse model and Tenax-improved physiologically-based extraction test. Chemosphere 186, 709–715.

Mayer, P., Hilber, I., Gouliarmou, V., Hale, S.E., Cornelissen, G., Bucheli, T.D., 2016. How to determine the environmental exposure of PAHs originating from biochar. Environ. Sci. Technol. 50 (4), 1941–1948.

Moser, G.A., Mclachlan, M.S., 2001. The influence of dietary concentration on the absorption and excretion of persistent lipophilic organic pollutants in the human intestinal tract. Chemosphere 45, 201–211.

Oomen, A.G., Sips, A., Groten, J.P., Sijm, D., Tolls, J., 2000. Mobilization of PCBs and lindane from soil during in vitro digestion and their distribution among bile salt micelles and proteins of human digestive fluid and the soil. Environ. Sci. Technol. 34 (2), 297–303.

Oomen, A.G., Rompelberg, C.J.M., Van de Kamp, E., Pereboom, D., De Zwart, L.L., Sips, A., 2004. Effect of bile type on the bioaccessibility of soil contaminants in an in vitro digestion model. Arch. Environ. Contam. Toxicol. 46 (2), 183–188.

Ounnas, F., Feidt, C., Toussaint, H., Marchand, P., Le Bizec, B., Rychen, G., Jurjanz, S., 2010. Polychlorinated biphenyl and low polybrominated diphenyl ether transfer to milk in lactating goats chronically exposed to contaminated soil. Environ. Sci. Technol. 44 (7), 2682–2688.

Pu, X.Z., Lee, L.S., Galinsky, R.E., Carlson, G.P., 2004. Evaluation of a rat model versus a physiologically based extraction test for assessing phenanthrene bioavailability from soils. Toxicol. Sci. 79 (1), 10–17.

Rodriguez, R.R., Basta, N.T., 1999. An in vitro gastrointestinal method to estimate bioavailable arsenic in contaminated soils and solid media. Environ. Sci. Technol. 33 (4), 642–649.

Rostami, I., Juhasz, A.L., 2011. Assessment of persistent organic pollutant (POP) bioavailability and bioaccessibility for human health exposure assessment: a critical review. Crit. Rev. Environ. Sci. Technol. 41 (7), 623–656.

Ruby, M.V., Davis, A., Schoof, R., Eberle, S., Sellstone, C.M., 1996. Estimation of lead and arsenic bioavailability using a physiologically based extraction test. Environ. Sci. Technol. 30 (2), 422–430.

Shanmukhan, N.K., Radhakishnan, A., Maniarasan, V., Kuppusamy, G., Gideon, S., 2018. Bioformulative concepts on intracellular organ specific bioavailability. Ther. Deliv. 9 (10), 775–796.

Smith, E., Weber, J., Rofe, A., Gancarz, D., Naidu, R., Juhasz, A.L., 2012. Assessment of DDT relative bioavailability and bioaccessibility in historically contaminated soils using an in vivo mouse model and fed and unfed batch in vitro assays. Environ. Sci. Technol. 46 (5), 2928–2934.

Tang, X.Y., Tang, L., Zhu, Y.G., Xing, B.S., Duan, J., Zheng, M.H., 2006. Assessment of the bioaccessibility of polycyclic aromatic hydrocarbons in soils from Beijing using an in vitro test. Environ. Pollut. 140, 279–285.

Venier, M., Hites, R.A., 2011. Flame retardants in the serum of pet dogs and in their food. Environ. Sci. Technol. 45, 4602–4608.

Wang, X.X., 2015. Comparison of Bioavailability and Bioaccessibility for Polybrominated Diphenyl Ethers in Soils (A dissertation for master degree). Shanghai University.

Wragg, J., Cave, M., Nathanail, P., 2007. A study of the relationship between arsenic bioaccessibility and its solid-phase distribution in soils from Wellingborough, UK. J. Environ. Sci. Health A Tox. Hazard. Subst. Environ. Eng. 42 (9), 1303–1315.

Wragg, J., Cave, M., Basta, N., Brandon, E., Casteel, S., Denys, S., Gron, C., Oomen, A., Reimer, K., Tack, K., Van de Wiele, T., 2011. An inter-laboratory trial of the unified BARGE bioaccessibility method for arsenic, cadmium and lead in soil. Sci. Total Environ. 409 (19), 4016–4030.

Wu, M.H., Pei, J.C., Zheng, M., Tang, L., Bao, Y.Y., Xu, B.T., Sun, R., Sun, Y.F., Xu, G., Lei, J.Q., 2015. Polybrominated diphenyl ethers (PBDEs) in soil and outdoor dust from a multi-functional area of Shanghai: levels, compositional profiles and interrelationships. Chemosphere 118, 87–95.

Xiong, J.K., An, T.C., Li, G.Y., 2015. Pollution profiles and risk assessment of PBDEs and phenolic brominated flame retardants in water environments within a typical electronic waste dismantling region. Environ. Geochem. Health 37 (3), 457–473.

Xiong, J.K., Li, G.Y., An, T.C., Zhang, C.S., Wei, C.H., 2016. Emission patterns and risk assessment of polybrominated diphenyl ethers and bromophenols in water and sediments from the Beijiang River, South China. Environ. Pollut. 219, 596–603.

Yu, Y.X., Wen, S., Feng, Y.L., Bi, X.H., Wang, X.M., Peng, P.A., Sheng, G.Y., Fu, J.M., 2006. Development of a compound-specific carbon isotope analysis method for atmospheric formaldehyde via NaHSO$_3$ and cysteamine derivatization. Anal. Chem. 78 (4), 1206–1211.

Yu, Y.X., Han, S., Zhang, D., Van de Wiele, T., Lu, M., Wang, D.Q., Yu, Z.Q., Wu, M.H., Sheng, G.Y., Fu, J.M., 2009. Factors affecting the bioaccessibility of polybrominated diphenyl ethers in an in vitro digestion model. J. Agric. Food Chem. 57 (1), 133–139.

Yu, Y.X., Li, J.L., Zhang, X.Y., Yu, Z.Q., Van de Wiele, T., Han, S.Y., Wu, M.H., Sheng, G.Y., Fu, J.M., 2010. Assessment of the bioaccessibility of polybrominated diphenyl ethers in foods and the correlations of the bioaccessibility with nutrient contents. J. Agric. Food Chem. 58 (1), 301–308.

Yu, Y.X., Pang, Y.P., Zhang, X.Y., Li, C., Yu, Z.Q., Fu, J.M., 2011. Optimization of an in vitro method to measure the bioaccessibility of polybrominated diphenyl ethers in dust using response surface methodology. J. Environ. Sci. 23 (10), 1738–1746.

Yu, Y.X., Pang, Y.P., Li, C., Li, J.L., Zhang, X.Y., Yu, Z.Q., Feng, J.L., Wu, M.H., Sheng, G.Y., Fu, J.M., 2012a. Concentrations and seasonal variations of polybrominated diphenyl ethers (PBDEs) in in- and out-house dust and human daily intake via dust ingestion corrected with bioaccessibility of PBDEs. Environ. Int. 42, 124–131.

Yu, Y.X., Zhang, S.H., Huang, N.B., Li, J.L., Pang, Y.P., Zhang, X.Y., Yu, Z.Q., Xu, Z.G., 2012b. Polybrominated diphenyl ethers and polychlorinated biphenyls in freshwater fish from Taihu lake, China: their levels and the factors that influence biomagnification. Environ. Toxicol. Chem. 31 (3), 542–549.

Yu, Y.X., Yang, D., Wang, X.X., Huang, N.B., Zhang, X.Y., Zhang, D.P., Fu, J.M., 2013. Factors influencing on the bioaccessibility of polybrominated diphenyl ethers in size-specific dust from air conditioner filters. Chemosphere 93 (10), 2603–2611.

Yu, Y.X., Wang, M.M., Zhang, K.Q., Yang, D., Zhong, Y.F., An, J., Lei, B.L., Zhang, X.Y., 2017. The transepithelial transport mechanism of polybrominated diphenyl ethers in human intestine determined using a Caco-2 cell monolayer. Environ. Res. 154, 93–100.

Zhang, S.H., Xu, X.J., Wu, Y.S., Ge, J., Li, W.Q., Huo, X., 2014. Polybrominated diphenyl ethers in residential and agricultural soils from an electronic waste polluted region in South China: distribution, compositional profile, and sources. Chemosphere 102, 55–60.

Zhang, Y.H., Liu, W.J., Cheng, F.F., Xiong, G.N., Yang, X.H., Wang, X., Tao, S., Xing, B.S., Liu, W.X., 2015. Variations and influencing factors of oral bioaccessibility of polybrominated diphenyl ethers in soils using an in-vitro gastrointestinal model. Environ. Sci. 36 (6), 2292–2299 (in Chinese).