10.1071/EN19061_AC

Environmental Chemistry 2019, 16(6), 419-429

Supplementary Material

Copper toxicity to *Folsomia candida* in different soils: a comparison between nano and conventional formulations

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Information on Agro-formulations used in the present study can be found below:

Bordeaux mixture® ($CuSO_4$)

http://jovagro.cdn.reage.pt/sites/default/files/Ficha%20tecnica%20Calda%20Bordalesa%20Azul.%20docx.pdf

Champion® $Cu(OH)_2$

http://www.nufarm.pt/produtos?t=Fungicidas&q=CHAMPION%20WP

Cupravit® $H_6Cl_2Cu_4O_6$

https://cropscience.bayer.pt/internet/produtos/produto.asp?id_produto=25

Nordox® Cu_2O

http://www.massoagro.com/pt/produtos/fungicidas/21136-cobre-nordox-75-wg-pt

Kocide 3000® $Cu(OH)_2$ NP

http://gcrec.ifas.ufl.edu/static/docs/pdf/strawberry-pathology/Fung-label/2008/kocide-3000.pdf

Table S1: Summary of Cu concentrations tested for the commercial formulations and in the pure forms (as mg Cu.kg $soil^{-1}$).

Copper concentrations (mg Cu.kg $soil^{-1}$)		
Commercial formulations	Bordeaux mixture® ($CuSO_4$)	500; 1000; 2500; 4000; 5500; 7000
	Champion® ($Cu(OH)_2$)	17; 50; 150; 450; 900; 1800; 3600
	Cupravit® ($H_6Cl_2Cu_4O_6$)	19; 56; 167; 500; 1500; 4500; 9000; 18000
	Nordox® (Cu_2O)	17; 50; 150; 450; 900; 1800
	Kocide® ($Cu(OH)_2$) NP	11; 33; 98; 239; 586; 1171
Pure forms	Copper sulfate ($CuSO_4$)	100; 200; 400; 800; 1600
	Copper hydroxide ($Cu(OH)_2$)	17; 50; 150; 450; 900; 1800

Table S2: Toxicity endpoints (NOEC, LOEC and EC_{50}) with standard error for *Folsomia candida* exposed to different copper pure forms for 28 days, in Lufa 2.2 and Lufa 2.1 with two spiking equilibrium time procedures. ± SE – Standard error.

Compound		**NOEC (mg Cu/kg)**			**LOEC (mg Cu/kg)**			**EC_{50}±SE (mg Cu/kg)**		
		Lufa 2.2		Lufa 2.1	Lufa 2.2		Lufa 2.1	Lufa 2.2		Lufa 2.1
Time for exposure after spiking		0h	48h	48h	0h	48h	48h	0h	48h	48h
Pure forms	Copper sulfate $CuSO_4$	200	<100	200	400	100	400	564±50.2	470±121.4	213±17.8
	Copper hydroxide $Cu(OH)_2$	<17	17	<17	17	50	17	57±50.8	18±29.9	6±2.9

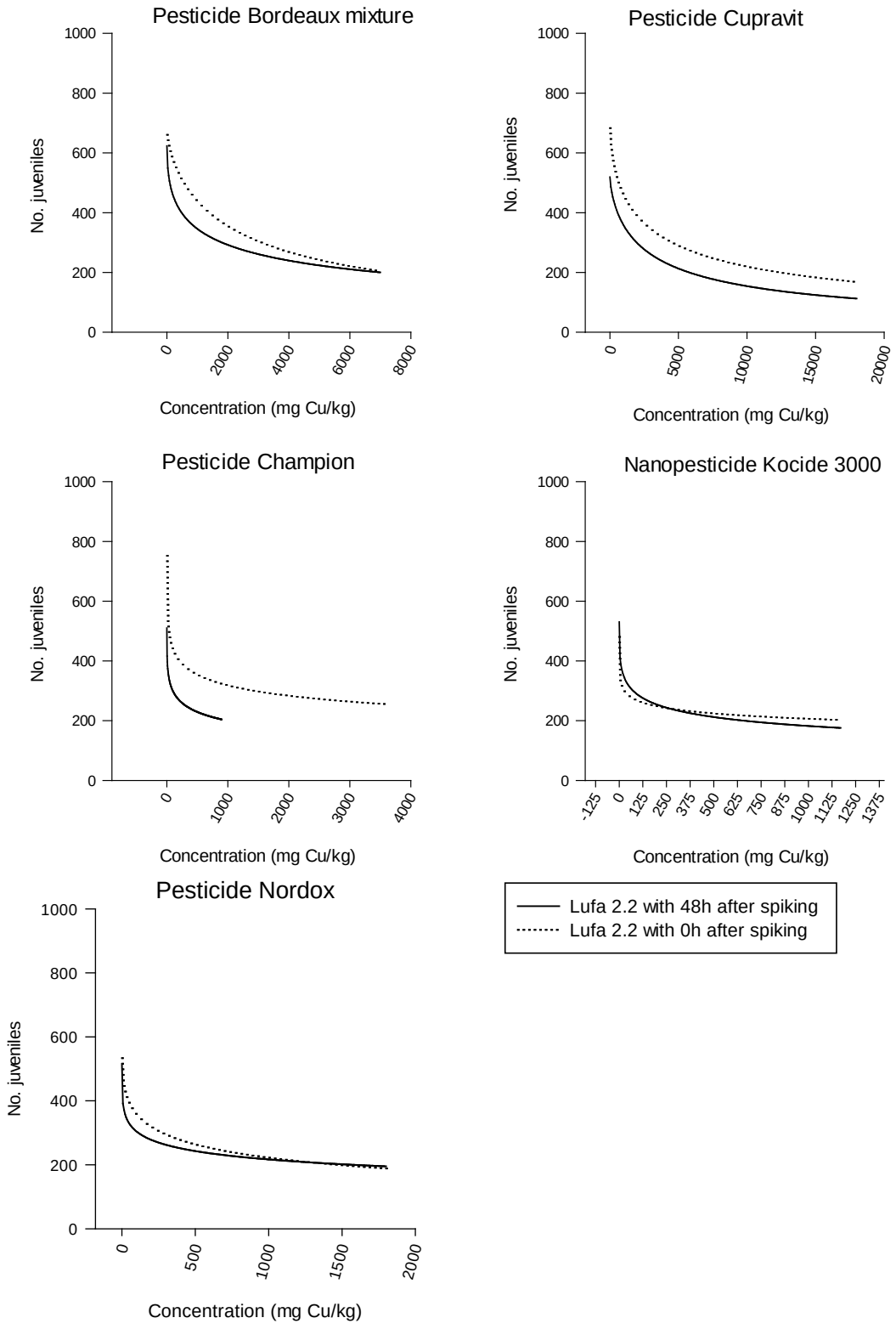


Figure S1: EC_{50} curves for reproductive output of *Folsomia candida* (juveniles average ±SD) exposed to five Cu pesticides: Bordeaux mixture®, Cupravit®, Champion®, Kocide 3000® and Nordox® testing two different spiking equilibrium times (0h and 48h).

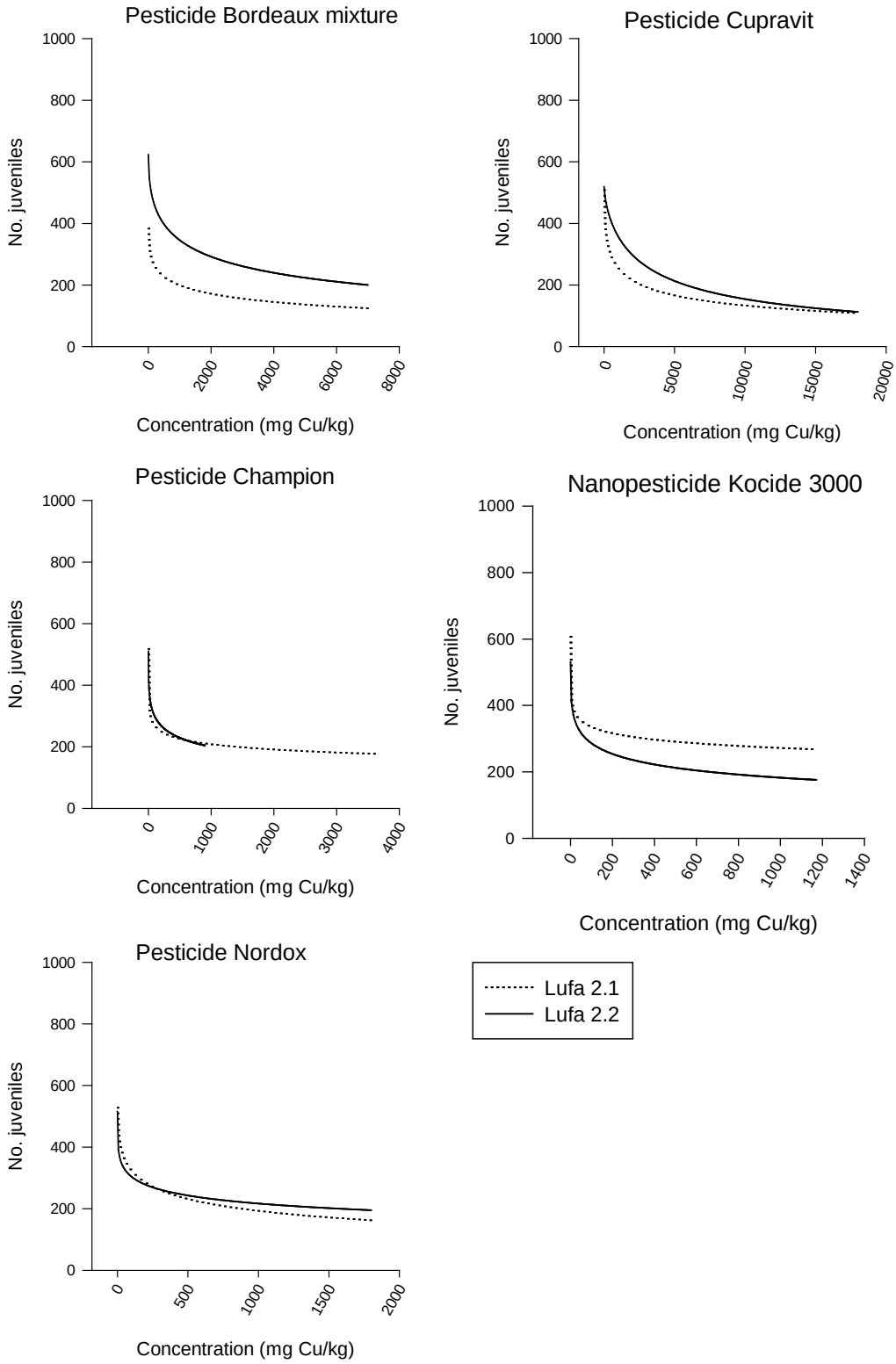


Figure S2: EC_{50} curves for reproductive output of *Folsomia candida* (juveniles average ±SD) exposed to five Cu pesticides: Bordeaux mixture®, Cupravit®, Champion®, Kocide 3000® and Nordox® in two different soils (Lufa 2.1 and Lufa 2.2).

Table S3: Copper concentrations in porewater from Lufa 2.2 and 2.1 soils spiked with copper pure forms and pesticides.

Bordeaux mixture®	Porewater concentrations (mg Cu/L)				
	Lufa 2.2			Lufa 2.1	
	Day 1	Day 1	Day 28	Day 1	Day 28
Time for equilibrium	48h	0h		48h	
Nominal concentrations in soil (mg Cu/kg)					
0	0.14	0.33	<0.05	0.17	0.42
500	1	0.85	1	7.5	130
1000	2	2.2	2.3	14	17
2500	3.4	4.6	3.9	15	17
4000	4.1	5.6	4.2	23	14
5500	7.1	8.9	7.5	31	16
7000	8.5		5.3		

Champion®	Porewater concentrations (mg Cu/L)				
	Lufa 2.2			Lufa 2.1	
	Day 1	Day 1	Day 28	Day 1	Day 28
Time for equilibrium	48h	0h		48h	
Nominal concentrations in soil (mg Cu/kg)					
0	<0.05	<0.05	<0.05	0.042	0.039
17	0.07	0.18	0.07	0.16	0.18
50	0.17	0.11	0.11	0.49	0.36
150	0.3	0.34	0.3	1	1.1
450	0.72	0.52	0.42	2.1	1.9
900	1.1	0.91	0.55	2.7	2
1800	1.6		0.75		
3600	2.6		1.1		

Cupravit®	Porewater concentrations (mg Cu/L)				
	Lufa 2.2			Lufa 2.1	
	Day 1	Day 1	Day 28	Day 1	Day 28
Time for equilibrium	48h	0h		48h	
Nominal concentrations in soil (mg Cu/kg)					
0	<0.05	<0.05	<0.05	0.04	0.039
19	0.11		0.05		
56	0.15		0.11		
167	0.22	0.34	0.18	0.94	0.79
500	0.38	0.52	0.86	1.8	2
1500	1.6	1.2	2.6	4.8	4.3
4500	2.8	3.4	3.8	14	13
9000	6.7	6.1	5.2	23	18

18000	12	8.9

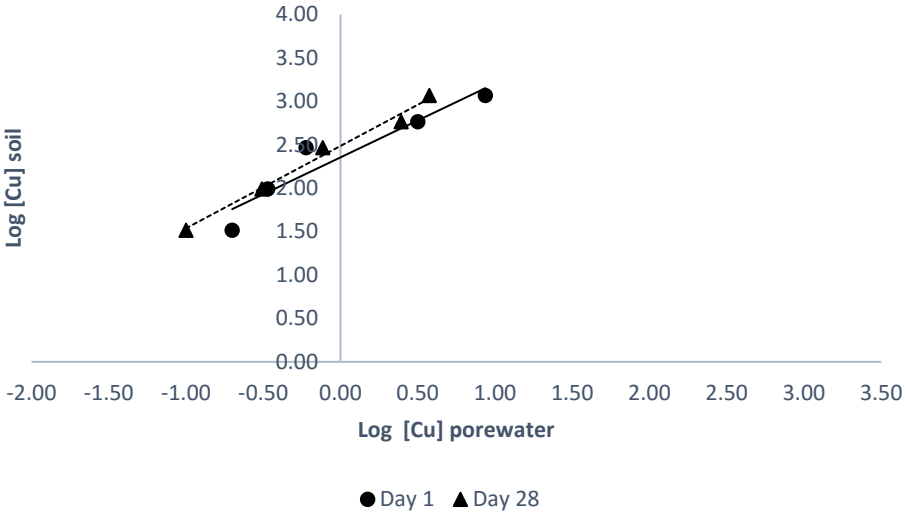
Nordox®	Porewater concentrations (mg Cu/L)				
	Lufa 2.2			Lufa 2.1	
	Day 1	Day 1	Day 28	Day 1	Day 28
Time for equilibrium	48h	0h		48h	
Nominal concentrations in soil (mg Cu/kg)					
0	0.16	<0.05	<0.05	0.14	0.022
17					
50	0.16	0.11	0.11	0.41	0.28
150	0.23	0.23	0.22	1.1	0.88
450	0.46	0.5	0.36	2.5	1.6
900	0.78	0.89	0.46	3.4	2.8
1800	1.7	1.9	0.9	5.7	4.2

Kocide®	Porewater concentrations (mg Cu/L)				
	Lufa 2.2			Lufa 2.1	
	Day 1	Day 1	Day 28	Day 1	Day 28
Time for equilibrium	48h	0h		48h	
Nominal concentrations in soil (mg Cu/kg)					
0	0.03	0.03	0.02	0.031	0.098
11					
33	0.11	0.42	0.13	0.23	0.13
98	0.23	0.35	0.29	0.37	0.34
293	0.47	0.42	0.51	0.63	0.8
586	2.4	4.8	1.9	3.2	2.5
1171	15	53	7.6	8.7	3.8

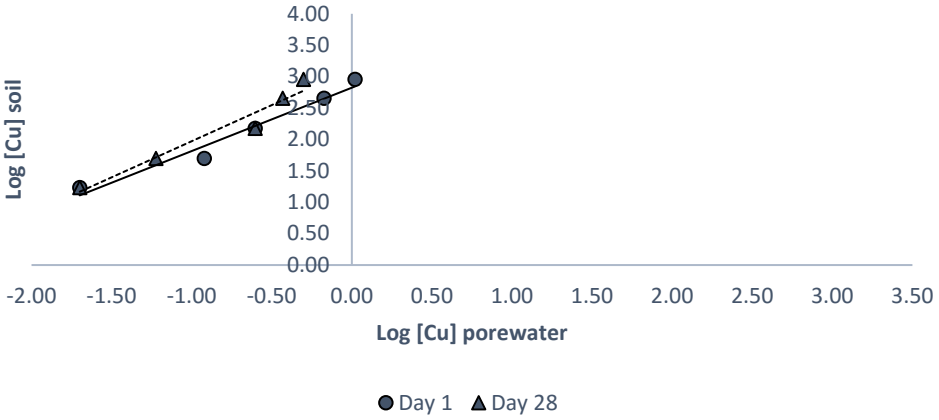
$CuSO_4$	Porewater concentrations (mg Cu/L)				
	Lufa 2.2			Lufa 2.1	
	Day 1	Day 1	Day 28	Day 1	Day 28
Time for equilibrium	48h	0h		48h	
Nominal concentrations in soil (mg Cu/kg)					
0	<0.05	0.015	<0.05	0.17	0.42
100	0.2	0.23	0.19	1.6	2.6
200	0.44	0.43	0.32	8	4.2
400	2.3	2.9	0.94	120	28
800	13	28	10	610	530
1600	586	1000	205	3100	1200

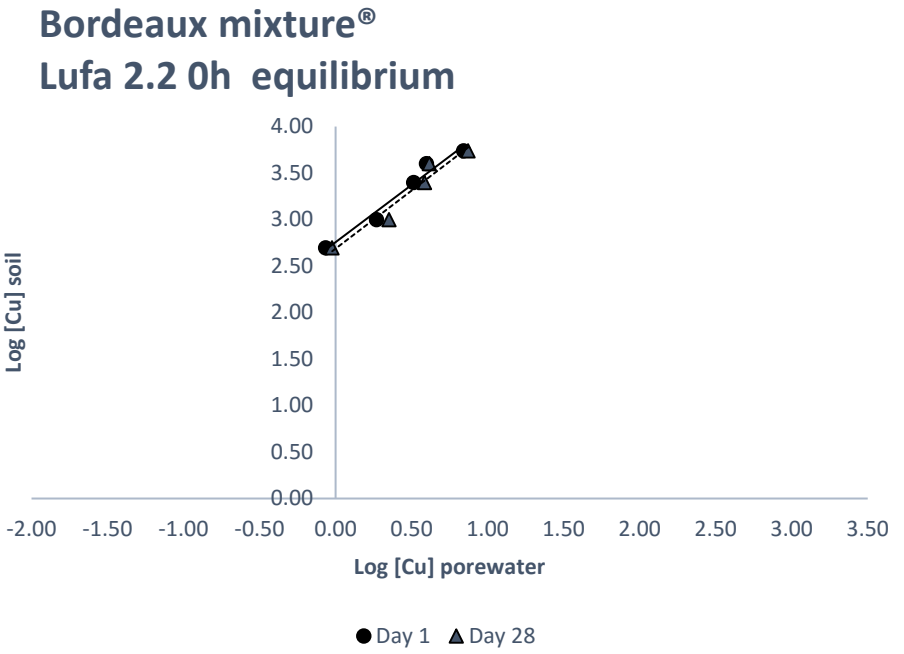
$Cu(OH)_2$	Porewater concentrations (mg Cu/L)				
	Lufa 2.2			Lufa 2.1	
	Day 1	Day 1	Day 28	Day 1	Day 28
Time for equilibrium	48h	0h		48h	
Nominal concentrations in soil (mg Cu/kg)					
0	0.026	0.18	0.02	0.091	0.12
17	0.08	0.12	0.11	0.17	0.13
50	0.2	0.18	0.14	0.54	0.32
150	0.26	0.26	0.36	1.5	0.67
450	1.3	1.1	0.83	1.6	1.2
900	0.51	1.5	0.59	1.8	1.8

Kocide®
Lufa 2.1 48h equilibrium



Champion®
Lufa 2.2 48 h equilibrium





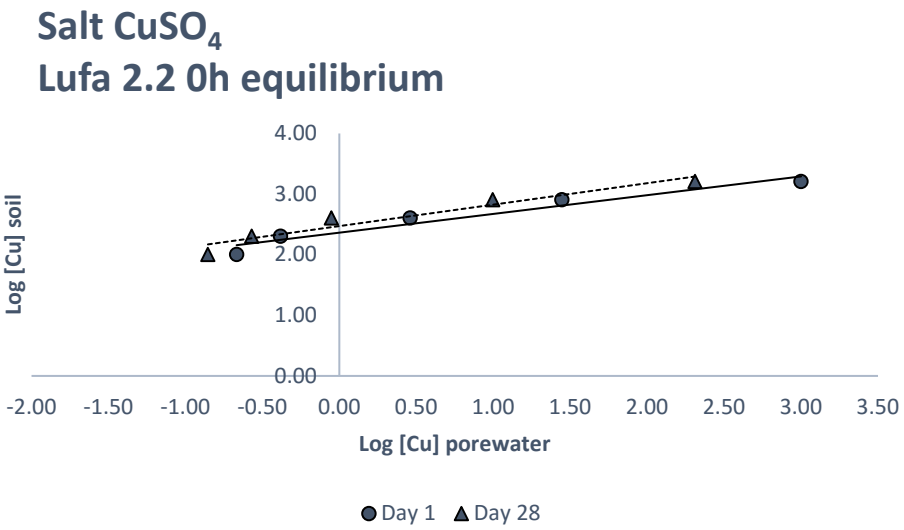


Figure S3: Examples of Freundlich isotherms in Cu formulations, Kocide®, Champion® and Bordeaux mixture® and salt CuSO4 in Lufa 2.1 and 2.2 in both days of the test, day 1 and day 28.