

Supplementary materials

Heavy metal exposures on freshwater snail *Pomacea insularum*: Understanding its biomonitoring potentials

Chee Kong Yap ^{1,*}, Bin Huan Pang ¹, Wan Hee Cheng ², Krishnan Kumar ², Ram Avtar ³, Hideo Okamura ⁴, Yoshifumi Horie ⁴, Moslem Sharifinia ⁵, Mehrzad Keshavarzifard ⁵, Meng Chuan Ong ^{6,7}, Abolfazl Naji ^{8,9}, Mohamad Saupi Ismail ¹⁰ and Wen Siang Tan ^{11,12}

¹ Department of Biology, Faculty of Science, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia

² Faculty of Health and Life Sciences, INTI International University, Persiaran Perdana BBN, Nilai 71800, Negeri Sembilan, Malaysia

³ Graduate School of Environmental Science, Hokkaido University, Sapporo, Hokkaido 060-0810, Japan

⁴ Graduate School of Maritime Sciences, Faculty of Maritime Sciences, Kobe University, Kobe 658-0022, Japan

⁵ Shrimp Research Center, Iranian Fisheries Science Research Institute, Agricultural Research, Education and Extension Organization (AREEO), Bushehr 7516989177, Iran

⁶ Faculty of Science and Marine Environment, Universiti Malaysia Terengganu, Kuala Nerus 21030, Terengganu, Malaysia

⁷ Ocean Pollution and Ecotoxicology (OPEC) Research Group, Universiti Malaysia Terengganu, Kuala Nerus 21030, Terengganu, Malaysia

⁸ Department of Fisheries, Faculty of Marine Science and Technology, University of Hormozgan, Bandar Abbas, 7C9Q+QC3, Iran

⁹ Leibniz Centre for Tropical Marine Research (ZMT), Wiener Str. 7, 28359 Bremen, Germany

¹⁰ Fisheries Research Institute, Batu Maung, Pulau Pinang 11960, Malaysia

¹¹ Laboratory of Vaccines and Biomolecules, Institute of Bioscience, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia

¹² Department of Microbiology, Faculty of Biotechnology and Biomolecular Sciences, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia

* Correspondence: yapckong@hotmail.com or yapchee@upm.edu.my

Table S1: Nominal and measured concentrations (mg/L) of Ni, Cd and Cu in the toxicity test for the *Pomacea insularum* of two different sized groups (Shell lengths, small 0.50-0.70cm; large: shell 1.50-2.20cm).

	Small		Large	
	Nominal	Measured	Nominal	Measured
Cd	Control	0.010	Control	0.010
	1.000	0.978	1.000	1.001
	2.000	1.935	5.000	4.982
	3.000	2.816	10.000	9.801
	4.000	3.819	15.000	14.82
	5.000	4.853	20.000	19.98
Cu	Control	0.010	Control	0.010
	1.000	1.013	1.000	0.965
	1.500	1.503	2.000	1.989
	2.000	1.945	3.000	3.056
	2.500	2.466	4.000	3.976
	3.000	3.013	5.000	4.912
Ni	Control	0.010	Control	0.010
	1.000	1.034	1.000	0.979
	4.000	4.072	5.000	4.789
	8.000	7.901	10.000	9.920
	12.000	11.49	15.000	14.31
	16.000	15.13	20.000	19.14
Pb	Control	0.010	Control	0.010
	1.000	1.054	1.000	0.987
	5.000	4.896	5.000	4.995
	10.000	9.991	10.000	10.01
	15.000	14.89	15.000	14.87
	20.000	19.98	20.000	19.58
Zn	Control	0.010	Control	0.010
	1.000	1.011	1.000	0.936
	5.000	4.857	5.000	4.977
	10.000	9.889	10.000	9.890
	25.000	24.71	30.000	29.32
	50.000	50.12	60.000	59.16

Table S2: Mortality of individuals (*Pomacea insularum*) for the small sized group (Shell length: 0.50-0.70 cm) collected after four different periods of exposure to a series of different concentrations of Pb, Ni, Cd, Zn and Cu.

	Number of dead individuals			
Cd	24 hrs	48 hrs	72 hrs	96 hrs
Control (0.010)	0	0	0	0
1.000 (0.978)	0	2	4	5
2.000 (1.935)	0	3	4	6
3.000 (2.816)	0	3	6	10
4.000 (3.819)	1	5	8	10
5.000 (4.853)	2	7	10	10
Cu	24 hrs	48 hrs	72 hrs	96 hrs
Control (0.010)	0	0	0	0
1.000 (1.013)	2	6	9	10
1.500 (1.503)	4	8	9	10
2.000 (1.945)	5	9	10	10
2.500 (2.466)	8	9	10	10
3.000 (3.013)	8	10	10	10
Ni	24 hrs	48 hrs	72 hrs	96 hrs
Control (0.010)	0	0	0	0
1.000 (1.034)	0	3	4	4
4.000 (4.072)	3	5	7	9
8.000 (7.901)	5	7	8	10
12.000 (11.49)	7	9	10	10
16.000 (15.13)	7	10	10	10
Zn	24 hrs	48 hrs	72 hrs	96 hrs
Control (0.010)	0	0	0	0
1.000 (1.011)	0	0	0	0
5.000 (4.857)	0	1	3	4
10.000 (9.889)	0	3	7	10
25.000 (24.71)	2	4	9	10
50.000 (50.12)	3	8	10	10
Pb	24 hrs	48 hrs	72 hrs	96 hrs
Control (0.010)	0	0	0	0
1.000 (1.054)	0	1	1	2
5.000 (4.896)	0	1	2	3
10.000 (9.991)	2	3	7	10
15.000 (14.89)	5	8	9	10
20.000 (19.98)	6	10	10	10

Note: All values in bracket are the measured metal concentrations and presented in mg/L.

Table S3: Mortality of individuals (*Pomacea insularum*) for the large-sized group (Shell length: 1.50-2.20 cm) collected after four different periods of exposure to a series of different concentrations of Ni, Cd and Cu.

	Number of dead individuals			
Cd	24 hrs	48 hrs	72 hrs	96 hrs
Control (0.010)	0	0	0	0
1.000 (1.001)	0	0	0	4
5.000 (4.982)	0	2	3	8
10.000 (9.801)	0	2	3	10
15.000 (14.82)	0	2	6	10
20.000 (19.98)	0	4	10	10
Cu	24 hrs	48 hrs	72 hrs	96 hrs
Control (0.010)	0	0	0	0
1.000 (0.965)	0	2	7	8
2.000 (1.989)	0	5	9	10
3.000 (3.056)	0	5	9	10
4.000 (3.976)	0	6	10	10
5.000 (4.912)	0	7	10	10
Ni	24 hrs	48 hrs	72 hrs	96 hrs
Control (0.010)	0	0	0	0
1.000 (0.979)	0	0	0	2
5.000 (4.789)	1	2	4	4
10.000 (9.920)	1	5	8	8
15.000 (14.31)	4	7	10	10
20.000 (19.14)	4	9	10	10
Zn	24 hrs	48 hrs	72 hrs	96 hrs
Control (0.010)	0	0	0	0
1.000 (0.936)	0	0	0	0
5.000 (4.977)	0	0	0	1
10.000 (9.890)	0	1	3	3
30.000 (29.32)	1	2	5	6
60.000 (59.16)	3	5	10	10
Pb	24 hrs	48 hrs	72 hrs	96 hrs
Control (0.010)	0	0	0	0
1.000 (0.987)	0	0	0	0
5.000 (4.995)	0	1	1	3
10.000 (10.01)	0	2	5	8
15.000 (14.87)	2	4	7	10
20.000 (19.58)	3	6	9	10

Note: All values in the bracket are the measured metal concentrations and presented in mg/L.