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In situ measurement of CuO and $Cu(OH)_2$ nanoparticle dissolution rates in quiescent freshwater mesocosms

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ABSTRACT

Recent studies have characterized copper-based nanoparticles (CBNPs) as relatively insoluble, raising potential persistence, accumulation, and toxicological concerns about their long-term application as agricultural pesticides. The dissolution *rates* of two CBNPs were measured in natural and artificial waters under both saturated and unsaturated conditions with respect to $CuO_{(s)}$ (total Cu<1 mg/kg). Kocide 3000®, an agricultural pesticide formulation with nano-scale $Cu(OH)_2$ particles, rapidly dissolved with an experimental half-life of less than eight (8) hours in natural water. Copper oxide nanoparticles were longer-lived, with an experimental half-life of seventy-three (73) hours in natural water. In contrast to prior reports of CuONP dissolution, our results suggest that even in moderately alkaline waters, CuO and $Cu(OH)_2$ NPs may persist as particles for days to weeks under quiescent conditions in a freshwater environment.

INTRODUCTION

Copper (Cu) salts and minerals have been extensively used as agricultural fungicides and algaecides for over 100 years.[1-3] Cu pesticides remain widely used today due to their low incidence of resistance development and accepted use in organic food production.[4-7] Several studies have shown that high concentrations of Cu in soils can lead to phytotoxicity in crops, toxicity in soil biota, and aquatic toxicity in bodies of water that receive runoff from agricultural lands.[8-15] Commercial pesticide formulations can contain nano-scale Cu minerals such as CuO or $Cu(OH)_2$.[16] Previous studies have indicated that Cu-based nanoparticles (CBNPs) are relatively insoluble and may have greater toxicity compared to their bulk or dissolved counterparts.[17-26] However, most previous studies determine the "% dissolved" Cu at the end of a specified time, and a NP concentration that is supersaturated with respect to CuO(s), typically by several orders of magnitude (Table S1).[22-27] This approach cannot provide reliable estimates of the *rate* of dissolution of nanomaterials needed for fate and exposure modeling.[28]

Only two studies have reported *rates* of dissolution of CuONPs, and indicate dissolution half-life times of 1.4 h and 2 h, respectively.[29, 30] However, Misra et al. used 750mg/L, well above saturation, and particles were 7nm spheres, smaller than those in most products. Kent and Vikesland used AFM measurements of particle height of $CuO/Cu(OH)_2$ grown from, and adhered to an AFM substrate to estimate a dissolution rate for flow through conditions in a stream.[29] These two reported rapid rates suggest extremely short lifetimes (hours) for CuONPs in the environment. However, neither situation accurately represents a realistic dissolution scenario at low particle concentration in a quiescent freshwater environment the particles will ultimately reside.

Several studies have indicated that pH, dissolved organic carbon (DOC), and inorganic ligands affect CBNP equilibrium solubility and can increase the number of dissolved Cu species in soil and water.[31-34] Thermodynamic modeling indicates that in redox-neutral waters, pH, DOC, and carbonate will largely determine Cu speciation at equilibrium (See SI Figure S1). Conway et al. found that Cu oxide nanoparticle (CuONP) equilibrium solubility correlated well with pH, total organic carbon, and ionic strength in several natural and artificial oxic waters.[23] However, it still remains unclear the extent to which these parameters affect the *rate* of dissolution.[35] Therefore, it is important to study the effects that pH, DOC, and transformations have on the rate of CBNP dissolution so that they may be used to predict the fate of CBNPs in the environment.

Here, we measure the *rates* of dissolution of dispersed CBNPs *in situ*, and at low concentration (1 mg/L) expected in the environemnt. This unique dataset provides critical dissolution rate data for CuONPs and a commercially used $Cu(OH)_2$ required for accurate environmental fate and exposure modeling. Additionally, we compare dissolution rates of CBNPs in ligand-free deionized water (pH=5.8) to that of natural water (pH=7.7) from a freshwater wetland mesocosm to qualitatively assess the combined effects of pH, organic, and inorganic ligands on dissolution rates. Using dissolution rate data, we estimated the dissolution half-life of CBNPs under these conditions.

MATERIALS AND METHODS

Thorough characterizations of the waters and nanoparticles used in this study are provided in the supporting information.

Dissolution Experiments. Dissolution experiments were carried out in both the laboratory (with and without mixing) and directly in the freshwater mesocosms. In all cases, Float-A-Lyzer G2 membrane dialysis devices (Spectrum Labs; Rancho Dominguez, CA) with a molecular weight cutoff of 8−10 kDa and a working volume of five milliliters were used to separate Cu nanoparticles from the dissolved Cu species as previously described.[30, 36] A stock solution of $Cu(NO_3)_2$ or particles (CuO or Kocide) was diluted to 1 mg/L with either ultrapure DI water or filtered mesocosm water. The suspensions were sonicated briefly, then five milliliters of the suspension or solution was put into the dialysis bag.

In laboratory experiments, the dialysis apparatuses were placed inside centrifuge tubes (50 ml) containing 20 mL of either ultrapure DI water or filtered mesocosm water. These were laid horizontally on an orbital shaker, and mixed at 200 rpm at 20 °C±2 °C. Each experiment was performed in duplicate. The media in the dialysate reservoir was replaced with fresh media after every sample was collected. Cu in the dialysate was determined by ICP-MS. The baseline copper concentration of the mesocosm water was also determined by ICP-MS, and ranged from 7 to 25 ug L^{-1}. This was then subtracted from the copper concentration measured in the mesocosm water experimental samples to determine the copper recovered due to dissolution and diffusion through the dialysis membrane. The pH of the DI and mesocosm water for the $Cu(NO_3)_2$, Kocide 3000® , and CuONP samples was measured after sample collection but prior to acidification and was found to be 5.8 ± 0.3 and 7.7 ± 0.3, respectively. A more thorough description of the dissolution experiments is provided in the supporting information.

Stagnant Dissolution Experiments. Dissolution was also determined under more environmentally relevant 'no-mix' conditions. One experiment was performed in a large tank of DI water to avoid buildup of Cu concentration in the dialysate (Figure S10). A second was performed *in-situ* at the Duke mesocosm facility (Figures S11 and S12). To determine the rate of Cu dissolution, a dialysis tube (Figure S13) was sacrificially sampled at specified time points over 72 hours. The concentration of Cu in the aqueous suspension aspirated from the tube and the mass of Cu attached to the membrane were both measured and the difference between the initial mass of Cu and the mass of Cu in the tube at the sampling interval was assumed to be the dissolved mass. A more thorough description of the stagnant dissolution experiments is provided in the supporting information.

Dissolution model. Fickian diffusion through the dialysis membrane was assumed and the theoretical diffusion model was fit to experimental data to calculate an effective first order diffusion rate constant, k_{diff}. For a cylinder, the fraction of dissolved Cu diffused at time t is given by:[37]

$$\frac{M_t}{M_\infty} = 1 - \frac{8}{\pi^2}\exp(-k_{diff}\pi^2 t) \quad (1)$$

$$\text{Where } k_{diff} = \frac{D}{L^2} \quad (2)$$

With M_t (µg) the mass of Cu diffused at time, t (hr), M_∞ (µg) the total mass in the system, L (m) the membrane thickness, and D the diffusivity coefficient ($m^2\ hr^{-1}$). The effective first order diffusion rate constant for Cu was determined from $Cu(NO_3)_2$. For particle dissolution scenarios, the calculated k^o_{diff} is a composition of the diffusion and dissolution processes in series:

$$\frac{1}{k^o_{diff}} = \frac{1}{k_{diff}} + \frac{1}{k_{diss}} \tag{3}$$

The reported k_{diss} (hr^{-1}) values for Cu NPs are determined from a fit of the dissolution data to yield k^o_{diff}, and k_{diff} determined from the $Cu(NO_3)_2$ experiments (diffusion only).

RESULTS AND DISCUSSION

Characteristics of CBNPs and Experimental Waters. A full discussion of the characteristics of the CuONPs can be found in Ma et al.[26] Briefly, TEM images indicated that the CuONPs are roughly spherical with diameters ranging from 30-50 nm and the XRD pattern of the CuONPs showed characteristic tenorite peaks at 2θ values of 36 and 39, which are typical of the pristine CuONPs used in previous studies.[24, 27, 30] The intensity-averaged HDD and ζ potential of the CuO and Kocide 3000® particles are presented in Table S4. X-Ray diffraction of Kocide 3000® indicated primarily $Cu(OH)_2$, consistent with the reported "active ingredient" in the product (Figure S3 in supporting information). TEM showed acicular particles that are typical of $Cu(OH)_2$[38] (Figures S5, S7, and S8 in supporting information). These particles were not stable in the TEM beam and transformed to small spheres after a short period of time (few tens of seconds).

Dissolution and Kinetic Parameters in "mixed" laboratory experiments. The percentage of dissolved Cu species recovered over time for dissolution of Cu $(NO_3)_2$, Kocide 3000®, and CuONP are presented in Figure 1. For $Cu(NO_3)_2$, Cu recoveries of greater than 95% were observed in DI and mesocosm water within 48 hours, indicating rapid diffusion of Cu species through the dialysis membrane. Similarly high levels of recovery were achieved for Kocide 3000®, with 97% of dissolved Cu recovered from DI water and 88% recovered from mesocosm water within 48 hours, indicating rapid dissolution of these particles and transport of dissolved Cu species across the membrane. The dissolution rate of the CuONPs in both waters was slower than for Kocide 3000 which is primarily $Cu(OH)_2$. The rates of dissolution for both

Kocide 3000® and CuONPs were slower in mesocosm water than in DI water. The pH disparity between the two waters, 5.8±0.3 for DI water and 7.7±0.3 for mesocosm water, was the factor most likely responsible for the divergence. These differences are discussed in detail later in the paper. The effective dissolution rate constant and half-lives of Kocide 3000® and the CuONPs were calculated from model fits of the dissolution data (Table S5). The dissolution half-life estimates indicate that, at low concentration (1 mg L^{-1}) Kocide 3000® should readily dissolve in water, with a half-life of 1-8 h. The CuONPs also readily dissolved, albeit more slowly with a half-life ranging from 30-73 h. This contrasts numerous reports of low solubility and perceived environmental persistence of CuONPs based on measurements made at higher concentrations of NPs.[22-26, 30] The data here suggests that the persistence of CBNPs in the water column will be a few days to a week, especially if the total Cu in the water remains under saturated with respect to $CuO_{(s)}$.

Effect of Water Quality Parameters on Dissolution. Both Kocide 3000® and CuONPs had greater dissolution rates in DI water than in mesocosm water. The difference can be attributed to the differences in the chemical characteristics of the water types. Namely, the lower pH of the DI water compared to the mesocosm water, divalent cations in the mesocosm water that increased the solutions ionic strength, and the presence of DOC in the mesocosm water.

Visual MINTEQ was used to estimate the Cu saturation concentrations with respect to $CuO_{(s)}$, 25 μg L^{-1} for mesocosm water and 7.6 mg L^{-1} for DI water, and model the Cu speciation for both waters. The results indicated that for total Cu concentrations of 800 μg L^{-1} and 400 μg L^{-1}, roughly equivalent to 1 mg L^{-1} CuONP and 1 ppm Kocide 3000® respectively, DI water (pH=5.8) is undersaturated with respect to $CuO_{(s)}$ whereas in mesocosm water (pH=7.7) is oversaturated. Only 25 μg L^{-1} of total Cu exists as dissolved Cu at equilibrium in mesocosm water, primarily Cu-DOM. The difference in speciation is the higher $[H^+]$ in DI water, which favors CuO dissolution (eqn 4).

$$\mathrm{CuO} + 2\mathrm{H}^+ \rightarrow \mathrm{Cu}^{2+} + \mathrm{H_2O};\quad \mathrm{K_{eq}} = \frac{\{\mathrm{Cu}^{2+}\}}{\{\mathrm{CuO}\}\{\mathrm{H}^+\}^2} \tag{4}$$

Despite the fact that the dialysis solution was changed at each sampling point to maintain a large driving force for dissolution, the differences in the dissolution rate in DI water compared to mesocosm water is likely due to accumulation of dissolved Cu between sampling events. The dissolution rate equation is given in eqn 5, where C_{sat} is the saturation Cu concentration and C_s is the Cu concentration at the particle-bulk solution interface.

$$\frac{dm}{dt} = k_{diss}(C_s - C_{sat}) \tag{5}$$

Dissolution in the undersaturated DI water proceeds at the maximum particle-specific rate until complete dissolution occurs. However, in mesocosm water the concentration of dissolved Cu rapidly approaches C_{sat}, which slows the rate of dissolution compared to the DI water condition. It is important to note that other factors may also have affected the dissolution rate. The higher $[H^+]$ in DI water may have also enhanced particle dissolution via surface protonation.[39] Wang et al. found that CuO underwent acid-promoted dissolution at similar pH values.[24] The higher pH and ionic strength of mesocosm water may have promoted aggregation as evidenced by the lower magnitude of ζ potential and higher HDD measured for CBNP suspensions in mesocosm water compared to DI water (Table S4). It is also possible that CBNPs in mesocosm water were coated by dissolved organic matter, potentially affecting dissolution. While passivating layers on metallic nanoparticles are typically oxide films,[40] Levard et al. suggested that organic coatings could also act as a passivating layer on Ag NPs , limiting ligand-enhanced dissolution.[41]

The observed differences in dissolution rates for CBNPs in both waters corroborates previous findings that while pH is the parameter that predominantly affects dissolution, other environmental factors (DOC, TDS, etc.) can influence dissolution under thermodynamic conditions in which CBNPs would otherwise be considered insoluble or poorly soluble.[23, 24, 26]

Effect of Mixing Conditions on Dissolution. Rates of CBNP dissolution under realistic stagnant conditions were lower the rates of the ‘well-mixed' laboratory experiments (Figure 2; Table S5).

Differences observed between the rate constants for the mesocosm water experiments may also be attributed to differences in environmental conditions. The 'well-mixed' laboratory experiments were conducted at a constant water temperature of 20°C while the water temperature of the *in-situ* experiments fluctuated from 10 to 16 °C, with direct implications for the dissolution rate. The pH of the mesocosm water in the well-mixed experiments was relatively constant at 7.7 ± 0.3 over the course of the experiment. In contrast, the natural diurnal pH variations of the mesocosm water *in-situ* rose sharply to 9 during the first 12 hours of the experiment, abruptly dropped to 7.5 in the subsequent 12 hours, and trended downward toward a final pH of 6.7 (Figure S14). It is expected that the higher initial pH inhibited dissolution over the first 12 hours and favored dissolution over the remainder of the experiment with respect to the controlled laboratory experiments do to dynamic changes in the saturation concentration of Cu with respect to the $CuO_{(s)}$ phase. For the 'well-mixed' laboratory experiments, samples of mesocosm water were centrifuged and filtered to remove suspended solids and microorganisms that might induce membrane fouling and inhibit Cu ion transport. The mesocosms contained those suspended solids and microorganisms during the *in-situ* experiments and membrane fouling may have contributed to the reduced dissolution rates during the final twenty-four hours of the experiment. Despite differences in temperature, pH, and biological activity, the stagnant *in-situ* and 'infinite dilution' experiments corroborate the 'well-mixed' laboratory experiments and also suggest CBNPs lifetimes of days to weeks.

Environmental Implications. These data suggest that, at relatively low aqueous concentrations, CBNPs are unlikely to become persistent pollutants in their nanoscale form, as has been previously suggested.[29] However, the dissolution half-life of eight (8) hours for Kocide 3000® and seventy-thee (73) hours for CuONP estimated in quiescent mesocosm water at low NP concentration is an order of magnitude slower than previously reported at high NP concentrations[30] and under flow conditions.[29] This difference in times scales (days to weeks vs. hours) suggests greater potential for uptake by organisms (e.g. plants) in the time that nanoparticles remain suspended in the water column. Long-term low dose chronic inputs of CBNPs in the water column may result is a steady-state concentration of particles that may behave differently than dissolved Cu species. Accumulation of dissolved or bulk Cu in agricultural soils may remain a potential source of ecotoxicity to sensitive, non-target organisms.[18, 21, 42-45]

CBNP dissolution appears to proceed at a slightly faster rate at lower pH values where the water is under saturated with respect to the $CuO_{(s)}$ phase despite a lack of ligand-enhanced Cu^{2+} ion release.[24] Even at higher pH where the water is oversaturated with respect to the $CuO_{(s)}$ phase, CBNP dissolution proceeds in natural waters, likely influenced by DOC, which is ubiquitous in agricultural runoff.[46-48] In addition, this work suggests that commercial formulations of nano-phase Cu minerals (primarily acicular $Cu(OH)_2$ particles) dissolve faster than CuONPs. Future investigations of CBNP transformation (e.g. sulfidation or phosphorylation) in soils and soil-pore water would further improve predictions of CBNP fate and transport in the environment. In particular, the dissolution rate of these materials in unsaturated soils with low water content where Cu may exceed its solubility limit with respect to $CuO_{(s)}$ must be determined as this may affect the persistence and bioavailability of these materials in unsaturated soils.

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ASSOCIATED CONTENT

Supporting Information Available: Tabular comparison of published studies on CBNP dissolution, a detailed description of the experimental methods, water characterization data, discussion of chemical equilibrium model and results, particle characterization data, XRD results, TEM and EDS results, and photographs of the ‘infinite dilution’ and *in-situ* dissolution experiments. This material is available free of charge via the Internet at http://pubs.acs.org.

The authors declare no competing financial interest.

REFERENCES

1. Smith, A. E.; Secoy, D. M., A compendium of inorganic substances used in European pest control before 1850. *J. Agric. Food. Chem.* **1976,** *24*, 1180-1186.
2. Taylor, E.; Holley, A. G.; Kirk, M., Pesticide development: a brief look at the history. *Southern Regional Extension Forestry* **2007,** *SREF-FM-010.*
3. Thornton, J. A.; Rast, W., *The use of copper and copper compounds as algicides*. 1997; p 123-142.
4. Fernandez-Cornejo, J.; Nehring, R.; Osteen, C.; Wechsler, S.; Martin, A.; Vialou, A., Pesticide Use in US Agriculture: 21 Selected Crops, 1960-2008. *Economic Research Service, United States Department of Agriculture* **2014**, 40-49.
5. Exemptions from the requirement of a tolerance. In *40 C.F.R. § 180.900*, 2004.
6. Synthetic substances allowed for use in organic crop production. In *7 C.F.R. § 205.601* 2000.
7. Brent, K. J.; Hollomon, D. W., *Fungicide resistance in crop pathogens: How can it be managed?* GIFAP Brussels: 1995.
8. Zhou, X.; He, Z.; Liang, Z.; Stoffella, P. J.; Fan, J.; Yang, Y.; Powell, C. A., Long-term use of copper-containing fungicide affects microbial properties of citrus grove soils. *Soil Sci. Soc. Am. J.* **2011,** *75*, 898-906.
9. Wang, Q.-Y.; Zhou, D.-M.; Cang, L., Microbial and enzyme properties of apple orchard soil as affected by long-term application of copper fungicide. *Soil Biol. Biochem.* **2009,** *41*, 1504-1509.
10. Wightwick, A. M.; Mollah, M. R.; Partington, D. L.; Allinson, G., Copper fungicide residues in Australian vineyard soils. *J. Agric. Food. Chem.* **2008,** *56*, 2457-2464.
11. Eijsackers, H.; Beneke, P.; Maboeta, M.; Louw, J.; Reinecke, A., The implications of copper fungicide usage in vineyards for earthworm activity and resulting sustainable soil quality. *Ecotoxicol. Environ. Saf.* **2005,** *62*, 99-111.
12. Van Zwieten, L.; Rust, J.; Kingston, T.; Merrington, G.; Morris, S., Influence of copper fungicide residues on occurrence of earthworms in avocado orchard soils. *Sci. Total Environ.* **2004,** *329*, 29-41.
13. Michaud, A.; Bravin, M.; Galleguillos, M.; Hinsinger, P., Copper uptake and phytotoxicity as assessed in situ for durum wheat (Triticum turgidum durum L.) cultivated in Cu-contaminated, former vineyard soils. *Plant Soil* **2007,** *298*, 99-111.
14. Kiaune, L.; Singhasemanon, N., Pesticidal copper (I) oxide: environmental fate and aquatic toxicity. In *Reviews of Environmental Contamination and Toxicology* Springer: New York, NY, 2011; Vol. 213, pp 1-26.
15. Kungolos, A.; Emmanouil, C.; Tsiridis, V.; Tsiropoulos, N., Evaluation of toxic and interactive toxic effects of three agrochemicals and copper using a battery of microbiotests. *Sci. Total Environ.* **2009,** *407*, 4610-4615.
16. Tegenaw, A.; Tolaymat, T.; Al-Abed, S.; El Badawy, A.; Luxton, T.; Sorial, G.; Genaidy, A., Characterization and Potential Environmental Implications of Select Cu-Based Fungicides and Bactericides Employed in US Markets. *Environ. Sci. Technol.* **2015,** *49*, 1294-1302.
17. Aruoja, V.; Dubourguier, H.-C.; Kasemets, K.; Kahru, A., Toxicity of nanoparticles of CuO, ZnO and TiO 2 to microalgae Pseudokirchneriella subcapitata. *Sci. Total Environ.* **2009,** *407*, 1461-1468.
18. Bondarenko, O.; Juganson, K.; Ivask, A.; Kasemets, K.; Mortimer, M.; Kahru, A., Toxicity of Ag, CuO and ZnO nanoparticles to selected environmentally relevant test organisms and mammalian cells in vitro: a critical review. *Arch. Toxicol.* **2013,** *87*, 1181-1200.
19. Kasemets, K.; Ivask, A.; Dubourguier, H.-C.; Kahru, A., Toxicity of nanoparticles of ZnO, CuO and TiO 2 to yeast Saccharomyces cerevisiae. *Toxicol. In Vitro* **2009,** *23*, 1116-1122.

20. Kaweeteerawat, C.; Chang, C. H.; Roy, K. R.; Liu, R.; Li, R.; Toso, D.; Fischer, H.; Ivask, A.; Ji, Z.; Zink, J. I., Cu Nanoparticles Have Different Impacts in Escherichia coli and Lactobacillus brevis than Their Microsized and Ionic Analogues. *ACS Nano* **2015,** *9*, 7215-7225.

21. Wang, Z.; Li, J.; Zhao, J.; Xing, B., Toxicity and internalization of CuO nanoparticles to prokaryotic alga Microcystis aeruginosa as affected by dissolved organic matter. *Environ. Sci. Technol.* **2011,** *45*, 6032-6040.

22. Adeleye, A. S.; Conway, J. R.; Perez, T.; Rutten, P.; Keller, A. A., Influence of extracellular polymeric substances on the long-term fate, dissolution, and speciation of copper-based nanoparticles. *Environ. Sci. Technol.* **2014,** *48*, 12561-12568.

23. Conway, J. R.; Adeleye, A. S.; Gardea-Torresdey, J.; Keller, A. A., Aggregation, Dissolution, and Transformation of Copper Nanoparticles in Natural Waters. *Environ. Sci. Technol.* **2015,** *49*, 2749-2756.

24. Wang, Z.; Von Dem Bussche, A.; Kabadi, P. K.; Kane, A. B.; Hurt, R. H., Biological and environmental transformations of copper-based nanomaterials. *ACS Nano* **2013,** *7*, 8715-8727.

25. Wang, L.-F.; Habibul, N.; He, D.-Q.; Li, W.-W.; Zhang, X.; Jiang, H.; Yu, H.-Q., Copper release from copper nanoparticles in the presence of natural organic matter. *Water Res.* **2015,** *68*, 12-23.

26. Ma, R.; Stegemeier, J.; Levard, C.; Dale, J. G.; Noack, C. W.; Yang, T.; Brown, G. E.; Lowry, G. V., Sulfidation of copper oxide nanoparticles and properties of resulting copper sulfide. *Environ. Sci.: Nano* **2014,** *1*, 347-357.

27. Mudunkotuwa, I. A.; Pettibone, J. M.; Grassian, V. H., Environmental implications of nanoparticle aging in the processing and fate of copper-based nanomaterials. *Environ. Sci. Technol.* **2012,** *46*, 7001-7010.

28. Dale, A. L.; Casman, E. A.; Lowry, G. V.; Lead, J. R.; Viparelli, E.; Baalousha, M., Modeling nanomaterial environmental fate in aquatic systems. *Environ. Sci. Technol.* **2015,** *49*, (5), 2587-2593.

29. Kent, R. D.; Vikesland, P. J., Dissolution and Persistence of Copper-Based Nanomaterials in Undersaturated Solutions with Respect to Cupric Solid Phases. *Environ. Sci. Technol.* **2016**, (50), 6772-6781.

30. Misra, S. K.; Dybowska, A.; Berhanu, D.; Croteau, M. N. l.; Luoma, S. N.; Boccaccini, A. R.; Valsami-Jones, E., Isotopically modified nanoparticles for enhanced detection in bioaccumulation studies. *Environ. Sci. Technol.* **2012,** *46*, 1216-1222.

31. Meador, J. P., The interaction of pH, dissolved organic carbon, and total copper in the determination of ionic copper and toxicity. *Aquat. Toxicol.* **1991,** *19*, 13-31.

32. Flemming, C.; Trevors, J., Copper toxicity and chemistry in the environment: a review. *Water, Air, Soil Pollut.* **1989,** *44*, 143-158.

33. Son, J.; Vavra, J.; Forbes, V. E., Effects of water quality parameters on agglomeration and dissolution of copper oxide nanoparticles (CuO-NPs) using a central composite circumscribed design. *Sci. Total Environ.* **2015,** *521*, 183-190.

34. Odzak, N.; Kistler, D.; Behra, R.; Sigg, L., Dissolution of metal and metal oxide nanoparticles in aqueous media. *Environ. Pollut.* **2014,** *191*, 132-138.

35. Misra, S. K.; Dybowska, A.; Berhanu, D.; Luoma, S. N.; Valsami-Jones, E., The complexity of nanoparticle dissolution and its importance in nanotoxicological studies. *Sci. Total Environ.* **2012,** *438*, 225-232.

36. Levard, C.; Mitra, S.; Yang, T.; Jew, A. D.; Badireddy, A. R.; Lowry, G. V.; Brown Jr, G. E., Effect of chloride on the dissolution rate of silver nanoparticles and toxicity to E. coli. *Environ. Sci. Technol.* **2013,** *47*, 5738-5745.

37. Crank, J., *The mathematics of diffusion*. Oxford university press: New York, NY, 1979.

38. Durand-Keklikian, L.; Matijević, E., Needle-type colloidal copper (II) hydroxide particles. *Colloid. Polym. Sci.* **1990,** *268*, 1151-1158.

39. Stumm, W.; Wollast, R., Coordination chemistry of weathering: Kinetics of the surface ‐ controlled dissolution of oxide minerals. *Rev. Geophys.* **1990,** *28*, 53-69.

40. Borm, P.; Klaessig, F. C.; Landry, T. D.; Moudgil, B.; Pauluhn, J.; Thomas, K.; Trottier, R.; Wood, S., Research strategies for safety evaluation of nanomaterials, part V: role of dissolution in biological fate and effects of nanoscale particles. *Toxicol. Sci.* **2006,** *90*, 23-32.
41. Levard, C.; Hotze, E. M.; Lowry, G. V.; Brown Jr, G. E., Environmental transformations of silver nanoparticles: impact on stability and toxicity. *Environ. Sci. Technol.* **2012,** *46*, 6900-6914.
42. Alva, A.; Huang, B.; Paramasivam, S., Soil pH affects copper fractionation and phytotoxicity. *Soil Sci. Soc. Am. J.* **2000,** *64*, 955-962.
43. Chatterjee, J.; Chatterjee, C., Phytotoxicity of cobalt, chromium and copper in cauliflower. *Environ. Pollut.* **2000,** *109*, 69-74.
44. McBride, M. B.; MartÍnez, C. E., Copper phytotoxicity in a contaminated soil: remediation tests with adsorptive materials. *Environ. Sci. Technol.* **2000,** *34*, 4386-4391.
45. Moore, J. D.; Stegemeier, J. P.; Bibby, K.; Marinakos, S. M.; Lowry, G. V.; Gregory, K. B., Impacts of pristine and transformed Ag and Cu engineered nanomaterials on surficial sediment microbial communities appear short-lived. *Environ. Sci. Technol.* **2016,** *50*, (5), 2641-2651.
46. Wilson, H. F.; Xenopoulos, M. A., Effects of agricultural land use on the composition of fluvial dissolved organic matter. *Nat. Geosci.* **2009,** *2*, 37-41.
47. Fujii, R.; Ranalli, A.; Aiken, G.; Bergamaschi, B., Dissolved organic carbon concentrations and compositions, and trihalomethane formation potentials in waters from agricultural peat soils, Sacramento-San Joaquin Delta, California: Implications for drinking-water quality. In *U.S. Geological Survey*, Sacramento, CA, 1998; Vol. 98-4147.
48. Royer, T. V.; David, M. B., Export of dissolved organic carbon from agricultural streams in Illinois, USA. *Aquat. Sci.* **2005,** *67*, 465-471.

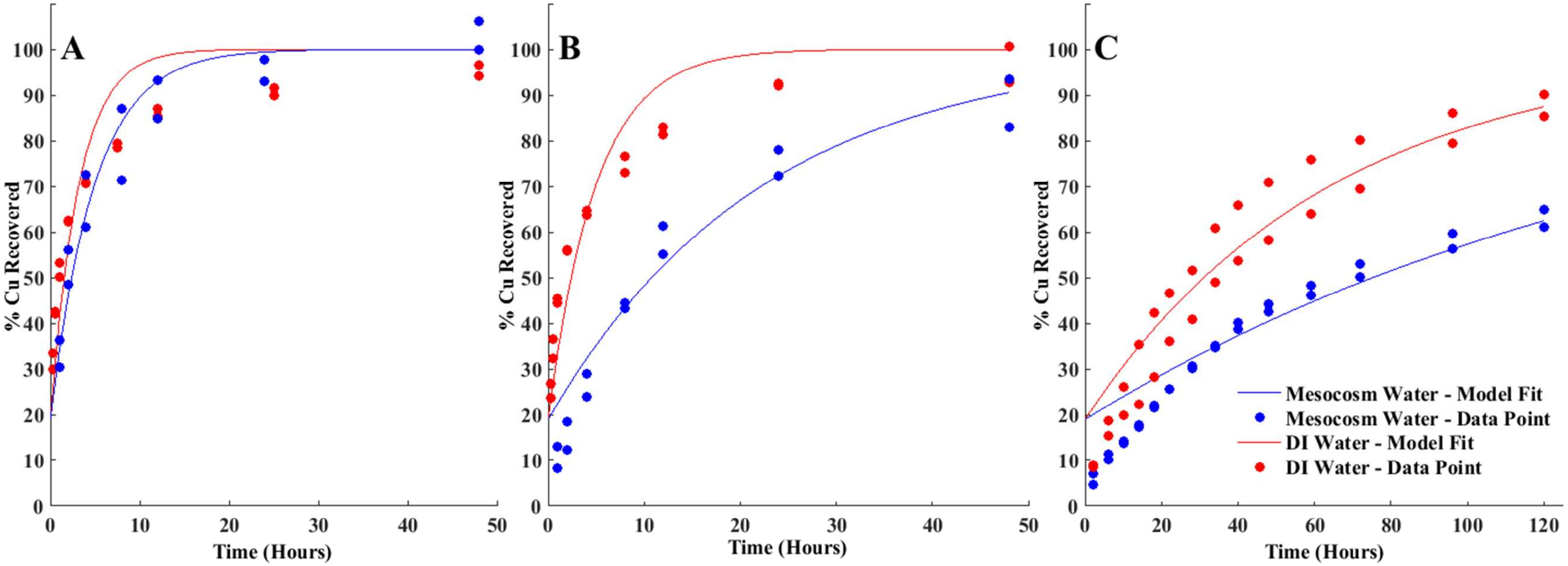

Figure 1. First-order dissolution model fit and duplicate experimental data for A) cupric nitrate, B) Kocide 3000®, and C) CuONPs in "mixed" DI and mesocosm water. The calculated half-life times from fits of eqn 1 are 1h and 8h for Kocide 3000®, and 30h and 73h for CuONPs, in DI and mesocosm water, respectively. The model fit for CuONPs suggest that there were two populations of particles, with faster initial dissolution followed by slower dissolution at later times. However, this range of dissolution rates is captured in the 95% confidence limits for the fitted value of k_{diss} (Table S5)

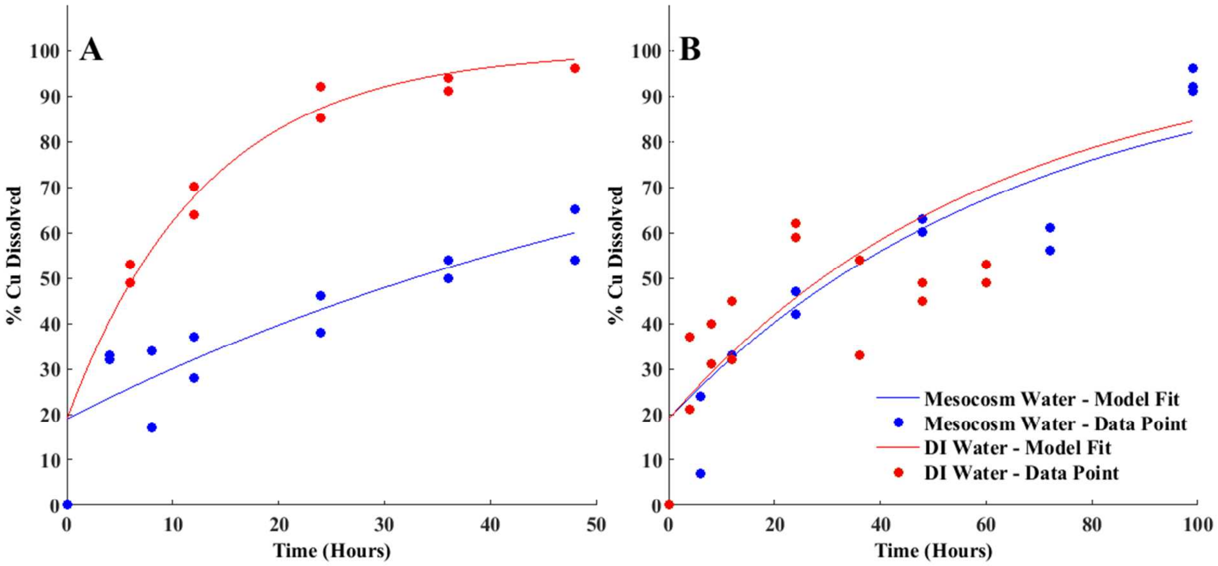

Figure 2. First-order dissolution model fit and duplicate experimental data for stagnant dissolution of A) Kocide 3000® and B) CuONPs in DI and mesocosm waters under 'infinite dilution' and *in*-situ conditions, respectively. The calculated half-life times from fits of eqn 1 are 6h and 33h for Kocide 3000®, and 32h and 29h for CuONPs, in DI and mesocosm water, respectively. Rate constants and 95% confidence limits for these data fits are provided in Table S5.

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