

Supporting Information for

Cellular internalisation of dissolved cobalt ions from ingested CoFe_2O_4 nanoparticles: *in vivo* experimental evidence

Environmental Science and Technology

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Spectrum	C	O	Mg	Si	Ca	Fe	Co	Pd	Au	Total
Spectrum 1	21.63	32.85		0.58	1.33	27.55	15.13	0.93		100.00
Spectrum 2	15.28	38.51	6.54	1.03	15.27	14.61	7.49	1.28		100.00
Spectrum 3	19.29	27.26	0.26	0.53	1.33	32.43	16.77	1.15	0.97	100.00
Spectrum 4	45.59	34.17	0.57	1.24	2.04	9.85	5.39	1.14		100.00
Spectrum 5		38.20	0.55	1.70	2.50	36.61	18.93	1.53		100.00
Max.	45.59	38.51	6.54	1.70	15.27	36.61	18.93	1.53	0.97	
Min.	15.28	27.26	0.26	0.53	1.33	9.85	5.39	0.93	0.97	

Table S1. EDX spectra analyses of abaxial leaf surface. EDX spectra analyses of abaxial leaf surface covered with suspension of CoFe_2O_4 nanoparticles (nominal concentration 5000 $\mu\text{g/g}$). Analytical results in weight % for analysed elements in all five spectra taken in areas marked in Figure 1B.

		CoFe_2O_4 (2000 μg Co/mL)	CoFe_2O_4 (5000 μg Co/mL)	CoCl_2 (2000 μg Co/mL)	CoCl_2 (5000 μg Co/mL)
Concentration of Co ions ($\mu\text{g/mL}$); mean \pm SD, n = 3	supernatant diluted with deionized water (1:1)	0,162 \pm 0,027	below detection limit	2096 \pm 14	4451 \pm 151
	supernatant diluted with 1 M HCl (1:1)	0,215 \pm 0,039	below detection limit	2127 \pm 48	4904 \pm 88
Concentration of Fe ions ($\mu\text{g/mL}$); mean \pm SD, n = 3	supernatant diluted with deionized water (1:1)	0,238 \pm 0,122	below detection limit	Not measured	Not measured
	supernatant diluted with 1 M HCl (1:1)	0,315 \pm 0,121	0,099 \pm 0,029	Not measured	Not measured

Table S2 Concentration of Co and Fe ions in supernatants. Concentration of Co and Fe ions in supernatants of centrifuged suspensions of CoFe_2O_4 NPs and CoCl_2 solution, as measured with flame AAS.

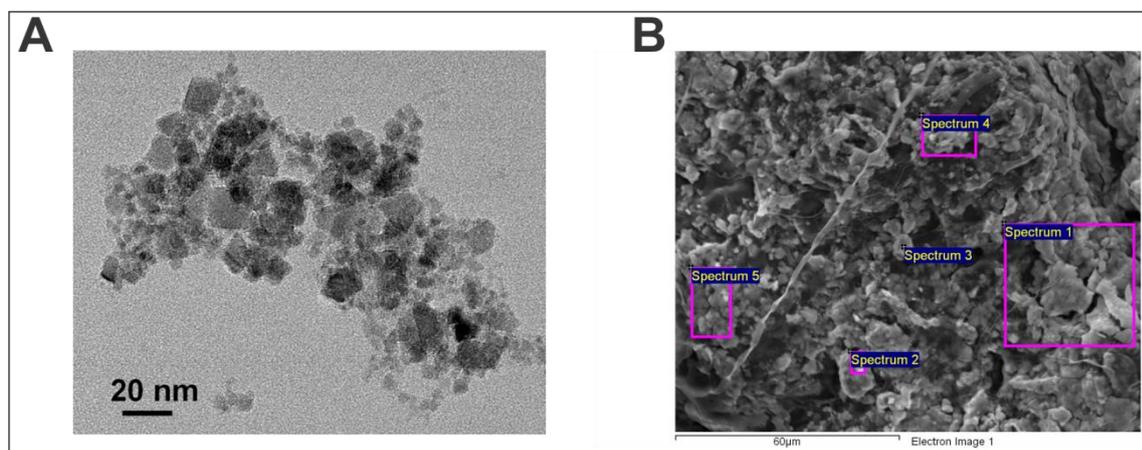


Figure S1. SEM/EDX analyses of nanoparticles on leaves. SEM characterization of CoFe_2O_4 nanoparticles and SEM/EDX analyses of CoFe_2O_4 nanoparticles on leaves from experiment D. (A) SEM micrograph of CoFe_2O_4 NPs. (B) Abaxial leaf surface covered with suspension of CoFe_2O_4 nanoparticles (nominal concentration $5000 \mu\text{g/g}$) with marked areas where EDX spectra were taken.

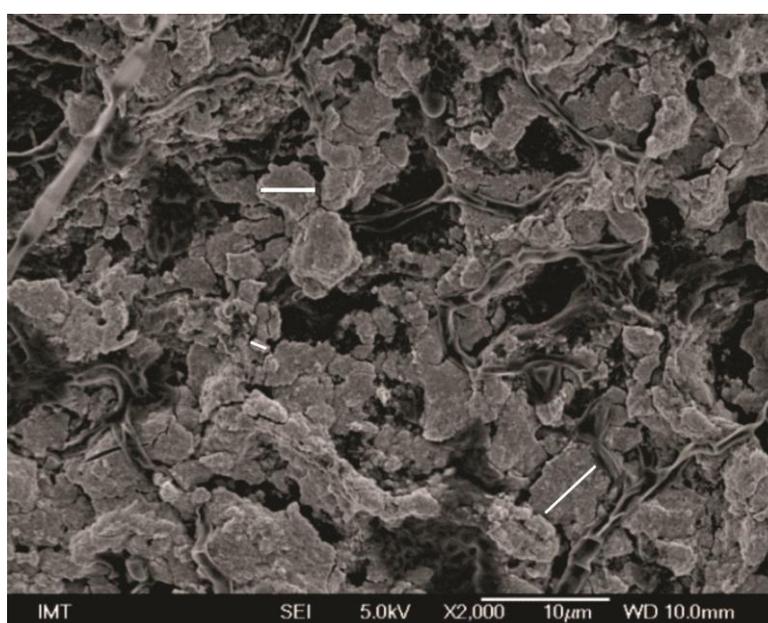


Figure S2. SEM image of the surface of leaf covered with CoFe_2O_4 nanoparticles. The scanning electron microscope (SEM) image of the surface of leaf (food) on which suspension of CoFe_2O_4 nanoparticles at a concentration $5000 \mu\text{g/ml}$ was applied. White lines

indicating some of agglomerates show that the size of individual agglomerates varied from 1 to 10 μm .

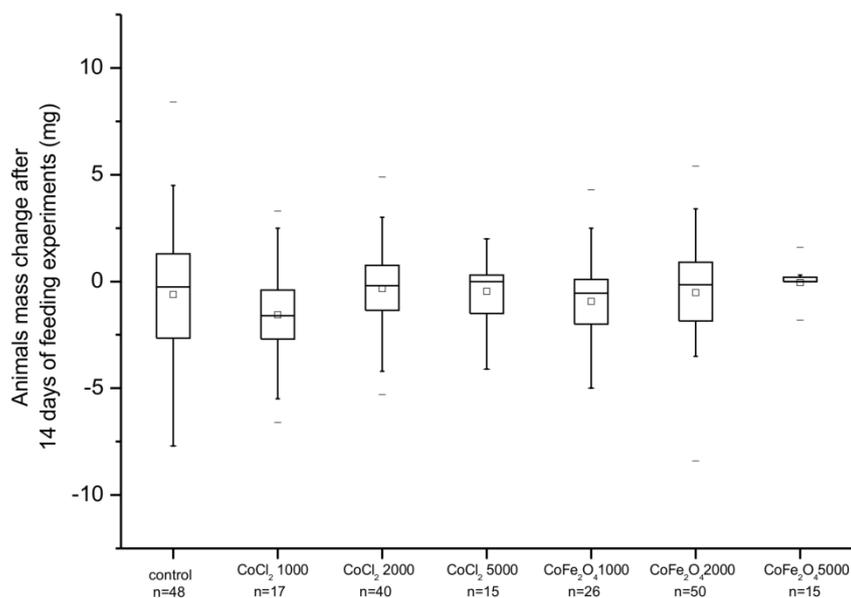


Figure S3. Mass change of animals after experiments. Mass change of animals (mg) fed for 14 days on food dosed with CoCl₂ or CoFe₂O₄ nanoparticles. Symbols on the box plot represent minimum and maximum data values (whiskers), mean value (\square), 75th percentile (upper edge of box), 25th percentile (lower edge of box), median (line in box) and max and min value (-). There was no statistically significant differences between exposed and control animals. Nominal exposure concentrations (1000, 2000 or 5000 μg CoCl₂ or nano-CoFe₂O₄/g of leaf) and the number of animals (n) per group are provided on the x-axis.

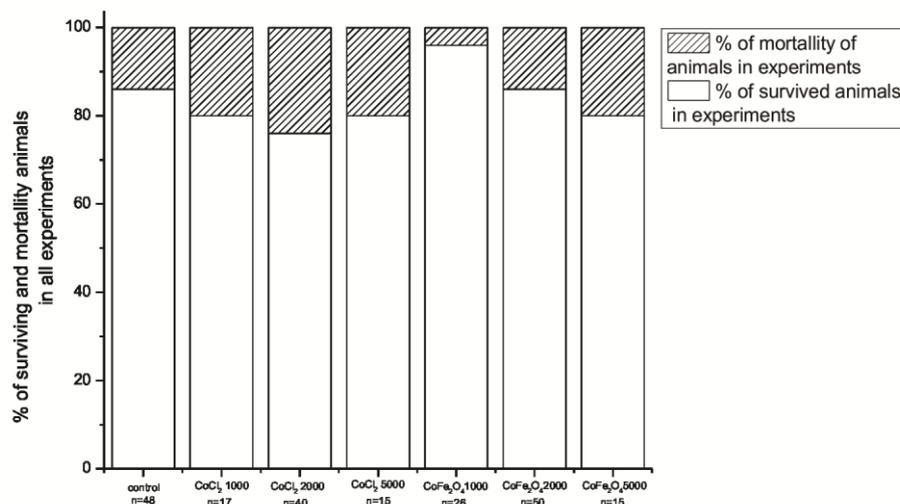


Figure S4. Survival of animals after feeding experiments. Survival of animals fed for 14 days on food dosed with CoCl₂ or CoFe₂O₄ nanoparticles. Nominal exposure concentrations (1000, 2000 or 5000 µg CoCl₂ or nano-CoFe₂O₄/g of leaf) and the number of animals (n) per group are provided on the x-axis.

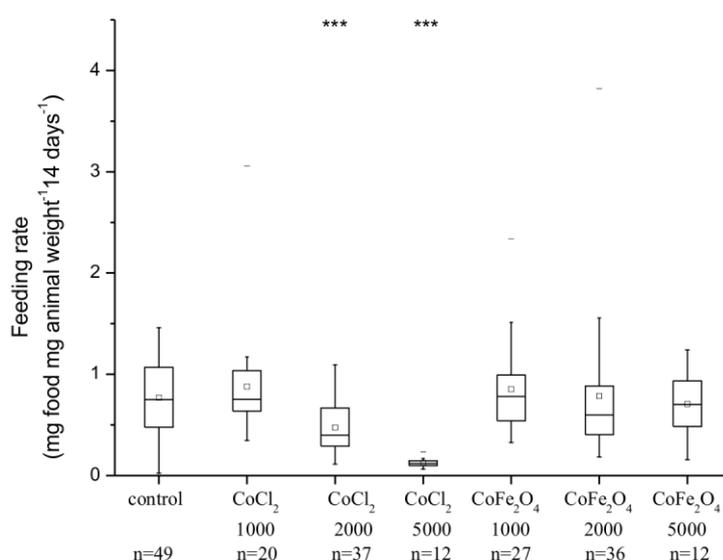


Figure S5. Feeding rate of animals fed on CoCl₂ or CoFe₂O₄ nanoparticles. Feeding rate (mg of consumed leaf per mg of animal mass) of animals fed for 14 days on food dosed with CoCl₂ or CoFe₂O₄ nanoparticles. Symbols on the box plot represent minimum and maximum

data values (whiskers), mean value (\square), 75th percentile (upper edge of box), 25th percentile (lower edge of box), median (line in box) and max and min value (-). Statistically significant differences between exposed and control animals are marked with *** ($p < 0.001$). Nominal exposure concentrations (1000, 2000 or 5000 $\mu\text{g CoCl}_2$ or nano- $\text{CoFe}_2\text{O}_4/\text{g}$ of leaf) and the number of animals (n) per group are provided on the x-axis.