Electronic Supplementary Material (ESI) for Environmental Science: Nano.

ELECTRONIC SUPPLEMENTARY INFORMATION

FOR:

Cerium (IV) oxide nanoparticles induce sublethal changes in honeybees after chronic exposure†

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3.1.4 SEM micrograph and EDX of $nCeO_2$ stock dispersion.

3.1.5 SEM micrograph and EDX of $nCeO_2$ stock dispersion.

1. CHARACTERISATION OF THE $nCeO_2$

Table S1: Supplement to the results of the characterisation of $nCeO_2$ dispersions in the 1.5 M sucrose solution. In parallel with the $nCeO_2$ dispersions we performed ultracentrifugation and ICP-MS measurements with Ce standard at concentrations 2, 10, 50, 250, and 500 mgL^{-1} in 1.5 M sucrose to evaluate the adsorption of Ce onto the quick-seal tubes and effectiveness of microwave assisted digestion. The table comprises the measured Ce concentration before and after ultracentrifugation in acid-digested samples, recovery (%), share of the Ce ions species in the supernatant (%), and the pH.

Ce standard, nominal conc. (mgL^{-1})	**Ce nominal conc.** (mgL^{-1})	**Ce measured conc.** ($mg\ L^{-1}$)	**Recovery** (%)	**Ce measured conc.** (%)	**pH**
Ce standard, before ultracentrifugation					
2	2	2.34	117.00	/	2.63
10	10	12.59	125.90	/	1.87
50	50	60.81	121.62	/	1.24
250	250	280.16	112.64	/	0.62
500	500	541.24	108.08	/	0.47
Ce standard, after ultracentrifugation					
2	2	2.35	117.50	100.43	1.94
10	10	11.57	115.70	91.90	1.76
50	50	58.21	116.42	95.72	1.21
250	250	288.00	115.20	102.80	0.65
500	500	526.64	105.33	97.30	0.41

2. FEEDING TESTS WITH SUMMER AND WINTER HONEYBEES

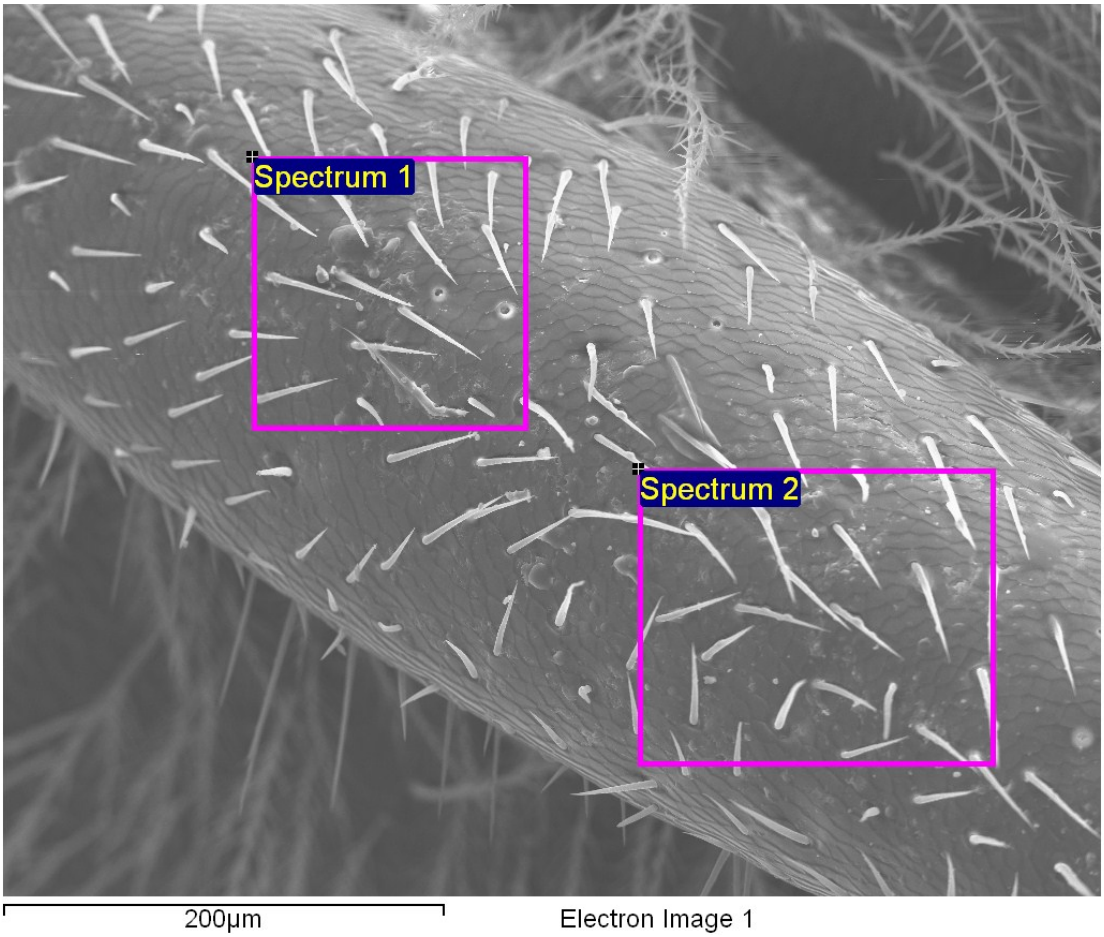
Table S2: Mortality (in %) of summer and winter honeybees *Apis mellifera carnica* that had been exposed for 9 days to $nCeO_2$ spiked food in the feeding test. Feeding test with winter honeybees was done in triplicates per treatment therefore mortality is given as a mean of the triplicates (± SE in %). Number (N) of the honeybees per treatment and number of dead honeybees per treatment are also given.

Treatment, $nCeO_2$ (mgL^{-1})	**N bees per treatment**	**N dead bees per treatment**	**Mortality** (± SE in %)
Chronic feeding test with summer bees			
0	27	0	0.00
2	28	2	7.14
10	24	1	4.17
50	24	3	12.50
250	22	1	4.55
500	25	1	4.00
Chronic feeding test with winter bees			
0	44	1	2.56±2.56
250	47	1	2.22±2.22

3. ANNEX 1 TO THE FIGURE 1

3.1 Results from the SEM EDX analysis

3.1.1 SEM micrograph and EDX of honeybee's antenna after 9 days of exposure to $nCeO_2$ spiked food (50 mgL^{-1}).

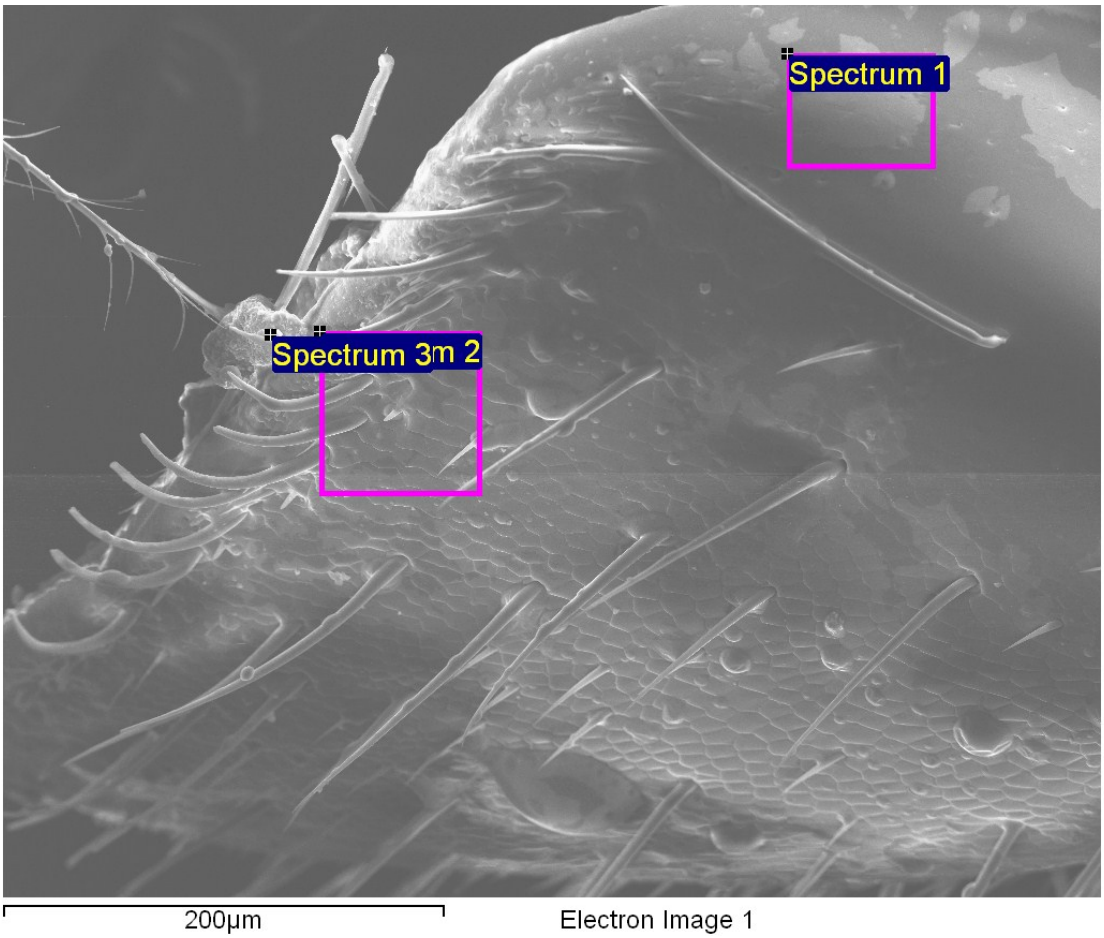


Processing option : All elements analysed (Normalised)

Spectrum	In stats.	C	O	K	Ca	Ce	Au	Total
Spectrum 1	Yes	62.80	23.20	1.36	0.62	0.00	12.01	100.00
Spectrum 2	Yes	63.94	23.88	0.95	0.57	0.00	10.66	100.00
Mean		63.37	23.54	1.15	0.60	0.00	11.34	100.00
Std. deviation		0.80	0.48	0.29	0.04	0.00	0.96	
Max.		63.94	23.88	1.36	0.62	0.00	12.01	
Min.		62.80	23.20	0.95	0.57	0.00	10.66	

All results in weight%

3.1.2 SEM micrograph and EDX of honeybee's mouth parts after 9 days of exposure to $nCeO_2$ spiked food (50 mgL^{-1}).

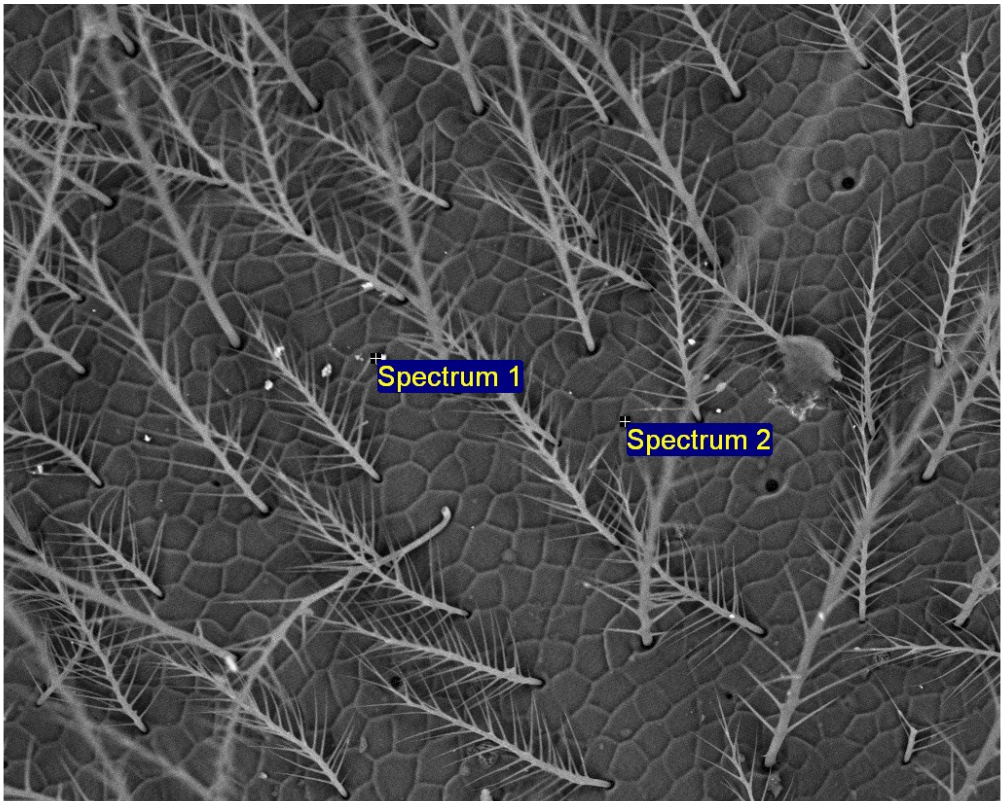


Processing option : All elements analysed (Normalised)

Spectrum	In stats.	C	O	Ca	Ce	Au	Total
Spectrum 1	Yes	62.07	30.85	0.00	0.00	7.08	100.00
Spectrum 2	Yes	59.46	31.91	0.37	0.00	8.26	100.00
Spectrum 3	Yes	44.18	35.54	20.28	0.00	0.00	100.00
Mean		55.24	32.77	6.88	0.00	5.11	100.00
Std. deviation		9.66	2.46	11.60	0.00	4.47	
Max.		62.07	35.54	20.28	0.00	8.26	
Min.		44.18	30.85	0.00	0.00	0.00	

All results in weight%

3.1.3 SEM micrograph and EDX of honeybee's head after 9 days of exposure to $nCeO_2$ spiked food (50 mgL^{-1}).

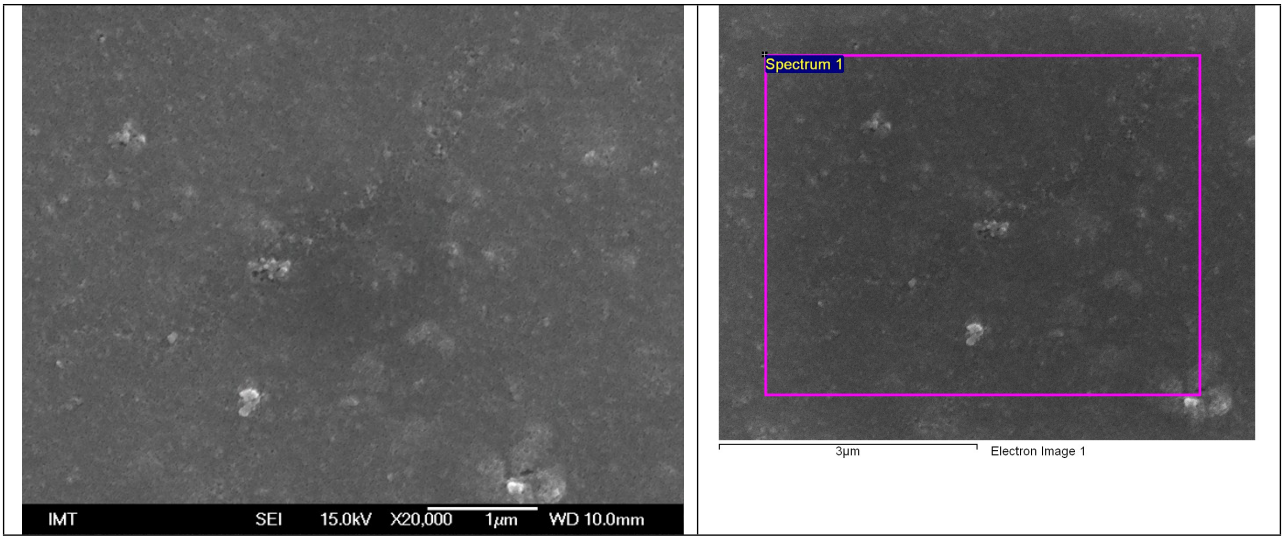


Processing option : All elements analysed (Normalised)

Spectrum	In stats.	C	O	Na	Mg	Al	Si	P	Cl	K	Ca	Ti	Total
Spectrum 1	Yes	40.53	47.10	0.34	0.27			0.20	0.27	0.19	11.11		100.00
Spectrum 2	Yes	54.42	38.12		0.20	0.75	0.88		0.36	0.23	4.38	0.66	100.00
Max.		54.42	47.10	0.34	0.27	0.75	0.88	0.20	0.36	0.23	11.11	0.66	
Min.		40.53	38.12	0.34	0.20	0.75	0.88	0.20	0.27	0.19	4.38	0.66	

All results in weight%

3.1.4 SEM micrograph and EDX results of $nCeO_2$ stock dispersion.

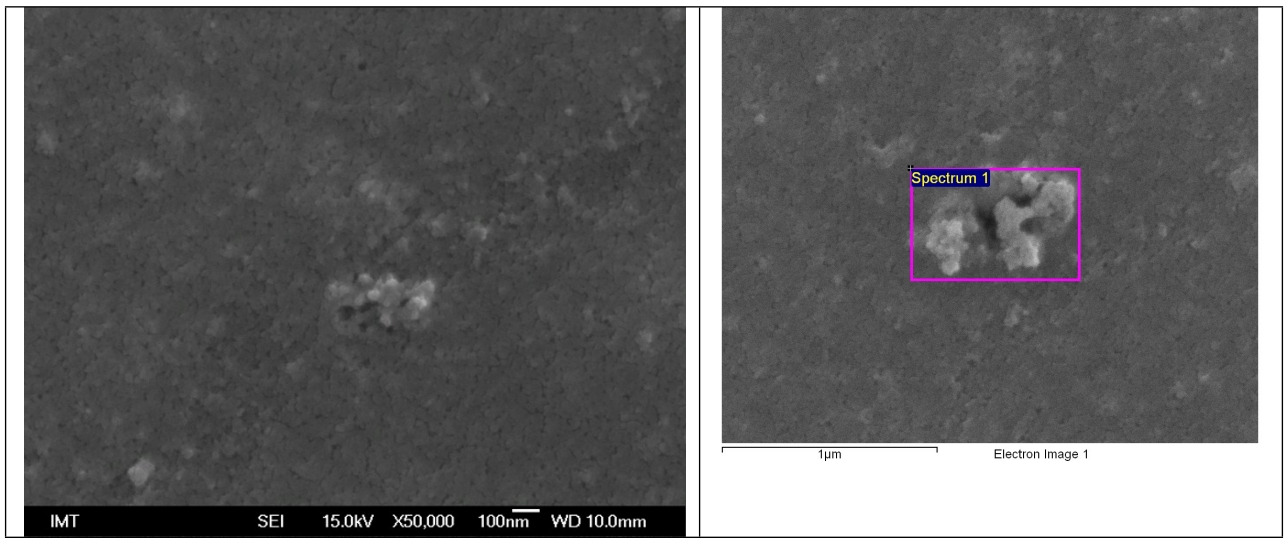


Processing option : All elements analysed (Normalised)

Spectrum	In stats.	O	Cl	Ca	Ce	Au	Total
Spectrum 1	Yes	15.76	6.37	2.94	68.16	6.77	100.00
Mean		15.76	6.37	2.94	68.16	6.77	100.00
Std. deviation		0.00	0.00	0.00	0.00	0.00	
Max.		15.76	6.37	2.94	68.16	6.77	
Min.		15.76	6.37	2.94	68.16	6.77	

All results in weight%

3.1.5 SEM micrograph and EDX of $nCeO_2$ stock dispersion.



Processing option : All elements analysed (Normalised)

Spectrum	In stats.	O	Cl	Ca	Ce	Au	Total
Spectrum 1	Yes	15.85	6.22	3.01	66.80	8.12	100.00
Mean		15.85	6.22	3.01	66.80	8.12	100.00
Std. deviation		0.00	0.00	0.00	0.00	0.00	
Max.		15.85	6.22	3.01	66.80	8.12	
Min.		15.85	6.22	3.01	66.80	8.12	

All results in weight%