Mercury accumulation in vegetable *Houttuynia cordata* Thunb. from two different geological areas in southwest China and implications for human consumption

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*Houttuynia cordata* Thunb. (*HCT*) is a common vegetable native to southwest China, and grown for consumption. The results suggested that THg contents in all parts and MeHg in underground parts of *HCT* in Hg mining areas were much higher than those in non-Hg mining areas. The highest THg and MeHg content of *HCT* were found in the roots, followed by the other tissues in the sequence: roots > leaves > rhizomes > aboveground stems (THg), and roots > rhizomes > aboveground stems > leaves (MeHg). The average THg bioaccumulation factor (BCF) of *HCT* root in the Hg mining area and in non-Hg mining areas could reach 1.02 ± 0.71 and 0.99 ± 0.71 respectively, indicating that *HCT* is a Hg accumulator. And the THg and MeHg contents in all tissues of *HCT*, including the leaves, were significantly correlated with THg and MeHg content in the soil. Additionally, preferred dietary habits of *HCT* consumption could directly affect the Hg exposure risk. Consuming the aboveground parts (CAP) of *HCT* potentially poses a high THg exposure risk and consuming the underground parts (CUP) may lead to a relatively high MeHg exposure risk. Only consuming the rhizomes (OCR) of the underground parts could significantly reduce the exposure risk of THg and to some extent of MeHg. In summary, *HCT* should not be cultivated near the Hg contaminated sites, such as Hg tailings, as it is associated with a greater risk of Hg exposure and high root Hg levels, and the roots should be removed before consumption to reduce the Hg risk.

Mercury (Hg) is a highly toxic, mobile, and chemically stable element. It presents health concerns because of the risk of exposure to humans through various pathways, including inhaling contaminated air, consuming contaminated food and drinking water, and direct skin contact. Guizhou province, situated in the center of the Circum-Pacific mercuriferous belt, is a globally important center of Hg production. Long-term mining activities in this region have caused serious Hg pollution, and also increased the exposure risks in the local population through the presence of a high Hg concentration in various environmental media, especially in food stuff for human consumption, including meat, crops (especially rice) and vegetables.

*Houttuynia cordata* Thunb. (*Saururaceae; HCT*), which is native to Asian countries, has heart-shaped leaves and stoloniferous rhizomes (Fig. S1 in the Supporting Information), and prefers warm, moist, and shady environments. As an extremely popular vegetable in southwest China, its consumption history could be date back to the Eastern Han dynasty (25–220 A.D.). It is also used among diverse cultures across Asia for medicinal purposes. According to an earlier report, Guiyang (the capital city of Guizhou province) has the largest...
consumption amount of HCT in China, with an average daily consumption amount over 15 tons (average daily per capita consumption at least 30.74 g/day/person). Another report suggested that the average daily per capita consumption was even up to 400 g/day/person in Guiyang City. Our investigation also showed the per capita intake of HCT by adult in Kaiyang county of Guizhou city was as high as 76 g/day/person. Moreover, previous studies have confirmed the ability of HCT to strongly uptake lead, arsenic, and cadmium. It is therefore a good candidate species for soil remediation at sites contaminated by heavy metals. It also exhibits a high Hg uptake. Qian, X. et al. investigated 259 wild plants belonging to 49 genera in 29 families growing on wasteland comprising tailings in Wanshan Hg mining area to screen for possible phytoremediation species, and HCT was the only one that has relatively high Hg accumulation ability that potentially consumed in large quantities by the local population. Previous studies have reported many other Hg accumulation plant, such as Pteris vittata, Sesbania drummondii and Jatropha curcas, while almost all such plants are not edible for human beings. Moreover, recently reports have paid close attention to health risks of associated with the consumption of local food items in contaminated environment. A research of our group indicated HCT has the highest Hg concentration and bioaccumulation factor (BCF) value among dozens of vegetables, fruits and crops in Hg mining areas. And the highest total mercury (THg) content of HCT was approximately 100 times higher than the upper limit for vegetables (10 μg/kg, fresh weight; FW) in China’s General Standard for Contaminants in Foods. Our previous study recognized that consuming HCT was an important human Hg exposure route in Kaiyang mercury mine in central Guizhou and HCT has the highest mercury content by compare with other five vegetables. Therefore, relatively high accumulation of Hg and large consumption amount of HCT may pose a significant risk of Hg exposure to local population.

However, previously studies didn't pay much attention on the Hg distribution among different tissues of HCT and their associated health risk implications, since the eaten part of HCT varied considerable among regions in China, and different dietary habits may lead to significant regional differences in Hg exposure. In Guizhou and Yunnan province, people prefer to consume the underground parts (roots and rhizomes) of HCT, in particular the tender rhizomes, whereas the aboveground parts (aboveground stems and leaves) are generally preferred in Sichuan province and Chongqing city (see Fig. S1 for a schematic diagram of HCT plant parts). Moreover, it has been suggested that the Hg content (dry weight basis, DW) in the aboveground parts is higher than in the underground parts, contrasting with the common acceptance that plant roots contained a higher Hg content than other plant parts. And there is still insufficient information about Hg concentrations in HCT growing in different areas, especially in locations with lower Hg contamination levels.

In present study, samples of HCT and rhizosphere soil were collected for analysis from an Hg mining area (Danzhai Hg mine, DZ) and a non-Hg mining area (Zhijin county, ZJ), both located in Guizhou province. Our objectives were to: (1) determine the Hg accumulation and distribution of different HCT tissues between the two locations; (2) analyze the effect of soil Hg content on HCT tissue content in different tissues; and (3) evaluate the Hg exposure risks presented by varying dietary habits related to HCT. Our overall aim was to provide insights into the potential risks associated with HCT consumption.

Material and methods

Study areas. The study areas were in the center-west and southeastern areas of Guizhou province, located in ZJ (area, 2868 km²; population, 1.231 million [2018]; altitude, 860–2262 m a.s.l.) and Danzhai (DZ; area, 940 km²; population, 0.174 million [2017]; altitude, 700–1100 m a.s.l.) counties, respectively (Fig. 1). The two counties are approximately 260 km apart; both have a well-developed karst landform and subtropical monsoon climate (average temperature, approximately 16.6–17.2 °C). The local climate and environmental conditions are very suitable for HCT growth.

Danzhai Hg mine, wherethin mining activity dates back to the 1950s, is located in the south of DZ (Fig. 1C). Its ores also contain Au and As, and it is sometimes referred to as Danzhai Au–Hg mining area. The mine closed in the 1990s. A rough estimate is that 186 million tons of waste were produced and deposited close to the mining site without treatment, causing severe pollution of the local environment. Zhijin (Fig. 1B) is one of the main anthracite coal-producing areas of Guizhou province, and forms part of the famous Zhijin-Nayong coalfield. The coal Hg content is relatively lower (<0.1 mg/kg) than in other coal-producing regions in Guizhou. And according to the Geochemical Atlas of China, ZJ is located in an area of a relatively lower Hg background in Guizhou province.

Sample collection. Local populations forage for HCT in the wild and also cultivate it as a vegetable crop. Therefore, HCT samples were both collected from vegetable gardens and from sites where it grows wild (Fig. 1). The sampling sites in DZ were mainly distributed around Danzhai city and near the Hg tailings, while those in ZJ county were mainly distributed in rural areas. Samples of HCT and rhizosphere soil (approximately 0.5 kg and 1 kg, respectively, from each site) were collected between June and September, 2019. In total, 14 HCT and 14 rhizosphere soil samples were collected in DZ, and 11 HCT and 11 rhizosphere soil sample were collected in ZJ, respectively, placed in polyethylene ziplock bags, and stored in a cooler at 4 °C for transportation. They were taken back to the laboratory, where the HCT samples were cleaned and separated into roots, rhizomes (underground stems), aboveground stems, and leaves (Figs. S1 and S2), according to previous studies and the standard for food stuff determination. The weights of different parts of each sample were recorded and then dried at 50 °C for approximately 5 days. The rhizosphere soil samples were also dried at 50 °C. All solid samples were then ground and passed through a 0.150-mm nylon sieve.

Analytical methods and quality assurance. THg and MeHg in HCT and rhizosphere soil samples. The THg concentrations in the HCT and rhizosphere soil samples were analyzed using a Milestone Direct Mer-
cury Analyzer with AMA 254 software (Model DMA-80) according to Environmental Protection Agency (EPA) method 7473. The basic steps of THg measurement consist in placing a known amount of milled solid sample in a nickel or quartz sample holder. The sample holder is then introduced in a quartz furnace, where it is heated up to 200 °C for 60 s for sample drying and 650 °C for 105 s for Hg reduction and volatilization. Air is used as combustion and carrier gas. Combustion gases containing released Hg0 are then flushed through a cobalt-manganese oxide catalyst, where interferents like halogen compounds, nitrogen oxides and sulfur oxides are retained. Hg0 is selectively trapped in a gold-coated sand amalgamator and then released from it by heating at 850 °C for 3 s. And then the released Hg0 is carried to an atomic absorption detection cell, where the absorbance from the radiation emitted by a mercury lamp is measured at 253.7 nm. MeHg concentrations in the soil and HTC tissue samples were measured using gas chromatography–cold vapor atomic fluorescence spectrometry (GC–CVAFS) (GC: Model TRACE 1300E, Thermo Fisher Scientific, USA; CVAFS: Model 2500, Tekran, Canada) after potassium hydroxide (KOH)-methanol/solvent extraction, ethylation, and purge-and-trap collection, according to the method used in a previous study.

Calculation of bioaccumulation factor (BCF) and chronic daily intake (CDI) value. The bioaccumulation factor (BCF) is an index of the ability of organisms to accumulate a particular metal with respect to its concentration in their environment, which in the case of our experiment is the soil substrate. The BCF values for THg and MeHg in different HTC tissues can be calculated according to Eq. (1).

$$\text{BCF} = \frac{C_{\text{HCT}}}{C_{\text{soil}}}$$

where $C_{\text{HCT}}$ is the concentration in different HTC tissues (DW, μg/kg), and $C_{\text{soil}}$ is the soil Hg content (μg/kg).

Hg exposure risk to HTC was evaluated by using chronic daily intake (CDI) values (Eq. (2)) for the general adult population, as recommended by US EPA.

$$\text{CDI} = \frac{(C \times IR \times EF \times ED)}{(BW \times AT)}$$

where $C$ is the THg and MeHg concentration in HTC (unit in μg/kg, fresh weight basis (FW)); IR is the ingestion rate (kg/person/day); EF is the exposure frequency (365 days/year); ED is the exposure duration (76.3 years, equivalent to the average life span); BW is the average body weight (55.9 kg); and AT is the average time of exposure for noncarcinogens (AT = 365 × ED). The value of average life span is 76.3 years in China.

Figure 1. Sampling locations and historical mercury (Hg) mining sites in the study areas.
Quality assurance and quality control. Quality control measures were in reference of several previous studies\textsuperscript{17, 48}, including method blanks, triplicates, and several certified reference materials. The limits of detection were 0.01 μg kg\textsuperscript{-1} for THg and 0.002 μg kg\textsuperscript{-1} for MeHg respectively. The certified reference materials (CRM) of GBW10020 (Orange foliage, THg: 150 ± 13 ng/g) and TORT-2 (Lobster Hepatopancreas, MeHg: 152 ± 13 ng/g) in this study were used for THg and MeHg analysis and the average recoveries were 106.3% and 91.7%. The recoveries on matrix spikes (MeHgCl solution) of MeHg for HCT and soil digest were in the range 91—121%. And the certified reference GBW07405 (Yellow–red soil, THg: 290 ± 40 ng/g) and ERMCC580 (Estuarine sediment, MeHg: 75.5 ± 3.7 ng/g) in this study were used for soil THg and MeHg analysis, and the average recoveries were 104.7% and 101.6%. The mean THg concentration of CRM GBW10020 was determined at 146 ± 11 ng g\textsuperscript{-1} (N = 6) and of GBW07405 at 297 ± 53 ng g\textsuperscript{-1} (N = 6), whereas the mean MeHg concentration of CRM TORT-2 was 154 ± 21 ng g\textsuperscript{-1} (N = 6) and of CRM ERMCC580 was 74.3 ± 4.1 ng g\textsuperscript{-1} (N = 6), which were all comparable well with its certified values. The relative standard deviation (RSD) of duplicate analysis for Hg concentration data in this study all less than 8%. Statistical analysis was performed with SPSS 21.0 software, and the figures were created using Origin 9.0.

Results and discussion

THg and MeHg in the underground and aboveground parts of HCT. Varying dietary habits between regions mean that different parts of HCT are often preferred: underground parts, including roots and rhizomes, or aboveground parts, including aboveground stems and leaves. The results of the statistical analysis of Hg concentrations in the underground and aboveground parts of Houttuynia cordata Thunb. (HCT) are shown in Table 1 and Fig. 2 (based on DW). The THg and MeHg concentrations in both HCT parts among different sampling sites are shown in Fig. S3. The results clearly showed that the THg and MeHg contents in HCT varied widely among different collection sites. By comparing with these two areas, significant difference (p < 0.05) was found for the THg content in both under- and aboveground parts and MeHg in the underground parts between DZ and ZJ, with the Hg mining area are higher than the non-Hg mining area. And no significant difference was found of MeHg content between the under- and aboveground parts in ZJ. It should also be noted that HCT samples collected near Hg tailings (#9–11; Fig. 1) contained a significantly higher content of THg and MeHg compared with other sample sites in DZ, and this indicated that the presence of Hg tailings can greatly increase HCT Hg content in the surrounding farmland.

Table 1. Total mercury (THg) and methylmercury (MeHg) concentration in the underground and aboveground parts of Houttuynia cordata Thunb. (HCT). AM Arithmetic mean, SD standard deviation. DW dry weight basis, FW fresh weight basis.

| Items     | Study areas | Underground part, μg/kg | Aboveground part, μg/kg |
|-----------|-------------|-------------------------|-------------------------|
|           |             | Range (DW)              | AM ± SD (DW)            | Range (FW)              | AM ± SD (FW) |
| THg       | DZ          | 71–272                  | 127 ± 61                | 15–56                   | 26 ± 13       |
|           | ZJ          | 19–83                   | 33 ± 20                 | 4–17                    | 7 ± 4         |
| MeHg      | DZ          | 0.45–3.49               | 1.59 ± 1.01             | 0.09–0.72               | 0.33 ± 0.21   |
|           | ZJ          | 0.33–0.68               | 0.45 ± 0.11             | 0.05–0.11               | 0.07 ± 0.02   |

Figure 2. Total mercury (THg) and methylmercury (MeHg) concentrations in different tissues of Houttuynia cordata Thunb. (HCT) (dry weight [DW]) in Danzhai (DZ) and Zhijin (ZJ).
| Resource of HCT | Underground parts (mg/kg, DW) | Aboveground parts (mg/kg, DW) | References |
|----------------|------------------------------|-----------------------------|------------|
| Danzhai, Guizhou | 0.127 ± 0.061 | 0.246 ± 0.115 | This study |
| Zhijin, Guizhou | 0.039 ± 0.020 | 0.036 ± 0.007 | |
| Kaiyang Guizhou | 0.081 ± 0.121 | - | Our previous study Wang et al. |
| Farmlands, Sichuan and Chongqing | 0.02–0.03 | 0.03–0.05 | Chen et al. |
| Wanshan mercury mining area, Guizhou | 1.5 ± 0.81 | 1.8 ± 0.72 | Qian et al. |
| A coal mining area, China | 0.009 | 0.003 | Li et al. |

Table 2. Concentrations of total mercury (THg, mg/kg) in Houttuynia cordata Thunb. (HCT) reported in the literature.

| Items | Study area | Sample number | Root (DW, μg/kg) | Rhizome (DW, μg/kg) | Aboveground stem (DW, μg/kg) | Leaf (DW, μg/kg) | Soil (DW, μg/kg) |
|-------|------------|---------------|------------------|--------------------|-----------------------------|-----------------|----------------|
|       | DZ         | 14            | 172–1295         | 523 ± 357          | 53–219                      | 109 ± 50        | 24–156         | 68 ± 34         | 169–659         | 260 ± 140       | 116–3440        | 756 ± 870       |
| THg   | ZJ         | 11            | 85–266           | 130 ± 59           | 15–76                       | 29 ± 19         | 13–30          | 21 ± 16         | 25–61           | 40 ± 10         | 50–779          | 199 ± 203       |
| MeHg  | DZ         | 14            | 0.5–6.3          | 2.2 ± 1.8          | 0.4–3.4                     | 1.6 ± 1.0       | 0.3–1.6        | 0.7 ± 0.1       | 0.2–1.5         | 0.6 ± 0.4       | 0.9–7.3         | 2.8 ± 1.8       |
|       | ZJ         | 11            | 0.4–1.4          | 0.8 ± 0.3          | 0.3–0.7                     | 0.4 ± 0.1       | 0.3–0.6        | 0.4 ± 0.1       | 0.1–0.6         | 0.3 ± 0.1       | 0.2–1.9         | 1.0 ± 0.5       |

Table 3. Total mercury (THg) and methylmercury (MeHg) content in soil and different tissues of Houttuynia cordata Thunb. (HCT) in Danzhai (DZ) and Zhijin (ZJ). AM arithmetic mean, SD standard deviation.

THg content in the aboveground parts of HCT collected from DZ was approximately twice than that of the underground parts (DW), but only slightly higher in comparison to material collected from ZJ. In DZ, the THg and MeHg contents (measured in DW) could be as high as 272 μg/kg and 3.49 μg/kg in the aboveground parts, and 541 μg/kg and 1.39 μg/kg in the underground parts, respectively. The average values for THg and MeHg content in the underground parts in DZ (127 ± 61 and 1.59 ± 0.10 μg/kg, DW, respectively), were about four and three times as high as in ZJ, respectively (33 ± 20 and 0.45 ± 0.11 μg/kg, DW). Furthermore, THg and MeHg in the aboveground parts in DZ (246 ± 116 and 0.62 ± 0.36 μg/kg, DW, respectively) were approximately six and two times as high as in ZJ (36 ± 7 and 0.30 ± 0.10 μg/kg, DW, respectively). THg concentrations in all samples (N = 14) collected from DZ exceeded the limit for vegetables specified in the national guidance (10 μg/kg, FW), while only 18.2% (N = 11) of the underground parts in ZJ exceeded this limit (Fig. 1). Moreover, our results showed that the highest MeHg content (1.59 ± 0.10 μg/kg) in the underground parts collected from DZ was higher than in other vegetables (0.02–2.5 μg/kg), meat (0.26–0.85 μg/kg), poultry (0.56–2.4 μg/kg), and corn (0.20 ± 0.34 μg/kg) collected from Hg mining areas in Guizhou. The lowest MeHg content (0.30 ± 0.10 μg/kg) in the aboveground parts of HCT collected from ZJ was only slightly higher than in corn (0.20 ± 0.34 μg/kg), which is considered to be a low MeHg staple cereal. This indicates that the Hg exposure risk vary both with different dietary habits and whether the HCT source area is in an Hg or non-Hg mining area.

It was commonly accepted that Hg is taken up/accumulate by the roots for most plants and less is translocated towards the shoot. Though some studies showed a high translocation of Hg towards the aerial parts, most of Hg was found in the root of a plant. In contrast with previous studies, THg concentration in the aboveground parts of HCT was significant higher that in the underground parts in both Hg (DZ) and non-Hg mine areas (ZJ). And this result also consisted with several previous studies shown in Table 2 that THg content in the aboveground parts of HCT collected from DZ and ZJ is much lower than in severely Hg-contaminated areas, such as mine tailings in Wanshan Hg mining area, but is much higher than in samples collected from farmland in Sichuan and Chongqing and a coal mining area in China. THg content in the underground parts of HCT in DZ is comparable with the results of our previous study in Kaiyang Hg mining area, Guizhou. This result once again confirmed that HCT is a plant with high translocation of Hg from root to the aerial parts just like Jatropha curcas and Cyrtomium macrophyllum. And for MeHg, previous studies showed that its high solubility in lipids and easily been taken up by plants. Our results showed that MeHg content of the underground parts was significantly higher than that of the aboveground parts in DZ and no significant difference was found between the under- and aboveground parts in ZJ. This also indicates that HCT has relatively high translocation ability of MeHg. In summary, although the aboveground parts of HCT demonstrated a less contamination than the underground parts in terms of MeHg, consumption of the former (mainly in Sichuan and Chongqing) could pose a greater risk of THg exposure, whereas the consumption of roots and rhizomes may pose a greater risk of exposure to MeHg.

Hg accumulation in HCT tissues. Table 3 and Fig. 3 showed the results of THg and MeHg content in HCT tissues. Hg content in different tissues and soils collected from the study area is shown in Figs. S4 and S5. The results clearly showed that THg and MeHg content in all four types of HCT tissues collected from DZ is much higher than that from ZJ. Mean THg concentrations in HCT tissues were ranked from high to low as follows:

| Resource of HCT | Underground parts (mg/kg, DW) | Aboveground parts (mg/kg, DW) | References |
|----------------|------------------------------|-----------------------------|------------|
| Danzhai, Guizhou | 0.127 ± 0.061 | 0.246 ± 0.115 | This study |
| Zhijin, Guizhou | 0.039 ± 0.020 | 0.036 ± 0.007 | |
| Kaiyang Guizhou | 0.081 ± 0.121 | - | Our previous study Wang et al. |
| Farmlands, Sichuan and Chongqing | 0.02–0.03 | 0.03–0.05 | Chen et al. |
| Wanshan mercury mining area, Guizhou | 1.5 ± 0.81 | 1.8 ± 0.72 | Qian et al. |
| A coal mining area, China | 0.009 | 0.003 | Li et al. |
roots > leaves > rhizomes > aboveground stems. Mean MeHg concentrations in HCT showed a different order, again from high to low: roots > rhizomes > aboveground stems > leaves. The results were in agree with previous studies on rice\textsuperscript{55} and corn \textsuperscript{38} that roots and leaves contained relatively high levels of THg.

Interestingly, although roots had the highest THg content (the highest DW value being 1295 μg/kg), the average THg content in the underground parts was still lower than in the aboveground tissues. This is partly because the fibrous roots account for only 4.29 ± 1.3% (DW) of the total weight of underground parts, and partly because the leaves contain a relatively high THg content. Previous studies \textsuperscript{56, 57} have suggested that inorganic Hg in leaves is mainly derived from the atmosphere and this results also confirmed by other studies about paddy rice (\textit{Oryza sativa} \textit{L.})\textsuperscript{58}, corn (\textit{Zea mays} \textit{L.}) \textsuperscript{38}, and other plants \textsuperscript{56}, though some study found that plant leaf may also be a potential sources of atmospheric Hg\textsubscript{0}\textsuperscript{59}. The average MeHg/THg ratio is 0.61 ± 0.45% for soil, which was comparable to the ratio in roots (0.52 ± 0.19%), and significantly higher (p < 0.05) than in leaves (0.41 ± 0.30%), and much lower than in the rhizomes (1.64 ± 0.69%) and the aboveground stems (1.52 ± 0.85%).

The BCF value results are showed in Table S1 and Fig. 4. It could see that the highest BCF value for THg and MeHg in roots could reach 2.88 and 2.91, respectively (Table 3 and Fig. 4). The average THg BCF value in roots is 1.02 ± 0.71 in DZ and 0.99 ± 0.71 in ZJ, and that for MeHg is 0.79 ± 0.30 in DZ and 0.94 ± 0.51 in ZJ. Previous research by Qian et al. \textsuperscript{17} shown that the highest BCF value of THg and MeHg among 259 wild plants could reached 5.5 and 18, respectively. Other research on rice \textsuperscript{60} suggested that the BCF value of roots is 4.2 ± 2.1, 1.1 ± 0.8 for stems, and 0.72 ± 0.82 for leaves. The study of Zhang et al. \textsuperscript{58} even showed that the BCF value for MeHg in rice could as much as 800 times higher than the value for inorganic Hg. This indicates that, for THg, HCT may be a high Hg accumulation plant, but its accumulation capacity for MeHg is relatively lower in comparison with other high MeHg accumulation plants. For both study areas of this study, the BCF value of THg followed the sequence of roots > leaves > rhizomes > aboveground stems, and MeHg followed the sequence of roots > rhizomes > aboveground stems > leaves. The sequence may confirm that part of inorganic Hg was transferred from soil through rhizomes to the aboveground stem and leaves and atmospheric Hg is another important Hg resources of foliage. It also suggested that soil MeHg was the only resources of MeHg in HCT.
Moreover, although HCT roots only account for a tiny share (4.29 ± 1.3%) of the weight of the underground parts, roots contributed 17.51 ± 7.83% (5.26–31.83%) and approximately 17.95 ± 3.47% (7.34–31.36%) of THg in the underground parts collected from DZ and ZJ, respectively. The equivalent values for MeHg were 5.82 ± 6.46% (3.42–10.10%) for DZ and 7.71 ± 3.47% (2.68–14.44%) for ZJ. Therefore, this level of potential Hg and MeHg exposure indicates that HCT roots should be removed before cooking.

| Mercury species | Correlation analysis | Regress equation | Pearson coefficients |
|-----------------|----------------------|-----------------|---------------------|
| THg Root y = 0.375x + 1161.30 $r^2 = 0.615$, $p < 0.01$, $N = 25$ |
| THg Rhizome y = 0.057x + 44.27 $r^2 = 0.499$, $p < 0.01$, $N = 25$ |
| THg Aboveground stem y = 0.038x + 27.75 $r^2 = 0.057$, $p < 0.01$, $N = 25$ |
| THg Leaf y = 0.169x + 96.80 $r^2 = 0.498$, $p < 0.01$, $N = 25$ |
| THg Underground parts y = 0.067x + 51.16 $r^2 = 0.490$, $p < 0.01$, $N = 25$ |
| THg Aboveground parts y = 0.140x + 81.82 $r^2 = 0.509$, $p < 0.01$, $N = 25$ |
| MeHg Root y = 0.828x - 0.042 $r^2 = 0.836$, $p < 0.01$, $N = 25$ |
| MeHg Rhizome y = 0.508x + 0.064 $r^2 = 0.816$, $p < 0.01$, $N = 25$ |
| MeHg Aboveground stem y = 0.165x + 0.251 $r^2 = 0.602$, $p < 0.01$, $N = 25$ |
| MeHg Leaf y = 0.131x + 0.189 $r^2 = 0.414$, $p < 0.01$, $N = 25$ |
| MeHg Underground parts y = 1.07 × 10^{-2}x + 0.49 $r^2 = 0.680$, $p < 0.01$, $N = 25$ |
| MeHg Aboveground parts y = 3.65 × 10^{-2}x + 0.272 $r^2 = 0.695$, $p < 0.01$, $N = 25$ |

Table 4. Correlations between total mercury (THg) and methylmercury (MeHg) in soils and in different tissues of *Houttuynia cordata* Thunb. (HCT) in Danzhai (DZ) and Zhijin (ZJ).

Risks of Hg exposure associated with HCT consumption. The parts of HCT considered edible differ among different areas of China: the underground parts are mainly consumed in Sichuan province and Chongqing city, while the underground parts are preferred in Guizhou, Yunnan, and Hunan provinces. Our results showed that HCT roots have the highest THg and MeHg contents (“Hg accumulation in HCT tissues”). Therefore, we assessed the Hg exposure risk from consuming HCT according to three scenarios: consuming the underground parts (CUP), the aboveground parts (CAP), or only the rhizomes (OCR). The ingestion rate (IR) value we choose was 76 g/person/day according to our previous study.21

CDI results clearly showed that substantial HCT consumption leads to relatively high health risks as a result of THg exposure (Fig. 5, Table S3); the highest CDI value reached over 1/3 of the reference dose (0.23 μg/kg/
day) recommend by USEPA\textsuperscript{68}. The exposure risk of THg in Hg mining area (DZ) was significant higher than that of non-Hg mining area (ZJ) (\( p < 0.05 \)), while for MeHg, only the expose risk of CAP slight high than that of ZJ (Fig. 5). Both in Hg and non-Hg mining area, the exposure risk of THg of CAP was higher than of CUP, whereas the risk of exposure to MeHg from CAP was lower than that of CUP. The THg CDI values of the under- and aboveground parts accounts for 13.6\% (range 7.6–29.2\%) and 19.56\% (range 10.9–42.9\%), and for MeHg only 0.39\% and 0.11\% of the reference dose, respectively. While, this values of the under- and aboveground parts for THg accounted for only 3.6\% and 2.9\% and for MeHg accounted for 0.38\% and 0.06\% of the reference dose, respectively. This indicated a greater risk of THg exposure from HCT consumption in Hg mining areas, and a lower risk of MeHg exposure in both Hg and non-Hg mining areas.

Moreover, in Guizhou province, the rhizome is the main part of HCT consumed, but the root was not removed before cooking; this is not recommended since the root contains the highest levels of THg and MeHg. Table S2 and Fig. 4 showed the CDI values yielded by only consuming the rhizomes (OCR), the results indicated CDI values for THg are 14.3\% (DZ) and 12.3\% (ZJ), and for MeHg are 1.52\% (DZ) and 23.1\% (ZJ) less than that consuming the entire underground parts. The fibrous roots attached to the rhizome should therefore be removed before consumption.

Conclusions

The risk of exposure to Hg by the consumption agricultural products is of great concern in Guizhou province because there are many Hg mining areas located. To our best knowledge, HCT is the only vegetable with high Hg-accumulation and been widely consumed in southwest China. Our results showed that THg contents in all parts and MeHg contents in underground parts of HCT in Hg mining area were significantly higher than that in the non-Hg mining area (control site). And the roots of HCT in Hg mining area were significantly higher than that of ZJ, while the shoots were comparable. This indicated a potential risk of THg and MeHg exposure from consuming HCT in Hg mining areas, and this risk was greater for THg than for MeHg. Moreover, the THg contents in all tissues, including the leaves, were significantly correlated with soil Hg content, indicating that Hg pollution may have a major effect on the safety consumption of HCT. Consuming HCT from Hg mining area could be associated with a higher exposure risk to Hg and MeHg than that of non-Hg mining area. Preferred dietary habits in HCT consumption could directly affect the Hg exposure risk. Consuming the underground parts (CAP) may lead to a relatively high MeHg exposure risk both in Hg and no-Hg mining areas. All of our results indicate that this Hg accumulation plant should not be cultivated in Hg-contaminated areas, such as areas close to Hg slag or tailings, and another important factor is which part of HCT to be consumed, and it is recommended to remove the roots before cooking to reduce the Hg exposure risk.

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References

1. Clarkson, T. W. & Magos, L. The toxicology of mercury and its chemical compounds. Crit. Rev. Toxicol. 36, 609–662 (2006).
2. Bjørklund, G., Dadar, M., Mutter, J. & Aaseth, J. The toxicology of mercury: Current research and emerging trends. Environ. Res. 159, 545–554 (2017).
3. Feng, X. & Qiu, G. Mercury pollution in Guizhou, Southwestern China—An overview. Sci. Total Environ. 400, 227–237 (2008).
4. Zhang, H., Feng, X., Larssen, T., Qiu, G. & Vogt Rolf, D. In inland china, rice, rather than fish, is the major pathway for methylmercury exposure. Environ. Health Perspect. 118, 1183–1188 (2010).
5. Qiu, G. et al. Methylmercury accumulation in rice (Oryza sativa L.) grown at abandoned mercury mines in Guizhou, China. J. Agric. Food. Chem. 56, 2465–2468 (2008).
6. Li, R. et al. Mercury pollution in vegetables, grains and soils from areas surrounding coal-fired power plants. Sci. Rep. 7, 46545 (2017).
7. Li, X. et al. Health risks of heavy metal exposure through vegetable consumption near large-scale Pb/Zn smelter in central China. Ecotoxicol. Environ. Saf. 161, 99 (2018).

Figure 5. Chronic daily intake (CDI) values of (a) total mercury (THg) and (b) methylmercury (MeHg) under three Houttuynia cordata Thunb. (HCT) consumption scenarios.
8. Yang, Y. Textual research on houttuynia cordata and historical change of its role as food. Agric. Archaeol. 4, 211–218 (2019) (in Chinese).
9. Kumar, M., Prasad, S. K., Laloo, D., Joshi, A. & Hemalatha, S. Pharmacognostical and phytochemical standardization of Houttuynia cordata Thunb: A potent medicinal herb of North-Eastern India and China. Phcog. J. 6, 34–42 (2014).
10. Wen, X., Zhongjie, S., Qing, L. & Bo, S. The status quo and standardized cultivation techniques for green food (A level) of Houttuynia cordata Thunb. in Guizhou province. Vegetables 10, 42–46 (2017) (in Chinese with English abstract).
11. Zhao, J. et al. Contents of heavy metals in some vegetables and their potential risks to human health in guiyang and wanshan areas. Asian J. Ecotoxicol. 4, 392–398 (2009) (in Chinese with English abstract).
12. Wang, Q. et al. Vegetable Houttuynia cordata Thunb as an important human mercury exposure route in Kaiyang county, Guizhou province, SW China. Ecotoxicol. Environ. Saf. 197, 110575. https://doi.org/10.1016/j.ecoenv.2020.110575 (2020).
13. Wu, J., Tang, Y., Meng, X. X., Li, H. X. & Wei, T. Lead absorption and accumulation in Houttuynia cordata Thunberg and hygienic safety evaluation, The 3rd International Conference on Bioinformatics and Biomedical Engineering(ICBBE), Beijing, China, 1–6 (2009).
14. Ha, N. T. H., Sakakibara, M., Sano, S. & Mai, T. N. Uptake of metals and metalloids by plants growing in a lead–zinc mine area, Northern Vietnam. J. Hazard. Mater. 186, 1384–1391 (2011).
15. Wu, L. et al. Phyto remediation of soil contaminated with cadmium, copper and polychlorinated biphenyls. Int. J. Phytorem. 10, 570–584 (2012).
16. Zhou, L., Chen, W., Hou, L. Notice of retraction the cadmium accumulation ability of houttuynia cordata enhanced by microorganisms in soil. In 5th International Conference on Bioinformatics and Biomedical Engineering(ICBBE), Wuhan, China, 1–6 (2011).
17. Qian, X. et al. Total mercury and methylmercury accumulation in wild plants grown at wastelands composed of mine tailings: Insights into potential candidates for phyto remediation. Environ. Pollut. 239, 757–767 (2018).
18. Manikandan, R., Sahi, S. V. & Venkatachalam, P. Impact assessment of mercury accumulation and biochemical and molecular response of menhda arvensis: A potential hyperaccumulator plant. Sci. World J. 2015, 751217. https://doi.org/10.1155/2015/751217 (2015).
19. Su, Y., Han, F. X., Chen, J., Sridhar, B. R. M. & Monts, D. L. Phytoextraction and accumulation of mercury in three plant species: Indian Mustard (Brassica Juncea), Beard Grass (Polygopon monospeliensis), and Chinese Brake Fern (Pteris vittata). Int. J. Phyto rem. 10, 547–560 (2008).
20. Venkatachalam, P., Srivastava, A. K., Raghothama, K. G. & Sahi, S. V. Genes induced in response to mercury-ion-exposure in heavy metal hyperaccumulator Sesbania drummondii. Environ. Sci. Technol. 43, 843–850 (2009).
21. Liu, Z. et al. A review on phyto remediation of mercury contaminated soils. J. Hazard. Mater. 400, 123138. https://doi.org/10.1016/j.jhazmat.2020.123138 (2020).
22. Chen, J. et al. Bioaccumulation and physiological effects of mercury in Pteris vittata and Nepheirops exallata. Ecotoxicology 18, 110 (2008).
23. Marrugo-Negrete, J., Marrugo-Madrid, S., Pinedo-Hernández, J., Durango-Hernández, J. & Diez, S. Screening of native plant species for phyto remediation potential at a Hg-contaminated mining site. Sci. Total Environ. 542, 809–816 (2016).
24. Antoine, J. M. R., Fung, L. A. H. & Grant, C. N. Assessment of the potential health risks associated with the aluminium, arsenic, cadmium and lead content in selected fruits and vegetables grown in Jamaica. Toxicol. Rep. 4, 181–187 (2017).
25. Augustsson, A. et al. Challenges in assessing the health risks of consuming vegetables in metal-contaminated environments. Environ. Int. 113, 269–280 (2018).
26. Raj, D. & Matti, S. K. Bioaccumulation of potentially toxic elements in tree and vegetable species associated with enhanced health and ecological risks: A case study from a thermal power plant, Chandrapur India. Rend. Lincei-Sci. Fis. 30, 649–665 (2019).
27. Yang, B., Gao, Y., Zhang, C., Zheng, X. & Li, B. Mercury accumulation and transformation of main leaf vegetable crops in Cambosol and Ferrosol soil in China. Environ. Sci. Pollut. R. 27, 391–398 (2020).
28. Yu, H., Li, J. & Luan, Y. Meta-analysis of soil mercury accumulation by vegetables. Sci. Rep. 8, 1261 (2018).
29. Ghaseemidekhordi, B. et al. Concentration of lead and mercury in collected vegetables and herbs from Markazi province, Iran: A non-carcinogenic risk assessment. Food Chem. Toxicol. 113, 204–210 (2018).
30. Xia, J. et al. Screening of native low mercury accumulation crops in a mercury-polluted mining region: Agricultural planning to manage mercury risk in farming communities. J. Clean. Prod. 262, 121324. https://doi.org/10.1016/j.jclepro.2020.121324 (2020).
31. NHFPC (National Health and Family Planning Commission of the People’s Republic of China) and CFDA (China Food and Drug Administration). National standard for food safety: limits of pollutants in food (GB2762–2017). Beijing, China: NHFPC and CFDA (2017).
32. Natasha, et al. A critical review of mercury speciation, bioavailability, toxicity and detoxification in soil-plant environment: Ecotoxicology and health risk assessment. Sci. Total Environ. 711, 134749. https://doi.org/10.1016/j.scitotenv.2019.134749 (2020).
33. Canario, J., Caezote, M., Vale, C. & Cesario, R. Evidence for elevated production of methylmercury in salt marshes. Environ. Sci. Technol. 41, 7376–7382 (2007).
34. Su, L. et al. Concentration and form analysis of heavy metals in soil and residues in Danzhi mercury mining areas in Guizhou. Guizhou Agric. Sci. 38, 202–204 (2010) (in Chinese).
35. Sun, X., Wang, J. & Feng, X. Distribution and potential environmental risk of mercury and arsenic in slag, soil and water of Danzhi mercury mining area, Guizhou province, China. Asian J. Ecotoxicol. 9, 1173–1180 (2014) (in Chinese).
36. Dai, S., Ren, D. & Tang, Y. Concentration and distribution of elements in Late Permian coals from western Guizhou Province China. Int. J. Coal. Geol. 61, 119–137 (2005).
37. Xie, X., Ren, T. & Sun, H. Geochanical atlas of China. Beijing: Geology Press, 56–57, (2012).
38. Sun, G. et al. Corn (Zea mays L.): A low methylmercury staple cereal source and an important biospheric sink of atmospheric mercury, and health risk assessment. Environ. Int. 131, 104971. https://doi.org/10.1016/j.envint.2019.104971 (2019).
39. NHFPC( National Health and Family Planning Commission of the People’s Republic of China, National food safety standards: determination of total mercury and organic mercury in food (GB5009.17–2014), Beijing,China: NHFPC (2014).
40. USEPA. Mercury in solids and solutions by thermal decomposition, amalgamation, and atomic absorption spectrophotometry(Method 7473 (SW-846)), Washington,US: EPA, (1998).
41. Melendez-Perez, J. J. & Foster, A. H. Assessment of direct mercury analyzer to quantify mercury in soils and leaf samples. J. Brazil. Chem. Soc. 24, 1880–1886 (2013).
42. Habte, G. et al. Elemental profiling and geographical differentiation of Ethiopian coffee samples through inductively coupled plasma-optical emission spectrometry (ICP-OES), ICP-mass spectrometry (ICP-MS) and direct mercury analyzer (DMA). Food Chem. 212, 512–520 (2016).
43. Liang, L., Horvat, M., Cernichiarli, E., Geleen, B. & Balogh, S. J. Simple solvent extraction technique for elimination of matrix interferences in the determination of methylmercury in environmental and biological samples by ethylation-gas chromatography–cold vapor atomic fluorescence spectrometry. Talanta 43, 1883–1888 (1996).
44. Zhuang, P., McBride, M. B., Xia, H., Li, N. & Li, Z. Health risk from heavy metals via consumption of food crops in the vicinity of Dabaoshan mine South China. Sci. Total Environ. 407, 1551–1561 (2009).
45. Liao, M. et al. Profiles and potential health risks of heavy metals in soil and crops from the watershed of Xi River in Northeast China. Ecotoxicol. Environ. Saf. 169, 442–448 (2019).
46. USEPA. Supplemental guidance for developing soil screening levels for superfund sites Washington, DC, US: Office of Solid Waste and Emergency Response (2001).
47. USEPA. Exposure factors handbook [EPA/600/R-09/032F]. Washington DC, US: National Center for Environmental Assessment (2011).
48. Zhao, H. et al. Mercury contents in rice and potential health risks across China. Environ. Int. 126, 406–412 (2019).
49. Ahammad, S. J., Sumithra, S. & Senthilkumar, P. Mercury uptake and translocation by indigenous plants. Rasayan J. Chem. 11, 1–12 (2018).
50. Marrugo-Negete, J., Durango-Hernandez, J., Pinedo-Hernandez, J., Olivo-Verbel, J. & Díez, S. Phytoremediation of mercury-contaminated soils by Jacaranda curcas. Chemosphere 127, 58–63 (2015).
51. Xu, Y., Feng, L., Li, Y. & Dong, H. Mercury accumulation plant Cyrtomium macrophyllum and its potential for phytoremediation of mercury polluted sites. Chemosphere 189, 161 (2017).
52. Chen, L., Gong, H. & Wu, W. Determination of As, Hg and 666, DDT of different Houttuynia cordata Accessions. J. Anhui Agric. Sci. 37, 14698–14700 (2009) (in Chinese).
53. Li, X., Su, J., Hui, B. & Gong, P. Heavy metal determination of Houttuynia cordata Thumb from coal mining fields. Sci. Technol. Food Ind. 37, 309–317 (in Chinese with English abstract) (2016).
54. Clayden, M. G. et al. Mercury biomagnification through food webs is affected by physical and chemical characteristics of lakes. Environ. Sci. Technol. 47, 12047–12053 (2013).
55. Meng, B., Feng, X., Qiu, G., Cai, Y. & Sommar, J. Distribution patterns of inorganic mercury and methylmercury in tissues of rice (Oryza sativa L.) plants and possible bioaccumulation pathways. J. Agric. Food Chem. 58, 4951–4958 (2010).
56. De Temmerman, L., Waegeneers, N., Claeys, N. & Roekens, E. Comparison of concentrations of mercury in ambient air to its accumulation by leafy vegetables: An important step in terrestrial food chain analysis. Environ. Pollut. 157, 1337–1341 (2009).
57. Shahid, M. et al. Foliar heavy metal uptake, toxicity and detoxification in plants: A comparison of foliar and root metal uptake. J. Hazard. Mater. 325, 36–58 (2017).
58. Zhang, H., Feng, X., Larsen, T., Shang, L. & Li, P. Bioaccumulation of Methylmercury versus Inorganic Mercury in Rice (Oryza sativa L.) Grain. Environ. Sci. Technol. 44, 4499–4504 (2010).
59. Canário, J. et al. Salt-marsh plants as potential sources of Hg in the atmosphere. Atmos. Environ. 152, 458–464 (2017).
60. Meng, B. et al. Localization and speciation of mercury in brown rice with implications for Pan-Asian Public Health. Environ. Sci. Technol. 48, 7974–7981 (2014).
61. Garcia-Sánchez, A., Marciego, A., Álvarez-Ayuso, E., Regina, I. S. & Rodríguez-González, M. A. Mercury in soils and plants in an abandoned cinnabar mining area (SW Spain). J. Hazard. Mater. 168, 1319–1324 (2009).
62. MEEP (Ministry of Ecology and Environmental Protection) and SAMR (State Administration for Market Regulation) Soil environmental quality risk control standard for soil contamination of agricultural land (GB15618–2018). Beijing, China: MEEP and SAMR (2018).
63. Yang, Y. & Qian, F. Heavy metal concentration and pollution assessment of different Houttuynia planting areas in Guizhou city. Journal of Guizhou Normal University (Natural Sciences) 31, 81–84 (in Chinese with English abstract) (2013).
64. Niu, Z., Zhang, X., Wang, Z. & Ci, Z. Field controlled experiments of mercury accumulation in crops from air and soil. Environ. Pollut. 159, 2684–2689 (2011).
65. Greger, M., Wang, Y. & Neuschütz, C. Absence of Hg transpiration by shoot after Hg uptake by roots of six terrestrial plant species. Environ. Pollut. 134, 201–208 (2005).
66. Wang, S., Feng, X., Qiu, G., Wei, Z. & Xiao, T. Mercury emission to atmosphere from Lanmuchang Hg–Tl mining area, Southwestern Guizhou China. Atmos. Environ. 39, 7459–7473 (2005).
67. Yin, R., Feng, X. & Meng, B. Stable mercury isotope variation in rice plants (Oryza sativa L.) from the Wanshan mercury mining district, SW China. Environ. Sci. Technol. 47, 2238–2245 (2013).
68. USEPA. Risk assessment guidance for superfund. in: Human health evaluation manual (Part A) USA EPA/540/1–89/002. Washington DC, US: USEPA 35–52 (1989).

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Author contributions
Q.W. designed the experiment, collected and analyzed the samples, interpreted the data and wrote the manuscript; A.W., D.W. and L.F. collected and analyzed the samples; X.L. prepared Figs. 1 and 2; Z.L. and X.F. supervised its analysis and edited the manuscript.

Competing interests
The authors declare no competing interests.

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