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Assessment of the Bioaccessibility of Polybrominated Diphenyl Ethers in Foods and the Correlations of the Bioaccessibility with Nutrient Contents

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Bioaccessibility of a contaminant in foods is the fraction of the contaminant mobilized from the food matrices during gastrointestinal digestion. In the present paper, the bioaccessibility of polybrominated diphenyl ethers (PBDEs) in 13 types of foods, including fish, meat, rice, flour, and vegetables, was determined using an in vitro digestion method. The bioaccessibility obtained ranged from 2.6% to 41.3%. It was found that the bioaccessibility of PBDEs exhibited positive correlations with fat and carbohydrate contents and negative correlations with protein and dietary fiber contents in the foods. Fat was the most important factor affecting the bioaccessibility of PBDEs. The mechanism underlying the correlations was discussed in view of partition of PBDEs between liquid and solid phases. To our knowledge, this is the first article systematically investigating the bioaccessibility of PBDEs in foods and demonstrating the influence of the food constituents on the bioaccessibility of PBDEs.

KEYWORDS: Bioaccessibility; PBDEs; food constituent; in vitro digestion; correlation

INTRODUCTION

Polybrominated diphenyl ethers (PBDEs) are organobromine compounds that are used as flame retardants in a variety of consumer products. The family of PBDEs consists of 209 congeners. PBDEs are highly stable and lipophilic and have been found to accumulate in the environment as well as in humans (*1*, *2*). Their chemical structures are similar to those of polychlorinated biphenyls (PCB), a class of persistent organic pollutants (POPs). Indeed, on May 8, 2009, the fourth meeting of the Convention of the Parties of the Stockholm Convention on POPs decided to list certain components of PBDEs, such as penta- and octabromodiphenyl ethers, as POPs (*3*).

PBDEs are endocrine disruptors, neuro- and developmental toxicants. In the family of PBDEs, lower brominated congeners, such as penta- and octabromodiphenyl ethers, are found to be the primary culprits to cause various toxic effects on humans, whereas higher brominated congeners, such as decabromodiphenyl ether, might be less toxic due to their large molecular weights, low vapor pressure, and high hydrophobicity (*4*−*6*).

Human exposure to PBDEs is caused mostly through digestion of food and ingestion of dust (*7*,*8*). Food is the primary source for lower brominated congeners (*9*, *10*), thus it is important to study human exposure to food-borne PBDEs. To accurately assess the exposure risk, two critical issues must be addressed. The first issue is bioaccessibility. Nutrition in foods is accessible for humans to absorb only after digestion. Likewise, to become accessible for uptake, PBDEs must first be released from the food matrices through digestion. The bioaccessibility of a contaminant in foods is thus defined as the fraction of the contaminant mobilized from the food matrices during gastrointestinal digestion (*11*). The second issue is bioavailability. After being absorbed by intestinal epithelium, a contaminant will undergo metabolism in cells, which can remove most or part of the contaminant. After first pass effect, the contaminant will enter the bloodstream and be transported to the target organs. The fraction of the contaminant in foods reaching the bloodstream is referred to as the bioavailable fraction that causes systemic toxic effects (*12*).

Bioavailability is an important parameter in the risk assessment. Human exposure to food-borne contaminants may be overestimated if bioavailability is not taken into account. However, the bioavailability data are limited as bioavailability is determined by in vivo experiments, which are usually costly and time-consuming. On the other hand, bioaccessibility is generally determined by in vitro experiments and thus is easier to be obtained. Consequently, bioaccessibility is often used as a substitute of bioavailability when estimating human exposure to food-borne contaminants (*13*).

To determine the bioaccessibility in foods, food samples need to be digested. In vitro digestion methods have been developed for this purpose (*14*, *15*). During the past two decades, several in vitro methods were employed on soil samples for human exposure

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assessment (*16*−*19*). On the basis of the methods in the literature, we developed a set of protocols, including an in vitro digestion process, to assess the bioaccessibility of PBDEs in foods (*20*). Totally, the bioaccessibility of PBDEs in 13 types of foods was determined and it was found that the constituents of foods exhibited great influence on the bioaccessibility of PBDEs. In fact, correlations were observed between the bioaccessibility of PBDEs and the contents of fat, protein, carbohydrate, and dietary fiber. To our knowledge, this is the first study systematically investigating the bioaccessibility of PBDEs in foods and demonstrating the correlations between the bioaccessibility of PBDEs and the contents of the food constituents.

MATERIALS AND METHODS

Materials. The PBDE standard consisting of 14 congeners (BDE17, 28, 47, 66, 71, 85, 99, 100, 138, 153, 154, 183, 190, and 209) was purchased from AccuStandard. ^{13}C-PCB208 and ^{13}C-PCB141 were purchased from Cambridge Isotope Laboratories. Bovine bile, D-(+)-glucose, mucin from porcine stomach (type Π), and starch from potato were obtained from Sigma. (+)-Arabinogalactan from larch wood, pectin from apples, and xylan from birch wood were purchased from Fluka. Peptone from poultry, pancreatin from porcine pancreas, pepsin from porcine gastric mucosa, and yeast extract were obtained from Merck. Silica gel (80−100 mesh) was purchased from Qingdao Haiyang Chemical Co., Ltd. (Qingdao, China). Acetone, dichloromethane, and *n*-hexane were obtained from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China) and redistilled prior to use. Other reagents were obtained from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China). Silica gel and alumina (100−200 mesh) were purified by Soxhlet extraction with *n*-hexane:dichloromethane (1:1, v/v) for 72 h and activated at 180 and 250 °C, respectively. The silica gel and alumina used in the experiments were prepared as follows. Deactivated silica gel (3%) or alumina: 3 g of water was added to 97 g of activated silica gel or alumina. NaOH-treated silica gel: 33 mL of 1 mol L^{-1} NaOH was added to 67 g of 3% deactivated silica gel. H_2SO_4-treated silica gel: 43.6 mL of concentrated H_2SO_4 was added to 100 g of 3% deactivated silica gel. The deactivated and treated silica gel/alumina were all stored in *n*-hexane until the time of use. Purified and sterilized water was obtained from a Milli-Q purifier (Millipore Co., USA). Ultrapure helium and nitrogen were obtained from Shanghai Spring Specialty Gases Co., Ltd.

Instrumentation. Analyses of samples containing PBDEs were performed on a Hewlett-Packard 6890N/5975 gas chromatography/mass spectrometry (GC/MS) equipped with an HP-5MS capillary column (30 m × 0.25 mm × 0.25 μm, J & W Scientific, USA), operated under negative chemical ionization (NCI) mode. Ultrapure helium was used as carrier gas under the constant flow mode at a flow rate of 1 mL min^{-1}. A 1 μL sample was used with splitless injection. The temperatures of the injector, interface, and ion source were set at 280, 290, and 250 °C, respectively. A linear temperature program was used, starting at 110 °C (1 min), 8 °C min^{-1} to 180 °C (1 min), 2 °C min^{-1} to 240 °C (5 min), 2 °C min^{-1} to 280 °C (5 min), 20 °C min^{-1} to 300 °C (15 min). The selective ion monitoring (SIM) mode was used and ions with *m*/*z* at 79 and 81 were monitored for tri- to heptabrominated congeners, while ions with *m*/*z* at 476/478 and 372/374 were monitored for ^{13}C-PCB208 and ^{13}C-PCB141, respectively.

General Procedures. Fat contents in food samples were measured gravimetrically with 2 g of food powders obtained after lyophilization and pulverization by Soxhlet extraction with *n*-hexane:acetone (1:1, v/v). The contents of protein, carbohydrate, and dietary fiber were measured by the Shanghai Institute of Quality Inspection and Technical Research of the National Center of Supervision and Inspection on Food Products Quality. Food samples were spiked with the PBDE standard. In all experiments, spiked food samples and the corresponding unspiked food samples were used in pairs. All experiments were performed in triplicate. The data were showed in the format of means ± s.d. All error bars and "±" values represent the standard deviations of at least three independent experiments.

Preparation of Digestion Solutions. The digestion solutions, including basic nutrition solution, gastric acid solution, gastric digestion solution, and intestinal solution, were prepared as described before (*19*, *20*). Briefly, the basic nutrition solution, autoclaved prior to use, contained (+)-arabinogalactan (1.0 g L^{-1}), pectin (2.0 g L^{-1}), xylan (1.0 g L^{-1}), potato starch (3.0 g L^{-1}), D-(+)-glucose (0.4 g L^{-1}), yeast extract (3.0 g L^{-1}), peptone (1.0 g L^{-1}), mucin (4.0 g L^{-1}), and L-cysteine (0.5 g L^{-1}). The gastric acid solution was prepared by dissolving pepsin in 0.1 mol L^{-1} HCl (0.0890 g L^{-1}) and sterilized by filtering through 0.22 μm filters. The gastric digestion solution with a pH at 3.0 ± 0.1 was prepared by mixing 400 mL of basic nutrition solution and 50 mL of gastric acid solution. The intestinal solution contained $NaHCO_3$ (12.5 g L^{-1}), bile bovine (6.0 g L^{-1}), and pancreatin (0.9 g L^{-1}). The solutions were stored at 4 °C until the time of use.

Sample Collection and Preparation. The foods, selected on the basis of Chinese diet, included rice and flour, four types of vegetables (spinach, cabbage, carrot, and tomato), four types of meat (pork, beef, chicken, and duck), and three types of fish (Grass carp (*Ctenopharyngodon idellus*), Crucian carp (*Carassius aumtus*), and pomfret (*Pampus argenteus*)). All food samples were randomly purchased from local markets in Shanghai, the largest city in China. Rice and the edible parts of vegetables were washed, air-dried, and then ground or mashed. The resulted rice powder or vegetable paste was lyophilized to dryness and were pulverized. The powders were sieved through a 100-mesh sieve. Meat and the fillets of fish were lyophilized to dryness and then pulverized. Flour was dried under vacuum. The samples spiked with the PBDE standard were prepared as follows. A solution of the PBDE standard in dichloromethane (the amount of the PBDE standard used was calculated to let the final concentration of the PBDE standard in the food powders reach 20−100 ng g^{-1}) was added to 0.2 g of food powder. The mixtures were shaken and the solvent was removed by gentle nitrogen blowing. The food powders containing the PBDE standard were stored at 4 °C until the time of digestion (no more than two days).

Determination of the Bioaccessibility of PBDEs. Digestion of food samples was performed using an in vitro digestion method based on the Simulator of the Human Intestinal Microbial Ecosystem developed by the Laboratory of Microbial Ecology and Technology of Ghent University in Belgium (*19*, *21*). The in vitro digestion of food samples was carried out by adding 12 mL of the gastric digestion solution to 0.2 g of food powder. The mixture was shaken and incubated at 37 °C for 2 h with gentle shaking. The intestinal solution (6 mL) was then added. The mixture, having a pH at 7.0 ± 0.2, was incubated at 37 °C for 6 h with shaking. The experiments were carried out under ultrapure nitrogen to simulate the anaerobic environment in human gastrointestinal tract and in the dark.

After digestion, the suspension of the food sample was centrifugated at 7000*g* for 10 min. The supernatant was filtered through a 0.45 μm membrane filter, and the surrogate and 20 mL of acetone was added. The mixture was extracted with *n*-hexane:dichloromethane (1:3, v/v, 30 mL × 3). The organic phase was concentrated to ca. 1 mL and then diluted with 30 mL of *n*-hexane. Concentrated sulfuric acid (10 mL) was added to the organic phase, the mixture was shaken violently and left at room temperature for several hours to remove biomolecules such as protein, chlorophyll, fat/lipid, and so on. The organic layer was washed with water for three times, dried over Na_2SO_4, and concentrated to 1 mL. Finally, the organic phase was cleaned up using a 10 mm i.d. silica−alumina column, which was packed, from bottom to top, with 3% deactivated neutral alumina (height: 6 cm), 3% deactivated silica gel (2 cm), NaOH-treated silica gel (5 cm), 3% deactivated silica gel (2 cm), and H_2SO_4-treated silica gel (6 cm). PBDEs were eluted with 70 mL of *n*-hexane:dichloromethane (1:1, v/v). The fraction was concentrated, and the internal standard (^{13}C-PCB208) was added. The samples were stored at −18 °C until the time of analysis by GC/MS.

Mass Balance Experiments. The mass balance experiments were carried out using four types of foods (Crucian carp, pomfret, rice, and carrot). After the in vitro digestion, the suspension of food samples was centrifugated. The supernatant was treated as described above. The pellet was resuspended in 10 mL of water and the surrogate (^{13}C-PCB141) was added. Acetone (20 mL) was added to the suspension, and the mixture was extracted with *n*-hexane:dichloromethane (1:3, v/v). After being treated with sulfuric acid and cleaned up on the silica gel-alumina column as described above, the organic phases obtained from both the supernatant and the pellet were subjected to GC/MS analysis. The mass balance value was calculated by dividing the combined quantity of PBDEs in the supernatant and the pellet (after subtracting the quantity of PBDEs in

the corresponding unspiked food sample) by the quantity of PBDE standard added to the food sample.

Quality Assurance and Quality Control (QA/QC). PBDEs were quantified on GC/MS by standard curves. Seven solutions of the PBDE standard in dichloromethane (from 0.5 to 100 ng mL^{-1}) were used. Excellent linearity for all PBDE congeners was observed ($R^2 > 0.998$). The limits of detection varied from 0.3 to 0.9 pg depending on individual PBDE congeners. The data were not corrected against the recovery because the surrogate showed that the recovery was excellent (97.5 ± 9.3%).

Calculation of Bioaccessibility. The bioaccessibility was calculated using the following equation (*20*):

$$Ba(\%) = \frac{m_{\text{PBDE-spiked-sample}} - m_{\text{PBDE-unspiked-sample}}}{m_{\text{spiked-PBDE}}} \times 100 \qquad (1)$$

where $Ba(\%)$ was the bioaccessibility of PBDE(s), and $m_{\text{PBDE-spiked-sample}}$ and $m_{\text{PBDE-unspiked-sample}}$ were the masses of PBDE(s) released from the spiked and unspiked food samples after digestion, respectively. The $m_{\text{spiked-PBDE}}$ was the mass of nominal spiked PBDE(s).

Statistical Analysis. The analysis of data was carried out using SPSS 11.5 for Windows and Metlab 7.0. The correlation between the two variables was considered statistically significant when $p < 0.05$.

RESULTS AND DISCUSSION

Methodology. Our method was developed on the basis of those to assess the bioaccessibility of contaminants in soil samples in the literature (*19*, *20*). In short, the food samples were spiked with a PBDE standard. Both the spiked and corresponding unspiked food samples were subjected to an in vitro digestion process and then centrifugated. The quantity of PBDEs in the supernatant of the unspiked sample was subtracted from that of the corresponding spiked sample. The obtained value was divided by the quantity of PBDEs added to the sample to give the bioaccessibility of PBDEs in the food sample. The quantity of PBDEs in the pellet was ignored because it was assumed that PBDEs in the pellet were excreted with feces.

It can be argued that the methodology has a flaw, that is, spiked PBDEs may be adsorbed on the surfaces of the food matrices rather than incorporated in cells and tissues of food samples, as expected in the naturally contaminated foods. Thus the spiked PBDEs are considered to be more easily extracted in comparison with the "natural" PBDEs incorporated in the contaminated foods, leading to overestimated bioaccessibility. However, this argument itself is in question because food samples are subjected to an in vitro digestion process. With a full digestion, this process will completely destroy tissues and cells and the "natural" PBDEs will be freed. Therefore, in principle there is no convincing reason to assume that the spiked PBDEs are easier to be extracted than the "natural" PBDEs. Moreover, while most foods are contaminated by absorbing PBDEs into cells and tissues, for some foods, such as fresh vegetables whose leaves are the edible parts, contamination also happens through adsorption of airborne PBDEs. In this situation, PBDEs are considered to be adsorbed on the surfaces of the food matrices. Apparently, the present status of PBDEs in foods is a complicated issue that has not been addressed yet. In addition, the bioaccessibility data obtained in the present study were comparable with the bioavailability data reported in the literature (see below), suggesting that the overestimate was not significant even though the spiking method did lead to overestimated bioaccessibility. Taken together, our method is a usable approach in spite of the above argument, although a better approach should be explored indeed.

Mass Balance. PBDEs, especially lower brominated congeners, are slightly volatile, which could lead to loss during the experiments. To exclude this possibility and validate our method, we performed mass balance experiments to investigate whether there were significant losses of PBDEs during the experiments. It was typical that the bioaccessibility of these investigated foods were very similar, therefore only four types of foods (Crucian carp, pomfret, rice, and carrot) were selected. The combined quantity of PBDEs in the supernatant and the pellet of an unspiked sample was subtracted from that of the corresponding spiked sample. The obtained value represented the total recovered quantity of the spiked PBDEs. The ratio of this value with the quantity of the PBDE standard added to the sample represented the recovery of the spiked PBDEs, which was an indicator how much PBDE standard was lost during the experiments. The recovery, listed in **Table 1**, ranged from 86.9% to 100.9%. Therefore, it indicated that no significant losses of PBDEs occurred during the experiments.

Bioaccessibility of PBDEs. As shown in **Figure 1**, the bioaccessibility of PBDEs in 13 types of foods ranged from 2.6% to 41.3%. In this paper, the bioaccessibility of PBDEs in a given food always referred to the mean of the bioaccessibility of 13 individual congeners in this food unless indicated otherwise. The bioaccessibility data were all listed in **Tables 1** and **2**. Generally, the bioaccessibility of PBDEs was the highest in fish (from 32.8% to 41.3%). The bioaccessibility was very high in rice (40.4%). However, the bioaccessibility of PBDEs in meat varied largely; the highest bioaccessibility was 7 times larger than the lowest one. The data in vegetables exhibited even larger variation; the

Table 1. Recoveries (%) of PBDEs in the Supernatants and Pellets after Digestion on Different Foods

	supernatant ($n = 3$)[a]				pellet ($n = 3$)				sum ($n = 3$)[b]			
	crucian carp	pomfret	rice	carrot	crucian carp	pomfret	rice	carrot	crucian carp	pomfret	rice	carrot
BDE17	29.2 ± 5.9	26.8 ± 6.3	39.9 ± 3.2	26.0 ± 2.4	61.2 ± 10.1	69.8 ± 14.7	61.0 ± 2.9	61.8 ± 8.2	90.4 ± 5.3	96.6 ± 10.0	100.9 ± 6.0	87.8 ± 3.8
BDE28	31.5 ± 5.6	29.4 ± 6.4	40.2 ± 2.0	26.4 ± 1.9	60.8 ± 10.7	67.9 ± 13.8	58.9 ± 3.6	61.2 ± 8.8	92.3 ± 8.2	97.3 ± 9.8	99.1 ± 1.6	87.6 ± 6.6
BDE71	29.4 ± 5.5	26.6 ± 5.9	41.7 ± 3.6	26.7 ± 2.2	60.2 ± 11.5	68.2 ± 13.5	58.8 ± 3.6	62.7 ± 9.1	89.6 ± 7.3	94.8 ± 9.7	100.5 ± 7.2	89.4 ± 13.0
BDE47	34.1 ± 3.8	31.6 ± 5.1	38.4 ± 3.5	29.7 ± 1.6	58.8 ± 10.2	65.5 ± 11.8	55.9 ± 3.4	59.7 ± 7.9	92.9 ± 9.6	97.1 ± 8.4	94.3 ± 6.8	89.4 ± 9.5
BDE66	32.4 ± 5.4	31.0 ± 7.4	40.3 ± 2.1	29.6 ± 2.8	58.7 ± 9.4	66.5 ± 10.9	57.0 ± 2.2	59.7 ± 7.8	91.1 ± 4.0	97.5 ± 9.0	97.3 ± 3.0	89.3 ± 9.0
BDE100	32.4 ± 5.4	31.4 ± 5.9	38.7 ± 4.5	30.7 ± 3.0	57.2 ± 11.5	63.7 ± 11.9	54.4 ± 4.4	57.3 ± 7.6	89.6 ± 7.0	95.1 ± 8.9	93.1 ± 8.9	88.0 ± 4.1
BDE99	34.8 ± 4.4	32.8 ± 6.0	40.9 ± 4.0	31.3 ± 2.3	56.6 ± 9.4	64.1 ± 11.2	53.6 ± 4.0	58.2 ± 6.9	91.4 ± 6.7	96.9 ± 8.3	94.5 ± 7.7	89.5 ± 6.8
BDE85	33.5 ± 5.5	32.2 ± 6.4	39.7 ± 3.2	31.4 ± 3.0	57.1 ± 9.0	63.0 ± 8.3	54.4 ± 3.9	58.8 ± 6.0	90.6 ± 5.0	95.2 ± 8.1	94.1 ± 6.7	90.2 ± 5.1
BDE154	34.7 ± 5.7	33.8 ± 6.5	40.6 ± 4.3	32.1 ± 3.4	54.7 ± 9.6	59.7 ± 10.4	52.6 ± 4.3	57.3 ± 6.5	89.4 ± 5.6	93.5 ± 8.8	93.2 ± 8.1	89.4 ± 4.6
BDE153	37.9 ± 4.5	36.0 ± 6.8	42.1 ± 3.9	33.2 ± 3.3	53.3 ± 7.3	58.1 ± 7.5	53.0 ± 4.1	56.5 ± 7.1	91.2 ± 5.8	94.1 ± 8.4	95.1 ± 7.4	89.7 ± 8.5
BDE138	36.1 ± 6.5	35.6 ± 6.8	42.4 ± 6.1	31.8 ± 3.1	53.0 ± 7.7	57.0 ± 6.0	54.3 ± 6.5	58.6 ± 6.1	89.1 ± 5.0	92.6 ± 7.9	96.7 ± 12.0	90.4 ± 6.3
BDE183	41.9 ± 5.6	39.7 ± 6.0	37.9 ± 5.5	28.7 ± 3.2	51.0 ± 7.1	54.8 ± 6.1	52.2 ± 5.5	58.9 ± 8.4	92.9 ± 13.0	94.5 ± 5.4	90.1 ± 11.0	87.6 ± 5.7
BDE190	45.7 ± 8.4	39.0 ± 10.0	42.3 ± 8.9	25.2 ± 4.5	43.9 ± 1.8	51.9 ± 3.9	51.3 ± 5.1	61.7 ± 11.0	89.6 ± 3.8	90.9 ± 8.8	93.6 ± 13.0	86.9 ± 11.0
Mean	34.9 ± 4.7	32.8 ± 4.1	40.4 ± 2.6	29.4 ± 2.6								

[a] The data represented the bioaccessibility of PBDEs. [b] Sum of the recoveries of PBDEs in the supernatant and the pellet.

difference between the highest and lowest bioaccessibility reached 11 times.

The bioavailability of lower brominated congeners (from tri- to heptaBDEs) on rats was reported to range from 25.1% to 76.4% with an average of 57.3% (*23*, *24*). The data were slightly higher than ours, probably being attributed to the fact that PBDEs were administered by dissolving in corn or peanut oil. Oil was expected to increase the bioaccessibility and thus bioavailability as well because PBDEs were highly lipophilic and oil was emulsified by bile in the gastrointestinal tract. In addition, the in vitro digestion method used in the present study was static and absorption was not taken into account. It was possible that PBDE release and thus bioaccessibility were underestimated if the rate of intestinal absorption was faster than release. In spite of the small difference, our data were comparable with the bioavailability data reported, demonstrating that bioaccessibility was a useful substitute to estimate bioavailability.

The Influence of the Constituents of Foods on the Bioaccessibility. It has been proposed that the composition of foods influenced strongly the bioaccessibility of nutrients and metals in foods (*25*−*27*). To explore if there was a correlation between the bioaccessibility of PBDEs and the composition of food, the linear regression analysis was performed between the bioaccessibility and the contents of fat, protein, carbohydrate, and dietary fiber, respectively. The contents of fat, protein, carbohydrate (starch and sugars, not including fiber), and dietary fiber in the foods were listed in **Table 3**.

1. Fat. As shown in **Figure 2A**, a good linear relationship ($R^2 = 0.92$) with a positive slope was observed between the bioaccessibility of PBDEs and the fat content. The correlation was statistically significant ($p < 0.01$). This was not surprising because PBDEs were highly lipophilic molecules. Both PBDEs and fat were freed from food matrices during digestion, and PBDEs were expected to accumulate in the fat. Fat containing PBDEs was then emulsified by bile and was thus brought into the liquid phase, becoming accessible for uptake. Therefore, it was logical that foods with higher fat content exhibited higher bioaccessibility of PBDEs. Similar examples were easily found in nutritional science. For example, it was reported that addition of olive oil to carrot samples during cooking and before the in vitro digestion could significantly improve bioaccessibility of carotenoid (*25*).

The bioaccessibility of individual PBDE congeners in a given food exhibited positive correlations with log K_{OW} (K_{OW} was the partition coefficient of an organic compound in octanol−water system) for the foods with high fat content (> 9%, **Figure 2B**). All correlations were statistically significant ($p < 0.01$). The log K_{OW} value of an organic compound was usually considered to be a

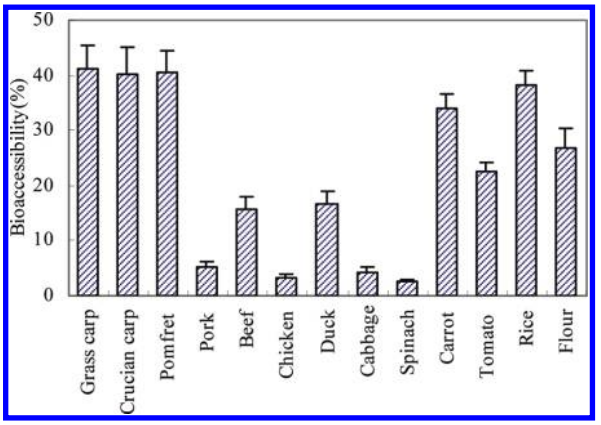

Figure 1. Bioaccessibility of PBDEs in 13 types of foods determined using an in vitro method. The bioaccessibility of PBDEs in a given food was the mean of that of 13 individual PBDE congeners.

Table 3. The Contents (%) of Food Constituents

	fat	protein[a]	hydrocarbon[a]	dietary fiber[a]
grass carp	17.0	70.3		
crucian carp	15.2	79.1		
pomfret	13.9	68.7		
duck	13.4	63.8		
beef	9.1	86.8		
chicken	6.6	72.9		
pork	5.0	71.2		
rice		9.1	88.2	1.1
flour		13.5	82.1	2.1
carrot		8.2	71.8	10.9
tomato		19.6	56.9	9.8
cabbage		28.3	48.3	13.3
spinach		30.4	31.6	17.7
nutrition in digestion solution[b]	1.6	22.8[c]	28.0[c]	2.2[c]

[a] The data were cited from the literature (*28*). [b] The values referred to the contents of the nutrients in a total 18 mL of digestion solution (based on the dried weight), consisting of 12 mL of gastric digestion solution and 6 mL of intestinal solution. [c] Measured values.

Table 2. Bioaccessibility (%) of PBDEs in Food Samples Determined Using the in Vitro Method

		bioaccessibility ($n = 3$)								
	log K_{OW}[a]	grass carp	pork	beef	chicken	duck	cabbage	spinach	tomato	flour
BDE17	5.74	34.2 ± 7.1	5.2 ± 1.2	16.5 ± 3.1	2.6 ± 1.5	18.6 ± 3.3	3.4 ± 0.3	2.3 ± 0.8	21.9 ± 3.7	22.4 ± 5.4
BDE28	5.94	36.5 ± 7.8	5.5 ± 1.2	18.7 ± 3.0	2.5 ± 1.5	19.0 ± 3.1	4.0 ± 0.1	2.6 ± 1.0	23.7 ± 4.8	24.6 ± 2.2
BDE71	6.60	36.0 ± 6.6	4.9 ± 0.7	22.4 ± 2.1	2.9 ± 1.4	20.0 ± 3.2	3.7 ± 0.3	2.7 ± 0.7	22.3 ± 3.0	26.7 ± 3.7
BDE47	6.81	41.5 ± 7.6	6.6 ± 1.7	17.4 ± 0.8	5.0 ± 2.3	19.6 ± 4.4	6.6 ± 0.5	2.7 ± 1.8	24.0 ± 3.3	29.3 ± 5.3
BDE66	6.60	40.0 ± 7.5	5.6 ± 1.0	22.0 ± 2.4	2.8 ± 1.7	23.0 ± 2.2	4.2 ± 0.5	2.8 ± 1.0	24.5 ± 3.2	25.5 ± 3.5
BDE100	7.24	40.9 ± 7.8	6.0 ± 0.9	20.1 ± 2.0	3.4 ± 1.7	21.9 ± 3.8	4.4 ± 0.5	2.7 ± 1.2	23.8 ± 2.9	30.2 ± 2.4
BDE99	7.32	42.2 ± 7.7	6.0 ± 0.8	20.8 ± 1.7	3.4 ± 1.9	23.3 ± 4.1	4.7 ± 0.6	2.9 ± 1.3	24.0 ± 3.6	30.4 ± 3.7
BDE85	7.37	41.3 ± 7.2	5.4 ± 1.0	22.0 ± 2.1	2.8 ± 1.6	23.5 ± 3.4	4.1 ± 0.9	2.7 ± 1.0	23.4 ± 4.3	24.7 ± 3.9
BDE154	7.82	42.2 ± 8.1	5.8 ± 0.9	20.4 ± 1.8	3.3 ± 1.5	22.4 ± 4.1	4.0 ± 0.6	2.9 ± 0.8	22.1 ± 4.5	31.2 ± 1.7
BDE153	7.90	44.1 ± 7.6	5.4 ± 0.8	21.2 ± 1.3	3.4 ± 1.3	24.4 ± 5.0	4.4 ± 0.7	2.8 ± 1.2	23.1 ± 5.0	32.9 ± 1.9
BDE138	7.85	43.9 ± 6.9	5.3 ± 1.1	24.0 ± 1.9	2.9 ± 1.4	24.6 ± 4.3	4.2 ± 0.4	2.9 ± 1.0	22.6 ± 5.9	23.9 ± 0.2
BDE183	8.27	45.9 ± 7.6	3.7 ± 2.7	22.4 ± 4.3	4.4 ± 1.9	22.9 ± 5.2	4.5 ± 1.0	2.5 ± 1.4	20.5 ± 5.6	25.3 ± 0.8
BDE190	8.47	48.4 ± 7.0	4.4 ± 2.8	23.4 ± 2.0	3.5 ± 2.2	25.2 ± 3.1	4.8 ± 0.7	2.2 ± 1.9	19.4 ± 5.1	23.2 ± 1.6
Mean	-	41.3 ± 4.0	5.4 ± 0.8	20.9 ± 2.3	3.3 ± 0.7	22.2 ± 2.2	4.4 ± 0.8	2.6 ± 0.2	22.7 ± 1.5	27.0 ± 3.4

[a] Cited from the literature or calculated using the reported formula (*22*).

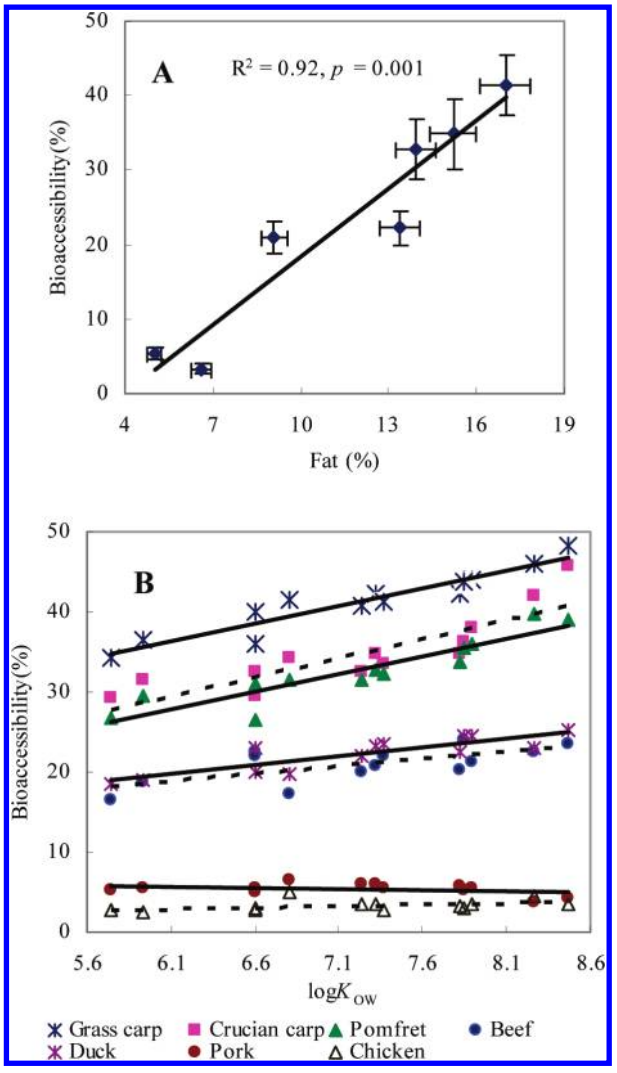

Figure 2. (**A**) Relationship between the bioaccessibility of PBDEs and the fat content in the food samples. (**B**) Relationship between the bioaccessibility and lipophilicity of individual PBDE congeners. K_{OW} is the partition coefficient in octanol−water system. The standard deviations of the constituent contents in foods (i.e., the *x* data) were calculated according to 5% of the contents.

measure on its hydrophobicity/lipophilicity. Thus, the correlations indicated that more lipophilic molecules had higher bioaccessibility in foods with high fat content (>9%). This observation was consistent with the above speculation, i.e., PBDEs accumulated in the fat during digestion. Similar correlations were reported between K_{OW} and the bioaccessibility of organochlorine pesticides in soil samples (*29*). However, it was different for the foods with low fat content (chicken, 6.6%, and pork, 5.0%) in which the bioaccessibility of individual PBDE congeners in a given food was virtually independent of their lipophilicity (**Figure 2B**). It might imply that other factors played important roles and thus partly offset the effect of fat.

Finally, a formula linked the bioaccessibility [*Ba*(%)], K_{OW}, and the fat content was established. The bioaccessibility of individual PBDE congeners and logK_{OW} was fitted using linear regression to obtain $Ba(\%) = k \log K_{OW} + A$ (A was constant) (**Figure 2B**). The obtained slope k was linearly fitted to the fat content (data not shown, $R^2 = 0.90, p < 0.01$). Thus, the formula linked *Ba*(%), K_{OW}, and the fat content (C_{fat}) was obtained as $Ba(\%) = (43C_{fat} - 2.38) \log K_{OW} + A$. When C_{fat} was 5.5%, the slope ($43C_{fat} - 2.38$) was zero. That is, in theory the bioaccessibility of individual PBDE congeners was positively correlated to log K_{OW} only when C_{fat} was more than 5.5%. However, for the foods with the fat content around 5.5%, the slope ($43C_{fat} - 2.38$) was so small that the bioaccessibility was virtually independent of log K_{OW}, consistent with the results observed on chicken (fat content 6.6%) and pork (fat content 5.0%). To observe the positive correlation in experiments, the fat content needed to be significantly higher than the theoretical threshold 5.5%, which was found on beef (9.1%), duck (13.4%), and three types of fish (from 13.9% to 17.0%) as shown in **Figure 2B**. It should be pointed out, however, that the formula was not suitable for the foods with the fat content less than 5.5%, because the bioaccessibility would be negatively correlated to log K_{OW} in this situation, which was not consistent with the data (**Tables 1** and **2**). As discussed below, when the fat content was too low, fat might not be the primary factor to influence the bioaccessibility. Instead, other factors (e.g., protein and carbohydrate) might play predominant roles, leading to different results from the prediction based on the above formula.

2. *Protein*. It appeared that the effect of protein contents on the bioaccessibility of PBDEs showed totally different profiles for animal-based and plant-based foods. The bioaccessibility showed a good negative correlation with the protein contents for plant-based foods ($R^2 = 0.91$, statistically significant, $p < 0.01$, **Figure 3A**), whereas no correlation was observed between the bioaccessibility and the protein contents for animal-based foods. Because plant-based foods selected in the present study had very low fat contents (data not shown), this phenomenon might be caused by fat in foods. PBDEs were highly lipophilic, and fat was expected to play a predominant role on the partition of PBDEs between liquid and solid phases when sufficient fat was present in foods. In other words, the effect of other food constituents on the bioaccessibility was masked when foods contained sufficient fat. Thus, the negative correlation between the bioaccessibility and protein contents might represent the "real" relationship between the two variables, which was possibly attributable to the effect caused by the ionic strength. Protein was hydrolyzed to amino acids during digestion. Most amino acids were soluble and carried positive or negative charge at physiological pH, i.e., they existed as a sort of salt, which increased the ionic strength of the aqueous phase. The increased ionic strength resulted in a lower solubility of the PBDEs (the "salting-out" effect) in the aqueous phase and hence lower bioaccessibility (it should be pointed out that the hypothesis only applied to free PBDEs; the PBDEs in micelles were probably not affected much by the increase in ionic strength). As a result, the bioaccessibility of PBDEs lowered down with increased protein contents.

A further analysis on the data from animal-based foods revealed a tendency that the bioaccessibility of PBDEs lowered down with the increased ratios of protein to fat ($R^2 = 0.79$, statistically significant, $p < 0.01$, **Figure 3B**). There was no correlation between the bioaccessibility and the protein content for the animal-based foods, but a correlation emerged if the protein content on the *x*-axis was substituted using the ratio of protein content to fat content. This was consistent with the above speculation that the effect of fat masked that of protein when sufficient fat was present.

3. *Carbohydrate*. A positive correlation ($R^2 = 0.87$, statistically significant, $p < 0.01$) was observed between the bioaccessibility of PBDEs and the carbohydrate contents (**Figure 4A**). This might be attributed to the fact that carbohydrate is capable of forming micelles, which can increase the partition of hydrophobic molecules in aqueous solution. A similar case, that the carbohydrate was found to self-assemble into micellar clusters that provided an enhanced carrying capacity for hydrophobic

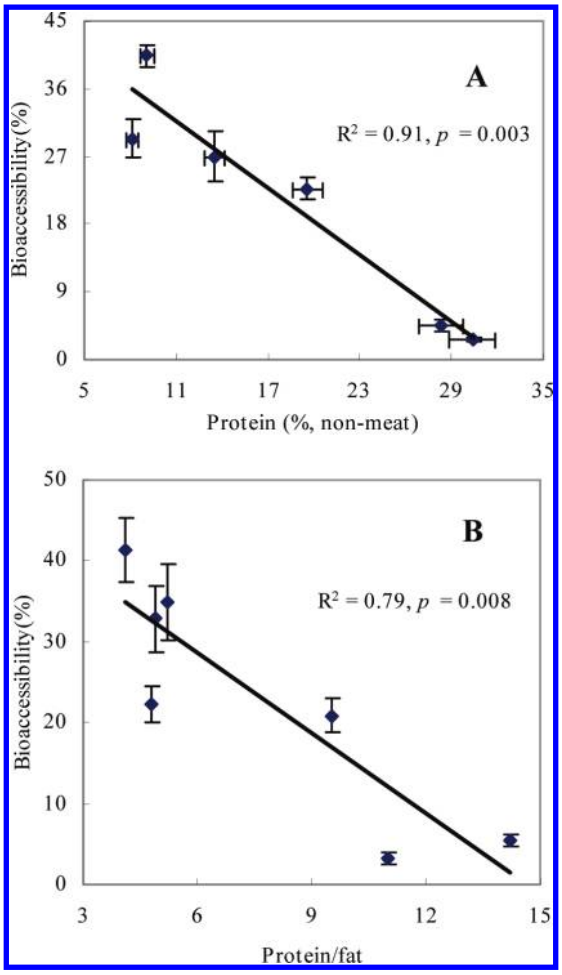

Figure 3. (**A**) Relationship between the bioaccessibility of PBDEs and the protein content in the plant-based foods. (**B**) Relationship between the bioaccessibility of PBDEs and the ratio of protein content to fat content in the animal-based foods. The standard deviations of the constituent contents in foods (i.e., the *x* data) were calculated according to 5% of the contents.

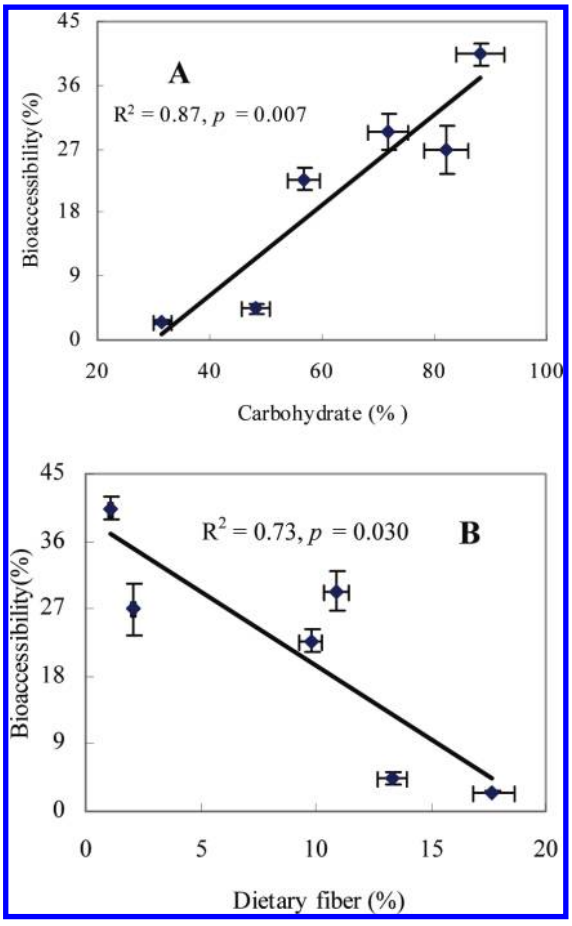

Figure 4. (**A**) Relationship between the bioaccessibility of PBDEs and the carbohydrate content in the food samples. (**B**) Relationship between the bioaccessibility of PBDEs and the dietary fiber content in the food samples. The standard deviations of the constituent contents in foods (i.e., the *x* data) were calculated according to 5% of the contents.

drug molecules, thus enhancing their bioavailability, was reported (*27*).

4. Dietary Fiber. A negative correlation was observed between the bioaccessibility of PBDEs and the content of dietary fiber ($R^2 = 0.73$, statistically significant, $p < 0.05$, **Figure 4B**).

Dietary fiber mainly consists of cellulose, which is insoluble and indigestible in the human gastrointestinal tract. Although it is not hydrophobic, dietary fiber forms the matrix of solid phase that insoluble and indigested substances bind to. Naturally, PBDEs that do not partition into liquid phase will bind to solid phase and finally be excreted with feces, thus becoming inaccessible for uptake. Dietary fiber may also decrease the micelle formation due to binding of bile acids and phospholipids, inhibition of enzyme (e.g., lipase) activity, and increased viscosity and volume of lumen contents (*30*). Thus, dietary fiber is expected to decrease the bioaccessibility of PBDEs. The bioavailability of β-carotene, lycopene, and lutein was reported to be markedly reduced by different kinds of dietary fiber (*31*).

Although the bioaccessibility of PBDEs in foods was observed to exhibit correlations with carbohydrate, protein, and dietary fiber contents, respectively, it must be pointed out that the correlations were not independent. As shown in **Table 3**, from spinach to rice, while carbohydrate content rose, both protein and dietary fiber contents lowered accordingly (the percentage was based on dried foods rather than fresh foods) due to the simple fact that the sum of the three nutrients was close to 100% for foods with little fat. Consequently, when it exhibited a positive correlation with carbohydrate content, the bioaccessibility inevitably exhibited negative correlations with protein and dietary fiber contents. The three variables (protein, carbohydrate, and dietary fiber contents) were simply inseparable, and what we observed was a combined result.

To further elucidate the inseparability of these variables, the effects of protein, carbohydrate, and dietary fiber on the bioaccessibility of PBDEs were able to be integrated into a formula to facilitate the understanding. The entire data set (average bioaccessibility of PBDE congeners and the nutrient contents) for the plant-based foods was fitted using quadratic surface model via Metlab 7.0 to obtain the formula as $Ba(\%) = 145.92 - 212.2C_{\text{protein}} - 96.23C_{\text{carbohydrate}} - 275.94C_{\text{fiber}}$, which produced $Ba(\%)$ values very close to the measured ones (**Figure 5**, $R^2 = 0.99$, $p = 0.000$). At a first glance at the formula, one probably got an impression that the bioaccessibility was negatively correlated to carbohydrate content, inconsistent with the experimental results. But the formula actually showed that the bioaccessibility was *positively* correlated to the carbohydrate content because when the carbohydrate content in food samples increased, both protein and dietary fiber contents decreased accordingly. The final output was higher bioaccessibility because the protein and dietary fiber contents possessed significantly greater coefficients. Similar analyses indicated negative correlations between the

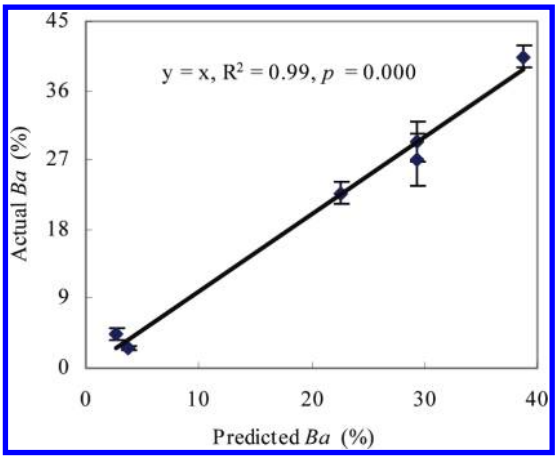

Figure 5. Relationship between the calculated and the measured bioaccessibility of PBDEs in plant-based foods.

bioaccessibility and protein or dietary fiber contents, consistent with the observed results. In addition, the above formula did not include the fat content. In other words, the formula was not suitable for foods with high fat content because the effect of fat on the bioaccessibility would mask those of other nutrients.

It should be mentioned that the digestion solutions used in our method contained small amounts of four nutrients (**Table 3**) because our method was developed to simulate the digestion process *after feeding*. When the contribution of the nutrients in the digestion solutions was taken into account, data analysis indicated that all above findings (positive or negative correlations) did not change although the regression equations changed (but the correlation coefficients R and the p values did not change).

In summary, in the paper we determined the bioaccessibility of PBDEs in 13 types of foods using an in vitro digestion method. The obtained data were comparable with the bioavailability of PBDEs reported in the literature, demonstrating the usability of our method and usefulness of bioaccessibility. The bioaccessibility of PBDEs exhibited positive correlations with the fat and carbohydrate contents in the foods and negative correlations with the protein and dietary fiber contents. The presence of fat and carbohydrate increased the bioaccessibility, which was possibly attributed to the facts that PBDEs were lipophilic and carbohydrate was capable of forming micelles. On the other hand, the influence of protein on the bioaccessibility might stem from the ionic strength effect of amino acids after being digested, which decreased the partition of PBDEs in aqueous phase. The effect of dietary fiber on the bioaccessibility might be the result of physical adsorption of PBDEs on the fiber. Among the four constituents, it seemed that fat played a predominant role in influencing the bioaccessibility. When fat and protein coexisted, only the effect of fat on the bioaccessibility was observed. On the other hand, the influence of protein, carbohydrate, and dietary fiber on the bioaccessibility of PBDEs was inseparable; the observed effects were the combined results of the three constituents. The findings may contribute to a better understanding of bioaccessibility of PBDEs in foods.

ABBREVIATIONS USED

Ba, bioaccessibility; BDE, brominated diphenyl ether; C_{fat}, the fat content in food; GC/MS, gas chromatography/mass spectrometry; K_{OW}, partition coefficient of an organic compound in octanol–water system; PBDEs, polybrominated diphenyl ethers; PCBs, polychlorinated biphenyls; POPs, persistent organic pollutants; QA/QC, quality assurance/quality control; SD, standard deviation.

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LITERATURE CITED

(1) Covaci, A.; Voorspoels, S.; de Boer, J. Determination of brominated flame retardants, with emphasis on polybrominated diphenyl ethers (PBDEs) in environmental and human samples—a review. *Environ. Int*. **2003**, *29* (6), 735–756.
(2) Hites, R. A. Polybrominated diphenyl ethers in the environment and in people: A meta-analysis of concentrations. *Environ. Sci. Technol*. **2004**, *38* (4), 945–956.
(3) UNEP/POPS/COP.4/17. Recommendations of the Persistent Organic Pollutants Review Committee of the Stockholm Convention to amend Annexes A, B or C of the Convention. *Stockholm Convention on Persistent Organic Pollutants*. 4 Feb **2009**.
(4) Costa, L. G.; Giordano, G. Developmental neurotoxicity of polybrominated diphenyl ether (PBDE) flame retardants. *NeuroToxicology* **2007**, *28* (6), 1047–1067.
(5) Darnerud, P. O. Toxic effects of brominated flame retardants in man and in wildlife. *Environ. Int*. **2003**, *29* (6), 841–853.
(6) McDonald, T. A. A perspective on the potential health risks of PBDEs. *Chemosphere* **2002**, *46* (5), 745–755.
(7) Oomen, A. G.; Janssen, P. J. C. M.; Dusseldorp, A.; Noorlander, C. W. *Exposure to Chemicals via House Dust*; RIVM rapport 609021064, **2008**, available at http://www.rivm.nl/en/.
(8) Harrad, S.; Hazrati, S.; Ibarra, C. Concentrations of polychlorinated biphenyls in indoor air and polybrominated diphenyl ethers in indoor air and dust in Birmingham, United Kingdom: Implications for human exposure. *Environ. Sci. Technol*. **2006**, *40* (15), 4633–4638.
(9) Domingo, J. L.; Marti-Cid, R.; Castell, V.; Llobet, J. M. Human exposure to PBDEs through the diet in Catalonia, Spain: temporal trend—a review of recent literature on dietary PBDE intake. *Toxicology* **2008**, *248*, 25–32.
(10) Akutsu, K; Hori, S. Polybrominated diphenyl ether flame retardants in foodstuffs and human milk. *J. Food Hyg. Soc. Jpn*. **2004**, *45*, 175–183.
(11) Oomen, A. G.; Sips, A. J. A. M.; Groten, J. P.; Sijm, D.; Tolls, J. 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*. **2000**, *34* (2), 297–303.
(12) Pouschat, P.; Zagury, G. J. In vitro gastrointestinal bioavailability of arsenic in soils collected near CCA-treated utility poles. *Environ. Sci. Technol*. **2006**, *40* (13), 4317–4323.
(13) Brandon, E. F. A.; Oomen, A. G.; Versantvoort, C. H. M. Human in vitro digestion models: powerful tools to predict maximum oral (relative) bioavailability. *Drug Metab. Rev*. **2006**, *38*, 37–38.
(14) Miller, D. D.; Schricker, B. R.; Rasmussen, R. R.; Van Campen, D. An in vitro method for estimation of iron availability form meals. *Am. J. Clin. Nutr*. **1981**, *34* (10), 2248–2256.
(15) Weber, C. W.; Kohlhepp, E. A.; Idouraine, A.; Ochoat, L. J. Binding capacity of 18 fiber sources for calcium. *J. Agric. Food Chem*. **1993**, *41* (11), 1931–1935.
(16) Oomen, A. G.; Hack, A.; Minekus, M.; Zeijdner, E.; Cornelis, C.; Schoeters, G.; Verstraete, W.; Van de Wiele, T.; Wragg, J.; Rompelberg, C. J. M.; Sips, A. J. A. M.; Van Wijnen, J. H. Comparison of five in vitro digestion models to study the bioaccessibility of soil contaminants. *Environ. Sci. Technol*. **2002**, *36* (15), 3326–3334.
(17) Wragg, J.; Cave, M. R. *In Vitro Methods for the Measurement of the Oral Bioaccessibility of Selected Metals and Metalloids in Soils: A Critical Review*; Technical Report. Environmental Agency, UK, **2002**.
(18) Dean, J. R.; Ma, R. L. Approaches to assess the oral bioaccessibility of persistent organic pollutants: a critical review. *Chemosphere* **2007**, *68* (8), 1399–1407.
(19) Laird, B. D.; Van de Wiele, T. R.; Corriveau, M. C.; Jamieson, H. E.; Parsons, M. B.; Verstraete, W.; Siciliano, S. D. Gastrointestinal microbes increase arsenic bioaccessibility of ingested mine tailings

using the simulator of the human intestinal microbial ecosystem. *Environ. Sci. Technol.* **2007**, *41* (15), 5542–5547.
(20) Yu, Y. X.; Han, S. Y.; Zhang, D. P.; Van de Wiele, T.; Lu, M.; Wang, D. Q.; Yu, Z. Q.; Wu, M. H.; Sheng, G. Y.; Fu, J. M. Factors affecting the bioaccessibility of polybrominated diphenyl ethers in an in vitro digestion model. *J. Agric. Food Chem.* **2009**, *57*, 133–139.
(21) Molly, K.; Vande Woestyne, M.; Verstraete, W. Development of a 5-step multichamber reactor as a simulation of the human intestinal microbial ecosystem. *Appl Microbiol. Biot.* **1993**, *39*, 254–258.
(22) Braekevelt, E.; Tittlemier, S. A.; Tomy, G. T. Direct measurement of octanol−water partition coefficients of some environmentally relevant brominated diphenyl ether congeners. *Chemosphere* **2003**, *51* (7), 563–567.
(23) Huwe, J. K.; Hakk, H.; Smith, D. J.; Diliberto, J. J.; Richardson, V.; Stapleton, H. M.; Birnbaum, L. S. Comparative absorption and bioaccumulation of polybrominated diphenyl ethers following ingestion via dust and oil in male rats. *Environ. Sci. Technol.* **2008**, *42* (7), 2694–2700.
(24) Huwe, J.; Hakk, H.; Lorentzsen, M. Bioavailability and mass balance studies of a commercial pentabromodiphenyl ether mixture in male Sprague−Dawley rats. *Chemosphere* **2007**, *66* (2), 259–266.
(25) Hornero-Méndez, D.; Mínguez-Mosquera, M. I. Bioaccessibility of carotenes from carrots: effect of cooking and addition of oil. *Innovative Food Sci. Emerging Technol.* **2007**, *8* (3), 407–412.
(26) Vitali, D.; Dragojević, I. V.; Šebečić, B. Bioaccessibility of Ca, Mg, Mn, and Cu from whole grain tea-biscuits: Impact of proteins, phytic acid and polyphenols. *Food Chem.* **2008**, *110* (1), 62–68.
(27) Qu, X. Z.; Khutoryanskiy, V. V.; Stewart, A.; Rahman, S.; Papahadjopoulos-Sternberg, B.; Dufes, C.; McCarthy, D.; Wilson, C. G.; Lyons, R.; Carter, K. C.; Schtzlein, A.; Uchegbu, I. F. Carbohydrate-based micelle clusters which enhance hydrophobic drug bioavailability by up to 1 order of magnitude. *Biomacromolecules* **2006**, *7* (12), 3452–3459.
(28) Beijing Epidemic Prevention Station. *Table of Food Composition in Beijing*; China Light Industry Press: Beijing, 1990; Vol. 1, no.1, pp 1−67.
(29) Tao, S.; Lu, Y.; Zhang, D. Y.; Yang, Y. F.; Yang, Y.; Lu, X. X.; Sai, D. J. Assessment of oral bioaccessibility of organochlorine pesticides in soil using in vitro gastrointestinal model. *Environ. Sci. Technol.* **2009**, *43* (12), 4524–4529.
(30) O'Connell, O.; Ryan, L.; O'Sullivan, L.; Aherne-Bruce, S. A.; O'Brien, N. M. Carotenoid micellarization varies greatly between individual and mixed vegetables with or without the addition of fat or fiber. *Int. J. Vitam. Nutr. Res.* **2008**, *78* (4/5), 238–246.
(31) Ried, J.; Linseisen, J.; Hoffmann, J.; Wolfram, G. Some dietary fibers reduce the absorption of carotenoids in women. *J. Nutr.* **1999**, *129* (12), 2170–2176.

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