ISSN: 0270-5060 (Print) 2156-6941 (Online) Journal homepage: https://www.tandfonline.com/loi/tjfe20

Bioaccumulation of Nickel, Copper, and Cadmium by *Spirodela polyrhiza* and *Lemna gibba*

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To cite this article: Fatih Duman , Zeliha Leblebici & Ahmet Aksoy (2009) Bioaccumulation of Nickel, Copper, and Cadmium by *Spirodela polyrhiza* and *Lemna gibba* , , 24:1, 177-179, DOI: 10.1080/02705060.2009.9664279

To link to this article: https://doi.org/10.1080/02705060.2009.9664279

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ABSTRACT

Lemna gibba and *Spirodela polyrhiza* were exposed to different concentrations of Ni, Cu, and Cd for 10 days. Bioaccumulation of the three metals for both plants were dependent on and positively correlated with the external metal concentrations. Both plants were hyperaccumulators of Cd but not of Ni or Cu. *L. gibba* was more effective in extracting all three metals than was *S. polyrhiza.*

Lemna and *Spirodela* species have been widely used as scavengers of heavy metals from aquatic environments, and they are being used in wastewater purification systems (Landolt and Kandeler 1987). *Lemna gibba* and *Spirodela polyrhiza* are free-floating and rapidly-growing aquatic macrophytes that exhibit relatively high tolerance to toxic metals and are suitable for accumulation of heavy metals against concentration gradients (Demirezen et al. 2007, Boniardi et al. 1999, Appenroth et al. 2001). Bioaccumulation of metals varies considerably among species and also among morphologically similar species grown in the same area (Brekken and Steinnes 2004). In order to find suitable plants to remove pollutants from the aquatic environment, a wide range of physiological and biochemical features of potential accumulator species is needed. Therefore, we examined the abilities of *L. gibba* and *S. polyrhiza* to accumulate Ni, Cu, and Cd from their external environment.

The accumulation capacities of these plants were determined in the laboratory by adding nickel nitrate [$Ni(NO_3)2{\cdot}6H_2O$], copper chloride ($CuCl_2$), and cadmium chloride ($CdCl_2$) to Hoagland medium (Eliasson 1978). Exposure chambers were cylindrical vessels (diameter: 15 cm), each containing 500 mL of Hoagland medium and 4 g of plant material. Each exposure treatment was run in duplicate. Correction for evaporation was made daily by adding tap water. The pH was monitored daily and averaged 7.0. On day 5, exposure treatment media were replaced with fresh media. After day 10, plant material was removed and assayed for Ni, Cu, and Cd.

Samples were dried at 70 °C to determine dry weight and then digested with 10 mL of HNO_3 using a CEM-Marsh 5 microwave digestion system (CEM Corp., Mathews, North Carolina, USA). Concentrations of Ni, Cu, and Cd were determined by inductively coupled plasma optical emission spectroscopy (Varian-Libery II, ICP-OES, Australia). All metal concentrations in plants were adjusted for metals in control (unexposed) plants.

All statistical analyses were performed with the software SPSS 15.0. Significant differences were calculated using Student's *t*-test wherever applicable. One-way analysis of variance was implemented to identify significant differences in metal concentrations in plants at different initial concentrations ($p \leq 0.05$). All pairwise mean comparisons were performed using the post hoc Duncan test with a degree of significance of 0.05.

Bioaccumulation of Ni, Cu, and Cd was concentration dependent. Ni and Cd concentrations in the plants increased with increased initial metal exposure concentration. In the case of Cu, however, the Cu concentration in plants peaked at a very low exposure concentration (0.3 $\mu g\ L^{-1}$) and then appeared to plateau even in the face of increased metal exposure concentration.

Reeves and Baker (1999) defined a Ni hyperaccumulator as a species that would contain at least 1,000 μg Ni g^{-1} (dry weight). By that standard, neither *L. gibba* nor *S. polyrhiza* was a hyperaccumulator of Ni. A hyperaccumulator of Cd has 100 μg Cd g^{-1} in

its tissue, compared to a normal level for most plants of 0.1 μg g^{-1} (Brooks 1998). Thus, both plants examined in this study were hyperaccumulators of Cd. Neither plant was a hyperaccumulator of Cu (≥5,000 μg g^{-1}), according to Brooks (1998).

The uptake of heavy metals may decrease with time and accumulation due to their toxic effects. Hou et al. (2007) indicated that the removal of low-level Cd from aqueous solution by *Lemna minor* was quite efficient. Nyquist and Greger (2007) stated that uptake of metals increases with increasing external metal concentration, but this is not a linear correlation. With time, the metal concentration in tissue increases, which causes saturation, and then the effective uptake decreases. Soltan and Rashed (2003) suggested that metal accumulation by aquatic plants at low concentrations in solution is more effective than at high concentrations. Under the conditions of our study, bioaccumulation of these metals by both *L. gibba* and *S. polyrhiza* was most efficient when external solutions of Ni, Cu, and Cd were less than 40.0, 1.0, and 2.4 μg L^{-1}, respectively.

Table 1. Mean (± SE) Ni, Cu, and Cd concentrations of *L. gibba* and *S. polyrhiza* (μg g^{-1} dry weight) after 10 days of cutivation with different initial metal concentrations in solution. Different letters in the same column indicate significant differences at $P<0.05$.

	Initial metal concentration (μg L^{-1})	*L. gibba* (n=2)	*S. polyrhiza* (n=2)
Ni	Control	2.6 ± (0.5)	1.3 ± (0.3)
	5.0	92.4 ± (2.8) a	56.5 ± (3.2) a
	10.0	118.0 ± (2.0) b	82.8 ± (4.5) b
	20.0	147.3 ± (3.5) c	100.1 ± (3.4) c
	30.0	154.2 ± (4.4) d	105.7 ± (3.0) c
	40.0	162.4 ± (5.8) d	114.5 ± (2.0) d
	50.0	189.1 ± (5.4) e	126.9 ± (3.1) e
Cu	Control	10.5 ± (0.4)	10.1 ± (0.3)
	0.1	111.7 ± (7.3) a	125.0 ± (6.3) a
	0.3	236.4 ± (14.1) c	238.7 ± (13.5) d
	0.5	222.8 ± (7.5) bc	152.4 ± (6.2) b
	1.0	214.4 ± (6.4) b	160.3 ± (4.8) bc
	1.5	234.1 ± (9.9) bc	174.2 ± (5.8) c
	2.0	240.3 ± (6.8) c	170.5 ± (4.1) c
Cd	Control	0.6 ± (0.1)	0.2 ± (0.1)
	0.1	136.8 ± (4.3) a	82.1 ± (2.9) a
	0.2	258.6 ± (7.8) b	145.6 ± (4.6) b
	0.6	305.1 ± (6.4) c	164.9 ± (3.2) c
	1.2	328.0 ± (4.5) d	233.9 ± (3.3) d
	2.4	339.6 ± (4.0) d	324.4 ± (3.3) e
	3.0	391.1 ± (3.5) e	261.0 ± (2.0) f

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Received: 29 May 2008 Accepted: 17 July 2008