The persistence and transformation of silver nanoparticles in littoral lake mesocosms monitored using various analytical techniques

Lindsay M. Furtado, ^A *Md Ehsanul Hoque*, ^A *Denise M. Mitrano*, ^{B,D} *James F. Ranville*, ^B *Beth Cheever*, ^{C,E} *Paul C. Frost*, ^C *Marguerite A. Xenopoulos*, ^C *Holger Hintelmann*^A *and Chris D. Metcalfe*^{A,F}

^ATrent University, Water Quality Center, 1600 Westbank Drive, Peterborough, ON, K9J 7B8, Canada.

^BColorado School of Mines, Department of Chemistry and Geochemistry, 1500 Illinois Street, Golden, CO 80401, USA.

^CTrent University, Department of Biology, 1600 Westbank Drive, Peterborough, ON, K9J 7B8, Canada.

^DPresent address: Empa – Swiss Federal Laboratories for Materials Science and Technology, Technology and Society Laboratory, Lerchenfeldstrasse 5, CH-9014 St Gallen, Switzerland.

^EPresent address: Michigan State University, Department of Microbiology and Molecular Genetics, 220 Trowbridge Road, East Lansing, MI 48824, USA.

^FCorresponding author. Email: cmetcalfe@trentu.ca



Fig. S1. Picture of a mesocosm used in the study installed in Lake 239 at the Experimental Lakes Area, ON, Canada.

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Fig. S2. Single particle inductively coupled plasma mass spectrometry (spICP-MS) results for fresh 60-nm polyvinylpyrrolidine (PVP)-capped particles (fresh), flash frozen 60-nm PVP-capped particles that were stored at - 80 °C for a week (frozen), and 60-nm PVP-capped particles that had remained at room temperature for a week (room). All concentrations were at 50 ppt.

Determination of extraction efficiency for cloud point extraction

Aliquots of polyvinylpyrrolidine (PVP)-silver nanoparticle (AgNP) or AgNO₃ stock solutions were spiked to the sample matrix immediately before performing the cloud point extraction (CPE) procedure described in the experimental section. The extraction efficiency was calculated as the ratio of the Ag concentration measured after extraction (adjusted for the $4\times$ dilution factor) and the initial Ag concentration of the sample. To determine the initial Ag concentration in the sample, sub-samples of the spiked AgNP or AgNO₃ solutions were taken for total Ag analysis before performing the CPE procedure. The initial Ag content measured before extraction was also adjusted for the percentage of dissolved silver (dAg) found in the AgNP stock solutions as determined by a separate analysis using 3-kDa ultrafiltration.

Table S1.Extraction efficiencies by cloud point extraction (CPE) of Ag⁺, 50-nmpolyvinylpyrrolidine (PVP) silver nanoparticles (AgNPs), and a mixture of Ag⁺ with 50-nm PVPAgNPs in deionised water (DI) and 0.2-μm pre-filtered Lake 239 water

Mean \pm s.d, is presented (n = 3). AgNP concentration before CPE was determined from the Ag

concentration measured before CPE, adjusted for the percentage of dissolved Ag (dAg) in AgNP stock

Matrix	Form of Ag spiked in sample	Ag concentration before CPE	AgNP concentration	Ag concentration extracted ($\mu g L^{-1}$)	Extraction efficiency (%)
	1 1	$(\mu g L^{-1})$	before CPE		
			$(\mu g L^{-1})$		
DI	AgNO ₃	35.7 ± 0.2	n.a	0.6 ± 0.1	1.7 ± 0.0
	AgNP	28.6 ± 0.1	27.5 ± 0.1	20.7 ± 0.6	75.3 ± 2.1
	$AgNO_3 + AgNP$	62.7 ± 0.5	27.5 ± 0.1	20.2 ± 1.1	$73.5\pm3.9^{\rm A}$
Lake	AgNO ₃	33.7 ± 0.4	n.a.	0.7 ± 0.2	2.1 ± 0.0
239	AgNP	26.3 ± 0.3	25.3 ± 0.3	10.3 ± 2.3	40.9 ± 9.2
	$AgNO_3 + AgNP$	62.8 ± 0.8	25.3 ± 0.3	10.1 ± 2.0	$40.1\pm7.8^{\rm A}$

solution. n.a, not apparent

^AApparent extraction efficiency with respect to initial concentration of AgNPs as measured in samples with only

AgNPs and assuming Ag⁺ was not extracted.

Table S2.	Transmission electron microscopy (TEM) operating conditions
zing mode	High resolution TEM low dose TEM nanonrobe free lens cou

Working mode	High-resolution TEM, low dose TEM, nanoprobe, free lens control		
Gun	LaB6-cathode thermionic gun		
Point-to-point resolution	0.24 nm		
Line resolution	0.17 nm		
Operation voltage	200 kV		

Table S3. Instrumentation and operating parameters for asymmetric flow field flow fractionation with on-line inductively coupled plasma mass spectrometry (AF4-ICP-MS) analysis

ICP-MS model	XSeries 2 (Thermo Scientific)
Nebuliser	Conikal U-series (isoSPEC; 1 mL min ⁻¹)
Spray chamber	Conical with impact bed
Cones	Ni
Analysis mode	Time resolved analysis
Isotopes monitored	¹⁰⁷ Ag, ¹¹⁵ In
Dwell time	20 000 µs

Table S4. Asymmetric flow field flow fractionation (AF4S) channel description and operating narameters

	parameters				
Spacer thickness	350 μm				
Membrane type	regenerated cellulose with 10 kDa MWCO ^A				
Injection volume	100 μL				
Carrier fluid	500 mg L^{-1} sodium dodecyl sulfate				
Channel flow	1.0 mL min^{-1}				
Cross flow	0.7 mL min^{-1}				
Focus flow	0.2 mL min^{-1}				
Rinse flow	Cross flow off, 1.0 mL min^{-1}				
Focussing time	10 min				
Elution time	35 min				
Rinse time	10 min				
Total analysis time	55 min				

^AMolecular weight cut-off

Table S5. Size information for polyvinylpyrrolidine (PVP) silver nanoparticle (AgNP) standards

Particles were sized by the manufacturer (NanoComposix, CA, USA) using transmission electron

microscopy (TEM) and dynamic light scattering (DLS) analysis

Nominal size (nm)	TEM diameter (nm)	DLS hydrodynamic diameter (nm)
20	20.0 ± 1.8	25.8
40	40.7 ± 4.1	49.3
50	48.3 ± 3.7	56.3
60	59.9 ± 3.9	77.1
80	82.1 ± 4.4	93.3



Fig. S3. Asymmetric flow field flow fractionation with on-line inductively coupled plasma mass spectrometry (AF4-ICP-MS) fractograms for various sizes of polyvinylpyrrolidine (PVP) silver nanoparticles (AgNPs) (100 μ g Ag L⁻¹). The corresponding hydrodynamic size is shown in parentheses.



Fig. S4. Asymmetric flow field flow fractionation with on-line inductively coupled plasma mass spectrometry (AF4-ICP-MS) fractograms for various concentrations of 50-nm polyvinylpyrrolidine (PVP) silver nanoparticles (AgNPs) and the resulting standard curve.

Table S6. Concentration of major anions and cations in Lake 239

Samples taken in August, 2012, mean \pm standard deviation, n = 3

Major Anions and Cations (mg L^{-1})					
Ca ²⁺	\mathbf{K}^+	Mg^{2+}	Na ⁺	Cl	$\mathrm{SO_4}^{2-}$
3.45 ± 0.04	0.43 ± 0.00	0.85 ± 0.01	1.10 ± 0.01	$0.27 \pm$	2.25 ± 0.01
				0.01	

Table S7.Extraction efficiencies by cloud point extraction (CPE) of 50-nm polyvinylpyrrolidine
(PVP) silver nanoparticles (AgNPs) at various concentrations in deionised (DI) water

Mean \pm standard deviation is presented (n = 3). AgNP concentration before CPE was determined from the Ag concentration measured before CPE, adjusted for the percentage of dissolved Ag (dAg) in AgNP stock solution

Ag concentration	AgNP	Ag concentration	Extraction
before CPE	concentration	extracted (µg L ⁻)	efficiency (%)
(µg L ⁻)	before CPE		• • •
	(µg L⁻)		
7.4 ± 0.1	6.6 ± 0.1	5.7 ± 0.1	85.4 ± 1.6
18.8 ± 0.4	17.0 ± 0.4	13.1 ± 0.5	77.3 ± 3.4
33.6 ± 0.5	30.2 ± 0.5	22.6 ± 0.3	74.9 ± 2.0
58.3 ± 0.3	52.5 ± 0.3	43.4 ± 2.6	82.7 ± 4.5
72.7 ± 0.4	65.4 ± 0.4	54.1 ± 1.0	82.6 ± 1.4

Table S8.Extraction efficiencies by cloud point extraction (CPE) of various sizes of
polyvinylpyrrolidine (PVP) silver nanoparticles (AgNPs) in deionised (DI) water

Mean \pm standard deviation is presented (n = 3). All sizes of PVP AgNPs were obtained from NanoComposix. AgNP concentration before CPE was determined from the Ag concentration measured before CPE, adjusted for the percentage of dissolved Ag (dAg) in AgNP stock solution

AgNP Size (nm)	Ag concentration before CPE (μ g L ⁻¹)	AgNP concentration before CPE (μ g L ⁻¹)	Ag concentration extracted ($\mu g L^{-1}$)	Extraction efficiency (%)
10	30.8 ± 0.3	18.3 ± 0.2	9.6 ± 0.4	52.4 ± 2.2
20	36.5 ± 0.7	26.3 ± 0.5	14.3 ± 1.1	54.2 ± 3.7
40	35.3 ± 0.2	33.3 ± 0.2	24.2 ± 0.5	72.7 ± 1.1
50	33.9 ± 0.5	30.5 ± 0.4	22.5 ± 0.0	73.9 ± 1.1
60	29.5 ± 0.0	23.4 ± 0.0	16.9 ± 0.4	72.3 ± 1.7
80	42.6 ± 0.5	40.7 ± 0.5	33.8 ± 0.8	82.9 ± 1.0



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Label A: std (Nrm.%= 38.86, 20.96, 34.83, 1.14, 3.84, 0.28)



Fig. S5. Transmission electron microscopy (TEM) images of nanoparticles in the TX-114 rich phase after cloud point extraction (CPE) of mesocosm sample (24 h, replicate 1) (top) and the corresponding energy dispersive X-ray spectroscopy (EDXS) image (bottom). The nanoparticles could not be confirmed as silver nanoparticles (AgNPs) by X-ray diffraction analysis.



Fig. S6. Concentration of silver nanoparticles (AgNPs) (particles L^{-1}) over time in mesocosm replicates as measured by single particle inductively coupled plasma mass spectrometry (spICP-MS).



Fig. S7. Asymmetric flow field flow fractionation with on-line inductively coupled plasma mass spectrometry (AF4-ICP-MS) fractograms and standard curve for injection of 40- and 80-nm polyvinylpyrrolidine (PVP) silver nanoparticle (AgNP) standards (100 μ g Ag L⁻¹) for daily size calibration of mesocosm samples. The calibration curve was extended through the origin.



Fig. S8. Asymmetric flow field flow fractionation with on-line inductively coupled plasma mass spectrometry (AF4-ICP-MS) fractograms for mesocosm samples. The *y*-axis displays the normalised 107 Ag intensity. Results from one sample analysed from mesocosm replicate 1 (black) and mesocosm replicate 2 (blue) are presented. Time points of 6 h and 21 days were not analysed.



Fig. S9. Single particle inductively coupled plasma mass spectrometry (spICP-MS) histograms for mesocosm samples. Average results from duplicate analysis of one sample from mesocosm replicate 1 (blue) and mesocosm replicate 2 (red) are presented.