

Eduardo Cervi ORCID iD: 0000-0003-2446-5550

METAL TOXICITY DURING SHORT-TERM SEDIMENT RESUSPENSION AND REDEPOSITION IN A TROPICAL RESERVOIR

Eduardo Cimino Cervi^{1*}, Michelle Hudson¹, Alison Jean Rentschler¹, G. Allen Burton^{2,3}

¹School for Environment and Sustainability, University of Michigan, Ann Arbor, Michigan, United States

²Natural Resources & Environment, University of Michigan, 440 Church St, Ann Arbor, Michigan, United States

³Department of Earth & Environmental Sciences, Wright State University, 3640 Colonel Glenn Highway, Dayton, Ohio, United States

ecervi@umich.edu

ABSTRACT

Billings Complex is the largest water-storage reservoir in São Paulo, Brazil and has been contaminated since the 1960s. Periodically, Billings sediments are subjected to currents causing resuspension and subsequent metals release. A short-term (4 h) resuspension was simulated using Sediment Flux Exposure Chambers (SeFEC) to better understand fate, bioavailability, and transport of iron (Fe), manganese (Mn), and zinc (Zn) during these events and possible organism toxicity. *Daphnia magna* and *Hyaella azteca* were exposed during the 4 h resuspension, after post exposure, and monitored for survival, growth, and

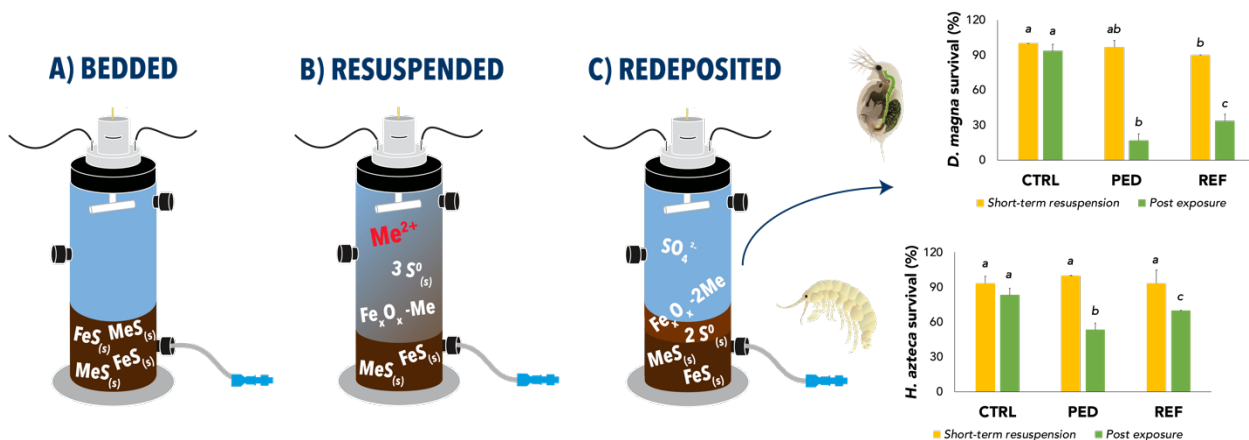
This is the author manuscript accepted for publication and undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the [Version of Record](#). Please cite this article as [doi: 10.1002/etc.4434](https://doi.org/10.1002/etc.4434).

This article is protected by copyright. All rights reserved.

reproduction. Resuspension rapidly deoxygenated overlying water, decreased pH, and resulted in elevated dissolved Zn above the US Environmental Protection Agency's (USEPA) criteria for acute toxicity ($120 \mu\text{g L}^{-1}$). However, Zn was scavenged (after 20 h) from solution as new sorption sites formed. Dissolved Mn increased during and after resuspension with maximum values 20 h post exposure. An initial release of Fe occurred, likely associated with oxidation of acid volatile sulfides, but decreased after 1 h of resuspension. The Fe decrease is likely due to precipitation as oxyhydroxides. No acute toxicity was observed during resuspension; however, mortality of *D. magna* and *H. azteca* occurred during the post exposure period. *Daphnia magna* also showed chronic toxicity with decreased neonate production from post exposures. This sub-lethal effect could lead to decreased zooplankton populations over a longer period in the reservoir.

GRAPHICAL ABSTRACT

Caption. Conceptual model of metal (Me) speciation under different sediment redox states. During bedded conditions (A) metals are mainly bounded as insoluble sulfides or associated with organic carbon (OC). When resuspended (B), sulfide species are oxidized, mobilizing metals (such as Zn) into the overlying water (OW). However, the mobilized metal is scavenged by OC and freshly-precipitated Fe_xO_x . As particles redeposit (C), and are returned to the benthic environment, further oxidation can occur in the aerobic sediment layer releasing Fe. High dissolved Fe concentrations in OW caused acute and chronic toxicity to *D. magna*. Ingestion of Zn caused growth inhibition and mortality to *H. azteca*.



KEYWORDS: Sediment toxicity; metal bioavailability; acid-volatile sulfide; water quality criteria; tropical ecotoxicology.

INTRODUCTION

Metals are the most common and significant sediment pollutants (Hill et al. 2013), representing a threat to benthic communities due to their toxicity, persistence nature and non-biodegradability (Duman et al. 2007; Varol and Şen 2012). During advective processes, such as resuspension events arising from high flows, dredging, or vessel propeller wash; anoxic depositional sediments may be disturbed and released into overlying oxygenated waters (Burton and Johnston 2010). Changes in the physical and chemical environment (e.g., dissolved oxygen, redox, pH, and oxidation of sulfides) may transform metals, directly affecting their mobility and bioavailability (Atkinson et al. 2007; Roberts 2012). This raises the question of whether there is resulting toxicity and potential ecological impacts to populations and communities of aquatic organisms (Carvalho et al. 1998; Fetters et al. 2016).

Differences in climate and geochemistry between temperate and tropical aquatic ecosystems may also be of great importance when comparing fate of metals (Silvério et al. 2005; Cervi et al. 2017; Wang et al. 2019). Metals can be in solution, sorbed or co-precipitated with mineral surfaces and organic matter; but, in anoxic sediments, iron sulfides may be the primary solid phases controlling metal availability (Charriau et al. 2011; Richards et al. 2018). When anoxic sediments are exposed to oxic conditions, acid-volatile sulfide (AVS) bound metals are released, followed by dissolved iron (Fe) and manganese (Mn) rapidly precipitating as amorphous and poorly crystalline Fe/Mn oxyhydroxides (Caetano et al. 2003). Not all metals may be re-adsorbed or not bound tightly to solids making them potentially toxic (Fetters et al. 2016).

Several studies have measured metals release from sediments under simulated field conditions (Calmano et al. 1994; Cantwell et al. 2002; Caetano et al. 2003; Carvalho et al. 1998). Most recent investigations used stirring and shaking methods to resuspend sediments (Hong et al. 2011; Durán et al. 2012; Pourabadehei and Mulligan 2016), with unrealistic suspension durations, or excessive levels of suspended particulate matter (SPM), ranging from 5 g L^{-1} to $>1000 \text{ g L}^{-1}$.

Toxicological effects of metals on aquatic organisms has mainly focused on temperate countries (Nguyen et al. 2011; De Jonge et al. 2012; Simpson and Spadaro 2016). This means tropical studies are using temperate species to assess toxicity (Carvalho et al. 1998; Rico et al. 2011). Several authors have hypothesized that extrapolation of temperate toxicity data for tropical assessments may place tropical ecosystems at undue risks, because differences in environmental conditions and species sensitivity are not normally considered (Castillo et al. 1997; Lacher Jr. and Goldstein 1997; Kwok et al. 2007; Daam and Van den Brink 2010).

There is also a paucity of studies on the influence of resuspension on the toxicity to tropical species, however the physicochemical processes of resuspension and metal release should be similar to temperate regions. Fetters et al. (2016) developed a Sediment Flux Exposure Chambers (SeFEC) for studying sediment resuspension. The SeFEC design allows a more representative metal exposure and associated toxicological effects under environmentally realistic conditions (0.2 g L^{-1} to 1.0 g L^{-1} of SPM). To assess resuspension effects on pelagic and epibenthic organisms, this exposure system

was used to better understand the fate, bioavailability, and toxicity of metals from sediments in a contaminated tropical reservoir.

MATERIAL AND METHODS

Site description

Billings Complex (23° 42' S and 45° 27' W) is the largest water-storage reservoir in São Paulo, Brazil covering an area of 127 km², with an approximate volume of 1.2 km³ and mean depth of 10 meters (Carvalho et al. 1998). Billings was built in the 1920s for hydropower generation and has been contaminated from sewage and industrial effluents since the 1960s by the reversal of Pinheiros river, a tributary of Tietê river. The reservoir supplies about 1.8 million people, and also is used for fishing and swimming.

Sediment and water characterization

Initial laboratory and field screening assessment involving physical, chemical, and toxicological characterizations were conducted along ten representative stations within Billings reservoir (Figure 1).

Water quality parameters (pH, redox potential, dissolved oxygen, conductivity, turbidity, and temperature) were measured at incremental depths (intervals of 1 m) using a calibrated Horiba® U-50 multiparameter water quality meter. Surface sediments were collected using a Petit Ponar grab sampler and sediments shipped at -4 °C to the University of Michigan. All sediment analyses were performed within 7 d following collection.

Sediment samples were processed for AVS content and simultaneously extracted metals (SEM; Simpson et al. 2005), dry weights, and loss on ignition (6 h combustion at 450 °C) for organic carbon content. Subsamples were digested and analyzed for metals cadmium (Cd), chromium (Cr), copper (Cu), iron (Fe), manganese (Mn), nickel (Ni), lead (Pb), and zinc (Zn). Organochlorine pesticides and semi volatile organic compounds were determined according to US Environmental Protection Agency (USEPA) methods SW-8270 (USEPA 2007) and SW-8081b (USEPA 2007), respectively.

Porewater was collected within the sediment bottles using a nitrogen-purged syringe attached to a Rhizon[®] sampler (0.22 µm built-in filter). Overlying water was collected using 500 mL HDPE bottles and filtered through 0.45 µm cellulose acetate (Sartorius Minisart[®]). The porewater and overlying water samples were acidified with concentrated HNO₃ (2% v/v, Merck[®]) and analyzed for dissolved metals (Cd, Cr, Cu, Fe, Mn, Ni, Pb, vanadium, and Zn) using an inductively coupled plasma atomic emission spectrometer (Optima 8000 Series).

Based on initial screening results, 2 stations were chosen for resuspension tests – Pedreira Pumping Station and Capivari River—to represent the most contaminated sediments and a reference station, respectively (Table 1). Pedreira pumping station is highly impacted by industrial activities and urbanization and was selected based on its high SEM/AVS ratio and total metal concentrations, which exceeded recommended probable effect concentration (PEC) or several effect level (SEL) for Cr, Cu, Fe, and Zn. Criteria for reference site selection (Capivari River) included similar water chemistry, but low metal and ΣSEM-AVS contents.

Experimental design

Short-term (4 h) sediment resuspension events were simulated using Sediment Flux Exposure Chambers (SeFEC) modified from chamber designs of others (Figure 2; Eggleston 2011; Fetters et al. 2016). SeFEC was constructed from polycarbonate core tubes (580 mL) with polycarbonate end caps to minimize absorption and desorption of metals during resuspension. Pairs of chambers were connected by a recirculating system, with 1 chamber as a resuspension chamber and the other as an organism exposure chamber.

The exposure chamber contained an inner chamber (9 cm height x 2 cm i.d.) made of polycarbonate in which test organisms were held during resuspension and bedded experiments. The inner chamber had a screen (26 cm²) composed of nylon mesh (pore size of 250 nm), allowing test organisms to be exposed to overlying water during resuspension (Fetters et al. 2016).

Sediment was resuspended using a motor – powered by a 6-volt battery – which rotates a polycarbonate propeller creating a vortex powerful enough to resuspend the surface layer sediment within the chamber. Two ports along the side of the chamber allow for sampling of both overlying water and pore water within settled sediment via Rhizon[®] samplers (Figure 2).

The experimental design for resuspension tests was conducted according to Fetters et al. (2016). Sediment samples were gently homogenized before each experiment, avoiding excess contact with oxygen. Resuspension events were simulated

by adding mixed sediment to the resuspension chamber to create a layer of 3.5 to 4.0 cm thick. Particulate matter in the exposure chamber was kept in suspension by the use of a stir plate and Teflon-coated stir bar at the bottom of the exposure chamber at 150 rpm to achieve $\sim 1.0 \text{ g L}^{-1}$ of sediment particulate material (SPM).

Comparison experiments using bedded sediment were conducted without induced shear stress; but, were otherwise identical. Water and suspended particles were recirculated between the resuspension and exposure chambers by a peristaltic pump at 30 rpm through polyvinyl chloride tubing (3.2 mm i.d.). Complete mixing of water column between the chambers was achieved within 2 h. All resuspension and bedded sediment experiments were 4 h in duration with 3 replicates. Physicochemical parameters (pH, dissolved oxygen, conductivity, and temperature) were monitored in overlying water throughout the experiment at 0, 15, 30, 60, 120, and 240 minutes. Dissolved Fe, Mn, and Zn concentrations were measured to determine the concentration of metals released.

After resuspension experiments, suspended sediments could settle over a 20 h period, after which redeposited sediments were subsampled (10 mm depth) to investigate effects of resuspension on AVS/SEM concentrations. Initial screening AVS/SEM results (Table 1) were considered as pre-resuspension condition. Dissolved Fe, Mn, and Zn were determined in both overlying water and porewater after the 20 h settling period to determine metal fluxes from the sediment.

Short-term and post exposure toxicity testing

Organisms cultured at University of Michigan were exposed during short-term (4 h) resuspension tests and after post exposure to elucidate survival, growth and reproduction endpoints (USEPA 1991). Overlying water and sediment/porewater toxicity were assessed with *Daphnia magna* (10 neonates/replicate, 4 d old) and *Hyalella azteca* (10 organisms/replicate, 8 d old), respectively.

Following short-term (4 h) resuspension, *H. azteca* were transferred into beakers containing 150 mL of culture water with a strip of unbleached paper towel and 3 preweighted, preconditioned 1 cm diameter disks of red maple leaf (*Acer rubrum*). *H. azteca* organisms were held for 7 d recovery period to monitor survival and growth.

Daphnia magna were placed into beakers containing 25 mL of culture water and held for 4 d recovery period to determine survival. At the start of post exposure monitoring and every other day, organisms were fed 0.25 mL day^{-1} per organism with Sel-cero, a mixture of *Raphidocelis subcapitata* (green algae) and cerophyl (1.0×10^7 algal cells mL^{-1}). After 4 d recovery period, *D. magna* were kept in the beakers until they reached 3 brood cycles to assess reproduction (R_0), which was counted as total number of neonates produced per adult.

Three representative groups (10 organisms each) were collected to estimate initial mass on day 0. Individual growth rate (IGR) was calculated according to Nedrich and Burton Jr. (2017) for both organisms as follows:

$$\text{IGR} = \frac{\left[\frac{\Sigma(\text{mass}_{\text{org}})_{\text{final}}}{n_{\text{org}}} = \frac{\Sigma(\text{mass}_{\text{org}})_{\text{initial}}}{n_{\text{org}}} \right]}{\text{time}}$$

where mass is in μg , n is the number of organisms per replicate, and time is days.

Following short-term resuspension, chambers containing sediment received 100% culture water change and were further used for post exposure sediment toxicity tests.

Daphnia magna and *H. azteca* were included together within the sediment chamber for 4 d and 7 d, respectively. These additional toxicity tests allowed for direct contact between test organisms and sediment. *Hyalella azteca* were not fed during exposure to promote sediment grazing.

Quality assurance/quality control

All plastic and glass-ware used during the experiment process were new or soaked in 20% (v/v) nitric acid for at least 24 h followed by 2 rinses with deionized water (prepared using a Milli-Q 18 m Ω cm). For quality control, all sediment samples were analyzed using a blank, control and triplicates.

To confirm the viability of organisms used in both short-term and post exposure toxicity tests, *D. magna* and *H. azteca* controls consisted of 3 additional replicates each that were not exposed to either contaminated water or sediment. Organisms were held in 25 mL of clean water without food during the 4 h tests but were otherwise treated identically through post exposure monitoring procedures.

Statistical analysis

All statistical analyses were conducted in Excel[®] using XLSTAT[®] statistical software package. For significance testing, Shapiro test for normality was first used to determine if data were normal or skewed. Levene's test was used to determine whether variances were equal among treatments. Most data were not normally distributed due to natural heterogeneity of organisms' endpoints. For nonparametric data, Kruskal-Wallis tests were used for multiple variable comparisons, and the median or sign test was used for paired analysis (control vs Pedreira pumping station and Capivari River stations). For normally distributed variables, the equivalent one- or two-way analysis of variances were used for multivariate comparisons of normally-distributed variables, followed up with Tukey's Honest Significant Difference post-hoc test when warranted.

RESULTS

Overlying water

Dissolved oxygen during bedded and resuspension experiments are shown in Figure 3. For resuspended results, T_0 corresponds to first minute of sampling after shear stress. dissolved oxygen was significantly consumed during resuspension in Pedreira pumping station, decreasing from 8.7 mg L⁻¹ to 1.3 mg L⁻¹ within 2 h. After redeposition (20 h) dissolved oxygen slightly increased in Pedreira pumping station. The same trend was observed in Capivari River, with significant decrease from 8.3 to 2.8 mg L⁻¹ within 2 h, and further decreasing to approximately 2 mg L⁻¹ over 20 h. In contrast, dissolved oxygen in bedded experiments remained consistent approximately 7.5 mg L⁻¹.

In bedded experiments, pH remained constant at approximately 7 although a sharp decrease of approximately 1 unit was observed within the first 15 min of resuspension (Figure 4). Consistent with O₂ consumption, the pH continued to slowly decrease over 20 h in Pedreira pumping station to 5.9, whereas a slight increase was observed in Capivari River.

Temperature in bedded and resuspended conditions remained constant ($p < 0.01$). Overlying water temperature in bedded sediments averaged 23 °C whereas resuspended overlying water remained at a temperature of 19 °C. This slight difference between bedded and resuspended conditions was due to sediment/overlying water mixing after resuspension, consequently decreasing overlying water temperature.

Conductivity in bedded experiments were consistent around 330 and 353 $\mu\text{S cm}^{-1}$ for Capivari River and Pedreira pumping station, respectively. After resuspension, conductivity decreased from 356 to 339 $\mu\text{S cm}^{-1}$ within 2 h in Capivari River, and further increased to approximately 355 $\mu\text{S cm}^{-1}$ over 20 h. Conductivity in Pedreira pumping station sediments increased from the beginning of short-term (4h) resuspension (361 $\mu\text{S cm}^{-1}$) up to their maximum values at 20 h post exposure to 381 $\mu\text{S cm}^{-1}$.

Dissolved metals

Short-term (4 h) resuspension resulted in metal mobilization to overlying water that was sediment and metal specific. A significant release of Fe (Figure 5) occurred in Pedreira pumping station during the first 30 min but decreased after 1 h of resuspension. Fe²⁺ was almost totally oxidized in Pedreira pumping station after short-term

resuspension, with concentrations similar to bedded conditions. The same trend was observed in Capivari River sediments. Dissolved Fe concentrations up to 1 h of resuspension were lower compared to bedded experiments.

After 20 h, Fe concentrations in the resuspended treatment increased in both Pedreira pumping station and Capivari River to approximately 1.3 mg L^{-1} and 0.6 mg L^{-1} , respectively. Dissolved Fe released during the first hour of resuspension and after 20 h post exposure was above USEPA (USEPA 2002) aquatic life criteria for chronic toxicity (i.e. $1,000 \text{ } \mu\text{g L}^{-1}$).

Dissolved Mn (Figure 6) remained below USEPA aquatic life criteria for chronic toxicity (i.e. $80 \text{ } \mu\text{g L}^{-1}$) during bedded conditions in both Capivari River and Pedreira pumping station ($35.0 \pm 3.4 \text{ mg L}^{-1}$ and $73.9 \pm 7.1 \text{ mg L}^{-1}$, respectively). Unlike Fe, dissolved Mn increased ($p < 0.01$) from the beginning of short-term (4h) resuspension up to its maximum values at 20 h post exposure in both Capivari River and Pedreira pumping station to $680.3 \pm 15.3 \text{ mg L}^{-1}$ and $726.4 \pm 17.9 \text{ mg L}^{-1}$, respectively.

Zinc was immediately released to overlying water after resuspension, reaching concentrations 17-fold bedded conditions for both Pedreira pumping station and Capivari River sediments (Figure 7). Trends for Zn were similar to Mn during short-term (4 h) resuspension and remained above the USEPA aquatic life criteria for acute toxicity (i.e. $120 \text{ } \mu\text{g L}^{-1}$). However, dissolved Zn decreased significantly in both sites after 20 h post exposure, decreasing to 33 and $36 \text{ } \mu\text{g L}^{-1}$ in Pedreira pumping station and Capivari River, respectively.

Porewater Fe and Mn decreased during resuspension in both stations (Table 2). Release of Fe^{2+} to overlying water occurred mostly during the first 30 min, providing a substrate for Fe oxides formation whereas porewater was oxidized. Porewater Mn was significantly released during resuspension in Pedreira pumping station, decreasing from 3319 to 1477 mg L^{-1} over 20 h. Lower Mn release was observed during resuspension in Capivari River, ranging between 1175 and 986 mg L^{-1} after 20 h. Meanwhile, porewater Zn increased during resuspension. A rapid release was observed during initial shear stress, probably as a result of AVS oxidation, degradation of particulate organic matter and/or diffusion/advection.

Acid-volatile sulfide and SEM were measured in both sediments under bedded and after 20 h post exposure (Table 3). When comparing sulfide concentrations in sediments, Pedreira pumping station and Capivari River had the same AVS on average (4.33). However, the molar sum of SEM concentrations was higher than the molar concentration of AVS in Pedreira pumping station during bedded and after 20 h post exposure. This is likely to higher Zinc extracted by AVS/SEM in Pedreira pumping station (~78% of SEM values) compared to Capivari River (~53% of SEM values).

When SEM-AVS is > 0 , there are more metals than the sulfide can bind, resulting in the likelihood of metals mobilization to dissolved phase, or in another metal-bound phase of the sediment. However, as normalized for organic carbon, SEM-AVS were generally low ($< 115 \text{ mmol g dry wt}^{-1}$) compared to reported toxicity thresholds for Zn of $\sim 500 \text{ mmol g dry weight}^{-1}$ (Burton et al. 2005). According to the USEPA recommended SEM/AVS model, these sediments would be considered “not toxic” (USEPA 2005).

Acute toxicity was not observed during short-term (4 h) resuspension for both *D. magna* and *H. azteca* compared to laboratory control ($p < 0.01$; Figures 8-A and 8-B, respectively). However, high mortality was observed when *D. magna* were held after post exposure. Acute effects were significantly higher in Pedreira pumping station (organism survival of $16.6 \pm 5.7\%$) compared to Capivari River ($33.3 \pm 5.7\%$) and control ($93.3 \pm 5.7\%$). *Hyalella azteca* survival was also affected after sediment post exposure in both Pedreira pumping station ($53.3 \pm 5.7\%$) and Capivari River ($70 \pm 0\%$) tests.

Short-term (4 h) resuspension did not affect IGR of *D. magna* and *H. azteca* ($p < 0.01$). However, *D. magna* growth was significantly affected after post exposure in Pedreira pumping station ($0.11 \pm 0.03 \mu\text{g d}^{-1}$) and Capivari River ($0.07 \pm 0.02 \mu\text{g d}^{-1}$), with IGR similar to water-only control ($0.09 \pm 0.02 \mu\text{g d}^{-1}$). *H. azteca* post exposure IGR was significantly higher in Pedreira pumping station ($0.05 \pm 0.00 \mu\text{g d}^{-1}$) and Capivari River ($0.05 \pm 0.01 \mu\text{g d}^{-1}$) compared to control ($0.02 \pm 0.00 \mu\text{g d}^{-1}$), but lower than bedded conditions.

Daphnia magna exhibited a slight decreased neonate production (R_0) in Pedreira pumping station (3.0 ± 0.5 neonates adult^{-1}) and Capivari River (2.5 ± 0.3 neonates adult^{-1}) compared to control (3.8 ± 0.5 neonates adult^{-1}) after short-term resuspension. However, post exposure significantly affected *D. magna* reproduction in Capivari River and Pedreira pumping station (0.5 ± 0.3 and 0.4 ± 0.1 neonates adult^{-1} , respectively) after post exposure when compared to control (3.2 ± 0.5 neonates adult^{-1}).

DISCUSSION

Dissolved oxygen significantly decreased in both stations within the first 2 h. A similar trend was reported by Fetters et al. (2016) and this decline was attributed to O₂ consumption via the formation of Fe oxyhydroxides and aerobic respiration. When anoxic contaminated sediments are disturbed, dissolved oxygen permeates the sediment which drives the oxidation of sulfide, Fe, and Mn complexes (Eggleton and Thomas 2004).

Sulfide oxidation implies that dissolved oxygen may also be depleted, as observed elsewhere (Burton et al. 2006), and could lead to anoxic and/or hypoxic conditions that could be detrimental for organisms (Luther et al. 2004). Even though dissolved oxygen in Pedreira pumping station was below the 2.5 mg L⁻¹ recommended by USEPA (2002) for toxicity test, we suggest sulfide oxidation was not detrimental, because no acute toxicity was observed during short term (4 h) resuspension. Oxygen consumption reflected a rapid oxidation of Fe on the surface of ferrous sulfide particles. The rate of O₂ consumption slowed once Fe on the particles surface became oxidized.

Similar to dissolved oxygen, pH in overlying water rapidly decreased within the first minutes of shear stress. The extent of pH reduction depends on the amount of sulfide in sediments and how much is oxidized (Roberts 2012). A change in chemical properties of the sediment–contaminant causes mobilization and transfer of metals, mainly from sulfide-bound phases (Calmano et al. 1993). A more pronounced pH decrease was observed after resuspension of Pedreira pumping station likely due to higher concentration of Fe and Mn. The 1-unit lower pH in overlying water of Pedreira pumping

station compared to Capivari River could be a possible explanation for different release patterns of Zn via desorption from resuspended sediments.

Decrease of pH during resuspension may also be related to lower buffering capacity of Billings' parent material. Parent material in temperate regions is often composed of primary minerals (i.e. carbonates and gypsum) which act as a buffer to mitigate acidity. On the other hand, in tropical regions such as Billings reservoir, weathering is associated with intensive leaching and strongly oxidizing conditions, such that only hydrous oxides of aluminum, ferric Fe (Fe^{3+}), and titanium persist (Sposito 2008).

Acid-producing capacity (APC) of a sediment/water system is counter-balanced by its acid-neutralization capacity (ANC). Quite often in tropical areas, sediments have a high APC due to the presence of lateritic soils. Tropical soils (such as Oxisols) are mainly composed of weathered soils, and they have the highest average clay contents, are the most acidic, and have the lowest organic carbon content, cation exchange capacity, and base saturation (Costa et al. 2004). In aquatic systems, ANC of solids is always greater than that related to the dissolved phase. With these assumptions, Calmano et al. (1993) proposed an index that could be used for sediment quality criteria: the effective acid-producing capacity (APC_{eff}), which is calculated from the oxidation of a known solid mass of sediments in a suspension volume, and the variation of suspension pH before and after oxidation.

For the present study, the calculated APC_{eff} was $44.5 \text{ mmol H}^+ \text{ kg}^{-1}$ (dry wt), which is 8.7 times greater than observed by Calmano et al. (1993) for Elbe river anoxic

sediment, also contaminated by metals. The high APC_{eff} of Billings sediments result in a significant release of metals to aqueous phase and enhanced bioavailability.

When reduced Fe enters oxic waters, Fe^{2+} is rapidly oxidized. This chemistry creates an amorphous ferric hydroxide floc, which is rapidly oxidized to elemental sulfur or sulfate (Jones-Lee and Lee 2005). Freshly precipitated ferric hydroxide is an effective scavenger of numerous other metals (such as Zn) in the water, an association that renders them unavailable. A similar phenomenon was described previously by Slotten and Reuter (1995) which suggested that resuspended Zn preferentially reabsorbs to newly-formed colloidal or particulate ferric hydroxide in overlying water and becomes redeposited onto the sediment.

Colloidal or particulate ferric hydroxide in the water column can develop sufficient mass to settle and become part of the bedded sediments (Cantwell et al. 2002). As these particles returned to freshly redeposited sediment layer, we suggest that further oxidation occurred in the water column or in the aerobic sediment layer up to 20 h, releasing labile species of Fe to the dissolved phase. Increase in dissolved Mn concentrations in overlying water after 20 h post exposure suggests that deposition of resuspended particles did not scavenge Mn from overlying water.

The kinetics of these reactions may also have an important role for trace element distributions during resuspension. Saulnier and Mucci (2000) observed that remobilized Fe was rapidly removed from dissolved phase during the first hour of sediment resuspension, whereas the removal processes of Mn became predominant after a week. In comparison, Gerringa (1990) had no data from first hour of resuspension and observed no

Fe remobilization. Similarly, Caille et al. (2003) highlighted various readsorption rates for different metals.

The most dominant driver for Zn release is the oxidation of sediment sulfide (to sulfate), releasing $\text{Zn}^{2+}(\text{aq})$ from $\text{ZnS}(\text{s})$. Dissolution and oxidation of metal sulfides concurrently decreases pH, thus increasing porewater Zn^{2+} via acidification of Zn-hydroxides (i.e. $\text{Zn}(\text{OH})_2$ or $\text{Zn}(\text{OH})^{-1}$) and carbonates (i.e. ZnHCO_3^{-1} or ZnCO_3). However, solubilization of Zn from suspended particles likely was not from dissolution of ZnS, given that metal sulfides can take more than 8 h to oxidize and that AVS concentrations were similar during bedded and after resuspension.

High Zn concentration in Pedreira pumping station sediments during bedded conditions (~ 77% of total SEM) indicated that Zn could be bioavailable for organism uptake and potentially cause toxicity. In the environment, it is likely that metals such as copper and nickel would be bound to the available AVS, and only a small portion of Zn may be bound. Simpson et al. (2000) observed high proportions of Zn in unmodified sediments may not be present as metal sulfides despite high AVS concentrations. Therefore, Zn would be the most likely metal to cause any observed toxicity.

The predominant means by which responses of organisms to chemical stressors are assessed and predicted by Sediment Flux Exposure Chambers (SeFEC). However, chemical characterization alone could underestimate the potential ecological implications of resuspended sediments in freshwater reservoirs. Due to the SeFEC inner chamber design, organisms are not exposed to the physical stress of an increase in total suspended solids (TSS) that comes with naturally resuspended sediments. We understand this could

be detrimental since physical stress also influences the aquatic ecosystem in terms of water quality and biological responses.

Relevant exposures for this study are through to overlying water (zooplankton) and surficial sediments (benthos) and are reflected by toxicity testing of *D. magna* and *H. azteca*. Our results agree with previous studies showing short resuspension periods are not likely to cause immediate toxicity but could cause chronic effects (Eggleton and Thomas 2004; Fetters et al. 2016). Although there was no significant acute toxicity to *D. magna* or *H. azteca* over the initial 4 h resuspension period, other long-term effects were noted such as increased mortality after post exposure and decreased neonate production in both resuspended Pedreira pumping station and Capivari River sediments.

Aggregation of particles into flocs may occur relatively quickly (Roberts 2012). After resuspension, water column conditions were favorable to formation of Fe and Mn oxyhydroxide flocs and may have influenced lower dissolved Zn concentrations after redeposition. For epibenthic organisms, such as *H. azteca*, exposure pathways include ingestion of sediment and water. Ingestion of Zn can lead to cellular damage and displacement of dietary Fe causing growth inhibition, reproductive issues, and mortality (Kubitz et al. 1995).

In addition, sediment porewater data reflects a migration of Zn from overlying water into the sediments after resuspension. Among benthic organisms, some of those exposed to porewater and sensitive to Zn include chironomids, Ephemeroptera, and some crustaceans (including amphipods; Brinkman and Johnston 2008).

Fetters et al. (2016) observed resuspension of 3 Zn-contaminated sediments resulted in minimal releases from suspended particles, likely because of scavenging onto organic matter and Fe oxides formed during sediment interaction with oxic water. Only *H. azteca* was affected by Zn dissolution after sediment redeposition.

High dissolved Fe concentrations in overlying water likely caused toxicity to *D. magna*. As described above, Fe was continually released into the overlying water via sulfide oxidation causing prolonged exposure. In addition, a rusty color was observed in *D. magna* bodies after post exposure tests, indicating Fe uptake. Once more relating this back to porewater data, there is a clear decrease of mobile Fe in sediments suggesting release into the overlying water. Elevated metals in overlying water can lead to competitive exclusion of dietary Ca, causing hypocalcaemia in *D. magna*, which decreases growth, affects organism movement, and decreases filtration rate (Muysen et al. 2006).

CONCLUSIONS

Although sediment resuspensions are not a constant threat, they are common and may be an important stressor for organisms. Although there was no significant acute toxicity to *D. magna* or *H. azteca* during a short-term resuspension period, long-term effects were noted. Adverse effects of resuspended contaminated sediments in hydrologically active systems, such as Billings reservoir, should be considered during environmental management decision-making.

REFERENCES

Atkinson C, Jolley R, Simpson S. 2007. Effect of overlying water pH, dissolved oxygen, salinity and sediment disturbances on metal release and sequestration from metal contaminated marine sediments. *Chemosphere* 69:1428–1437.

Brinkman SF, Johnston WD. 2008. Acute Toxicity of Aqueous Copper, Cadmium, and Zinc to the Mayfly *Rhithrogena hageni*. *Arch Environ Contam Toxicol* 54:466–472.

Burton ED, Bush RT, Sullivan LA. 2006. Acid-Volatile Sulfide Oxidation in Coastal Flood Plain Drains: Iron-Sulfur Cycling and Effects on Water Quality. *Environ Sci Technol* 40:1217-1222.

Burton GA, Johnston EL. 2010. Assessing contaminated sediments in the context of multiple stressors. *Environ Toxicol Chem* 29:2625–2643.

Caetano M, Madureira M, Vale C. 2003. Metal remobilisation during resuspension of anoxic contaminated sediment: short-term laboratory study. *Water Air Soil Pollut* 143:23–40.

Caille N, Tiffreau C, Leyval C, Morel JL. 2003. Solubility of metals in an anoxic sediment during prolonged aeration. *Sci Total Environ* 301:239–250.

Calmano W, Hong J, Forstner U. 1993. Binding and Mobilization of Heavy Metals in Contaminated Sediments Affected by pH and Redox Potential. *Water Sci Technol* 28:223-35.

Calmano W, Forstner U, Hong J. 1994. Mobilization and Scavenging of Heavy Metals Following Resuspension of Anoxic Sediments from the Elbe River'. In Alpers C and Blowes D, eds, *Environmental Geochemistry of Sulfide Oxidation*, American Chemical Society, Washington, DC, United States, pp 298–321.

Cantwell MG, Burgess RM, Kester DR. 2002. Release and Phase Partitioning of Metals from Anoxic Estuarine Sediments during Periods of Simulated Resuspension. *Environ Sci Technol* 36:5328-5334.

Carvalho PSM, Zanardi E, Buratini SV, Lamparelli MC, Martins MC. 1998. Oxidizing effect on metal remobilization and *Daphnia similis* toxicity from a Brazilian reservoir sediment suspension. *Wat Res* 32:193-199.

Castillo LE, de la Cruz E, Ruepert C. 1997. Ecotoxicology and pesticides in tropical aquatic ecosystems of Central America. *Environ Toxicol Chem* 16:41–51.

Cervi EC, Fernandes F, Miranda RB, Mauad FF, Michalovicz L, Poletto C. 2017. Geochemical speciation and risk assessment of metals in sediments of the Lobo-Broa Reservoir, Brazil. *Manag Environ Qual Int J* 28:430-443.

Charriau A, Lesven L, Gao Y, Leermakers M, Baeyens W, Ouddane B, Billon G. 2011. Trace metal behavior in riverine sediments: Role of organic matter and sulfides. *Appl Geochem* 26:80-90.

Costa ACS, Bigham JM, Tormena CA, Pintro JC. 2004. Clay mineralogy and cation exchange capacity of Brazilian soils from water contents determined by thermal analysis.

Thermochim Acta 413:73-79.

Daam MA, Van den Brink PJ. 2010. Implications of differences between temperate and tropical freshwater ecosystems for the ecological risk assessment of pesticides.

Ecotoxicol 19:24–37.

De Jonge M, Teuchies J, Meire P, Blust R, Bervoets L. 2012. The impact of increased oxygen conditions on metal-contaminated sediments part I: effects on redox status, sediment geochemistry and metal bioavailability. *Water Res* 46:2205-2214.

Duman F, Aksoy A, Demirezen D. 2007. Seasonal variability of heavy metals in surface sediment of Lake Sapanca, Turkey. *Environ Monit Assess* 133:277–283.

Durán I, Sánchez-Marín P, Beiras R. 2012. Dependence of Cu, Pb and Zn remobilization on physicochemical properties of marine sediments. *Mar Environ Res* 77:43–49.

Eggleston MR. 2012. Impact of Sediment Resuspension Events on the Availability of Heavy Metals in Freshwater Sediments. Master's thesis. University of Michigan, Ann Arbor, MI, United States.

Eggleton J, Thomas KV. 2004. A review of factors affecting the release and bioavailability of contaminants during sediment disturbance events. *Environ Int* 30:973–980.

Fetters KJ, Costello DM, Hammerschmidt CR, Burton Jr GA. 2016. Toxicological effects of short-term resuspension of metal-contaminated freshwater and marine sediments.

Environ Toxicol Chem 35:676-686.

Gerringa LJA. 1990. Aerobic degradation of organic matter and the mobility of Cu, Cd, Pb, Ni, Zn, Fe and Mn in marine sediment slurries. *Mar Chem* 29:355–374.

Hill NA, Simpson SL, Johnston EL. 2013. Beyond the bed: Effects of metal contamination on recruitment to bedded sediments and overlying substrata. *Environ Pollut* 173:182-191.

Hong YS, Kinney KA, Reible DD. 2011. Acid volatile sulfides oxidation and metals (Mn, Zn) release upon sediment resuspension: Laboratory experiment and model development. *Environ Toxicol Chem* 30:564–575.

Hwang KY, Kim HS, Hwang I. 2011. Effect of resuspension on the release of heavy metals and water chemistry in anoxic and oxic sediments. *Clean-Soil Air Water* 9:908-915.

Jones-Lee A, Lee G. 2005. Role of iron chemistry in controlling the release of pollutants from resuspended sediments. *Remed* 16:33-41.

Kubitz JA, Besser JM, Giesy JP. 1996. A twostep experimental design for a sediment bioassay using growth of amphipod *Hyalella azteca* for the test endpoint. *Environ Toxicol Chem* 15:1783-1792.

Kwok KWH, Leung KMY, Lui GSG, Chu VKH, Lam PKS, Morritt D, Maltby L, Brock TCM, Van den Brink PJ, Warne MSJ, Crane M. 2007. Comparison of tropical and temperate freshwater species sensitivities to chemicals: implications for deriving safe extrapolation factors. *Integr Environ Assess Manag* 3:49–67.

Lacher Jr TE, Goldstein MI. 1997. Tropical ecotoxicology: status and needs. *Environ Toxicol Chem* 16:100–111.

Luther GW, Ma S, Trouwborst R, Glazer B, Blickley M, Scarborough RW, Mensinger MG. 2004. The roles of anoxia, H₂S, and storm events in fish kills of dead-end canals of Delaware inland bays. *Estuaries* 27:551–560.

Muyssen BTA, De Schamphelaere KAC, Janssen CR. 2006. Mechanisms of chronic waterborne Zn toxicity in *Daphnia magna*. *Aquat Toxicol* 77:393-401.

Nedrich SM, Burton Jr GA. 2017. Sediment Zn-release during post-drought re-flooding: Assessing environmental risk to *Hyalella azteca* and *Daphnia magna*. *Environ Pollut* 230:1116-1124.

Nguyen LTH, Burton Jr GA, Schlegel CE, Janssen CR. 2011. Field measurement of nickel sediment toxicity: role of acid volatile sulfide. *Environ Toxicol Chem* 30:162–172.

Pourabadehei M, Mulligan CN. 2016. Resuspension of sediment, a new approach for remediation of contaminated sediment. *Environ Pollut* 213:63-75.

Richards CM, van Puffelen JL, Pallud C. 2018. Effects of Sediment Resuspension on the Oxidation of Acid-Volatile Sulfides and Release of Metals (Iron, Manganese, Zinc) in Pescadero Estuary (CA, USA). *Environ Toxicol Chem* 37(4):993-1006.

Rico A, Waichman AV, Geber-Corrêa R, van den Brink P. 2011. Effects of malathion and carbendazim on Amazonian freshwater organisms: comparison of tropical and temperate species sensitivity distributions. *Ecotoxicol* 20:625-634.

Roberts DA. 2012. Causes and ecological effects of resuspended contaminated sediments (RCS) in marine environments. *Environ Int* 40:230-243.

Saulnier I, Mucci A. 2000. Trace metal remobilization following the resuspension of estuarine sediment: Saguenay Fjord, Canada. *Appl Geochem* 15:191–210.

Silvério PF, Fonseca AL, Botta-Paschoal CMR, Mozeto AA. 2005. Release, bioavailability and toxicity of metals in lacustrine sediments: A case study of reservoirs and lakes in Southeast Brazil. *Aquat Ecosyst Health Manag* 8:313–322.

Simpson SL, Apte SC, Batley GE. 2000. Effect of short-term resuspension events on the oxidation of cadmium, lead, and zinc sulfide phases in anoxic estuarine sediments. *Environ Sci Technol* 34:4533–4537.

Simpson SL, Batley GE, Chariton AA, Stauber JL, King CK, Chapman JC, Hyne RV, Gale SA, Roach AC, Maher WA. 2005. *Handbook for Sediment Quality Assessment*. CSIRO, NSW, Banger, 117p.

Simpson SL, Spadaro DA. 2016. Bioavailability and chronic toxicity of metal sulfide minerals to benthic marine invertebrates: implications for deep sea exploration, mining and tailings disposal. *Environ Sci Technol* 50:4061-4070.

Slotten DG, Reuter JE. 1995. Heavy metals in intact and resuspended sediments of a California reservoir, with emphasis on potential bioavailability of copper and zinc. *Mar Freshwater Res* 46:257-265.

Sposito G. 2008. *The chemistry of soils*, 2nd ed, Oxford University, New York, NY, United States, 342p.

[USEPA] US Environmental Protection Agency. 1991. Compendium of ERT Toxicity Testing Procedures. EPA 540/P-91/009. Technical Report. US Environmental Protection Agency, Washington, DC.

[USEPA] US Environmental Protection Agency. 2001. Methods for Collection, Storage and Manipulation of Sediments for Chemical and Toxicological Analyses. EPA 823/B-01/002. Technical Manual. US Environmental Protection Agency, Washington, DC.

[USEPA] US Environmental Protection Agency. 2002. National recommended water quality criteria: 2002. EPA 822/R-02/047. Technical Report. US Environmental Protection Agency, Washington, DC.

[USEPA] US Environmental Protection Agency. 2007. Method 8081B – Organochlorine Pesticides by Gas Chromatography, Feb. 2007, rev. 02, 57 p.

[USEPA] US Environmental Protection Agency. 2007. Method 8270D – Semivolatile Organic Compounds by Gas Chromatography/Mass Spectrometry (GC/MS), Feb. 2007, rev. 04, 72 p.

Varol M, Şen B. 2012. Assessment of nutrient and heavy metal contamination in surface water and sediments of the upper Tigris River, Turkey. *Catena* 92:1-10.

Wang Z, Knok KWH, Leung KMY. 2019. Comparison of temperate and tropical freshwater species' acute sensitivities to chemicals: An update. *Integr Environ Assess Manag* (in press).

Figures

Figure 1. Initial screening sampling sites within Billings reservoir with highlighted stations (in bold) chosen for short-term (4) resuspension tests.

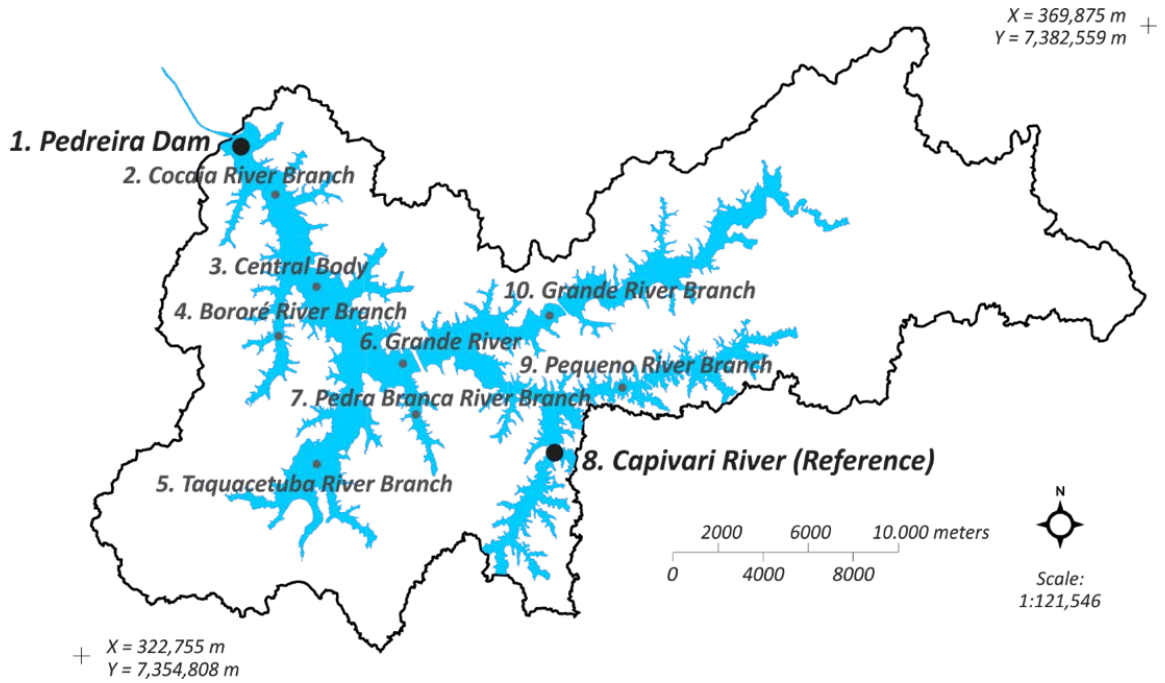


Figure 2. Concept model of Sediment Flux Exposure Chambers (SeFEC) setup for short-term (4 h) resuspension tests in laboratory.

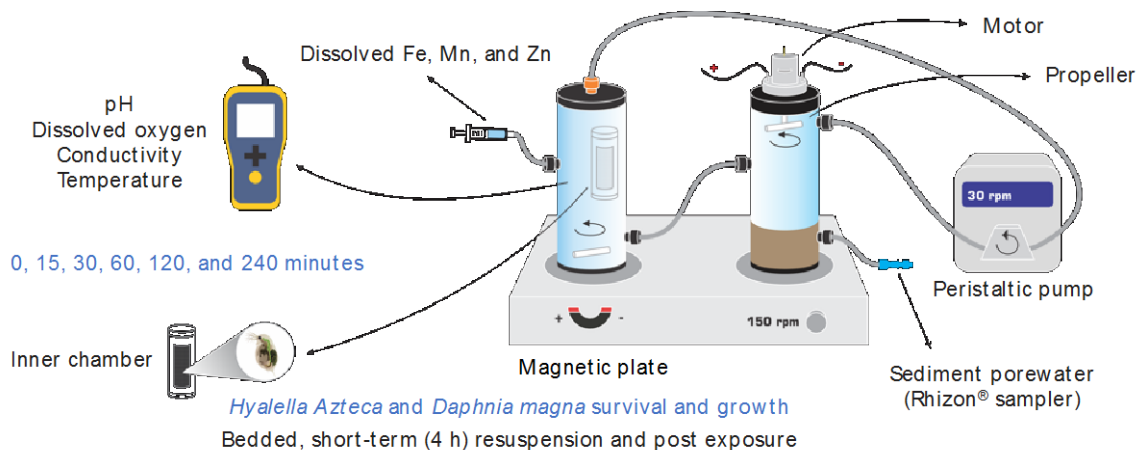


Figure 3. Dissolved oxygen (DO) concentrations in overlying water of bedded and short-term (4 h) resuspension tests in Pedreira Pumping Station (PED) and Capivari River (REF) samples (\pm SD, $n = 3$). Statistical significance ($p < 0.01$) is denoted with different letters.

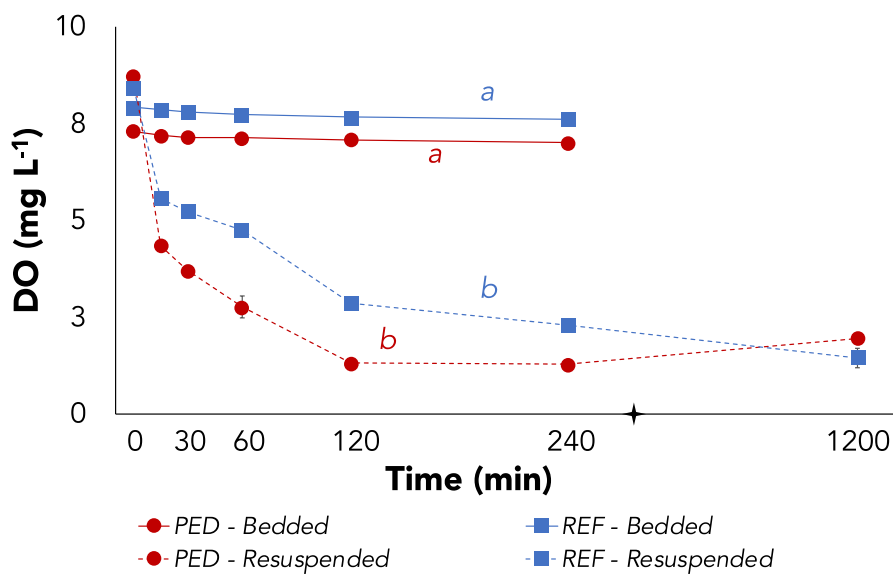


Figure 4. Overlying water pH of bedded and short-term (4 h) resuspension tests in Pedreira Pumping Station (PED) and Capivari River (REF) samples (\pm SD, $n = 3$). Statistical significance ($p < 0.01$) is denoted with different letters.

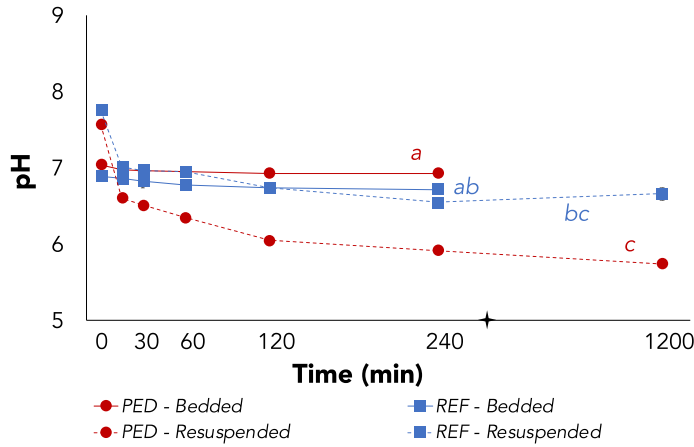


Figure 5. Dissolved iron (Fe) concentrations in overlying water of bedded and short-term (4 h) resuspension tests in Pedreira Pumping Station (PED) and Capivari River (REF) samples (\pm SD, $n = 3$). Statistical significance ($p < 0.01$) is denoted with different letters.

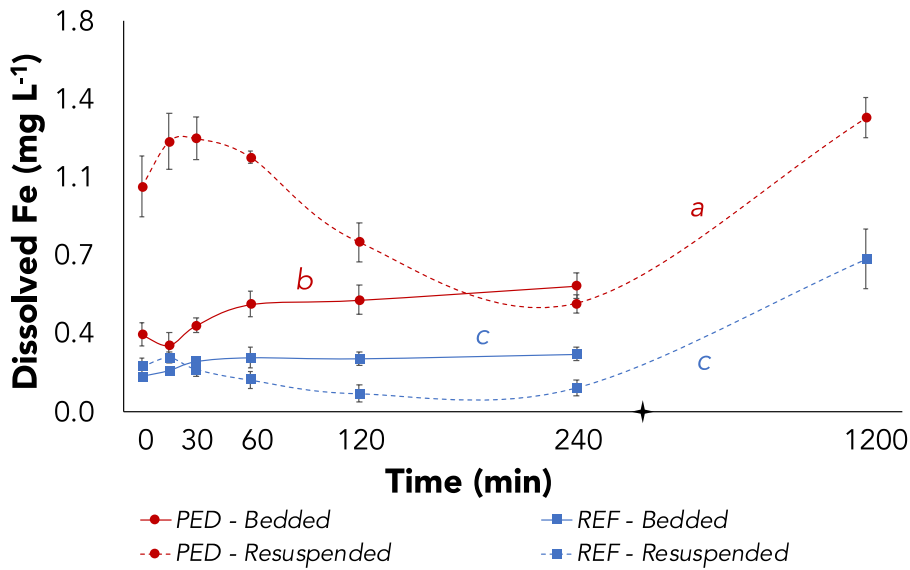


Figure 6. Dissolved manganese (Mn) concentrations in overlying water of bedded and short-term (4 h) resuspension tests in Pedreira Pumping Station (PED) and Capivari River (REF) samples (\pm SD, $n = 3$). Statistical significance ($p < 0.01$) is denoted with different letters.

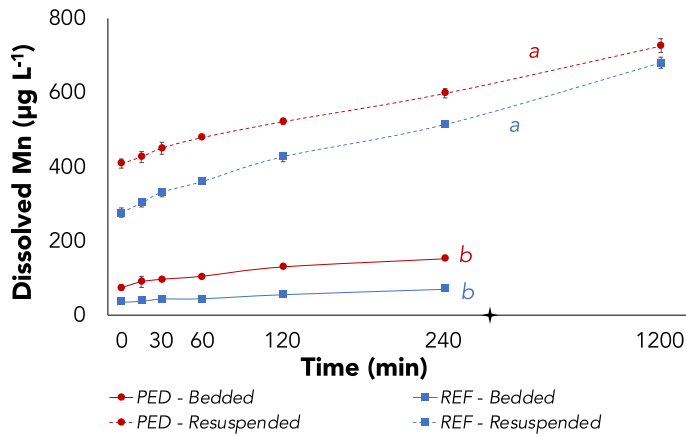


Figure 7. Dissolved zinc (Zn) concentrations in overlying water of bedded and short-term (4 h) resuspension tests in Pedreira Pumping Station (PED) and Capivari River (REF) samples (\pm SD, $n = 3$). Statistical significance ($p < 0.01$) is denoted with different letters.

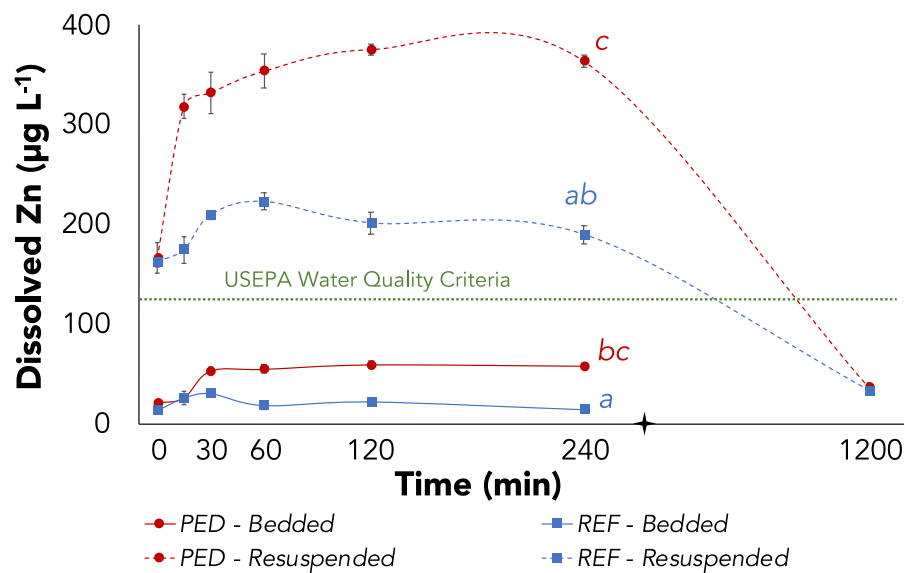
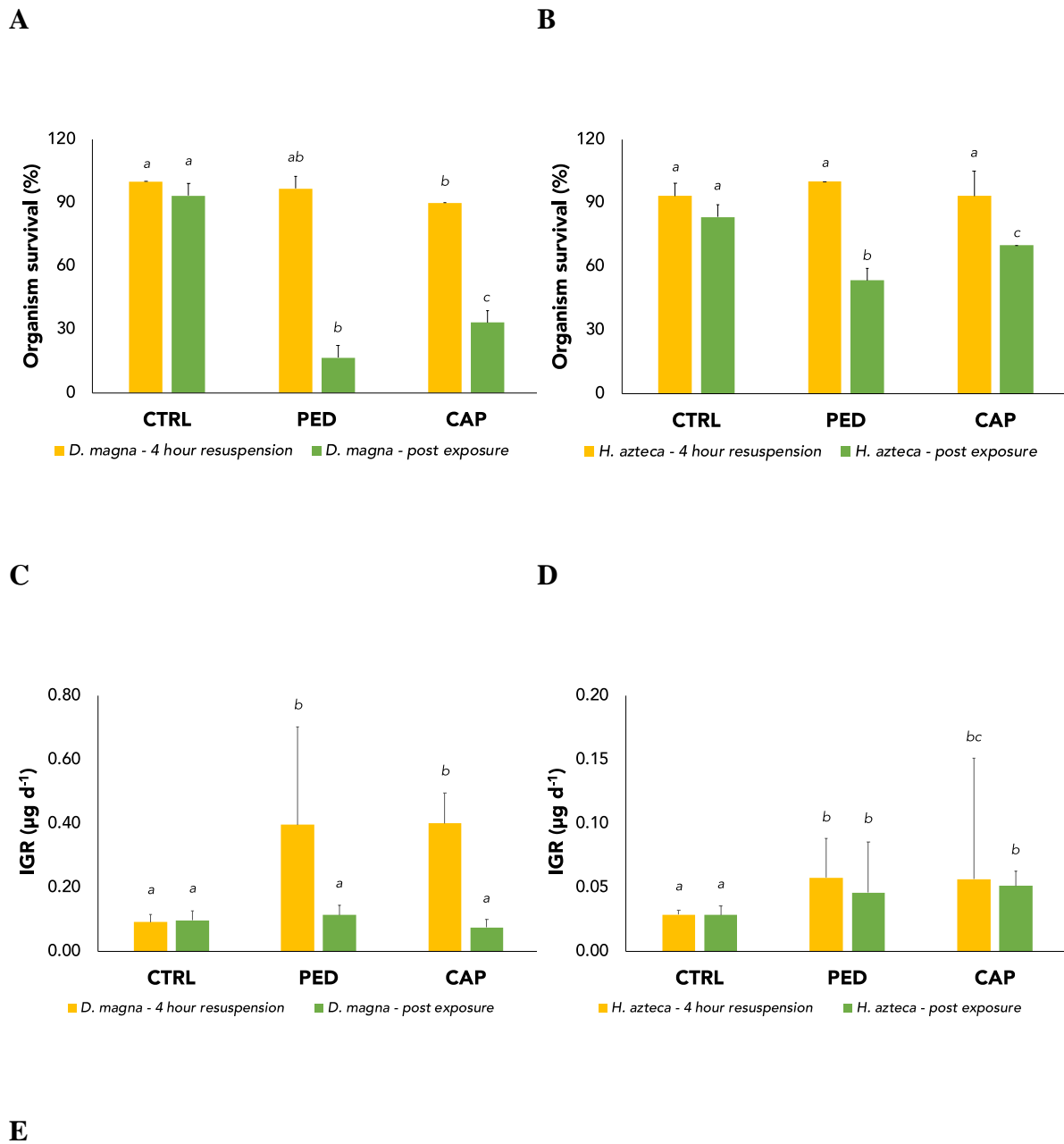
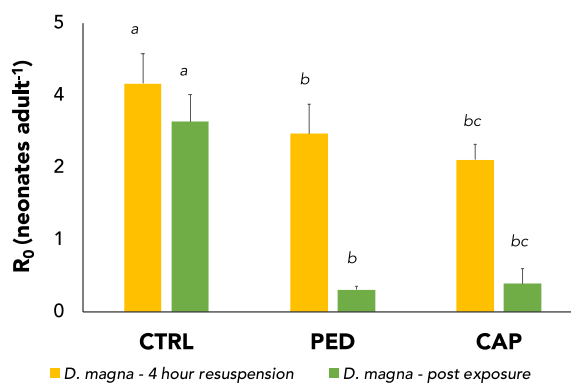


Figure 8. Organism survival, individual growth rate (IGR), and reproduction (R_0) for *Daphnia magna* and *Hyalella azteca* after short-term (4 h) resuspension and post exposure in Pedreira Pumping Station (PED) and Capivari River (REF) compared to control (CTRL; \pm SD, $n = 3$), where A) *D. magna* survival, B) *H. azteca* survival, C) *D. magna* IGR, D) *H. azteca* IGR, and E) *D. magna* R_0 . Different letters indicate statistically significant differences between groups ($p < 0.05$).





TABLES

Table 1. Summary of physical and chemical properties of overlying water and sediment samples (\pm SD, $n = 3$) at Pedreira Pumping Station and Capivari river

Surface water			Surface sediment		
Parameter	PED	REF	Parameter	PED	REF
Depth (meters)	10	10	Texture	Silty clay	Silty clay
pH	7.5 \pm 0.2	8.4 \pm 0.4	pH	6.6 \pm 0.1	6.5 \pm 0.0
ORP (mV)	66 \pm 32	87 \pm 25	E _H (mV)	-103 \pm 7	-105 \pm 8
DO (mg L ⁻¹)	5.0 \pm 1.2	5.4 \pm 2.0	LOI (% C)	19.4 \pm 4.2	17.7 \pm 3.4

Conductivity (S m ⁻¹)	184 ± 11.9	210 ± 0.9	dw/ww (%)	14.5 ± 0.5	12.4 ± 0.5
Turbidity (NTU)	15.4 ± 2.4	13.1 ± 1.1	AVS (μmol g ⁻¹ dw)	4.3 ± 0.3	4.3 ± 0.3
Temperature (°C)	24.6 ± 0.2	24.9 ± 0.2	ΣSEM-AVS	6.1 ± 0.4	-2.2 ± 0.6
			ΣSEM/AVS	2.4 ± 0.1	0.4 ± 0.1
			[SEM-AVS]/f _{OC}	77.2 ± 5.9	-37.1 ± 11.2

PED = Pedreira Pumping Station; REF = Capivari River; pH = potential of hydrogen; ORP = oxidation reduction potential; DO = dissolved oxygen; NTU = nephelometric turbidity unit; E_H = redox potential; LOI = loss-on-ignition; dw/ww = dry weight/wet weight ratio; AVS = acid volatile sulfide; SEM = simultaneously extracted metals; f_{OC} = organic carbon fraction.

Table 2. Porewater concentrations of dissolved Fe, Mn, and Zn (± SD, *n* = 3) during short-term (4 h) resuspension and after 20 h post exposure in Pedreira Pumping Station (PED) and Capivari River (REF)

Condition	Fe (mg L ⁻¹)		Mn (μg L ⁻¹)		Zn (μg L ⁻¹)	
	PED	REF	PED	REF	PED	REF

Bedded	44a ± 9	42b ± 7	3,319a ± 181	1,175b ± 116	118a ± 6	16b ± 6
20 h post exposure	17a ± 2	21b ± 3	1,477a ± 84	986b ± 107	206a ± 5	82b ± 8

*Different letters indicate statistically significant differences between groups ($p < 0.01$).

Table 3. Acid-volatile sulfide (AVS) and simultaneously extracted metals (SEM) in Pedreira Pumping Station (PED) and Capivari River (REF) sediments (\pm SD, $n = 3$) during bedded and after 20 h post exposure conditions

Variable ($\mu\text{mol g}^{-1}$ dw)	Bedded		20 h post exposure*	
	REF	PED	REF	PED
AVS	4.33 ± 0.37	4.33 ± 0.33	3.29 ± 0.52	3.22 ± 0.36
Σ SEM-AVS	-2.23 ± 0.68	6.18 ± 0.48	-0.64 ± 0.26	9.16 ± 0.93
SEM/AVS	0.49 ± 0.15	2.43 ± 0.11	0.81 ± 0.05	3.88 ± 0.61
[SEM-AVS]/ f_{OC}	-37.18 ± 11.27	77.25 ± 5.95	-10.62 ± 4.37	114.45 ± 11.59

*SEM/AVS analysis for 20 h post exposure was determined in duplicates.