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SHORT RESEARCH AND DISCUSSION ARTICLE

Oral bioaccessibility and human exposure assessment of cadmium and lead in market vegetables in the Pearl River Delta, South China

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Abstract A systematic investigation into cadmium (Cd) and lead (Pb) concentrations and their oral bioaccessibility in market vegetables in the Pearl River Delta region were carried out to assess their potential health risks to local residents. The average concentrations of Cd and Pb in six species of fresh vegetables varied within 0.09–37.7 and 2.3–43.4 μg kg^{-1}, respectively. Cadmium and Pb bioaccessibility were 35–66 % and 20–51 % in the raw vegetables, respectively, and found to be significantly higher than the cooked vegetables with 34–64 % for Cd and 11–48 % for Pb. The results indicated that Cd bioaccessibility was higher in the gastric phase and Pb bioaccessibility was higher in the small intestinal phase (except for fruit vegetables). Cooking slightly reduced the total concentrations and bioaccessibility of Cd and Pb in all vegetables. The bioaccessible estimated daily intakes of Cd and Pb from vegetables were far below the tolerable limits.

Keywords Cooking · Bioaccessibility · Vegetables · Estimated daily intake · Physiologically based extraction test (PBET) · Heavy metal

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Introduction

Food safety is a growing public concern worldwide, and food consumption has been identified as the major pathway for human exposure to certain environmental contaminants, accounting for >90 % of intake through daily food (Fries 1995). Cadmium (Cd) and lead (Pb) are among the most common toxic heavy metals. The excessive levels of these metals in vegetables are associated with etiology of a number of diseases, especially cardiovascular, kidney, nervous as well as bone diseases (Jarup 2003; WHO 2011). Human beings are encouraged to consume more vegetables and fruits, which are a good source of vitamin C, thiamine, niacin, pyridoxine, folic acid, minerals, and dietary fiber and also beneficial to human nutrition and health (Wargovich 2007). In Asia, especially the developing countries, people consume more vegetables than meat. More specifically, some population groups are highly exposed, especially vegetarians. Therefore, it is of great importance to determine the oral bioavailability of metals (the absorbed dose by human body in reality) in vegetables to assess human health risk rather than the traditional methods based on total concentrations of metals.

Oral bioaccessibility refers to the fraction of contaminant that is released from food matrix into the digestive juice chime and becomes available for intestinal absorption, i.e., entering the blood stream (Oomen et al. 2002). Therefore, this fraction is used as an indicator of maximal oral bioavailability of the contaminant in food. In recent years, several in vitro digestion methods have been developed to estimate the bioaccessibility of contaminants in different foods, such as seafood (Houlbrèque et al. 2011), rice (Signes-Pastor et al. 2012; Zhuang et al. 2016), vegetables (Intawongse and Dean 2008), and food (Lei et al., 2016). In most of the studies on the dietary intake of heavy metals, measurements were performed in uncooked/raw vegetables. It has been evidenced

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that the amount and bioaccessibility of heavy metals in foods can be widely changed by cooking processes (Atta et al. 1997; Sun et al. 2012). To date, only a few studies have investigated the effects of cooking on metal bioaccessibility in contaminated vegetables (Fu and Cui 2013; Pelfrêne et al. 2015), but the information regarding the effects of cooking on the bioaccessibility of heavy metals in market vegetables in the Pearl River Delta (PRD) is rather limited.

The PRD region in South China covers an area of 54,733 km^2, which is one of the most developed regions in China. As reported by the Statistics Bureau of Guangdong Province (SBG 2012), the PRD region is the largest vegetable production base in Guangdong Province and produced more than 11 million tons of vegetables in 2011. About 78.3 × 10^4 tons fresh vegetables were directly exported to Hong Kong, Japan, Korea, Malaysia, and Russia. With the rapid urbanization and industrialization and increasing reliance on fertilizers and agrochemicals, many agricultural soils have been contaminated with heavy metals in the PRD region in recent decades. Hence, the safety of vegetables is a concern of not only the local government but also the international community due to the increasing export and import trade. There has been a lack of comprehensive regional studies on heavy metal bioaccessibility in market vegetables in the PRD region, except for a few reports on vegetables from Hong Kong (Hu et al. 2013) and various food matrices from Guangzhou (Wang et al. 2014).

In this study, we conducted a systemic investigation into several types of vegetables from public markets in the PRD region. The present study was carried out (1) to determine concentrations and bioaccessibility of Cd and Pb in market vegetables in the PRD, (2) to study the influences of cooking on Cd and Pb bioaccessibility, and (3) to calculate daily intake, bioaccessible fraction, and target hazard quotients (THQs) of Cd and Pb to the general public via consumption of market vegetables.

Materials and methods

Sampling, preparation, and chemical analysis

A total of 270 vegetable samples were collected from a range of supermarkets and street markets throughout nine cities of the Pearl River Delta (PRD), South China (Fig. 1). Quintuple samples for each of 6 species of edible vegetables commonly consumed in PRD were randomly acquired for a better estimation of exposure. Around 100 g of vegetables was boiled in approximately 600 mL of doubly deionized water for 8 min (for leafy and fruit vegetables) and 12 min (for root vegetables). Once the vegetable samples had been boiled, the cooking water was separated for further analysis. The dry biomass was also measured after oven-drying at 70 °C for

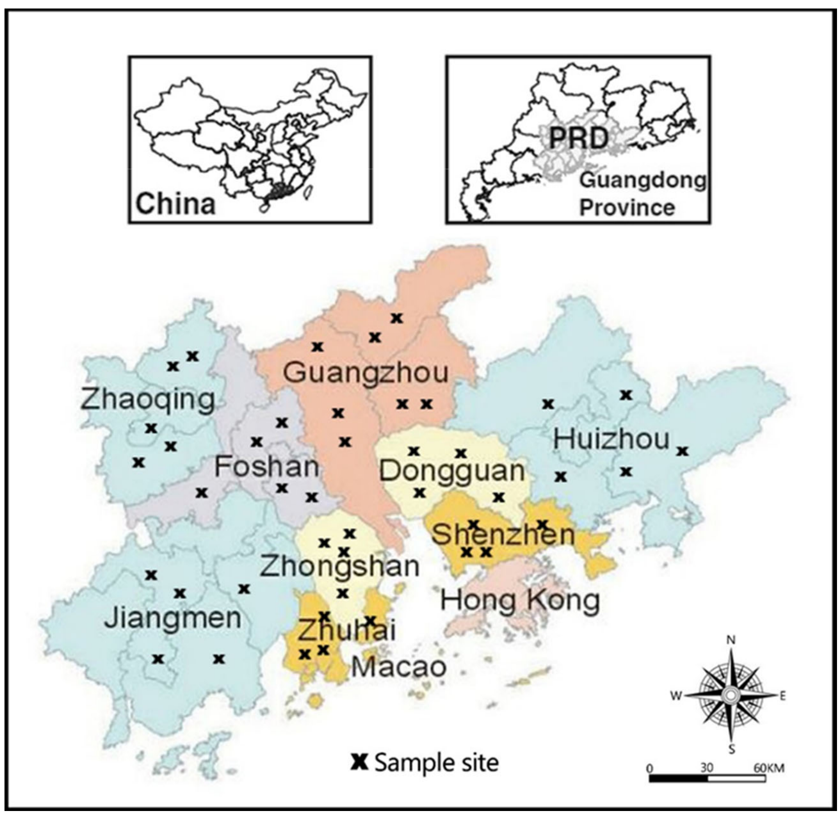

Fig. 1 Locations of sample sites in the Pearl River Delta area, Guangdong, China

48 h. Subsamples (0.5 g) of dried and ground vegetables were digested in a microwave oven (Anton-Paar PE Multiwave 3000) by conc. nitric acid, followed by inductively coupled plasma mass (ICP-MS) spectrometry (Agilent 7700×, Agilent Scientific Technology Ltd., USA) to measure tissue Cd and Pb concentrations. Both blank and standard reference materials (Spinach leaves GBW10015 (GSB-6) and Carrot GBW10047 (GSB-25)) were included for quality assurance. The recovery rates of Cd and Pb ranged from 92 to 108 %. Metal concentrations were expressed on a fresh weight basis by correcting for water content in the samples.

In vitro evaluation of bioaccessibility

Bioaccessibility measurements of Cd and Pb were performed using physiologically based extraction test (PBET) method, which was modified from the procedures described by Ruby et al. (1993) and Intawongse and Dean (2008). Overall, it mimics human digestion, which consists of two phases simulating the digestive processes in the stomach and small intestines. The gastric solution contained 1.25 g L^{-1} pepsin, 0.50 g L^{-1} citric acid, 0.50 g L^{-1} maleic acid, 420 μl L^{-1} DL-lactic acid, and 500 μl L^{-1} acetic acid dissolved in water, and the pH was adjusted to 1.5 with HCl. In the gastrointestinal stage, the amounts of 52.5 mg bile salts and 15 mg pancreatin were added in the sample tube and the pH of the mixture was raised to pH 7 with saturated $NaHCO_3$. For the bioaccessibility experiments, all of the samples were incubated at 37 °C with orbital–horizontal shaking and the obtained supernatant was centrifuged before analysis. The gastric and intestinal bioaccessibility of Cd and Pb in each sample was defined as the ratio of the bioaccessible fraction to the total concentration.

$$\text{Bioaccessibility }(\%) = \frac{\text{Bioaccessible metal concentration}}{\text{Total metal concentration in rice}} \times 100$$

Health risk assessment

In order to assess the potential human exposure to Cd and Pb via consumption of vegetables by the consumers, the estimated daily intake (EDI) and target hazard quotient (THQ) for Cd and Pb were calculated following the method described by Zhuang et al. (2009) assuming consumption of 100 g each vegetable per day.

$$\text{EDI} = \frac{\text{RC} \times \text{BC}}{\text{BW}}$$

$$\text{THQ} = \frac{\text{EF} \times \text{ED} \times \text{EDI}}{\text{RfD} \times \text{AT}} \times 10^{-3}$$

where RC is daily rice consumption (g $person^{-1}$ day^{-1}), BC is bioaccessible concentrations of metals in ingestion, BW is average body weight (60 kg for adults), ED represents the exposure duration (70 years), EF is exposure frequency (365 days per year), AT is average time for non-carcinogens (365 days $year^{-1}$ × number of exposure years, assuming 70 years in this study), 10^{-3} is the unit conversion factor, and RfD represents corresponding oral reference dose (1 and 3.6 μg kg^{-1} day^{-1} for Cd and Pb, respectively), as suggested by USEPA (2007).

Statistical analyses

All statistical analyses were performed using SPSS software (Ver 18.0; SPSS, Chicago, IL, USA) and Excel 2013. All data were reported as the mean or mean with standard deviation (SD) from several samples of each type of vegetable. The means were considered to be significantly different if p values were <0.05.

Results and discussion

Cadmium and Pb concentrations in raw and cooked market vegetables

The concentrations of Cd and Pb in lettuce, spinach, tomato, cucumber, carrot and potato are presented in Fig. 2. The average concentrations in the fresh vegetable samples collected from 9 cities in the PRD region were 37.7, 62.7, 6.6, 0.9, 24.5, and 36.1 μg kg^{-1} for Cd and 13.9, 43.4, 8.4, 2.3, 9.3, and 12.0 μg kg^{-1} for Pb, respectively (Table 1). Leafy vegetables showed relatively high levels of accumulation of Cd and Pb, with the highest in spinach. Moreover, the metal concentrations in cucumber were generally low, which were comparable with those recorded in leafy vegetables. Most selected samples from Dongguan city had higher Cd and Pb concentrations, whereas Pb concentrations in all vegetable samples from Jiangmen were the lowest compared with the other cities. All these Cd and Pb concentrations were acceptable according to the maximum allowable concentration in vegetable established by China (MHPRC 2012), except for 6.9 and 3.4 % the carrot samples which were contaminated by Cd and Pb respectively.

The mean Cd concentrations measured in vegetables were comparable with values published by Cui et al. (2012), ranging from 6 to 40 μg kg^{-1}. The concentrations of Pb in vegetables collected from 9 cities were lower or similar when compared with the values reported by Cui et al. (2012). For example, in the present study, the Cd concentrations in vegetables varied from 2.3 to 43 μg kg^{-1}, while in Cui et al. (2012), the values from 4 cities ranged between 30 and 60 μg kg^{-1}. The

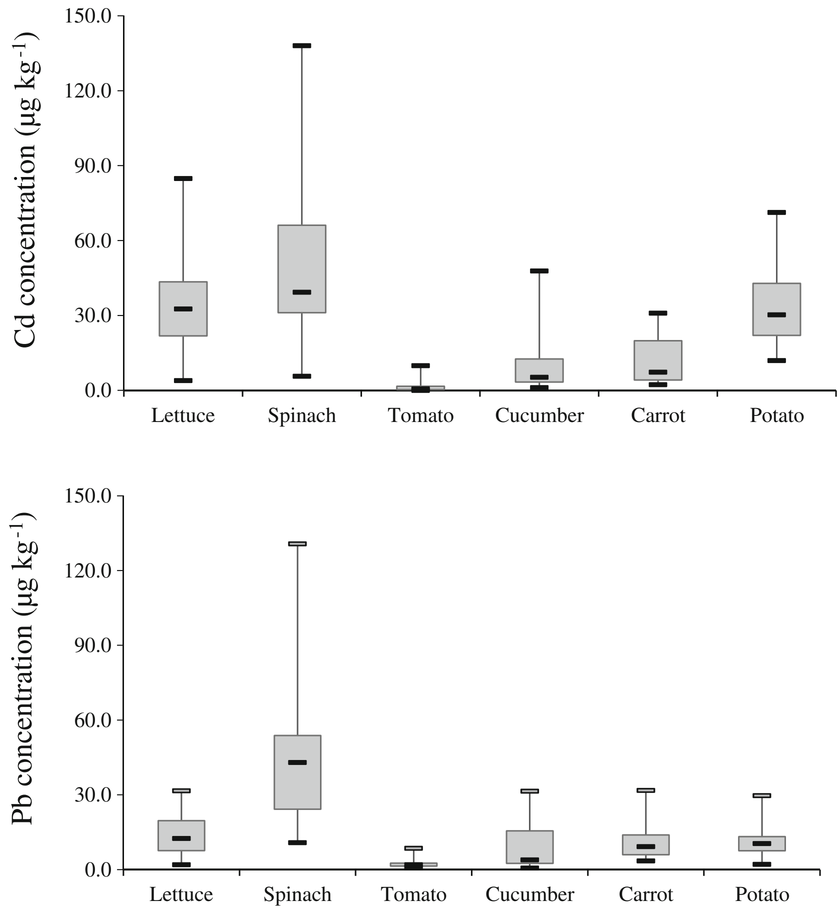

Fig. 2 Cadmium and Pb concentrations (μg kg^{-1}, fresh weight) in the six species of fresh vegetables collected from markets in Pearl River Delta

results showed a strong variation among vegetable species in Cd and Pb concentration, which were consistent with previous findings (Zhuang et al. 2009), with leafy, root, and tuber vegetables possessing a higher risk for Cd accumulation than fruit vegetables (Table 1). Huang et al. (2005) also reported that the Cd and Pb concentrations in ten species of vegetables for Zhongshan and Dongguan cities were 96.2 and 25.7 μg kg^{-1}, with 13.2 % of the vegetable samples exceeding the limit. Regarding regional distribution, Dongguan and Zhongshan cities situated in the central part of PRD economics zone, a large amount of heavy metals were transported continuously into the agricultural soils directly and indirectly (Huang et al. 2005). Especially, the relative low soil pH and the high percentages of DTPA-extractable metals in the agricultural soils resulted in the high metal phytoavailability. So the vegetables sampled from those studied market showed the relatively higher level of heavy metals than other sampling area (cities). On the other hand, the accumulation of metal by vegetable is also dependent on the vegetable species and/or cultivars.

The effects of cooking on Cd and Pb concentrations were evaluated and the results are shown in Fig. 3. The effect of cooking on concentrations of Cd and Pb was different among the vegetable species analyzed. Cooking caused significant changes ($p < 0.05$) of Cd in all vegetables, with a decrease of around 11–24 % in the cooked vegetables in comparison to the corresponding raw samples, except for cucumber. Cooking, meanwhile, decreased Pb concentration by 19 to 41 % in cooked spinach, tomato, and potato, with significant differences ($p < 0.05$) from the raw vegetables. However, cooking resulted in a slight increase in total Pb concentration in cucumber and carrot. Our results were in agreement with several studies, in which cooking decreased Cd concentration in *Agaricus blazei Murill* (Sun et al. 2012), in seafood (Atta et al. 1997) and in rice (Naseri et al. 2014). However, Wang et al. (2014) reported that there were no statistical differences of Cd concentration in microwave-cooked and raw food matrices. Naseri et al. (2014) reported that cooking can reduce the concentration of Pb in rice grains.

Bioaccessibility of Cd and Pb in raw and cooked vegetables

The oral bioaccessible concentrations of Cd and Pb measured in the gastric and gastrointestinal fractions for raw and cooked vegetables defined by the in vitro PBET methods are presented in Fig. 4. Significant differences were found in gastric and also in gastrointestinal extracts between the six vegetables

Table 1 Concentrations of cadmium and lead in fresh vegetables collected from markets in the Pearl River Delta, South China (mean ± SD, μg kg-1, n = 5, on fresh weight basis)

Cities	Cd						Pb					
	Lettuce	Spinach	Tomato	Cucumber	Carrot	Potato	Lettuce	Spinach	Tomato	Cucumber	Carrot	Potato
Zhuhai	36.6 ± 8.12	59.3 ± 25.0	5.65 ± 2.24	0.77 ± 0.44	13.0 ± 8.13	28.0 ± 13.1	10.3 ± 3.98	48.8 ± 5.31	3.65 ± 0.99	1.60 ± 0.39	12.8 ± 5.67	12.4 ± 5.44
Dongguan	43.8 ± 13.5	97.3 ± 38.6	9.98 ± 4.39	1.53 ± 0.72	25.4 ± 4.21	27.6 ± 11.2	24.6 ± 4.11	87.1 ± 26.1	7.09 ± 3.43	4.75 ± 2.29	15.2 ± 6.81	11.1 ± 1.64
Huizhou	32.1 ± 8.63	37.6 ± 1.78	3.11 ± 1.19	0.21 ± 0.13	9.25 ± 5.26	44.1 ± 13.9	19.4 ± 3.95	30.1 ± 12.2	22.4 ± 6.25	2.40 ± 1.05	7.24 ± 2.58	18.2 ± 7.68
Shenzhen	24.8 ± 7.56	46.7 ± 20.1	4.60 ± 1.93	0.39 ± 0.17	3.13 ± 0.47	27.7 ± 3.43	14.9 ± 6.23	49.9 ± 14.1	24.9 ± 8.62	3.04 ± 0.91	7.01 ± 2.02	20.1 ± 7.43
Zhongshan	43.7 ± 12.6	42.0 ± 18.7	6.16 ± 3.23	0.87 ± 0.37	7.10 ± 3.13	30.3 ± 10.3	15.0 ± 6.03	42.9 ± 21.9	3.77 ± 1.11	1.56 ± 0.31	7.57 ± 2.84	10.6 ± 1.69
Fushan	45.5 ± 23.2	55.0 ± 27.5	2.24 ± 0.98	0.44 ± 0.21	12.2 ± 4.81	62.2 ± 0.29	12.6 ± 5.86	26.9 ± 10.3	7.81 ± 3.46	1.60 ± 0.62	7.39 ± 0.83	5.11 ± 1.08
Guangzhou	33.3 ± 14.8	45.8 ± 25.0	7.00 ± 3.01	1.08 ± 0.53	9.69 ± 4.06	26.9 ± 6.49	12.9 ± 4.68	28.3 ± 12.7	2.48 ± 0.82	2.16 ± 0.61	13.6 ± 6.63	13.7 ± 5.47
Jiangmen	44.1 ± 20.3	–	10.1 ± 4.94	1.60 ± 0.68	8.57 ± 3.48	25.3 ± 3.06	5.82 ± 0.11	-	1.88 ± 0.72	1.71 ± 0.63	4.14 ± 0.54	3.96 ± 0.79
Zhaoqing	35.5 ± 14.9	55.5 ± 10.8	10.1 ± 5.10	1.58 ± 0.78	132 ± 40.9	53.2 ± 12.9	9.87 ± 1.71	33.6 ± 7.59	1.18 ± 0.39	2.24 ± 0.23	8.77 ± 1.01	12.7 ± 4.79
Average[a]	37.7 ± 12.1	54.9 ± 23.4	6.6 ± 2.83	0.9 ± 0.38	24.5 ± 20.1	36.1 ± 13.7	13.9 ± 4.72	43.4 ± 16.7	8.4 ± 3.69	2.3 ± 0.94	9.3 ± 6.86	12.0 ± 3.95
Threshold	200	200	50	50	100	100	300	300	100	100	100	100
Samples exceeded the limit (%)	0	0	0	0	6.9	0	0	0	0	0	3.4	0

– no data

[a] The average concentration of each type of market vegetables from nine cities

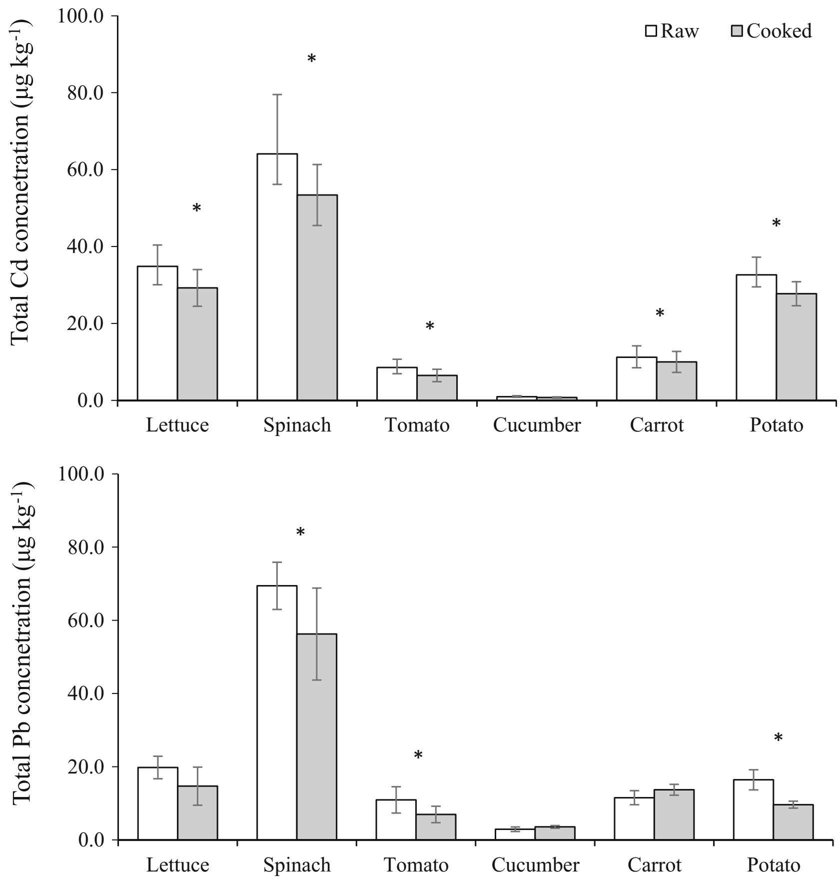

Fig. 3 Cadmium and Pb concentrations in the six species of raw and cooked vegetables collected from markets in the Pearl River Delta. Significant difference between raw and cooked vegetables ($p^* < 0.05$, $p^{**} < 0.01$) determined by a pairwise t test

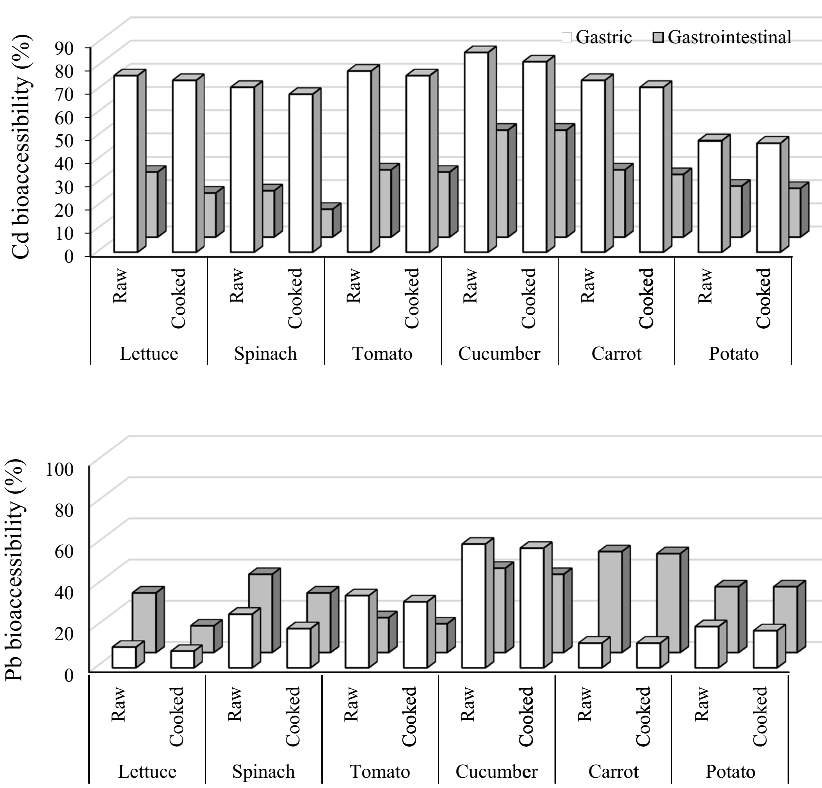

Fig. 4 Bioaccessibility of Cd and Pb (%, in gastric and gastrointestinal phase) in the six species of raw and cooked vegetables

species studied, because of the difference in the concentrations in their raw samples.

The bioaccessibility of Cd varied between 48 and 86 % in the gastric fraction, with the highest in raw cucumber and the lowest in raw potato (Fig. 4). In the gastrointestinal phase, the bioaccessibility of Cd in six vegetables varied within 20–46 %. These results here were higher or similar to Cd bioaccessibility from vegetables reported by Hu et al. (2013) and Fu and Cui (2013). Compared with the values in raw vegetables by Pelfrêne et al. (2015), the bioaccessibility values for the gastric and gastrointestinal fractions were lower, in which Cd bioaccessibility varied from 81 to 89 % in the gastric phase and from 63 to 72 % in the gastrointestinal phase. The higher Cd bioaccessibility percentages observed could be due to the fact that most Cd accumulates in the vacuoles of plant cells, except what is absorbed by the cell wall, so Cd is easily released from plant tissues during in vitro digestion (Hall 2002; Fu and Cui 2013; Pelfrêne et al. 2015). As reported in the literature, the amount of Cd released under gastrointestinal phases is less than that released under gastric phases, because cadmium tends to form bonds with dietary fiber sources, including cellulose, lignin, and pectin, which are all present in vegetables. In addition, Waisberg et al. (2004) found that a higher pH in the digestive tract resulted in lower Cd bioaccessibility after Cd was absorbed by the lettuce at high pH values, and under the typical pH of the gastrointestinal tract (pH = 7), Cd may form insoluble complexes with the phytates in the human diet (Schroder et al. 2003). With respect to the same food matrices, such as carrot and potato, differences in metal bioaccessibility percentages between the present results and previous finding (i.e., Hu et al. 2013 and Pelfrêne et al. 2015) were observed, depending not only on the food matrix but also on the experimental conditions and in vitro digestion methods used.

Lead bioaccessibility in the gastric phase followed the order: cucumber (60 %) > tomato (35 %) > spinach (26 %) > potato (20 %) > carrot (12 %) > lettuce (10 %). In raw vegetables, the Pb bioaccessibility in the gastric phase was lower than that in the small intestinal phase, except for tomato and cucumber. These observations were similar to the results reported by Intawongse and Dean (2008) and Fu and Cui (2013). In the literature, the values ranged from 15.6 % (in the gastric phase) to 23.9 % (in the gastrointestinal phase) for Malabar spinach (Fu and Cui 2013) and varied between approximately 7–27 % (in the gastric phase) and 20–61 % (in the gastrointestinal phase) in vegetables (Intawongse and Dean 2008). The bioaccessible fractions of Pb in market vegetables from Hong Kong under gastric and gastrointestinal phases have also been evaluated by Hu et al. (2013), with the values of 16–42 % and 0.7–26 %, respectively. In the gastric phase, enzymes (such as pepsin) were unable to release most of Pb. The decomposition effects of pancreatin on the integrity of cell wall increased the release of Pb into intestinal juice. The percentages of Pb bioaccessibility were lower than those of Cd, but its total concentrations were equal to those of Cd, except for lettuce and potato. Some types of food matrices exhibited relatively low Pb bioaccessibility percentages, as the analytes may bind with some slightly soluble or insoluble components of the matrix, such as cellulose, which is the most abundant compound in nature and is the most common component in cellular walls of vegetables (Das and Singh 2010).

The cooking process can alter metal bioaccessibility from food matrices. Specifically, heating can accelerate the change of the cellular structure or protein degradation which may affect the bioaccessibility of the remaining components in foods. Cadmium bioaccessibility in all cooked vegetables varied from 47 to 82 % and 12 to 46 % for gastric and gastrointestinal fractions, respectively (Fig. 4). Cooking significantly decreased Cd bioaccessibility of leafy vegetables in gastrointestinal phases and Pb bioaccessibility of leafy vegetables in both gastric and gastrointestinal phases. Fu and Cui (2013) found that cooking significantly decreased the Cd and Pb bioaccessibility in the two leafy vegetables. It was found that Cd bioaccessibility was significantly reduced after cooking *Agaricus blazei murill* (Sun et al. 2012). This may be explained that the heating process destroys tissues thoroughly, leading to more adsorption during the ingestion process and lowering the metal bioaccessibility, because some functional groups such as lysine, methionine,

Table 2 Calculated values of bioaccessible established diary intake (BEDI) and target hazard quotient (BTHQ) for Cd and Pb based on the average bioaccessibility data

Vegetables	Cd			Pb		
	BEDI	% of PTDI	BTHQ	BEDI	% of PTDI	BTHQ
Lettuce	0.029	2.92	2.92E-02	0.002	0.06	6.77E-04
Spinach	0.037	3.66	3.66E-02	0.017	0.43	4.83E-03
Tomato	0.006	0.57	5.68E-03	0.003	0.08	8.90E-04
Cucumber	0.001	0.10	1.00E-03	0.002	0.05	5.20E-04
Carrot	0.020	2.00	2.00E-02	0.005	0.12	1.29E-03
Potato	0.021	2.08	2.08E-02	0.005	0.12	1.39E-03
Total	0.113	11.3	0.113	0.035	0.86	0.010

phenylalanine, histidine, and cystine have an affinity for metal ions (Chou and Shen 2007). Compared to the data for raw vegetables, there was no or slight effect on the Cd and Pb bioaccessibility in cooked tomato, cucumber, carrot, and potato in both phases of digestion. Significant differences on bioaccessible fractions were observed after cooking between the leafy vegetables and non-leafy vegetables, probably due to the differences between the cell wall materials of vegetables and different biological structures and/or ligands in root/tube or fruit vegetables that might impede solubilization of Cd during digestion. Most likely, pectin composition and the presence of other polysaccharides in the cell wall may affect the metal bioaccessibility by interacting differently with the target compounds (Palmero et al. 2013; Pelfrêne et al. 2015).

Human exposure assessment of Cd and Pb in vegetables

The average bioaccessible estimated daily intakes (BEDIs) and target hazard quotients (BTHQs) of Cd and Pb via consumption of vegetables for Pearl River Delta consumers are presented in Table 2. The provisional tolerable daily intake (PTDI) of the FAO/WHO is 0.83 and 3.6 μg kg^{-1} day^{-1} for Cd and Pb, respectively. Based on the bioaccessibility data, assuming consumption of 100 g of each vegetable per day, the average BEDIs of the vegetables followed the order: spinach > lettuce > potato = carrot > tomato > cucumber for both Cd and Pb, which were all far below the corresponding tolerable limits. The BEDI of Cd accounted for 2.9, 3.6, 0.56, 0.1, 2.0, and 2.1 % of the PTDI value via consumption of 100 g lettuce, spinach, tomato, cucumber, carrot, and potato, respectively. The average BEDIs of Pb from six vegetables were less than 0.5 % of the PTDI value. The BTHQs of Cd and Pb from vegetables were all less than 1. In addition, spinach was the major risk contributor for the Pearl River Delta consumers with the highest BTHQ values of Cd and Pb.

The findings regarding the BEDI and BTHQ of Cd and Pb suggest that the consumption of the vegetables from the public markets in the PRD imposes low risks of these two pollutants (Table 2). However, the leafy vegetables are the dominant consumption for local residents in the PRD and Hong Kong, the planting area is about 51.7 % of the total arable area (SBG 2012). Taking these aspects into consideration, to reduce health risks from vegetables, those consumers can adapt their food selection to balance their diet in a healthy manner and avoid excessive consumption of leafy vegetables, such as edible amaranth and spinach. Therefore, in the future, we may need to investigate the possible mechanisms responsible for the decrease of metal bioaccessibility with combination of different food matrices.

Conclusions

The concentrations of Cd and Pb in all the vegetable samples collected from public markets in the Pearl River Delta region were within the Chinese National Standards, but the Cd and Pb levels in 6.9 and 3.4 % of carrot samples exceeded the thresholds. The average bioaccessibility of Cd and Pb from the six vegetables varied within 34–64 % and 11–48 %, respectively. Leafy vegetables possessed higher health risks compared to fruit and root/tube vegetables. The BEDIs of Cd and Pb via consumption of market vegetables for the local residents were all far below the tolerable limits, while the TBTHQ was 0.11 for Cd and 0.01 for Pb through consumption of 100 g vegetables, with Cd in spinach as the major risk contributor.

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References

Atta MB, El-Sebaie LA, Noaman MA, Kassab HE (1997) The effect of cooking on the content of heavy metals in fish (*Tilapia nilotica*. Food Chem 58:1–4

Chou KC, Shen HB (2007) Recent progress in protein subcellular location prediction. Anal Biochem 370:1–16

Cui XF, Li SY, Ding XD, Wang DH, Liao XR, Xue SL, Wang RP (2012) Contents of heavy metals in soil and vegetables at typical vegetable plot in the Pearl River Delta. Ecol Environ Sci 21:130–135

Das H, Singh SK (2010) Useful byproducts from cellulosic wastes of agriculture and food industry-a critical appraisal. Crit Rev Food Sci Nutr 44:77–89

Fries GF (1995) A review of the significance of animal food products as potential pathways of human exposures to dioxins. J Anim Sci 73:1639–1650

Fu J, Cui YS (2013) In vitro digestion/Caco-2 cell model to estimate cadmium and lead bioaccessibility/bioavailability in two vegetables: the influence of cooking and additives. Food Chem Toxicol 59:215–221

Hall JL (2002) Cellular mechanisms for heavy metal detoxification and tolerance. J Exp Bot 53:1–11

Houlbrèque F, Hervé-Fernández P, Teyssié J, Oberhaënsli F, Boisson F, Jeffree R (2011) Cooking makes cadmium contained in Chilean mussels less bioaccessible to humans. Food Chem 126:917–921

Hu J, Wu F, Wu S, Cao Z, Lin X, Wong MH (2013) Bioaccessibility, dietary exposure and human risk assessment of heavy metals from market vegetables in Hong Kong revealed with an in vitro gastrointestinal model. Chemosphere 91:455–461

Huang Y, Guo QR, Ren H, Yang GY, Wan HF, Luo W (2005) Investigation of heavy metal pollution in vegetables in the Pearl River delta: a case study of Zhongshan and Dongguan. Ecol Environ Sci 14:559–561

Intawongse M, Dean JR (2008) Use of the physiologically-based extraction test to assess the oral bioaccessibility of metals in vegetable plants grown in contaminated soil. Environ Pollut 152:60–72

Jarup L (2003) Hazards of heavy metal contamination. Brit. Med Bull 68:167–182

MHPRC (Ministry of Health of the People's Republic of China) (2012) National Food Safe Standard. Maximum levels of contaminants in foods (GB 2762–2012). Beijing: Ministry of Health of the People's Republic of China

Naseri M, Rahmanikhah Z, Beiygloo V, Ranjbar S (2014) Effects of two cooking methods on the concentrations of some heavy metals (cadmium, lead, chromium, nickel and cobalt) in some rice brands available in Iranian Market. J Chem Health Risks 4:65–72

Oomen AG, Hack A, Minekus M, Zeijdner E, Cornelis C, Schoeters G, Verstraete W, Van de Wiele T, Wragg J, Rompelberg CJM, Sips AJAM, Van Wijnen JH (2002) Comparison of five in vitro digestion models to study the bioaccessibility of soil contaminants. Environ Sci Technol 36:3326–3334

Palmero P, Lemmens L, Ribas-Agusti A, Sosa C, Met K, de Dieu Umunoti J, Hendrickx M, Van Loey A (2013) Novel targeted approach to better understand how natural structural barriers govern carotenoids in vitro bioaccessibility in vegetable-based systems. Food Chem 141:2036–2043

Pelfrêne A, Waterlot C, Guerin A, Proix N, Richard A, Douay F (2015) Use of an in vitro digestion method to estimate human bioaccessibility of Cd in vegetables grown in smelter-impacted soils: the influence of cooking. Environ Geochem Health 37:767–778

Ruby MV, Davis A, Link TE, Schoof R, Chaney RL, Freeman GB, Bergstrom P (1993) Development of an in vitro screening test to evaluate the in vivo bioaccessibility of ingested mine-waste lead. Environ Sci Technol 27:2870–2877

SBG (Statistics Bureau of Guangdong Province) (2012) Guangdong Statistical Yearbook 2012. Beijing: Statistics Bureau of Guangdong Province, China Statistic Press. http://www.gdstats.gov.cn/tjfx/t20110104_83252.htm accessed in 20/11/2015

Schroder JL, Basta NT, Si J, Casteel SW, Evans T, Payton M (2003) In vitro gastrointestinal method to estimate relative bioavailable cadmium in contaminated soil. Environ Sci Technol 37:1365–1370

Signes-Pastor AJ, Al-Rmalli SW, Jenkins RO, Carbonell-Barrachina ÁA, Haris PI (2012) Arsenic bioaccessibility in cooked rice as affected by arsenic in cooking water. J Food Sci 77:T201–T206

Sun LP, Liu GX, Yang MZZ, Zhuang YL (2012) Bioaccessibility of cadmium in fresh and cooked *Agaricus blazei Murill* assessed by in vitro biomimetic digestion system. Food Chem Toxicol 50:1729–1733

USEPA (2007) Integrated Risk Information System-database. Philadelphia PA; Washington, DC

Waisberg M, Black WD, Waisberg CM, Hale B (2004) The effect of pH, time and dietary source of cadmium on the bioaccessibility and adsorption of cadmium to/from lettuce (*Lactuca sativa* L. *Cv. Ostinata)*. Food Chem Toxicol 42:835–842

Wang C, Duan HY, Teng JW (2014) Assessment of microwave cooking on the bioaccessibility of cadmium from various food matrices using an in vitro digestion model. Biol Trace Elem Res 160:276–284

Wargovich MJ (2007) Anticancer properties of fruits and vegetables. Hortscience 35:573–575

WHO (World Health Organization) (2011) Evaluation of certain food additives and contaminants. Seventy-third Report of the Joint FAO/WHO Expert Committee on Food Additives. Technical Report Series. WHO, Geneva

Zhuang P, McBride MB, Xia HP, Li NY, Li ZA (2009) Health risk from heavy metals via consumption of food crops in the vicinity of Dabaoshan mine, South China. Sci Total Environ 407:1551–1561

Zhuang P, Zhang CS, Li YW, Zou B, Mo H, Wu KJ, Wu JT, Li ZA (2016) Assessment of influences of cooking on cadmium and arsenic bioaccessibility in rice, using an in vitro physiologically-based extraction test. Food Chem 213:206–214