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Source attributions of heavy metals in rice plant along highway in Eastern China

Jinfei Feng[1,**], Yinxi Wang[2,**], Jian Zhao[1], Liqun Zhu[1],
Xinmin Bian[1], Weijian Zhang[1,3,*]

1. Institute of Applied Ecology, Nanjing Agricultural University, Nanjing 210095, China. E-mail: jffeng22@163.com
2. Center of Modern Analysis, Nanjing University, Nanjing 210093, China. E-mail: yxwang@nju.edu.cn
3. Institute of Crop Science, Chinese Academy of Agricultural Sciences/Key Laboratory of Crop Ecology,
Physiology & Production, Ministry of Agriculture, Beijing 100081, China

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Abstract

Air and soil pollution from traffic has been considered as a critical issue to crop production and food safety, however, few efforts have been paid on distinguish the source origin of traffic-related contaminants in rice plant along highway. Therefore, we investigated metals (Pb, Cd, Cr, Zn and Cu) concentrations and stable Pb isotope ratios in rice plants exposed and unexposed to highway traffic pollution in Eastern China in 2008. Significant differences in metals concentrations between the exposed and unexposed plants existed in leaf for Pb, Cd and Zn, in stem only for Zn, and in grain for Pb and Cd. About 46% of Pb and 41% of Cd in the grain were attributed to the foliar uptake from atmosphere, and there were no obvious contribution of atmosphere to the accumulations of Cr, Zn and Cu in grain. Except for Zn, all of the heavy metals in stem were attributed to the root uptake from soil, although significant accumulations of Pb and Cd from atmosphere existed in leaf. This indicated that different processes existed in the subsequent translocation of foliar-absorbed heavy metals between rice organs. The distinct separation of stable Pb isotope ratios among rice grain, leaf, stem, soil and vehicle exhaust further provided evidences on the different pathways of heavy metal accumulation in rice plant. These results suggested that further more attentions should be paid to the atmospheric deposition of heavy metals from traffic emission when plan crop layout for food safety along highway.

Key words: source origin; traffic; heavy metal; stable Pb isotope; rice plant

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Introduction

Increasing evidences have showed that highway is a main source of heavy metal pollution for crop plants (e.g., vegetables and cereal crops) growing along the roadside field (Othman et al., 1997; Nabulo et al., 2006; Kalavrouziotis et al., 2007; Vissikirsky et al., 2008). Recently, human exposure to traffic-emitted heavy metals (e.g., Pb, Cd, Cr, Zn and Cu) via the food chain is a critical concern in the primary cropping regions with intensive highway traffic in Asian (Zhang et al., 1996; Yang et al., 2009). Rice is the most important staple crop in China, and the East China Plain is one of the key areas of rice cropping (about 9688 ha) with the most highway density in China (Bureau of Statistics of China, 2009). With rapid economy development, the traffic mileage has increased about 15,235 km from 1997 to date in this region (Bureau of Statistics of China, 2009). Heavy metals from highway traffic have significantly affected rice grain quality and food safety in this region (Yang et al., 2009). Moreover, due to the great pressures of insufficient arable land and quick increments of population, rice should be still closely sown along the roadsides of these highways. Thus, to learn the specific sources of heavy metals in rice plant will help to plan rice cropping layout along the roadsides for public health.

Traffic-emitted heavy metals may be accumulated by roadside crops from soil or atmosphere. For example, previous evidences from roadside leafy vegetable (Nabulo et al., 2006) and grass (Tjell et al., 1979) suggested atmospheric deposition to leaves rather than uptake from soils was the main source of Pb. Meanwhile, heavy metals derived from atmosphere via leaf could subsequently transport to unexposed organs (Chamberlain, 1983; Harrison and Chirgawi, 1989b; Greger et al., 1993). By using growth cabinets with filtered air, Harrison and Chirgawi (1989a) found that the relative atmosphere contribution of heavy metals to the storage root of different vegetables (e.g.,

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* Corresponding author. E-mail: zhangweij@caas.net.cn
** The authors contributed equally to this work.

radish, turnip and carrot) were 32%–88% for Pb, 4%–47% for Cd, 18%–72% for Cr, and 2%–39% for Zn. Haar (1970) reported that Pb in roadside rice grain was mainly derived from the soil; the contribution of foliar uptake was not obvious. However, more recent evidence reported that cereal grain could accumulate substantial amounts of Pb through foliar absorption (Bi et al., 2009; Colle et al., 2009). Bi et al. (2009) reported that maize grains could accumulate substantial amounts of Pb via foliar absorption by examining the Pb isotopic compositions of different organs. Besides Pb, considerable amounts of Cd, Cr, Zn and Cu were also observed in the roadside atmospheric particles, which may accumulate in crop plant along roadsides (Hewitt and Rashed, 1991; Johansson et al., 2009). Previous field investigations indicated that concentrations of these metals in roadside plants were related to roadside deposition (Lagerwerff, 1970; Flanagan et al., 1980; Sharma et al., 2008), however, the specific accumulation pathways of these metals, from soil or atmosphere, still remain poorly understand.

To our knowledge, the majority works carried out near busy roads about heavy metal pollution on rice plant has been concerned with root uptake rather than foliar uptake (Lin et al., 2002; Yang et al., 2009), little was known about the contribution of foliar uptake to the concentrations of traffic-related metals in rice grain. Therefore, we conducted field investigation and a pot experiment with the same soil and rice seedlings in the roadside paddy locations to: (1) differentiate the soil and air sources of traffic-related heavy metals (Pb, Cd, Cr, Zn, and Cu) in rice plants grown in roadside field, (2) estimate the relative contributions of foliar uptake and root uptake to the heavy metals concentrations in different organs (leaf, stem and grain) of rice plants. The Pb isotope compositions in roadside soil and rice plants were also analyzed to further identify the source origin and accumulation pathway of Pb in rice plants along roadsides.

1 Materials and methods

1.1 Field investigation

Field investigation was conducted at a section (31°57′47.08″N, 119°41′48.65″E; with a traffic of 62,000 vehicles per day) of Shanghai-Nanjing Highway which is mainly located in Jiangsu Province, China. The field is surrounded by paddy fields. Mean annual temperature and precipitation in this area were approximately 15.3°C and 1106.5 mm, respectively. At rice seedling stage, three replicated paddy soil and seedling samples were collected in the roadside paddy field at the distances of 10, 40, 100, 200, 300, and 450 m perpendicular from the road edge. The soil samples were separately put into pots and the seedlings were transplanted into the pots (25 cm in diameter and 30 cm in height, four plants per pot). Meanwhile, the sampling locations in each site were recorded with a Trimble GeoExplorer CE (GeoXT, Trimble, USA) for field investigation of plant growing *in situ* and plant and soil sampling at harvest. The irrigation scheduling and fertilizer application were recorded during the rice growing season (from July 8 to October 14, 2008).

1.2 Pot experiment

All the soil and seedling samples in pots were carried to the greenhouse in Nanjing Agricultural University, Nanjing, Jiangsu Province, China, which is located in the same climatic area with the sampling fields. Compared with the rice plants in roadside fields, pot plants were grown in the clean greenhouse without atmospheric heavy metal pollution from vehicle emission under natural sunlight at ambient temperatures (18–38°C). Similar agronomic practices, such as fertilization and irrigation regimes, were conducted according to the alongside field. The plants were harvested at the same time as the roadside rice after growing for 97 days.

1.3 Sample preparation and analysis

The leaf, stem and grain (polished rice) samples were washed with deionized water, and then over-dried at 80°C for 24 hr. The dried samples were ground separately in stainless steel mill to fine powder prior to chemical analysis. Subsamples of the dry plant (1 g) were digested in a high-walled beaker with 10 mL mixture solution of HNO_3-$HClO_4$ (4:1, V/V) (Lu, 2000). Each soil sample was air-dried, sieved and stored at room temperature. The digest of the soil samples were carried out in a PTEE crucible. Approximately 0.5 g of each soil samples was dissolved in a mixture solution of HNO_3-$HCLO_4$-HF (1:1:2, $V/V/V$) (Lu, 2000). The concentrations of Pb, Cd, Cr, Zn and Cu were determined using flame or graphite furnace atomic absorption spectrometry (AA220, Varian, USA). Certified reference materials GBW 10010 Chinese rice flour and GBW 07402 and GBW 07404 Chinese soils were used to validate the analysis. The recoveries of Pb, Cd, Cr, Zn and Cu in standard reference materials were in the range of 85%–113%.

Pb isotopic ratios were determined using ICP-MS (SCIEX Elan 9000, Perkin-Elmer, USA) as already described in the literature (Bindler et al., 2004; Hu and Ding, 2009). Precision and accuracy were verified using standard reference material from the National Institute of Standards and Technology (SRM 981 common lead isotopic material). The analytical precision of lead isotope ratio in SRM 981 was 0.21% for $^{206}Pb/^{207}Pb$, and 0.33% for $^{208}Pb/^{206}Pb$.

1.4 Data analysis

Statistical analysis was analyzed with SPSS statistical software (V# 11.5). Contribution coefficient was presented to quantify the relative contributions of root and foliar uptake to heavy metals in rice plants. The contribution coefficients of root uptake (CCRU, %) and foliar uptake (CCFU, %) were calculated as follows:

$$\mathrm{CCFU} = \frac{C_{\text{exposed rice}} - C_{\text{unexposed rice}}}{C_{\text{exposed rice}}} \times 100\% \quad (1)$$

$$\mathrm{CCRU} = (100 - \mathrm{CCFU})/100 \times 100\% \quad (2)$$

where, $C_{\text{exposed rice}}$ (μg/g) is the heavy metal content in the rice plant grown in the roadside paddy field; $C_{\text{unexposed rice}}$

(μg/g) is the heavy metal content in the rice plant grown in the pot. If the heavy metal contents in roadside rice plants are not significant higher than that in potted rice plants, set CCFU = 0%, and CCRU = 100%.

2 Results

2.1 Heavy metals contents in rice plant

Figure 1 shows the concentrations of Pb, Cd, Zn, Cr, and Cu in different aboveground organs of rice obtained from the field (exposed rice) and greenhouse (unexposed rice). In leaves, the concentrations of Pb, Cd and Zn were significantly higher in exposed rice than in unexposed rice. While for Cr and Cu, there was no significant difference between the exposed and unexposed plants. These results indicated that Pb, Cd and Zn in leaves of exposed rice plants should be partly derived from the atmosphere via foliar uptake, while Cr and Cu were mainly from the soil via root uptake. The concentrations of these five metals in leaves of exposed rice firstly increased to the highest value at 50 or 100 m and then decreased gradually with the increased distance from the road edge.

In stems, the concentration of Zn was significantly higher in exposed rice than in unexposed rice within 200 m from the highway edge, while no significant differences were found on Pb, Cd, Cr and Cu (Fig. 1), indicating that the Zn contents in stem of exposed rice should be partly from the atmosphere through foliar absorption, while the contents of Pb, Cd, Cr and Cu in the stem of exposed rice were mainly from the soil. The concentrations of Pb, Cd, Zn and Cu in the stem of exposed rice also firstly increased to the highest value at 50 or 100 m and then decreased gradually except Cr.

In grains, the concentrations of Pb and Cd were significant higher in the exposed rice than the unexposed rice within 300 and 200 m, but there was no significant difference for Zn, Cr and Cu (Fig. 1), indicating that Pb and Cd in the grain of exposed rice should be partly from the atmosphere, while Zn, Cr, and Cu were mainly from the soil. The concentrations of Pb, Cd, and Cr in the grain of exposed rice increased firstly to the highest value at 50 or

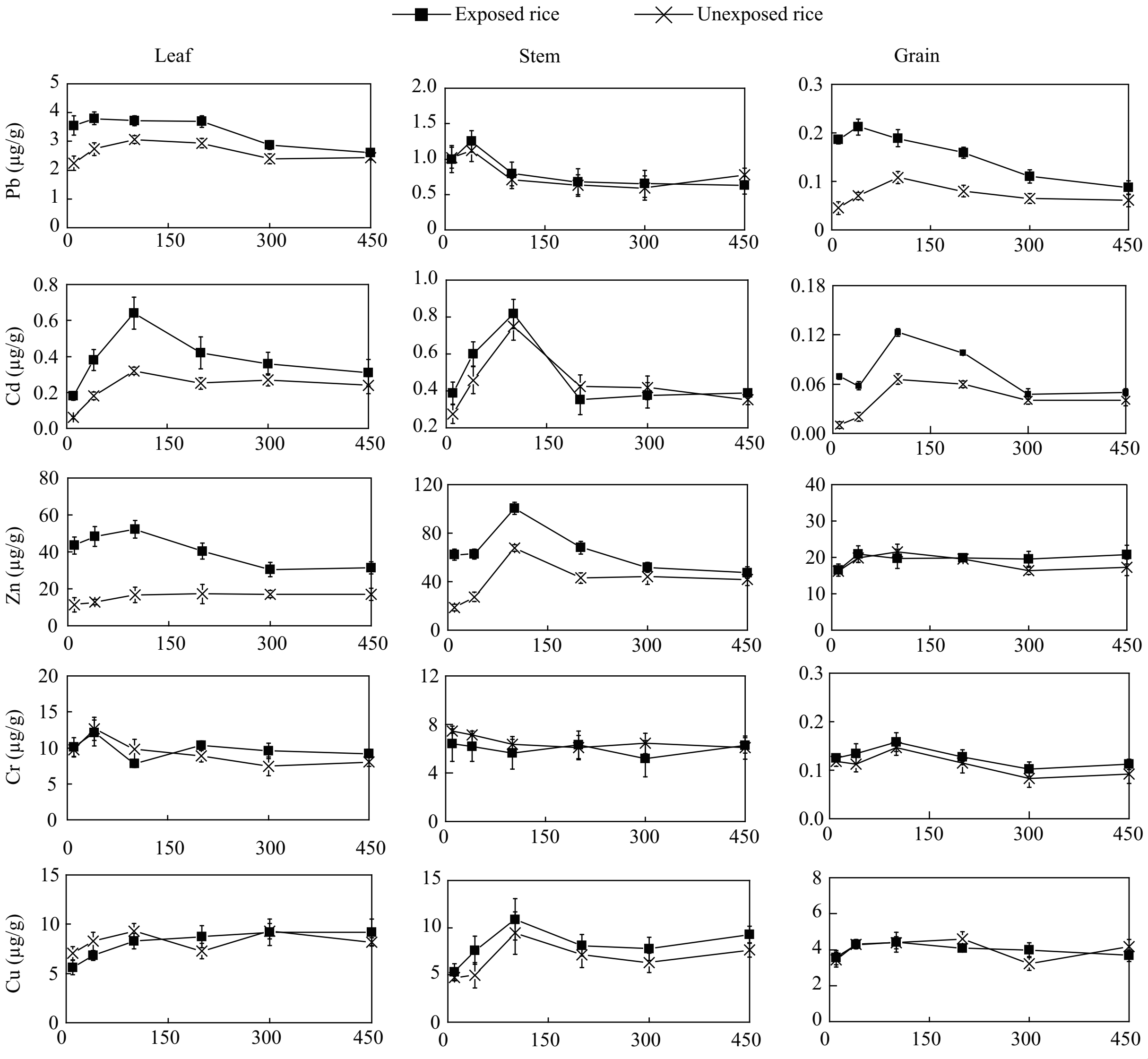

Fig. 1 Concentrations of Pb, Cd, Cr, Zn and Cu in leaf, grain and stem of exposed and unexposed rice plants.

100 m and then decreased gradually, while Zn and Cu kept on relative constant levels with increasing distance from the highway.

2.2 Contributions of foliar and root uptake to heavy metals in roadside rice

The contribution coefficients of foliar uptake and root uptake for different heavy metals in exposed rice plants are presented in Table 1. In leaves, 20% of Pb, 35% of Cd, and 60% of Zn were derived from the atmosphere via leaf absorption, while Cr and Cu were mainly from the soil via root absorption and translocation. In stems, root absorption and translocation was the dominant pathway for the accumulation of these metals except Zn, where 49% of which in the stem attributed to the leaf absorption and subsequent translocation. As in grain, 46% of Pb and 41% of Cd were attributed to the foliar uptake, while Cr, Zn and Cu were attributed to the root uptake.

Table 1 Average CCFU and CCRU of heavy metals in exposed rice plants

Rice organ	Heavy metal	CCFU (%)	CCRU (%)
Leaf	Pb	20	80
	Cd	35	65
	Cr	0	100
	Zn	60	40
	Cu	0	100
Stem	Pb	0	100
	Cd	0	100
	Cr	0	100
	Zn	49	51
	Cu	0	100
Grain	Pb	46	54
	Cd	41	59
	Cr	0	100
	Zn	0	100
	Cu	0	100

CCFU: contribution coefficient of foliar uptake; CCRU: contribution coefficient of root uptake.

The CCFU and CCRU of Pb, Cd and Zn in different rice organs exhibited wide ranges and depended on the distance from the road edge (Fig. 2). Along with the increased distance from the highway edge, the CCFUs of Pb and Cd in leaf and grain, and Zn in leaf and stem reached the highest value at 10 m from the road edge and then decreased. The contributions of foliar uptake for Pb, Cd and Zn in rice plants were observed within different distances from the highway. For Cd and Zn, the impact of foliar uptake existed within 200 m, and for Pb, within 300 m.

2.3 Lead isotope compositions in roadside soil and rice plant

The lead isotope compositions of roadside surface soil and rice samples are presented in Fig. 3. There was a clear distinction in Pb isotope ratios between soils and

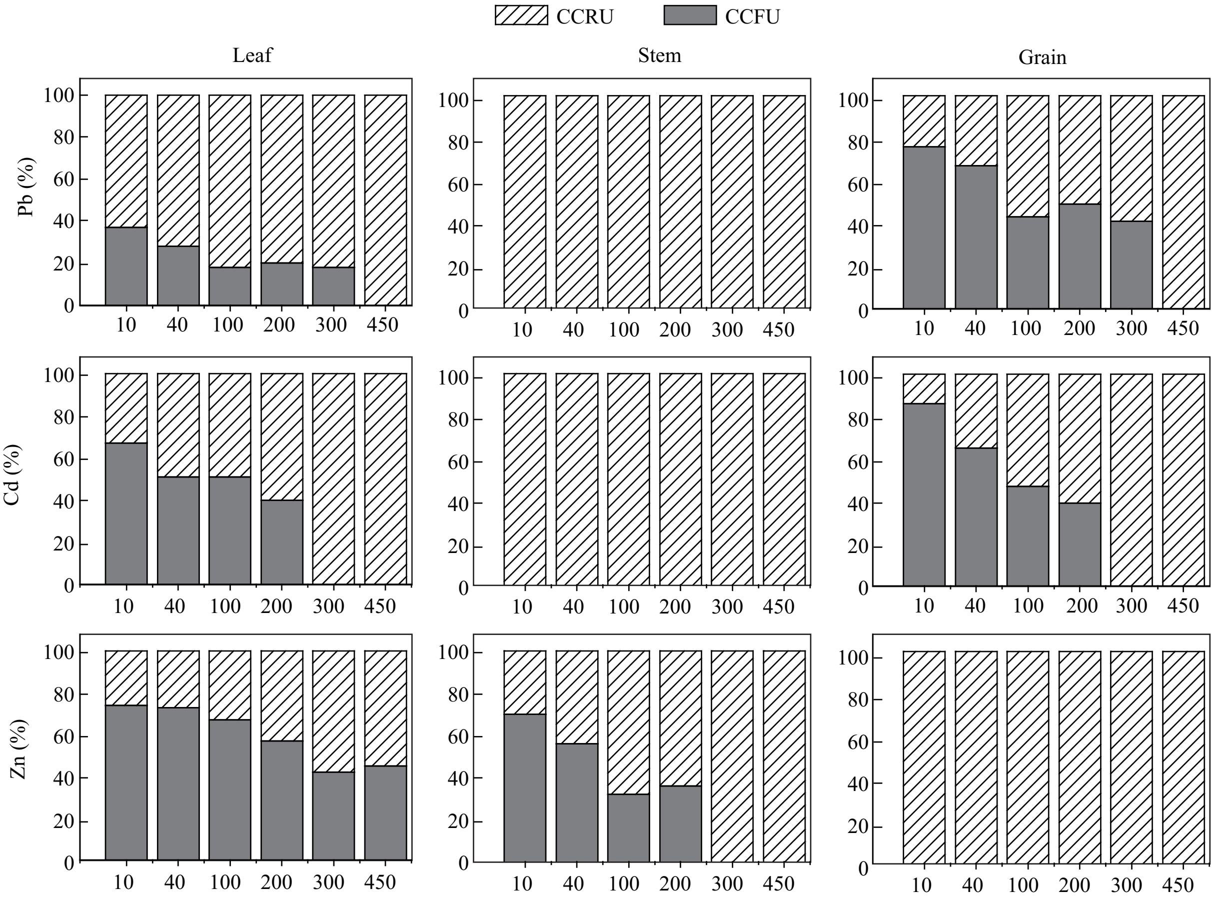

Fig. 2 Changes of CCFU (contribution coefficient of foliar uptake) and CCRU (contribution coefficient of root uptake) of Pb, Cd and Zn in exposed rice plants along with the field sampling distances from the highway edge.

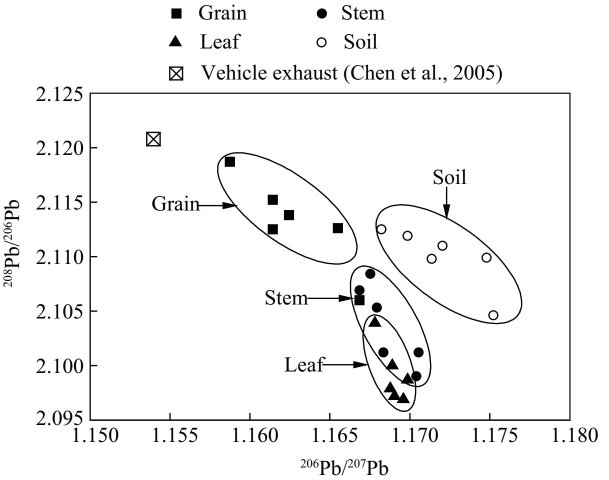

Fig. 3 Lead isotope ratio plot in the surface soil and different organs of exposed rice plant.

plants. Lead isotope ratios in rice grain distributed between the soil and vehicle exhaust, indicating that Pb stored in roadside rice grain may originate both from soil and atmospheric deposition from vehicle exhaust. The ^{206}Pb/^{207}Pb ratios in stem and leaf were between that in vehicle exhaust and in soil, suggesting mixing source of Pb in rice leaf and stem. While the ^{208}Pb/^{206}Pb ratios were lower than soil and vehicle exhaust. Relatively, the lead isotope ratios of rice stem distributed more closely to the soil.

3 Discussion

3.1 Rice leaf absorption of heavy metals along highway

Rice plant exhibited various uptake characteristics of traffic-related heavy metals (Fig. 1). Considerable amount of Pb, Cd and Zn were derived from atmosphere via foliar uptake, while Cr and Cu were mainly from root uptake. This can be explained by the dust particle size and mobility of different metals (Colle et al., 2009). Traffic emitted Pb and Cd were mainly in fine particles, Zn and Cu in medium particles and Cr in coarse particles, and Zn is more mobile than Cu (Zereini et al., 2005). Additionally, previous studies reported that the ratios of exchangeable contents of these five metals in roadside particulate were in the order of Zn (30.5%–31.8%) > Cd (26.4%–29.4) > Pb (1.1%–1.7%) > Cu (0.63%–1.02%) > Cr (0.24%–0.96%) (Xiang et al., 2010). It can be concluded that Pb, Cd and Zn in roadside particles were presented greater leaf penetration ability than Cr and Cu. Thus, the significant higher contribution coefficients of foliar uptake of Cd, Pb and Zn in rice leaf (Table 1 and Fig. 2) indicated that heavy metal accumulation in the leaf through foliar absorption not only depends on the particle size emitted by the traffic but also the metal availability.

In this study, the average foliar contribution of Pb in rice leaf was lower than that in leaf vegetables and grasses (Tjell et al., 1979; Chamberlain, 1983; Harrison and Chirgawi, 1989a), while the contribution of Cd was consistent with Harrison and Chirgawi's (1989a) finding about leaf vegetables. These differences were probably due to the discrepancies of atmospheric metal contents, humidity, and exposed time in different experiments (Watmough et al., 1999). The different leaf permeabilities of plant species also cannot be excluded. Previous experiments of foliar-applied radioisotopes proved that Pb, Cd and Zn ions could enter into plant leaves through the stomata or cuticle (Dollard, 1986; Greger et al., 1993; Brambilla et al., 2002). Plant species have different cuticles with different compositions of epicuticular and intracuticular lipids, which can result in dissimilar permeabilities (Prasad, 2004). Further efforts should be paid on the specific mechanisms underlying the differences in foliar contribution of heavy metals between crop varieties.

3.2 Subsequent translocation of foliar-absorbed heavy metals to other tissues

The significant higher contents of Pb and Cd in grain and Zn in stem in the exposed plants than that in the unexposed (Fig. 1) indicated that foliar-absorbed heavy metals could be transferred to other plant tissues. The distribution pattern of Pb isotope ratios of rice grain further provided the evidence on this translocation pattern of foliar-absorbed heavy metals (Fig. 3), which was consistent with previous observation (Greger et al., 1993). Meanwhile, the subsequent translocation of foliar Pb, Cd and Zn were different. Pb and Cd were mainly subsequent transferred to the grain, while Zn to the stem (Fig. 1). This difference was probably attributed to the biological properties of each metal (Colle et al., 2009). Foliar-absorbed Pb was subsequently transferred with essential nutrients towards the actively growing regions, such as grains (Watmough et al., 1999; Bi et al., 2009), resulting in a great contribution rate of foliar Pb to the grain (Fig. 2). Leaf-absorbed Cd was mainly transferred from the source leaf to phloem sink organs (Greger et al., 1993; Cakmak et al., 2000). Harris and Taylor (2001) founded that leaf-absorbed Cd could be subsequent transferred to wheat grain through phloem. Thus, similar contribution rate of foliar Cd with Pb to the grain occurred in the exposed rice (Fig. 2). Previous evidences about spinach (Harrison and Chirgawi, 1989a), tomato (Brambilla et al., 2002) and wheat (Harris and Taylor, 2001) revealed that foliar-absorbed Zn could be transferred to the stalk, fruit and grain. However, recently study reported that more than 90% of ^{65}Zn from the source leaf was transferred to other vegetative organs of rice plants, only less than 1% of leaf-absorbed ^{65}Zn were translocated to the grain (Jiang et al., 2007). Some unclear barriers inhibited the translocation of Zn from rachis into grain in rice plants under sufficient or surplus Zn conditions (Jiang et al., 2007). The concentrations of Zn in roadside particulate and soils, which were mainly from the abrasion of vechile tyres (Hjortenkrans et al., 2006), were far higher than that in unpolluted region (Shi et al., 2008; Christoforidis and Stamatis, 2009). Therefore, even though the Zn contents in the leaf of exposed rice were significant higher than in that of unexposed rice, but not significant difference of Zn contents in the grain of exposed and unexposed rice were observed.

3.3 Lead isotope composition in roadside rice plant

The distinct separation of lead isotope compositions in different organs of roadside rice plant suggested that lead accumulation in different parts originated from various sources (Fig. 3). In grain, Pb isotope ratios were distributed between the soil and vehicle exhaust (Fig. 3), indicating that Pb in rice grain may have originated both from atmosphere deposition of the vehicle emission and soil, which was consistent with the results of pot experiment (Fig. 2 and Table 1). In stem and leaf, medium ratio of ^{206}Pb/^{207}Pb exhibited an apparently mixing source of the vehicle emission and surface soil. However, the ^{208}Pb/^{206}Pb ratio in stem and leaf were lower than in the vehicle emission and soil. This is likely because that rice root is not only distributed in the surface soil (0–20 cm), but also in the layers deep than 20 cm (Yoshida et al., 1982). The ^{208}Pb/^{206}Pb ratio in soil below 20 cm is often lower than the surface soil (Hansmann and Koppel, 2000). Thus, the translocation of Pb from the deep root uptake to the leaf and stem may decrease the ratios of ^{208}Pb/^{206}Pb.

4 Conclusions

We have attempted to differentiate the sources of traffic-related heavy metals in rice plants grown in the field along the highway, and estimate the relative contribution of foliar and root uptake. Our results revealed that Pb, Cd and Zn in rice plants were partly from the foliar uptake, Cr and Cu were mainly derived from the root uptake. Considerable amount of foliar absorbed Pb and Cd were transported to rice grain. While most foliar absorbed Zn were transported to rice stems, the Zn contents in rice grains were mainly from the root uptake. Foliar absorption and transportation was a significant pathway for the accumulation of Pb and Cd in rice grains. This suggests that mitigation strategies should be applied to curtail both the foliar uptake and root uptake of traffic-emitted heavy metals for rice cropping along roadsides.

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