



Captive pandas are at risk from environmental toxins

Citation

Chen, Yi#ping, Lorraine Maltby, Qiang Liu, Yi Song, Ying#juan Zheng, Aaron M. Ellison, Qing# yi Ma, and Xiao#min Wu. "Captive pandas are at risk from environmental toxins." Frontiers in Ecology and the Environment 14, no. 7 (2016): 363-367.

Published Version

10.1002/fee.1310

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Accessibility

1	Captive pandas are at risk from environmental toxins
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19	Ex situ conservation efforts are the last resort for many critically endangered
20	species and captive breeding centers are thought to provide a safe
21	environment in which to produce individuals for eventual re-introduction to the
22	wild. The giant panda (Ailuropoda melanoleuca) is one of the most
23	endangered animals in the world, and it is recognized worldwide as a symbol
24	for conservation. Here, we report that captive pandas of the Sichuan and
25	Qinling subspecies are exposed to high concentrations of persistent organic
26	pollutants, including polychlorinated dibenzo-p-dioxins (PCDDs),
27	dibenzofurans (PDCFs), and biphenyls (PCBs), as well as heavy metals
28	(arsenic, cadmium, chromium, and lead). Further analysis of the ex situ
29	environment of the Qinling subspecies demonstrated that contaminated food
30	supplies exposed captive Qinling pandas to high concentrations of PCDD,
31	PCDFs, PCBs, As, Cd, Cr, and Pb). In the short term, these endangered
32	animals should be relocated to breeding centers in less contaminated areas.
33	Their long-term survival, however, depends on reducing emissions of toxic
34	pollutants throughout China.
35	

Key words: Endangered species, *ex situ* conservation, Giant panda, heavy
 metals, persistent organic pollutants.

39	The giant panda (Ailuropoda melanoleuca) is one of the most endangered
40	animals in the world, and it is recognized worldwide as a symbol for
41	conservation. The panda lineage is at least 11.6 million years old (Abella et al.
42	2012); fossils > 2 million years old and historical records have revealed that
43	pandas once were distributed in at least 18 of China's 23 provinces (Zhu and
44	Long 1983). Until the mid-19 th century, giant pandas still inhabited most of
45	eastern and southern China (Hunan, Hubei, Sichuan, Shaanxi and Gansu
46	provinces), but their range has declined in recent years as a result of hunting,
47	habitat destruction, logging, resource exploitation, and tourism (Zhang et al.
48	2013). Giant pandas now survive only in small, fragmented conservation
49	zones in the Qinling, Bashan and Qionglai Mountains (Zhang et al. 2013) and
50	in ex situ breeding centers including the zoos of Beijing and the breeding
51	centers of Wolong and Chengdu.
52	It is generally assumed that the conservation areas and the captive
53	breeding centers protect giant pandas from the adverse impacts of human
54	activities. However, their presumed safety may be compromised by the
55	dissemination of widespread pollutants into conservation zones or the
56	proximity of breeding centers to more heavily-polluted urban areas. For
57	example, perfluorinated compounds used in consumer and industrial products
58	as surfactants, surface protectors, and fire-fighting foams have been found in
59	serum samples taken from giant pandas in the Beijing zoo as well as from red
60	pandas (Ailurus fulgens) in a number of other zoos and wild animal parks in

China (Dai et al. 2006). However, the extent to which either wild pandas or
pandas in breeding centers are exposed to persistent organic pollutants (POPs)
and heavy metals that can accumulate in their tissues, compromise their
health, and potentially affect the success of ongoing conservation programs
remains unknown.

Here, we present data illustrating that giant pandas in *ex situ* breeding centers are exposed to much greater concentrations of POPs and heavy metals than their wild counterparts. Our data suggest that the bamboo fed to the pandas is the proximate source of these compounds. Consequently, urgent action is needed to safeguard these conservation icons, both in captivity and in the wild.

72

73 Materials and Methods

Faecal droppings, which can be used as non-invasive indicators of pollutant 74 75 exposure (Christensen et al. 2013), were collected from wild pandas in the Wolong and Foping National Nature Reserves, and from captive pandas 76 77 housed in China Conservation and Research Center for the Giant panda (CCRCGP) and the Shaanxi Wild Animal Research Center (SWARC) (Fig.1). 78 The CCRCGP is the largest captive panda breeding center for the Sichuan 79 subspecies of giant panda, and SWARC is the only breeding center for the 80 Qinling subspecies. Samples of bamboo, the primary food for giant pandas, 81 were collected in the wild from Foping and from plants grown at SWARC. 82

Mixed feedstuff, fed to pandas as a nutrient supplement, was also sampled
from SWARC. Additional details on sample collection are provided in the
Supplemental Online Material.

The faecal droppings, plant tissue, and feedstock samples all were dried 86 to constant mass, digested, and analyzed using standard methods. 87 Determination of concentrations of POPs in the samples was done using 88 (high-resolution mass spectrometry (Liu et al. 2006; Li et al. 2008) at the 89 90 Research Center for Eco-environmental Sciences of the Chinese Academy of 91 Sciences. Concentrations of heavy metals were determined using atomic absorption or fluorescence spectrometry at the Institute of Earth Environment 92 of the Chinese Academy of Sciences. Complete details on analytical methods, 93 94 including QA/QC protocols, can be found in the Supplemental Online Material). Data were analyzed using the SPSS software, version 19.0 (IBM SPSS) 95 Inc.). Contaminant concentrations in droppings from wild and captive giant 96 97 pandas among and between the two subspecies were compared using *t*-tests. 98

99 **Results and discussion**

It is generally thought that pandas in captive breeding centers are better
protected from human activities than are wild pandas in nature conservation
zones, primarily because *in situ* conservation zones have become more
fragmented and less suitable for giant pandas (Liu *et al.* 2001). However, *ex situ* breeding centers usually are close to urban areas and there is an

105 increasing concern that *ex situ* conservation efforts may be being

106 compromised due to environmental pollution associated with urbanization.

107 With China's rapid industrialization and urbanization, environmental pollution is

¹⁰⁸ increasing in seriousness and following as it follows a trajectory similar to that

109 previously traversed by developed countries (Seinfeld 2004). This pollution

trajectory is having major impacts on public health, as seen in, for example,

111 the > 200 "cancer villages" in China (Yang 2013).

Among the many pollutants, POPs and heavy metals are of significant 112 113 environmental concern because they may be transported over long distances in air and water (Lohmann et al. 2007), are very persistent in the environment, 114 115 accumulate readily in fatty tissues, and are highly toxic to humans and other 116 mammals (e.g., Qiu 2013; Adriano et al. 2014; Fernandez-Rodriguez et al. 2015; Syed Ali et al. 2015). Three classes of POPs - PCDDs (polychlorinated 117 dibenzo-p-dioxins), PCDFs (polychlorinated dibenzofurans), and PCBs 118 119 (polychlorinated biphenyls) were found in much higher concentrations in faecal droppings of captive giant pandas than in wild pandas (Fig. 2, WebTables1, 2). 120 121 POPs were also found at elevated levels in the bamboo fed to captive pandas and their nutrient-supplement feedstock (WebFigures 1, 2). A variety of forms 122 ("congeners") of PCDDs and PCDFs are generated as by-products from 123 various combustion and chemical processes, whereas polychlorinated 124 125 biphenyls (PCBs) were widely used as dielectric fluids in transformers and capacitors, heat exchange fluids, and as additives in pesticides, adhesives, 126

plastics, and paints because of their insulating and nonflammable properties
(Fiedler 2007). Although production of PCBs ceased in 1974, they are still
released from old capacitors and transformers and can still be found in various
environmental components and in human tissues (Mai *et al.* 2005; Imamura *et al.* 2007).

Because PCDDs, PCDFs, and PCBs occur as congeners that differ in 132 toxicity and toxic equivalency factors, the World Health Organization has 133 defined a single toxic equivalent (WHO-TEQ) that can be calculated to 134 135 determine total POP exposure (Van den Berg et al. 2006). Both total concentrations and the WHO-TEQ for PCDDs, PCDFs, and POPs were higher 136 in droppings collected from captive pandas than they were in wild pandas (Fig. 137 138 3). These results are paralleled by total concentrations and WHO-TEQs for the bamboo fed to the pandas and their nutrient-supplement feedstock 139 (WebFigure 2). 140 141 Four heavy metals with known toxicity – arsenic (As), cadmium (Cd), chromium (Cr), and lead (Pb) (Brahmia et al. 2013; Neal and Guilarte 2013; 142 Uddh-Soderberg et al. 2015) – also were found at elevated levels in droppings 143 of captive pandas relative to wild ones (Fig. 3), as well as in their food and their 144 nutrient-supplement feedstock (WebFigure 3). Unlike POPs, these heavy 145 metals occur in the natural environment, but they are readily mobilized by 146 147 human activities such as mining, automobile use, and overuse of chemical fertilizer. 148

149	Our results provide direct evidence that giant pandas are exposed to
150	PCDDs, PCDFs, PCBs, and heavy metals in both ex situ captive breeding
151	centers and in situ conservation areas, but concentrations of these toxins in
152	pandas are far greater for pandas in captivity. Previous studies have shown
153	that PCDDs and PCDFs are associated with developmental toxicity,
154	immunotoxicity, and reproductive toxicity. PCBs and their breakdown products
155	are known endocrine disrupters, cause the loss of renal cell viability, and are
156	associated with increased risk of chloracne, goiter, anemia, and cancer
157	(Lohmann et al. 2007; Qiu 2013; Adriano et al. 2014; Fernandez-Rodriguez et
158	al. 2015; Gustavson et al. 2015; Syed Ali et al. 2015). Heavy metal exposure
159	has been associated with increased incidence of cancer (Cr and As),
160	nephrotoxicity and bone damage (Cd), and reduced reproductive function (Pb)
161	(Neal and Guilarte 2013; Brahmia <i>et al</i> . 2013; Uddh-Soderberg <i>et al</i> . 2015).
162	We conclude that our results belie the notion that captive breeding centers and
163	zoos provide a safe haven from human impacts.
164	Our results also illustrate that dietary exposure is the dominant, proximal
165	pathway through which giant pandas are exposed to POPs and heavy metals
166	(WebFigures 1-3). Although the food of both captive and wild pandas was
167	enriched in POPs (WebFigures 1, 2) and heavy metals (WebFigure 3), the
168	concentrations of both POPs and metals, and WHO-TEQs of POPs were
169	significantly greater in bamboo eaten by captive pandas (WebFigures 1–3).
170	We note that the nutrient-supplemented feedstock (baked into steamed bread

for the pandas) was enriched only in Cd, Cr, and Pb, but not in As, relative tofresh bamboo.

173 In sum, our data provide clear evidence that giant pandas both in the wild and in captivity are exposed to PCDDs, PCDFs, PCBs, and heavy metals 174 175 through their diet, and that exposure to these environmental toxins is greater in ex situ breeding centers than in in situ nature reserves. Because exposure to 176 these environmental toxins is likely to impact negatively the health of these 177 animals, we suggest that urgent action is needed to safeguard these 178 179 conservation icons. In the short-term, captive breeding centers should be relocated to areas less impacted or contaminated by environmental toxins, and 180 the food provided to captive pandas should be strictly monitored to ensure that 181 it lacks POPs and heavy metals, and is of consistent high quality. In the long 182 term, however, a more sustainable solution will rely on improving air quality 183 through reducing emissions of toxic pollutants. 184

185

186 Acknowledgments

187 This work was supported by the IEECAS fund. We thank the China

188 Conservation and Research Center for Giant panda (CCRCGP) for helping

with this research. Here we also thank Professor An for valuable advice during
the course of this study.

191

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259	

260 Figure legends

Figure 1. Sites of sample collection (a); typical dropping of wild pandas (b); and captive pandas (c) at the Shaanxi Wild Animal Research Center (SWARC).

264	Figure 2. Concentrations of 12 PCB congeners (top) and 17 CDD/F congeners
265	(bottom) in the droppings of wild and captive pandas of the Sichuan and
266	Qinling subspecies of giant pandas. In each of these star plots, the radius is
267	equal to the maximum observed concentration, and concentrations of each
268	individual congeners are scaled to the maximum. The conclusion from these
269	plots is that captive pandas have both more congeners and higher
270	concentrations of them in their faecal samples than wild pandas. Tabular data
271	(actual mean concentrations and the standard errors of the means) are given
272	in WebTables 1 and 2.
273	
273 274	Figure 3. Concentrations of (a) all (summed) PCDDs and PCDFs; (b) all
	Figure 3. Concentrations of (a) all (summed) PCDDs and PCDFs; (b) all (summed) PCBs; (c) WHO-TEQs of PCDDs and PCDFs; and (d) WHO-TEQ of
274	
274 275	(summed) PCBs; (c) WHO-TEQs of PCDDs and PCDFs; and (d) WHO-TEQ of
274 275 276	(summed) PCBs; (c) WHO-TEQs of PCDDs and PCDFs; and (d) WHO-TEQ of PCBs in faecal samples collected from two subspecies of wild (blue) and
274 275 276 277	(summed) PCBs; (c) WHO-TEQs of PCDDs and PCDFs; and (d) WHO-TEQ of PCBs in faecal samples collected from two subspecies of wild (blue) and captive (red) giant pandas. Bars (means \pm 1 SE of the mean from $N = 4$
274 275 276 277 278	(summed) PCBs; (c) WHO-TEQs of PCDDs and PCDFs; and (d) WHO-TEQ of PCBs in faecal samples collected from two subspecies of wild (blue) and captive (red) giant pandas. Bars (means \pm 1 SE of the mean from $N = 4$ independent replicates comprising three or four pooled samples) with different

283	Figure 4. Concentrations of heavy metals in faecal samples collected from two
284	subspecies of wild (blue) and captive (red) giant pandas. (a) Arsenic (As); (b)
285	Cadmium (Cd); (c) Chromium (Cr); (d) Lead (Pb). Bars (means \pm 1 SE of the
286	mean from $N = 4$ independent replicates comprising three or four pooled
287	samples) with different letters between the wild and captive pandas for the
288	same subspecies (A or B), or between Sichuan and Qinling subspecies (X or Y)
289	are significantly different (P < 0.05, <i>t</i> -test).
290	

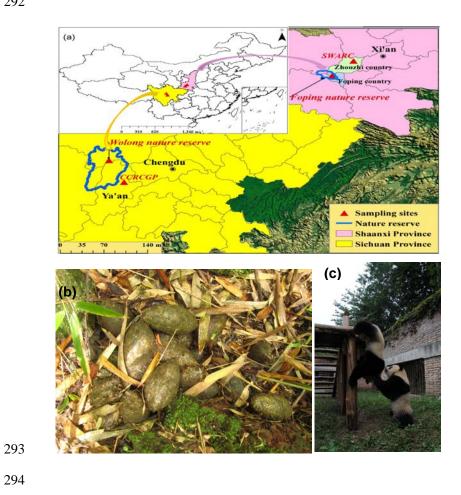
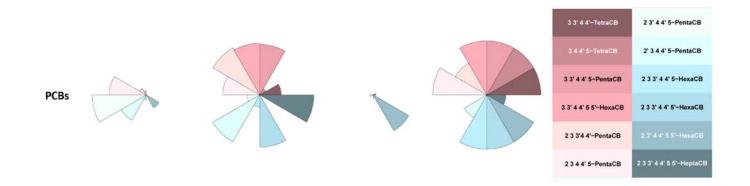


Figure 1



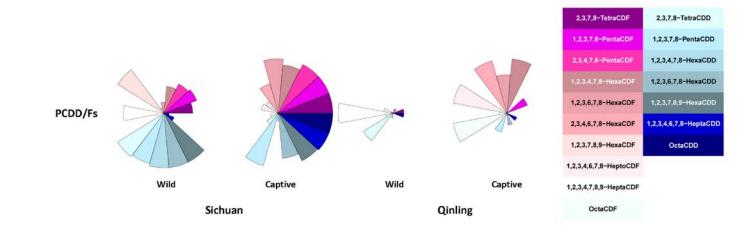
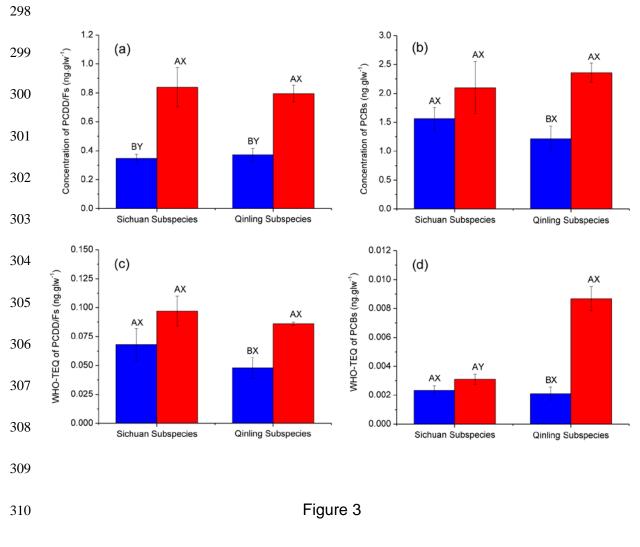
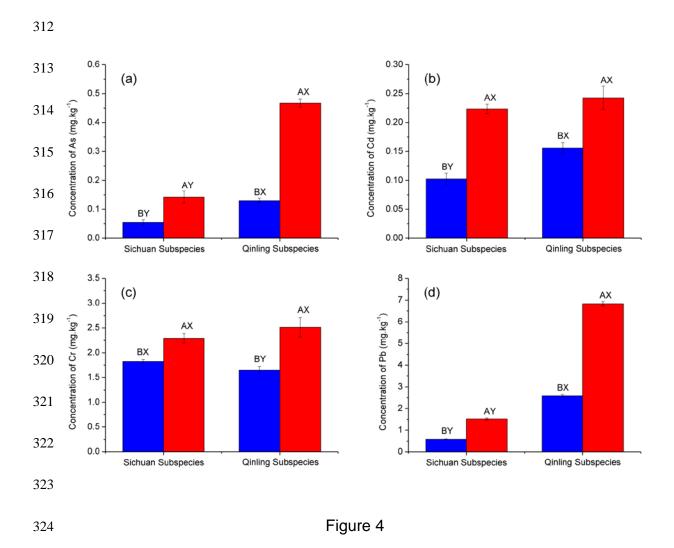


Figure 2





1	Captive pandas are at risk from environmental toxins
2	Yi-ping Chen, Lorraine Maltby, Qiang Liu, Yi Song, Ying-juan Zheng, Aaron M. Ellison,
3	Qing-yi Ma, Xiao-min Wu
4	
5	Supplemental Online Material
6	Additional Materials and Methods
7	Additional References
8	WebTables 1, 2
9	WebFigures 1 – 3
10	
11	
12	Additional Materials and Methods
13	
14	Sample collection
15	All faecal, plant, and feedstock samples were collected from the Wolong National
16	Nature Reserve in the Qionglai Mountains ("Wolong NNR": 30°45'-31°25'N,
17	102°52'-103°25'E), the Foping National Nature Reserve in the Qinling Mountains
18	("Foping NNR": 33°33'-33°46'N, 107°40'-55'E), the China Conservation and
19	Research Center for Giant Panda ("CCRCGP": 30°04'N, 102°59'E) and the Shaanxi
20	Wild Animal Research Center ("SWARC": 34°06'N, 108°32' E). The CCRCGP is the

largest captive breeding center for the Sichuan subspecies of the giant panda. It was
relocated to its current location in Bifengxia Ya'an city from Wolong after the 2008
Wenchuan earthquake. SWARC is located in Louguantai, Zhouzhi County, Xi'an city.
It was established in 1987 and is the only center for conservation of the Qinling
subspecies of the giant panda.

Faecal droppings of wild pandas were collected from 12 sites within the Wolong 26 NNR and 16 sites within the Foping NNR. Sampling locations were 10 km apart and 27 samples were pooled to give three samples/replicate from the Wolong NNR and four 28 29 samples/replicate from the Foping NNR. Droppings of captive pandas of the Sichuan subspecies were collected at CCRCGP whereas droppings of captive pandas of the 30 Qinling pandas were collected at SWARC. Droppings from either 12 individuals 31 32 (CCRCGP) or 16 (SWARC) individuals were sampled and pooled into four replicates each comprising of three or four independent samples. 33

34

35 Source of the Environmental Toxins

To investigate the source of the pollutants detected in panda droppings, the Qingling subspecies was studied in more detail. This subspecies was selected because there are about 350 individuals left (State Forestry Administration of the People's Republic of China.2015), so its conservation is much more urgent than that of the Sichuan subspecies. Further, as noted in the Results, the faecal droppings of the Qinling pandas contained significantly higher concentrations of As, Cd and Pb than droppings of the Sichuan subspecies. Fresh leaves of the primary bamboo fed to these pandas (*Fargesia qinlingensis*, *Bashania fargesii*) and mixed feedstock used to make nutrient-supplements for the Qingling subspecies were collected from the Foping NNR, from plants cultivated at SWARC, and from feedstock at SWARC. Twelve samples of each food type were collected per location and pooled to produce four replicates each consisting of three samples.

49

50 Heavy metal analysis

51 All samples were dried to constant mass at 60°C before being homogenized using a ball mill. Dried samples (500 mg) were placed into Teflon bombs to which were added 52 5 ml of HNO₃ for digestion with a microwave system (CEM, Mars 6, CEM, USA). After 53 54 digestion, samples were diluted to 50 mL with deionized water. Concentrations of cadmium (Cd), chromium (Cr), and lead (Pb) were measured using a graphite 55 furnace atomic absorption spectrometer (220-FS; Varian Company, USA.) with a 56 hollow cathode lamp (Vigorous Instruments Co., Ltd., Beijing, China) (Yu et al. 2001). 57 Concentrations of arsenic (As) were measured using an Atomic Fluorescence 58 Spectrometer (AF-7500; Beijing Dongxi Instruments Co., Ltd., China) with a hollow 59 cathode lamp (Vigorous Instruments Co., Ltd., Beijing, China) (Rahman et al. 2000). 60 61

62 Analysis of PCDDs, PCDFs, and PCBs

Samples (dropping, bamboo and feedstuff) were freeze-dried before being spiked
 with ¹³C-labeled surrogate standards (Environmental Protection Agency [EPA]

method 1613B and 1668A) and underwent accelerated solvent extraction with
dichlorinmethene: hexane (1:1). After determining the lipid content of each sample,
the extract was adjusted to 50 ml with hexane; 15 g of acid silica (30% w/w) was
added to remove lipids. The acid silica was stirred for 2 h and the extract was poured
through 5 g of anhydrous sodium sulfate. All of the extracts were concentrated to 2 ml
by rotary evaporation.

All solvents were purchased from Fisher (Fairlawn, NJ, USA). Silica gel was 71 obtained from Merck (silica gel 60; Darmstadt, Germany). Basic alumina was 72 73 obtained from Aldrich (Brockmann I, standard grade; Milwaukee, USA). Florisil was obtained from Riedel-de Haën (60-100 mesh ASTM; Seelze, Germany). Calibration 74 standard solutions, ¹³C₁₂-labeled surrogate standards, and ¹³C₁₂-labeled injection 75 standards were purchased from Wellington Laboratories (Guelph, Canada). 76 PCBs, PCDDs, and PCDFs were analyzed at the POP laboratory of the Research 77 Center for Eco-environmental Sciences, Chinese Academy of Sciences; all 78 79 concentrations were corrected for lipid weight. Sample extraction, cleanup, and chemical analysis followed established methods with some modifications (Liu et 80 al.2006; Li et al. 2008). Twenty-five PCB congeners, including 12 dioxin-like 81 congeners, were quantified by an isotope dilution method using high-resolution gas 82 chromatography coupled with high-resolution mass spectrometry (HRGC/HRMS). 83 Total organic carbon (TOC) concentration was analyzed on a TOC Analyzer (O.I 84 Analyzer; College Station, TX, USA). A 0.1-g sample was weighed and loaded into 85 the combustion cup, which was packed with quartz wool. Prior to combustion, the 86

samples were wetted with 5% phosphoric acid and heated to 250°C for 1 min to 87 purge inorganic carbon. The signal was detected by non-dispersed infrared (NDIR) 88 detection when flashed at 900 °C for 6 min in the combustion house. 89 The guantification of 17 PCDD/PCDF homologues was done using 90 HRGC/HRMS on an Agilent 6890 gas chromatograph coupled with an Autospec 91 92 Ultima mass spectrometer (Waters Micromass, Manchester, UK) operating in the EI mode at 35 eV with the trap current was 600 IA. The GC was equipped with a CTC 93 PAL autosampler. One or two µL samples were injected in splitless mode (splitless 94 95 time, 2 min for PCDD/Fs) in a DB-5MS fused silica capillary column (60 m for PCDD/Fs and PCBs) with helium as carrier gas at a constant flow rate of 1.2 ml/min. 96 The oven temperature programs were as follows: for PCDD/Fs, start 150°C held for 3 97 min, 150-230°C at 20°C min⁻¹ held for 18 min, 230-235°C at 5°C min⁻¹ held for 10 min, 98 235-320°C at 4°C min⁻¹ held for 3 min; for PCBs, start 120°C held for 1 min, 99 120-150°C at 30 °C min⁻¹, 150-300°C at 2.5°C min⁻¹ held for 1 min. 100

101

102 **Quality control and quality assurance**

All data were subject to quality control and quality assurance. All glassware was
 washed two times with distilled water, and then with dichloromethane after use. After
 washing, glassware was dried for 6 hours at 400 °C in a muffle furnace.

All performance criteria required for the analysis of PCBs and PCDD/PCDFs
 followed US EPA methods (1668A and 1613B).¹³C-labeled surrogated standards

(1668A-LCS and 1613-LCS) were spiked in the sample for qualification and 108 quantification, and ¹³C-labeled injection standards (EPA 68A-IS and 1613-IS) were 109 added for recovery calculation. The recoveries of the surrogate standards ranged 110 from 76.7±25.2% and 49.2±13.6% for PCB sand PCDD/PCDFs, respectively, which 111 met the requirements of US EPA methods 1668 A and 1613 B. Limit of detection (LOD) 112 in the sample was defined as a signal to noise (S/N) ratio = 3. The LOD values were 113 in the range of 0.01–0.82pg g^{-1} for PCBs and 0.04–8.40 pg g^{-1} for PCDD/PCDFs. 114 Laboratory blanks were analyzed with samples quality control at set intervals, and 115 there was no detection of target compounds in the blanks. 116

118 Additional References

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WebTable 1. Concentrations of PCDD and PCDF congeners in faecal droppings from 128 wild and captive pandas of both Sichaun (SS) and Qinling (QS) subspecies. Values 129 are means ± 1 standard error of the mean for N = 4 independent replicates each 130 comprising three or four pooled subsamples. 131

Congeners (pg.glw ⁻¹)	Wild SS	Captive SS	Wild QS	Captive QS
2,3,7,8-TetraCDF	7.802 ± 2.509	14.278 ± 4.946	3.402 ± 1.244	0.536 ± 0.473
1,2,3,7,8-PentaCDF;	10.098 ± 2.402	14.536 ± 5.413	2.074 ± 1.278	7.117 ± 7.117
2,3,4,7,8-PentaCDF;	11.585 ± 2.027	18.100 ± 3.694	3.214 ± 1.565	1.816 ±1.816
1,2,3,4,7,8-HexaCDF	11.002 ± 1.477	15.934 ± 6.152	4.868 ± 2.764	17.300 ± 9.700
1,2,3,6,7,8-HexaCDF	8.900 ± 3.097	19.125 ± 5.736	6.352 ± 3.043	15.397 ± 8.576
2,3,4,6,7,8-HexaCDF;	6.568 ± 3.119	15.621 ± 5.895	4.507 ± 2.000	24.320 ± 8.676
1,2,3,7,8,9-HexaCDF	6.582 ± 4.347	2.186 ± 2.001	0.800 ± 0.289	0.579 ± 0.579
1,2,3,4,6,7,8-HeptoCDF	42.796 ± 14.568	65.054 ± 13.461	54.471 ± 17.196	113.048 ± 37.687
1,2,3,4,7,8,9-HeptaCDF	4.365 ± 2.732	0.566 ± 0.382	5.661 ± 3.843	1.283 ± 1.222
OctaCDF	34.857±5.274	72.098 ± 31.329	67.689 ± 19.035	291.502 ± 165.188
2,3,7,8-TetraCDD	2.521 ± 1.456	0.631 ± 0.494	1.591 ± 1.334	
1,2,3,7,8-PentaCDD;	2.846 ± 1.582	2.952 ± 1.450	0.356 ± 0.235	1.289 ± 1.289
1,2,3,4,7,8-HexaCDD	7.514 ± 3.456	1.051 ± 0.698	0.680 ± 0.324	0.053 ± 0.053
1,2,3,6,7,8-HexaCDD	5.676 ± 2.729	4.926 ± 2.046	1.247 ± 0.428	2.216 ± 2.174
1,2,3,7,8,9-HexaCDD	6.654 ± 4.893	6.483 ± 3.772	\Leftrightarrow	0.288 ± 0.288
1,2,3,4,6,7,8-HeptaCDD	37.442 ± 5.204	87.879 ± 24.617	24.736 ± 8.696	37.214 ± 22.064
OctaCDD	148.753 ± 8.656	469.836 ± 97.490	189.186 ± 29.664	115.741 ± 92.914

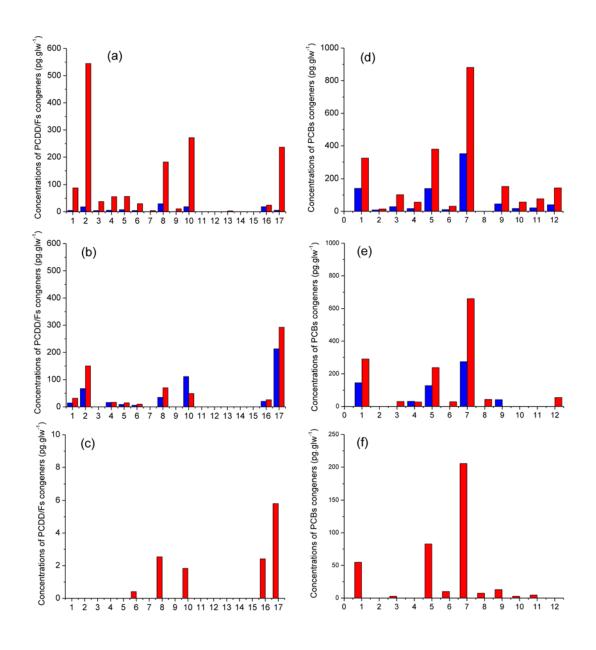
 \bigstar less than the limit of determination. 133

134

WebTable 2. Concentrations of PCB congeners in faecal droppings from wild and captive pandas of both Sichaun (SS) and Qinling (QS) subspecies. Values are means ± 1 standard error of the mean for N = 4 independent replicates each comprising three or four pooled subsamples.

Congeners(pg.glw ⁻¹)	Wild of SS	Captive of SS	Wild of QS	Captive of QS
3,3',4,4'-TetraCB	72.745 ± 10.721	151.170 ± 22.013	82.564 ± 32.295	270.217 ± 44.520
3,4,4',5-TetraCB	\overleftrightarrow	5.768 ± 4.476	Δ	45.409 ± 19.320
3,3',4,4',5-PentaCB	$7.910{\pm}3.856$	155.379 ±101.403	9.495 ± 5.967	165.775 ± 99.209
3,3',4,4',5,5'-HexaCB	1.721 ± 1.629	21.159 ± 14.862	${\leftrightarrow}$	22.145 ± 22.145
2,3,3'4,4'-PentaCB	295.723 ± 31.734	716.133 ±246.199	230.065 ± 57.408	556.994 ± 204.311
2,3,4,4',5-PentaCB	54.729 ± 6.431	54.830 ± 15.421	21.003 ± 8.061	70.455 ± 9.945
2,3',4,4',5-PentaCB	976.121 ±57.739	788.122 ± 446.010	744.270 ± 135.106	731.055 ± 238.280
2',3,4,4',5-PentaCB	54.104 ± 5.305	79.313 ±19.382	25.294 ± 3.305	51.228 ±12.658
2,3,3',4,4',5-HexaCB	64.191 ± 4.816	68.530 ±25.965	61.105 ± 4.694	94.781 ± 24.436
2,3,3',4,4',5'-HexaCB	11.179 ± 2.846	35.653 ±13.606	12.693 ± 9.960	36.634 ± 6.900
2,3',4,4',5,5'-HexaCB	24.245 ± 7.676	18.822 ± 10.687	33.975 ± 5.332	38.804 ± 13.105
2,3,3',4,4',5,5'-HeptaCB	${\sim}$	4.982 ± 4.860	☆	1.763 ± 1.763

141 \Leftrightarrow less than the limit of determination.





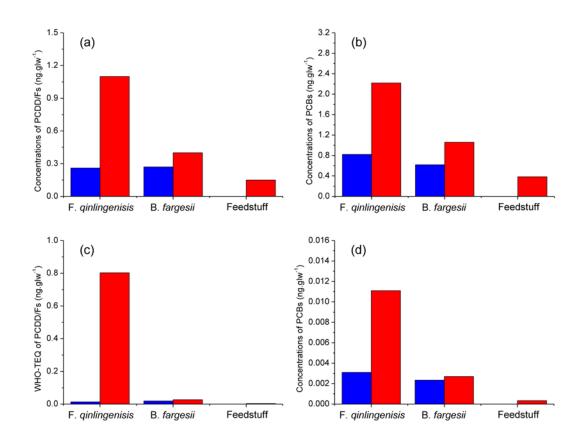
144 WebFigure 1. The concentrations of congeners of (a, b, c) PCDDs and PCDFs; and

145 (d, e, f) PCBs in the bamboos *Fargesia qinlingensis* and *Bashania fargesii*, and

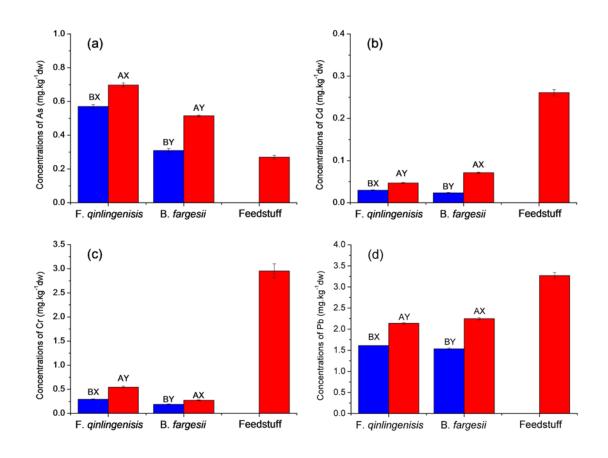
146 feedstuff of wild (blue) and captive (red) pandas. Numbers on the *x*-axis of panels (a),

- (b) and (c) denote different congeners of PCDD/F. 1=2,3,7,8-TetraCDF;
- 148 2=1,2,3,7,8-PentaCDF; 3=2,3,4,7,8-PentaCDF; 4=1,2,3,4,7,8-HexaCDF;
- 149 **5=1,2,3,6,7,8-HexaCDF**; **6=2,3,4,6,7,8-HexaCDF**; **7=1,2,3,7,8,9-HexaCDF**;
- 150 8=1,2,3,4,6,7,8-HeptoCDF; 9=1,2,3,4,7,8,9-HeptaCDF; 10=OctaCDF;

- 151 **11=2,3,7,8-TetraCDD**; **12=1,2,3,7,8-PentaCDD**; **13=1,2,3,4,7,8-HexaCDD**;
- 152 14=1,2,3,6,7,8-HexaCDD; 15=1,2,3,7,8,9-HexaCDD; 16=1,2,3,4,6,7,8-HeptaCDD;
- 153 17=OctaCDD; Numbers on the x-axis of panels (d), (e) and (f) denote different
- 154 congeners of PCBs. 1=3,3',4,4'-TetraCB; 2=3,4,4',5-TetraCB; 3=3,3',4,4',5-PentaCB;
- 155 **4=3,3',4,4',5,5'-HexaCB**; **5=2,3,3'4,4'-PentaCB**; **6=2,3,4,4',5-PentaCB**;
- 156 **7=2,3',4,4',5-PentaCB**; **8=2',3,4,4',5-PentaCB**; **9=2,3,3',4,4',5-HexaCB**;
- 157 10=2,3,3',4,4',5'-HexaCB; 11=2,3',4,4',5,5'-HexaCB; 12= 2,3,3',4,4',5,5'-HeptaCB.



- 160 WebFigure 2. Concentrations of (a) **PCDDs** and **PCDFs**, (b) **PCBs**, (c) WHO-TEQ
- 161 of PCDDs and PCDFs, and (d) WHO-TEQ of PCBs in the bamboos Fargesia
- *qinlingensis* and *Bashania fargesii*, and from panda feedstuff. Bars are the value from
- a single replicate comprising five pooled samples.



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WebFigure 3. Concentrations of heavy metals in Qinling subspecies food of wild (blue) and captive (red) from bamboo species (*Fargesia qinlingensis* and *Bashania fargesii*) and feedstuff. (a) Arsenic (As); (b) Cadmium (Cd); (c) Chromium (Cr); (d) Lead (Pb). Bars (means \pm 1 SE of the mean from N = 4 independent replicates from three pooled samples) with different letters between the two bamboo species (*Fargesia qinlingensis* and *Bashania fargesii*) (A and B) or between the bamboos fed to wild and captive (X or Y) are significantly different (*P* < 0.05, t-test).