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CHEMICAL SPECIATION OF SILVER(I) IN FRESHWATER

By

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Abstract

Recently, background silver concentrations in freshwater have been shown to be only 0.001-0.01 nanomolar. However, locations with significant sewage input have concentrations at least two orders of magnitude greater. Although geochemical processes appear to regulate background silver at picomolar levels, there is concern that point-discharges of silver may produce acute and/or chronic toxic responses in aquatic organisms. How silver speciates in these waters will largely determine its bioavailability and toxicity. This thesis project investigated silver(I) speciation in freshwater.

Silver binds much more strongly to sulfide, S(-II), containing ligands compared with ligands containing nitrogen and oxygen. Determinations of silver-sulfide formation constants gave log K values of 11.0 to 13.0 compared with log K values of <6.0 for ligands containing nitrogen and oxygen. Thermodynamic calculations suggest that silver in freshwaters is over-saturated relative to $Ag_2S(s)$ solubility (K_{sp} = 35.94, 25°C, $\mu = 0.0$) and that the predominant silver species in freshwater should be $AgHS^{\circ}$ and Ag-thiolates. However, field studies at a sewage treatment plant, Dundas, Ontario and at an old mining camp, Cobalt, Ontario, demonstrated that silver is 90-100% colloidally bound, which explains silver's apparent over-saturation in natural waters. Concentrations of aquo silver species ($AgHS^{\circ}$ and Ag-thiolates) represent <10% of total silver in these waters. The field studies also demonstrated that inorganic sulfide is present in oxic waters at 10-200 nM (80-100% colloidally bound) while thiol concentrations are below 1 nM. Using a competitive ligand equilibration-solvent extraction technique we showed that silver is bound to the inorganic sulfide. This inorganic sulfide is likely present as metal sulfide clusters that are bound to natural organic matter (NOM). The complexation of sulfide by metals would explain its persistence even in oxic waters.

In sediments silver likely forms $Ag_2S(s)$ as long as the silver to sulfide ratio is <1. A laboratory study demonstrated that silver reacts rapidly with sedimentary FeS and behaves thermodynamically like a distinct acanthite, $Ag_2S(s)$, phase. Silver measurable in the porewaters is probably associated with colloidal metal sulfides, as was observed for the surface waters and sewage treatment plant effluents.

In summary, the findings of this thesis research showed for the first time that sulfides are important to the speciation of silver not only in anoxic sediments but also in oxic surface waters and sewage treatment plant effluents. Based on these findings, we recommend that the SEM/AVS approach, developed as a sediment quality criterion, could also be used as a water quality criterion for silver. However, before adopting this approach, toxicity studies need to be carried out using well-characterized silver-sulfide species to verify that sulfide protects aquatic organisms from silver bioaccumulation and toxicity.

Key Words: Silver, sulfide, speciation, equilibrium constants, sewage treatment plants and sediment porewaters.

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AVS	acid volatile sulfides
MATC	maximum acceptable toxicant concentration
SEM	simultaneously extracted metals
WTP	municipal wastewater treatment plant

1. Thesis Objective

The bioavailability and toxicity of metals are integrally linked to their physico-chemical form (species). In order to conduct meaningful toxicity experiments and to develop intelligent regulations, we must, therefore, have knowledge of how silver speciates in the environment. To date, few studies exist which investigate silver speciation in natural waters.

The objective of this thesis is to investigate several important questions relating to silver speciation in freshwaters. These questions are: (1) what are the important chemical forms (species) of silver in the aqueous phase, (2) how is silver bound in sediments, (3) how does silver partition between sediments and porewaters, and (4) how stable are the different forms of silver?

These questions are addressed by drawing on the results and conclusions of the five papers presented in Appendices I-V along with findings from the literature. In the text the papers are referred to by their Roman numerals.

I. Reactivity of Ag^+ Ion with Thiol Ligands in the Presence of Iron Sulfide. Adams N.W.H. and Kramer J.R. (1998) *Environ. Toxicol. Chem.* **17**:625-629.

II. Potentiometric Determination of Silver Thiolate Formation Constants Using a Ag_2S Electrode. Adams N.W.H. and Kramer J.R. (1999) *Aquatic Geochem.* **5**:1-11.

III. Silver Speciation in Wastewater Effluent, Surface Waters and Porewaters. Adams N.W.H. and Kramer J.R. *Environ. Toxicol. Chem.* Accepted February 10, 1999.

IV. Determination of Silver Complexation in Wastewater and Receiving Waters by Competitive Ligand Equilibration/Solvent Extraction. Adams N.W.H. and Kramer J.R. *Environ. Toxicol. Chem.* Accepted February 10, 1999.

V. Silver at an Old Mining Camp, Cobalt, Ontario, Canada. Kramer J.R., Adams N.W.H., Manolopoulos H. and Collins P.V. (1999) *Environ. Toxicol. Chem.* **18**:23-29.

2. General Introduction

2.1 Silver Concentrations in Freshwater

There is limited historical data available regarding the distribution, fate and reactivity of silver in the aquatic environment. Reliable data on silver concentrations in freshwaters have been provided by a few recent studies, conducted using ultra-clean sampling and analytical practices (Paper III; Hurley et al., 1996; Benoit et al., 1997). Background silver concentrations in freshwater are extremely low (0.001-0.01 nanomolar) (Table 1a). However, higher concentrations (at least two orders of magnitude) have been documented in locations with significant sewage input (Table 1b). Although geochemical processes appear to regulate background silver to picomolar levels, there is concern that point discharges of silver may produce acute and/or chronic toxic responses in aquatic organisms.

2.2 Toxicity of Silver to Aquatic Organisms

Silver is a trace element with no known biological function (Fisher and Wang, 1998). As a class B metal, silver has a very high affinity for sulfur(-II) ligands. This affinity for sulfur(-II) leads to its association with proteins and the potential for interference with protein metabolism in organisms (Fisher and Wang, 1998). For this reason, silver is among the most toxic metals for aquatic organisms; toxicity has been reported at sub-nanomolar concentration levels.

Metal toxicity is dependent upon bioavailability and bioaccumulation in aquatic organisms. In turn, bioavailability is dependent on the physico-chemical form (species) of a metal (Kramer et al., 1997; Connell et al., 1991). Often the free metal ion is the most toxic form, and metals complexed with inorganic anions and dissolved organic compounds display lower degrees of

Table 1a Silver Concentrations in Uncontaminated Natural Waters

Marine Waters

0.001-0.002 nM	northeast Pacific coastal waters	(Sanudo-Wilhelmy and Flegal, 1992)

Freshwaters

0.001-0.005 nM	river water in Wisconsin, Michigan and Minnesota	(Shafer et al., 1994)
0.003-0.004 nM	Lake Michigan	(Shafer et al., 1995)
<0.01 nM	river water in Connecticut	(Benoit et al., 1997)
<0.0001-0.005 nM	river water in Texas	(Wen et al., 1997)
0.01-0.07 nM	near old mining site in Ontario	(Paper V)

Table 1b Silver Concentrations in Contaminated Natural Waters

Marine Waters

0.01-0.1 nM	San Francisco Bay	(Sanudo-Wilhelmy et al., 1996)

Freshwaters

0.01-0.03 nM	river water in Wisconsin, Michigan and Minnesota	(Shafer et al., 1994)
0.25-1 nM	river water in Connecticut	(Rozan et al., 1995)
0.005-0.15 nM	river water in Texas	(Wen et al., 1997)
0.1-1.5 nM	river water in Ontario	(Paper III)
1-2 nM	old mining site in Ontario	(Paper V)

bioavailability and toxicity (Kramer et al., 1997). This general rule, however, is not always valid. Some metal complexes may have enhanced bioavailability. For example, it has been suggested that the low polarity of the neutral complex $AgCl^{\circ}$ may facilitate diffusion across biological membranes (Luoma et al., 1995). The situation is further complicated by the fact that high-affinity binding sites, present in organisms, compete with ligands in the water for metal ions. Thus not all ligands can serve to protect organisms from bioaccumulating metals.

Studies of acute toxicity using $AgNO_3$ on a variety of freshwater fish have given 96-h LC_{50} values of 50-650 nM, making $AgNO_3$ one of the most toxic metal salts, more potent than copper or cadmium salts (Hogstrand and Wood, 1998). However, these LC_{50} values are at least 2-3 orders of magnitude greater than silver concentrations measured even in polluted waters. Furthermore, tests with $AgNO_3$ are not environmentally relevant, since in natural waters, only an extremely small portion of the total silver in the water will exist as free ionic Ag^+. Although the information on acute toxicity of complexed silver is more sparse, studies conducted with thiosulfate, chloride, sulfide and DOC suggest that complexed silver is far less toxic than Ag^+. 96-h LC_{50} values are at least two- to four-fold greater than 96-h LC_{50} values obtained from studies with $AgNO_3$ (Hogstrand and Wood, 1998).

Chronic thresholds are much lower than acute thresholds for tests based on $AgNO_3$. Based on chronic mortality, a maximum acceptable toxicant concentration (MATC) of between 0.8 and 1.5 nM has been reported for rainbow trout. Importantly, the protective effects of complexation observed for acute toxicity appear to hold true for chronic toxicity as well (Hogstrand and Wood, 1998).

Although there has been limited research investigating uptake of silver from diet, the one controlled study of juvenile rainbow trout demonstrated slow silver uptake from biologically incorporated silver, but no impairment of physiology (Galvez et al., 1996).

Although fish have been used as the model organism for most of the studies on the bioavailability and toxicity of silver, it has become apparent that silver concentrations in natural waters are likely too low to cause toxicity in fish. However, recent studies by Fisher and Hook (1998) suggest that aquatic organisms of lower trophic level may be more sensitive to silver. Fisher and Hook examined silver toxicity in freshwater zooplankton. The freshwater cladoceran, *Simocephalus* sp., was exposed to silver via the dissolved phase and through phytoplankton food. LC_{50} values using dissolved $AgNO_3$ indicated acutely toxic levels to be ≥100 nM, similar to values obtained for freshwater fish. However, sublethal effects were observed when cladoceran were fed with algae which had been exposed to 0.1 nM silver. There was a significant decline in egg production at 1.7 times normal body-burden concentrations (0.172 $\mu g\ g^{-1}$ dry wt). At 2.4 times normal levels (0.24 $\mu g\ g^{-1}$ dry wt), egg production decreased by >70%. Interestingly, silver concentrations as high as 15 times normal levels had no effect on egg production when silver was obtained from the dissolved phase.

The effects of silver have been investigated in a variety of other aquatic organisms. Silver is strongly bioaccumulated by many of these organisms, in particular, bivalve mollusks. The effects on the different classes of organisms along with references to the original works have been tabulated (Eisler, 1996). As with zooplankton, uptake from food sources can be a significant pathway for silver accumulation for many benthic organisms (Fisher and Wang, 1998). However, it appears that silver must be in solution before it is assimilated. The assimilation of silver from ingested sediment particles and phytoplankton cells in mussels occurs through the uptake of silver dissolved in the cytoplasm. The nutritional value of the ingested material strongly influences silver absorption because it affects the residence time of the material in the gut. Thus, organic coatings on sediment particles significantly increase the assimilation of silver (Fisher and Wang, 1998). The redox conditions and acidity of the gut also affect the fate of ingested silver.

Silver is also strongly absorbed from the dissolved phase. This high dissolved uptake rate is probably due to the strong binding of silver with protein ligands (Fisher and Wang, 1998). It is still not known whether these protein ligands are distributed within the biological membrane or the cytoplasm. Studies by Hempe and Cousins (1990 and 1992) suggest that metals such as zinc are first bound to cysteine-rich proteins in membranes and are then sequestered by metallothioneins within the cytoplasm. The physical size and chemical form of silver in the dissolved phase will have a significant impact on bioaccumulation rates. The kinetics of exchange between silver bound to solution species and proteins in the membrane will also be an important factor.

The study of silver's toxic effects and, in particular, the mechanisms underlying toxicity are still under investigation. One important area of ongoing research is to better understand the link between bioaccumulation and toxicity. There have been suggestions that up to 80% of the silver accumulated by bivalves is bound to sulfides (Berthet et al., 1992). Deposition of silver as an insoluble sulfide would greatly decrease its toxicity, and decouple the link between bioaccumulation and toxicity.

2.3 Regulatory Issues

In Canada, silver criteria for the protection of freshwater aquatic life is 0.1 µg total recoverable silver (i.e., nitric acid soluble) per litre (1 nM) (CCREM, 1987). In the U.S., silver criteria for the protection of freshwater aquatic life during acute exposure now range from 1.2 to 13.0 µg total recoverable silver per litre (10 to 120 nM) (depending on hardness) (U.S.Environmental Protection Agency (EPA), 1980). For freshwater aquatic life protection during chronic exposure, the suggested criterion is less than 0.13 µg total recoverable silver per litre (1.2 nM) (U.S.Environmental Protection Agency (EPA), 1980). If all total recoverable silver was in the ionic form, these criteria would overlap the range found lethal to sensitive aquatic species (Eisler, 1996).

Silver criteria in aquatic ecosystems are under revision by regulatory agencies. For example, total recoverable silver is no longer recommended by the U.S. Environmental Protection Agency (U.S.Environmental Protection Agency (EPA), 1995). There have been suggestions that dissolved silver more closely approximates the bioavailable fraction of silver than does total recoverable silver (U.S.Environmental Protection Agency (EPA), 1995). Others propose that the route of silver uptake and body-burden concentrations (rather than dissolved silver concentrations) should be considered in assessing toxicity (Fisher and Wang, 1998). Currently, U.S. EPA is considering criteria based on the Biotic Ligand Model (BLM), where toxicity is assessed by estimating the potential for silver complexation with binding sites on the gills of fish.

More information is needed on the chemical speciation of silver in order to better understand the risks associated with silver pollution (U.S.Environmental Protection Agency (EPA), 1987; Berthet et al., 1992). The merging of geochemical knowledge on silver with the toxicological data will allow regulators to better predict the impact of silver on the aquatic environment. Moreover, it will help to provide a sound basis for developing better regulations for silver.

2.4 Introduction to Chemical Speciation

Metals entering the aquatic environment will partition between various compartments: a portion will be associated with dissolved organic and inorganic ligands in solution whereas another fraction will become associated with colloidal or particulate matter following adsorption, precipitation or co-precipitation reactions, or uptake by living organisms. Thus metals entering a natural water system can occur in a considerable number of physico-chemical forms or species (Figure 1).

Speciation can be determined by direct or indirect methods. Direct methods involve identifying and quantifying the metal species, usually by electrochemical techniques. However, direct methods of determining metal species are unavailable, as is the case for silver, where concentrations in natural waters are prohibitively low. In these cases, a chemical modelling approach may be used to indirectly determine the metal species.

Chemical modelling is based on the concept of thermodynamic equilibrium. The distribution of a metal between the various physico-chemical forms is calculated from known equilibrium constants for the metal (M) with all ligands (L_i) present in the system. In most cases equilibrium constants are reported as follows:

$$m\,M + n\,L_i \rightleftharpoons M_mL_{i,n} \qquad \beta'_{nm} = [M_mL_{i,n}]/[M]^m[L_i]^n \qquad (\text{at } \mu, T)$$

where β'_{nm} is the apparent equilibrium constant valid for a specific ionic strength (μ) and temperature (T) and where [] refers to concentration.

The appeal of this technique is in its predictive ability (i.e., the consequences of changes in metal or ligand concentrations can be foreseen). However, for chemical modelling to be successful, a number of conditions must be met. First, the system must be in equilibrium, or at least a pseudo-equilibrium must be established. Second, the important ligands in the system must be correctly identified. Finally, accurate equilibrium constants must be available for these ligands.

3. Introduction to the Chemistry of Silver

Silver occurs in several oxidation states: Ag(I), Ag^o and less commonly, Ag(II) and Ag(III). The reduction potential for Ag(I) to Ag^o is -1.8 V; however, Ag(I) readily undergoes photo-reduction. Based on the Ag(I)/Ag^o reduction potential, Ag(I) should be the predominant oxidation state in aerobic environments. However, there is evidence suggesting that Ag(I) may also predominate over Ag^o under reducing conditions, as a result of complexation by strong ligands (Miller and Bruland, 1995).

Silver forms a variety of coordination compounds. Although three- and four-coordinate complexes are possible, the preferred coordination is one- and two-fold coordination. This preference has been explained through the hybridization of d, p and s orbitals which allows for the formation of a pair of strong linear covalent bonds (Cotton and Wilkinson, 1972; Bell and Kramer, 1999).

Silver is a group B metal and therefore forms strong complexes with soft bases (Cl, Br, I and S). Table 2 is a summary of silver formation constant values for a variety of ligand classes. From this summary table, it is evident that silver forms extremely strong complexes with S(-II)

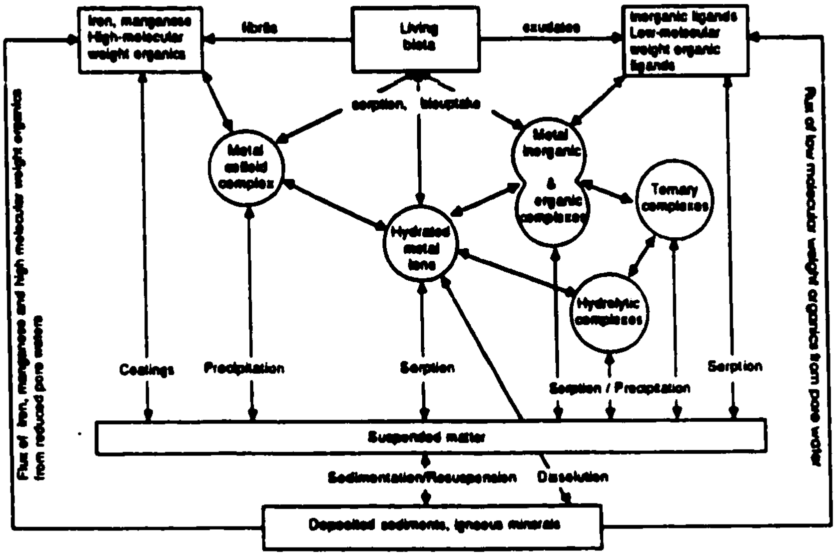

Figure 1. The physico-chemical forms (species) of metal ions in nature. (From Öhman and Sjöberg, 1988)

containing ligands compared with other ligands.

Silver forms polynuclear complexes with both inorganic and organic sulfides above total silver concentrations of ~ 10^{-5} M (Bell and Kramer, 1999; Paper II). Using this criterion, however, silver concentrations found in natural aquatic environments are too low for the formation of polynuclear species.

4. Chemical Speciation of Silver

From the available thermodynamic data on silver and estimates of ligand concentrations, several authors have made predictions of silver speciation in marine waters (Kramer, 1995; Cowan et al., 1985) and in freshwaters (Kramer, 1995). Cowan et al. predicted silver chloride complexes to predominate in marine waters, but did not include sulfides in their calculations. Kramer demonstrated that sulfides would predominate in marine waters at > 10^{-9} M total sulfide, S_T, and in freshwaters at > 10^{-13} M S_T.

To further demonstrate the potential importance of S(-II) ligands to silver speciation in freshwater, I used the constants from Table 2 along with estimates of ligand concentrations to predict speciation. As shown in Table 2, silver forms an extremely insoluble sulfide phase, $Ag_2S(s)$. Predictions of aqueous silver species concentrations can be made based on $Ag_2S(s)$ solubility. Figure 2 shows silver species concentrations calculated using the computer code Mineql+ (Westall et al., 1976) for the Ag-S(-II)-H^+-$Ag_2S(s)$ system at pH 8 ($\mu = 0.1$ M, 25°C) for a range of total sulfide concentrations, S_T. Other ligands were included at concentrations typical for freshwaters. Chloride was included at a concentration of 1 mM. Cysteine was chosen as a model thiol compound, assuming a concentration of 1 μM. This should be an upper

Table 2 Silver Formation Constants for Different Ligand Classes

Hydroxide ion, OH^-	$[ML]/[M][L]$	log K = 2.0	25°C; $\mu = 0.0$[a]
Phenol	$[ML]/[M][L]$	log K = 0.18	25°C; $\mu = 0.1$[a]
Acetic acid	$[ML]/[M][L]$	log K = 0.73	25°C; $\mu = 0.0$[a]
Benzoic acid	$[ML]/[M][L]$	log K = 0.60	25°C; $\mu = 0.1$[a]
		log K = 0.91	25°C; $\mu = 0.0$[a]
Methylamine	$[ML]/[M][L]$	log K = 3.06	25°C; $\mu = 0.1$[a]
Glycine	$[ML]/[M][L]$	log K = 3.20	25°C; $\mu = 0.1$[a]
		log K = 3.51	25°C; $\mu = 0.0$[a]
	$[ML_2]/[M][L]^2$	log K = 6.63	25°C; $\mu = 0.1$[a]
Glutamic acid	$[ML]/[M][L]$	log K = 3.79	25°C; $\mu = 0.1$[a]
Ethylenedinitrilotetraacetic acid (EDTA)	$[ML]/[M][L]$	log K = 7.22	25°C; $\mu = 0.1$[a]
Fulvic acid	$[ML]/[M][L]$	log K = 5.67 (pH 8)	25°C; $\mu = 0.1$[b]
Hydrogen cyanide	$[ML_2]/[M][L]^2$	log K = 20.45	20°C; $\mu = 0.1$[a]
Chloride	$[ML]/[M][L]$	log K = 3.08	25°C; $\mu = 0.1$[a]
		log K = 3.31	25°C; $\mu = 0.0$[a]
	$[ML_2]/[M][L]^2$	log K = 5.08	25°C; $\mu = 0.1$[a]
		log K = 5.25	25°C; $\mu = 0.0$[a]
	$[ML_3]/[M][L]^3$	log K = 5.20	25°C; $\mu = 0.0$[a]
	$[ML_4]/[M][L]^4$	log K = 6.04	20°C; $\mu = 4.0$[a]
	$[M][L]/[ML(s)]$	log K = -9.42	25°C; $\mu = 0.1$[a]
		log K = -9.75	25°C; $\mu = 0.0$[a]
Iodide	$[ML]/[M][L]$	log K = 6.6	18°C; $\mu = 0.0$[a]
2-Aminoethanesulfonic acid (Taurine)	$[ML]/[M][L]$	log K = 2.97	25°C; $\mu = 0.5$[a]
2-(Aminomethyl)thiophene	$[ML]/[M][L]$	log K = 2.87	30°C; $\mu = 1.0$[a]
Thiocarbamide	$[ML]/[M][L]$	log K = 7.11	25°C; $\mu = 0.5$[a]
Dimethylsulfide	$[ML]/[M][L]$	log K = 3.70	35°C; $\mu = 0.1$[a]
S-methylcysteine	$[ML]/[M][L]$	log K = 5.23	25°C; $\mu = 0.1$[a]
Methionine	$[MHL]/[M][HL]$	log K = 3.15	25°C; $\mu = 0.1$[a]
Hydrogen sulfite	$[ML]/[M][L]$	log K = 5.60	25°C; $\mu = 0.0$[a]
Thiosulfate	$[ML]/[M][L]$	log K = 8.82	20°C; $\mu = 0.0$[a]
Hydrogen sulfide	$[MHL]/[M][HL]$	log K = 14.0	25°C; $\mu = 0.0$[c] *

	$[M(HL)_2]/[M][HL]^2$	log K = 17.67	25°C; μ = 0.0[c] *
	$[M]^2[HL]/[H][M_2L(s)]$	log K = -35.94	25°C; μ = 0.0[d] *
Polysulfides			
$Ag(S_4)_2^{3-}$	$[ML_2]/[M][L]^2$	log K = 20.87	25°C; μ = 0.0[e] *
$Ag(HS)S_4^{2-}$		log K = 20.2	25°C; μ = 0.0[e] *
2-Mercaptoethanol	$[ML]/[M][L]$	log K = 13.2	25°C; μ = 0.1[a,g] *
Monothiopentaerythritol	$[ML]/[M][L]$	log K = 13	20°C; μ = 0.1[a,g] *
Penicillamine	$[MHL]/[M][HL]$	log K = 12.4	25°C; μ = 0.1[a,g] *
3-Mercaptopropionic acid	$[ML]/[M][L]$	log K = 12.0	20°C; μ = 0.01[f,g] *
	$[ML_2]/[M][L]^2$	log K = 14.0	
L-cysteine	$[MHL]/[M][HL]$	log K = 11.9	20°C; μ = 0.01[f,g] *
	$[M(HL)_2]/[M][HL]^2$	log K = 15.2	
Glutathione	$[MHL]/[M][HL]$	log K = 12.3	20°C; μ = 0.01[f,g] *
	$[M(HL)_2]/[M][HL]^2$	log K = 14.3	

a. Smith and Martell (1997)
b. Varshal et al. (1995)
c. Schwarzenbach and Widmer (1966)
d. Dyrssen and Kremling (1990)
e. Cloke (1963)
f. Adams and Kramer (Paper II)

g. thiols
* strong complexes with S(-II) ligands

boundary on thiol concentrations in freshwater. Glycine was chosen as a model for compounds containing amine and carboxylate groups. Mintrop and Duinker (1994) showed 10 μM is an upper limit on amino acid concentrations, measured at the water-sediment interface. An equilibrium constant for fulvic acid isolated from the Moscow River (Table 2) was used as a model for natural organic matter at a concentration of 5 mg/L (or 1 μeq/L). Polysulfides were assumed to be in equilibrium with rhombic S, and pε was established from the HS^-/S^0 redox couple. Formation constants for the silver complexes are shown in Table 2. Other constants were present in the Mineql+ database, except for the polysulfide constants which were taken from Wang et al. (1998). Corrections for ionic strength were made using the Davies equation (Stumm and Morgan, 1981).

These calculations demonstrate the potential importance of S(-II) in silver binding. The calculations predict that $AgHS^\circ$ should be the predominant silver species at $S_T > 10^{-8.5}$ M and that Ag-thiolates should be predominant at $S_T < 10^{-8.5}$ M. Although sulfur geochemistry has been studied in greater detail for marine systems (Luther et al., 1985; Boulegue et al., 1979; Boulegue et al., 1982), several studies are available for S(-II) in freshwaters. Wang et al. (1998) investigated sulfur species in lake sediment porewaters. Wang et al. (1998) showed that polysulfide concentrations were about half an order of magnitude greater in concentration than concentrations calculated assuming equilibrium with rhombic S. However, calculations based on sulfur species measured by Wang et al. (1998) also predict that $AgHS^\circ$ should be predominant and that polysulfides are probably not important in silver speciation. Silver speciation appears to differ from copper speciation in this respect, since Tessier (personal communication) has predicted that polysulfides are important in copper speciation, particularly in anoxic sediments. Wang et al.

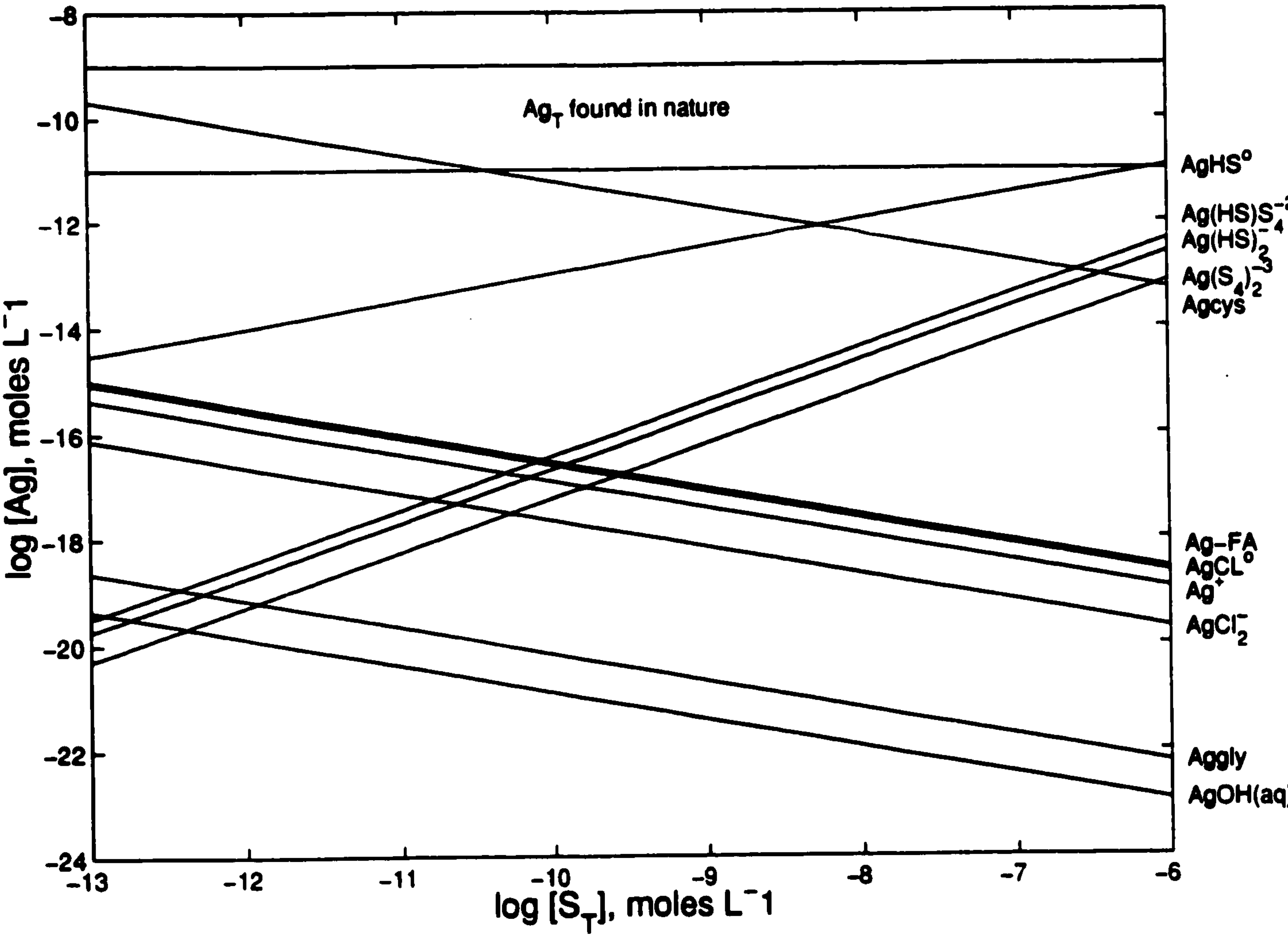

Figure 2. Silver sulfide solubility as a function of total aqueous sulfide concentration, S_T (= $[H_2S]$ + $[HS^-]$ + $\sum[S_n^{2-}]$ + $\sum[HS_n^-]$ + $\sum[H_2S_n]$), at a pH of 8.0 (25°C; μ=0.1). AgHS° and Ag-cys define the total soluble silver concentration, Ag_T. The range of Ag_T found in nature is also shown. Ag-cys, Ag-gly and Ag-FA are abbreviations for complexes with cysteine, glycine and fulvic acid. Cl_T = 1 mM, cys_T = 1 μM, gly_T = 10 μM and FA_T = 5 mg/L (1 μeq/L).

(1998) also observed no thiols in the porewaters they investigated. In the absence of thiols, AgHS° should predominate silver speciation at low S_T concentrations as well.

Adams and Kramer (Paper III) and Rozan and Benoit (1998) recently measured sulfides at nanomolar concentrations in oxygenated river waters. This finding suggests that sulfides bind silver(I) in oxic freshwaters also.

A comparison of predicted total silver, Ag_T, (the sum of the individual species concentrations) with a typical range of measured Ag_T values is also shown in Figure 2. In general, predictions underestimate measured Ag_T concentrations by several orders of magnitude. The discrepancy between the predicted and measured Ag_T may be explained by one, or a combination, of the following considerations: (a) important ligands may have been omitted in the above calculations, (b) concentration(s) of the ligand(s) in the above calculations were underestimated, (c) the assumption of $Ag_2S(s)$ equilibrium is somehow incorrect, or (d) the determination of Ag_T includes more than aqueous phase species (i.e., colloidally bound silver). These issues are explored in the following sections. In the following sections, I demonstrate that measured Ag_T values can be explained by silver binding to particulate and colloidal metal sulfides which was not considered in the above calculations.

4.1 Silver Speciation in Wastewater and Surface Waters

The previous section concluded that S(-II) species form strong complexes with silver(I) and showed through thermodynamic calculations that silver-S(-II) should be the predominant species in natural waters. This section examines silver-S(-II) complexation in the aqueous phase.

Silver is elevated in municipal wastewater treatment plant (WTP) effluents and therefore receiving waters represent one aquatic environment which may be impacted by silver pollution. The Dundas WTP, Dundas, Ontario, was chosen as one study site to examine silver speciation (Paper III). Mining sites also represent environments which may be impacted by silver pollution. An old silver mining camp in Cobalt, Ontario, was another study site where silver speciation was investigated (Paper V).

Dundas WTP

Based on the thermodynamic calculations shown in the previous section, silver should be complexed to S(-II) over other ligands present in natural waters. Table 2 summarized many of the possible S(-II) species which might be important complexants for silver. The speciation calculations shown in Figure 2 suggest that $AgHS^{\circ}$ and Ag-thiolate complexes are likely to be the predominant silver species. To evaluate this, inorganic sulfide and thiol concentrations were measured in the treated effluent and in the receiving waters downstream of the plant. Details of the sampling methodology for the determination of silver, sulfide and thiol concentrations at the Dundas WTP are given in Paper III.

Silver and sulfide concentrations measured in the plant and downstream from the plant are summarized in Tables 4, 5, 6 and 7 of Paper III. Measurable inorganic sulfide concentrations (at sub-micromolar concentrations) were present in both the wastewater effluent samples, as well as the fully oxic downstream samples. Inorganic sulfide concentrations were always in excess of silver, present at ultra-trace concentrations (<1 nM). This is shown by a plot of total silver versus total inorganic sulfide for all the samples collected during the study (Figure 3a).

Surprisingly, thiol compounds were not measurable in any of the wastewater or surface water samples (with a detection limit for most thiol compounds of 1-2 nM). Thiol compounds were also not measurable after reacting unfiltered samples with $NaBH_4$ to liberate thiols bound through disulfide linkages and with ethylenediaminetetraacetic acid (EDTA) to liberate thiols bound to metals (see Paper III). This finding suggests that simple thiols (e.g., methanethiol, ethanethiol, 3-mercaptopropionic acid, cysteine (amino acid), and glutathione (tripeptide)) are not present at significant concentrations relative to inorganic sulfide concentrations. However, Caron and Kramer (1989) did measure a number of volatile sulfur compounds in the Desjardin Canal at low ng/L levels (COS, MeSH, CS_2, DMS and DMDS). The methanethiol (MeSH) concentration was found to be 36 (±12) ng/L. It is possible that the procedures used in my analysis of thiol compounds resulted in significant losses of this more volatile thiol. Several chromatograms did suggest the presence of methanethiol in the Desjardin Canal samples, but concentrations were below the 3σ detection limit.

It appears from this study that organic sulfide (i.e., thiols) are present in low concentrations compared with inorganic sulfide and therefore silver speciation should be dominated by inorganic sulfide complexes.

Silver Mining Camp in Cobalt, Ontario

Silver and inorganic sulfide concentrations were measured in waters flowing through old mining tailings and are summarized in Table 5 of Paper V. As with the samples taken at the Dundas WTP, inorganic sulfide was present in these waters at low to high nanomolar concentrations. Here as well, inorganic sulfide concentrations were in excess of silver, except in samples collected at Cart Lake where water flows over inorganic tailings (Figure 3b). This is

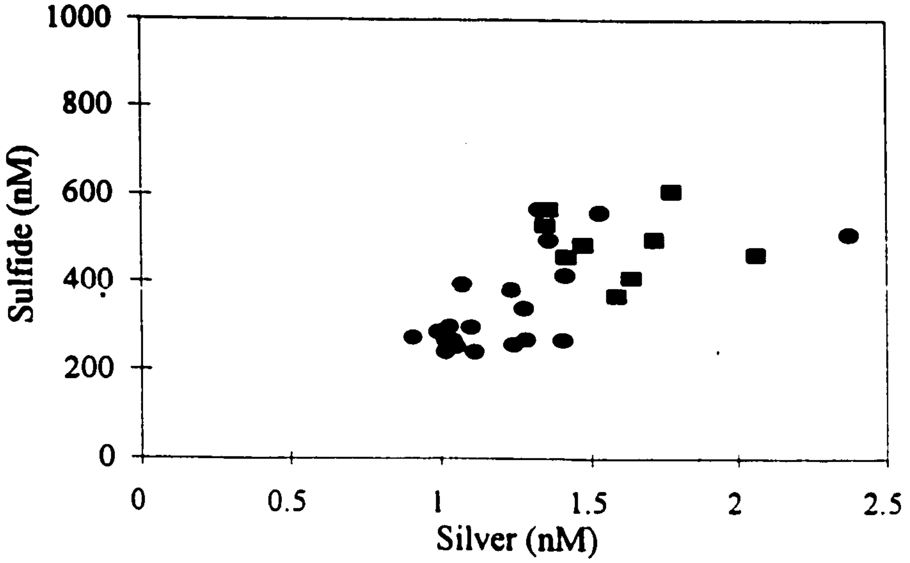

Figure 3a. Total sulfide versus total silver concentrations for the Dundas WTP (squares) and downstream in the Desjardin Canal (circles).

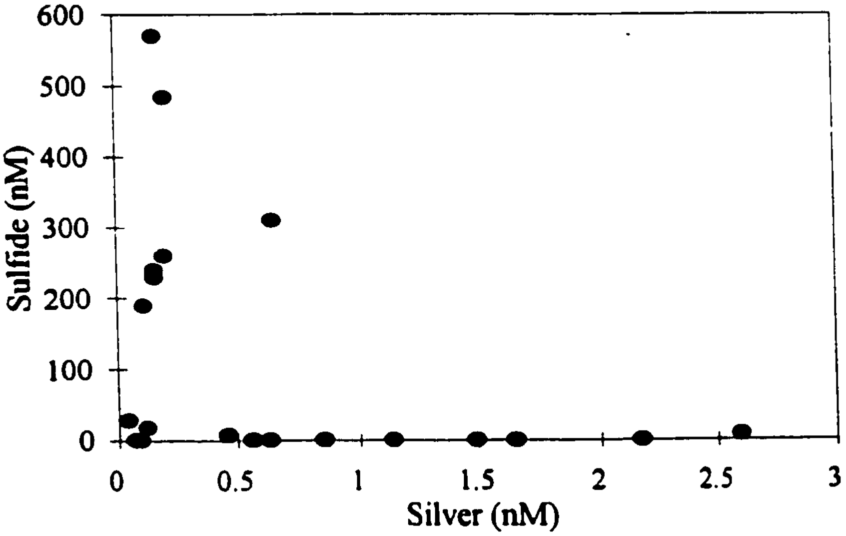

Figure 3b. 0.45 µm filtered sulfide versus 0.2 µm filtered silver concentrations for samples collected at an old silver mining camp in Cobalt, Ontario.

indicated by a plot of 0.2 μm filtered silver versus 0.45 μm filtered inorganic sulfide concentrations. Although thiols were not determined in these waters, it is likely that inorganic sulfide dominates silver speciation in these waters as well.

Aqueous Ag_T predicted based on $Ag_2S(s)$ solubility, assuming inorganic sulfide measured in these waters is HS^- (HS^- predominates at pH ~8), is approximately 100 times less than measured Ag_T at both the WTP and mining site (Figure 4). Clearly either important ligands have been neglected, the assumption that the aqueous silver species are in equilibrium with $Ag_2S(s)$ is incorrect, or measured Ag_T is including other species besides aqueous phase species.

Samples were fractionated into a particulate fraction (> 0.45 μm), a colloidal fraction (10 kDa - 0.45 μm) and a dissolved fraction (< 10 kDa) in an attempt to compare aqueous silver with total silver. This size fractionation at the Dundas WTP showed that for most samples at least 85% of silver was in the particulate or colloidal phase and at the mining site in Cobalt 90-99% was in the particulate or colloidal phase. This observation agrees with the findings of Shafer et al. (1998) and Pham and Garnier (1998). Shafer et al. found 98% of silver was in the particulate or colloidal phase in outfalls from several WTPs in Wisconsin, USA, and in river water samples investigated by Pham and Garnier at least 75% of silver was in the particulate or colloidal phase. Size fractionation of the inorganic sulfide samples at the Dundas WTP showed that 80-90% of the inorganic sulfide was in the particulate or colloidal fraction. These size fractionation studies demonstrate the importance of colloids to silver speciation and explain why Ag_T can not be predicted from $Ag_2S(s)$ solubility.

Sulfide concentrations determined by the Cline colorimetric method include the sulfide species: S^{2-}, HS^- and $H_2S(aq)$ and S(-II) of polysulfides (i.e., $S_n\underline{S}^{2-}$) (Wu and Kramer, 1998).

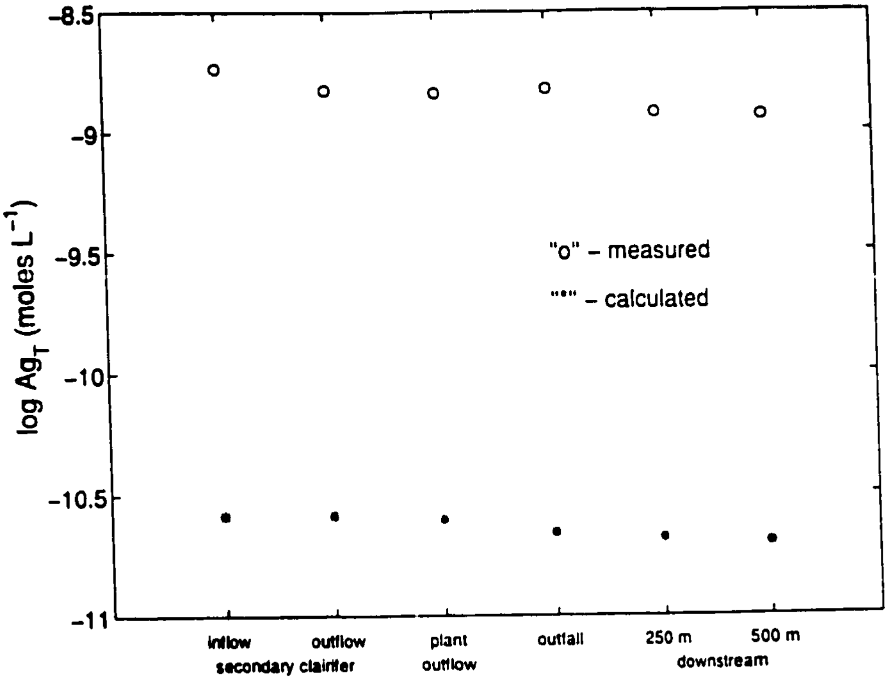

Figure 4a. Measured Ag_T versus calculated Ag_T (from $Ag_2S(s)$ solubility) for the Dundas WTP.

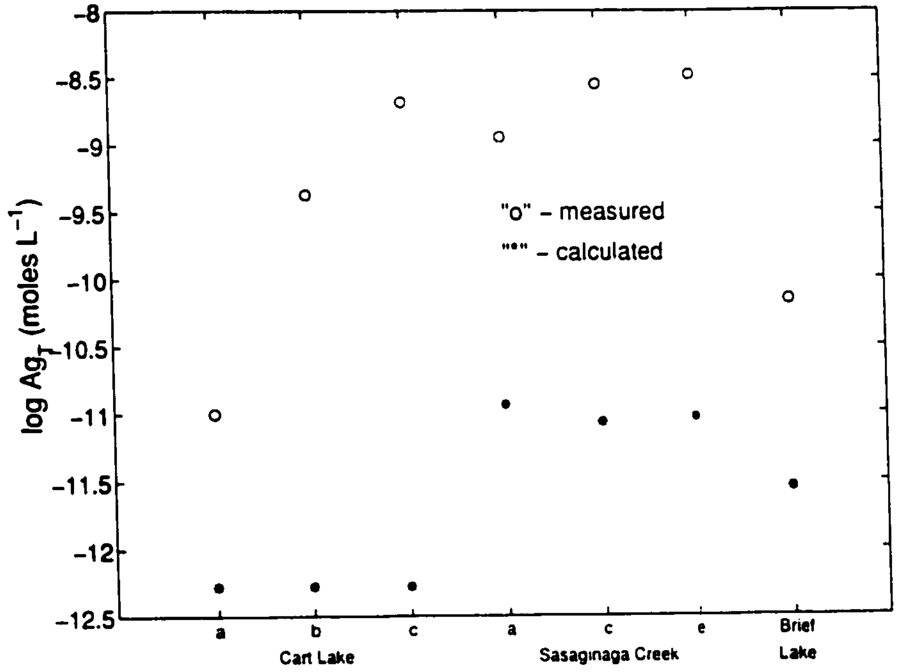

Figure 4b. Measured Ag_T versus calculated Ag_T (from $Ag_2S(s)$ solubility) for an old silver mining site in Cobalt, Ontario (letters refer to sampling locations in Paper V).

However, the addition of the acidic reagent solution to the sample decreases the pH and causes a variety of colloidal metal sulfides to decompose, allowing liberated sulfide to react with the colorimetric reagents. Amorphous FeS reacts quantitatively under these conditions ($FeS \rightarrow Fe^{2+} + \sum S$), as does ZnS and CuS, although at different rates (CuS<FeS~ZnS) (Wu and Kramer, 1998).

Combining the fact that a significant fraction of the inorganic sulfide is found in the particulate and colloidal phase with the fact that sulfide in the form of metal sulfides can be measured by the Cline method suggests that a significant amount of inorganic sulfide present in the WTP effluent and oxic waters downstream may be in the form of metal sulfides. Binding of silver to these colloidal metal sulfides would explain the discrepancy between measured and predicted Ag_T. As well, simple inorganic sulfide (present as HS^- or $H_2S(aq)$) would not be stable under the oxic conditions of the WTP and surface waters examined. However, certain metal sulfides are stable enough under oxic conditions to survive for periods of hours or days (Luther and Tsamakis, 1989; Simpson et al., 1998).

Complexation Experiments Using Competitive Ligand Equilibration/Solvent Extraction

The competitive ligand equilibration/solvent extraction (CLE/SE) technique developed by Miller and Bruland (1994 and 1995) was used to examine silver complexation in samples collected from the Dundas WTP and Desjardin Canal. Complexant concentrations and conditional complexation constants were determined in the different size fractions: unfiltered, <0.45 µm filtered and <10 kDa filtered. From this information it was possible to show a link between the inorganic sulfide measured in the WTP and Desjardin Canal and silver complexation in these waters.

A complete description of the methodology for the CLE/SE technique is given in Paper IV. Diethyldithiocarbamate was added to the samples in order to establish a competition for silver with the natural ligands. Competitive ligand equilibrations were conducted on sample aliquots spiked with increasing amounts of silver to generate titration curves which were then used to estimate the silver complexation constants and natural complexant concentrations. By choosing additions of silver which were near ambient concentrations, it was possible to infer the speciation for the original silver in the samples. This is important, since titrations at higher silver concentrations will saturate the strong ligands which complex the trace levels of silver present in natural waters.

In order to verify that the CLE/SE technique gave accurate estimates of the complexation of silver with strong ligands (i.e., sulfide ligands) at low concentrations, titrations were performed on 3-mercaptopropionic acid (3-MPA). These titrations gave an accurate estimate of the 3-MPA concentration and an estimate of log K'_{AgL} which was similar to the value obtained potentiometrically (Paper II).

Samples were collected immediately upstream of the secondary clairifier outflow at the Dundas WTP and 250 m downstream of the plant in the Desjardin Canal. Samples were processed by sequential cross-membrane filtration to produce a 0.45 µm and 10 kDa filtrate. However, due to problems with the reliability of the unfiltered and 0.45 µm filtered samples from the Dundas WTP, only the 10 kDa titration data were examined. Log K'_{AgL} and $[L_T']$ values from these samples along with samples collected on subsequent sampling dates are given in Tables 1 and 2 (Paper IV, pp. 20-21). A one-ligand model adequately described the complexation in these samples. Figure 5 shows a plot of log K'_{AgL} versus ligand concentration, $[L_T']$, for the Dundas

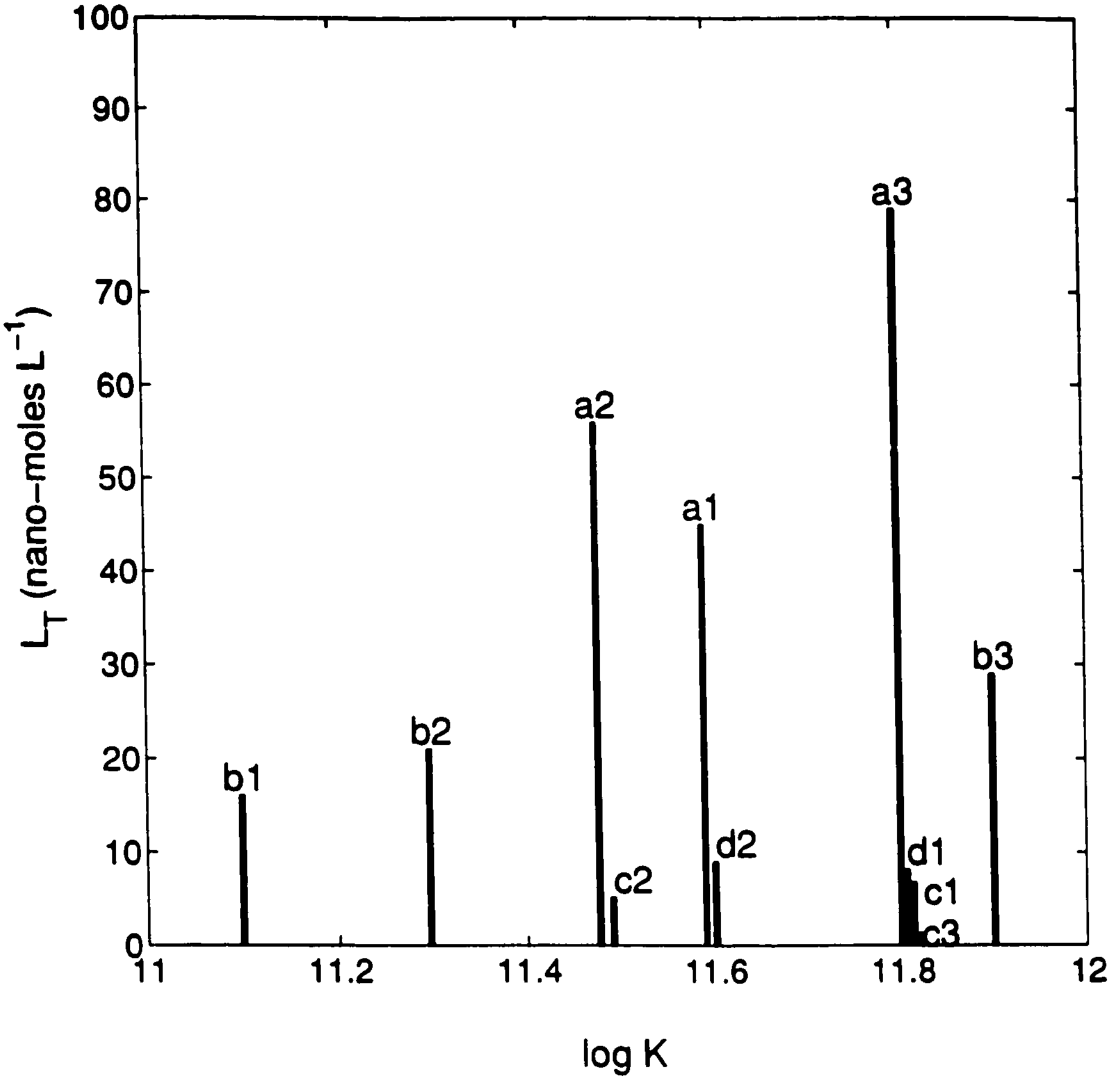

Figure 5. Silver complexation constants (log K) and natural ligand concentrations, $[L_T]$, for (a) unfiltered, (b) <0.45 µm filtered, (c) <10 kDa filtered samples from 250 m downstream of the plant (1- April 1998, 2- May 1998 and 3- June 1998) (e.g., a1, b1, ...) and (d) <10 kDa filtered sample from the outflow of the secondary clairifier at the Dundas WTP (1- April 1998 and 2- June 1998).

WTP and Desjardin Canal samples. Log K'_{AgL} values were different for the unfiltered, <0.45 µm filtered and <10 kDa filtered fractions from the Desjardin Canal samples. Filtration removed a portion of the ligands, which resulted in a new average K'_{AgL} value for the remaining ligands. Ligand concentrations, $[L_T']$, for the Desjardin Canal decreased 62-64% between the unfiltered and <0.45 µm filtered samples (for the three sampling dates), and decreased 58%, 76% and 96% between the <0.45 µm filtered and <10 kDa filtered samples. Ligand concentrations in the <10 kDa samples from the Desjardin Canal were 16% (April, 1998) and 86% (June, 1998) lower relative to the WTP samples.

This study demonstrates that there is excess complexation capacity for silver by strong ligands (log K'_{AgL} ~ 11-12). The similarity between silver complexation constants determined for the Dundas WTP and Desjardin Canal and complexation constants of known organic and inorganic sulfide species (log K = 11.9-13.0) (Table 2) supports the idea that silver in WTP effluents and receiving waters is complexed to sulfide.

Differences observed in $[L_T']$ between the unfiltered and <0.45 µm filtered and between the <0.45 µm filtered and <10 kDa filtered samples from the Desjardin Canal were similar to differences in inorganic sulfide concentrations between filter fractions (45% and 80%, respectively). As well, $[L_T']$ decreased in the <10 kDa samples between the WTP and Desjardin Canal by a similar amount compared to inorganic sulfide (30%). The similarities between inorganic sulfide concentrations and $[L_T']$ suggest that inorganic sulfide ≡ L_T and that silver is therefore complexed to inorganic sulfide in these waters. However, $[L_T']$'s determined by the CLE/SE technique were in general 70-90% lower than inorganic sulfide concentrations measured by the Cline method (Cline, 1969). Earlier I suggested that sulfide likely exists predominantly as colloidal metal sulfides. While sulfide existing as colloidal metal sulfides can be measured quantitatively by the Cline method, sulfide in the interior of metal sulfide colloids may be slow to react with silver during CLE/SE titrations. Substitution of silver into the interior of the metal sulfide colloids would likely take much longer than the three hour equilibration time of the experiments. This kinetic effect is likely responsible for the discrepancy between inorganic sulfide concentrations measured by the Cline method and [L_T'] determined by the CLE/SE technique.

The results presented in the previous section suggested that organic sulfides (i.e., thiols) were not important to silver speciation in the Dundas WTP and Desjardin Canal waters. To further investigate the relative contribution of organic versus inorganic sulfides to silver complexation, the CLE/SE technique was applied to UV-irradiated samples of the <0.45 µm filter fraction from the Desjardin Canal. The contribution to silver complexation by organic sulfides should be reduced in these UV-irradiated samples, since UV-irradiation should destroy all but the most recalcitrant organic material (Donat and Bruland, 1988). Only a very small decrease in the ligand concentration for the UV-irradiated samples (2-4%) was found compared to non-irradiated samples. This decrease was insignificant when the uncertainties in the estimated ligand concentrations were taken into consideration. As well, log K'_{AgL} values remained the same for UV-irradiated samples compared to non-irradiated samples. It can be concluded from these findings that (a) organic ligands (i.e., thiols) are not important to silver complexation in these waters, and since this experiment was conducted on the <0.45 µm filter fraction, (b) that concentrations of sulfhydryl sites in higher-molecular weight substances are insignificant as well (<1 nM). From the chromatographic analyses (Paper III), it was only possible to conclude that low-molecular weight thiols (<1000 MW) did not appear to be important in these waters.

4.2 Silver Speciation in Sediments and Porewaters

Since many of the biota sensitive to silver are benthic feeders, it is critical to understand the physico-chemical nature of silver in sediments and how silver partitions between sediments and porewaters. As well, it is important to know how silver speciates in the porewaters and overlying waters. These questions were explored in a laboratory study (Paper I) and in a field study on sediments from Hamilton Harbour (Paper III).

Silver Speciation in Sediments

It is important to have knowledge of the form of trace metals in the solid phase in order to understand their geochemistry (mobility, diagenesis and bioavailability) in sediments. Unfortunately it is difficult to obtain information on the chemical form of silver in sediments by direct methods (e.g., using scanning electron microscopy or x-ray powder diffraction techniques) since silver is present at such low levels in sediments (0.1 µg/g). However, we can infer the form of silver in sediments by examining thermodynamic equilibria.

The solubility product of acanthite, crystalline $Ag_2S(s)$, is very low.

$$Ag_2S(s) + H^+ \rightleftharpoons 2Ag^+ + HS^- \quad K(Ag_2S) = [Ag^+]^2[HS^-]/[H^+] = 10^{-35.94}$$

In fact, the order of metal sulfide solubilities for transition and group B metals is $Hg<Cu^+<Ag<Cu^{2+}<Pb<Zn<Ni<Fe$. Therefore, silver should replace the more abundant transition metals in metal sulfide phases, e.g.,

$$FeS(s) + 2Ag^+ \rightarrow Ag_2S(s) + Fe^{2+}$$

Philips and Kraus (1965) demonstrated that precipitated metal sulfides of higher solubility (Fe(II), Zn(II), Pb(II), Cd(II) and As(III)) exchange rapidly with Ag(I), Cu(II) and Hg(II). In sediments containing acid-volatile sulfides (AVS) (amorphous FeS, mackinawite and greigite) and other less stable metal sulfide phases, as long as total sulfide concentrations, S_T, are greater than total silver concentrations, Ag_T, we expect sedimentary silver to exist as $Ag_2S(s)$. This, of course, assumes thermodynamic equilibrium has been established. Paper III demonstrated that due to silver existing at such low concentrations in the aquatic environment, there is always an excess of sulfide relative to silver, even in oxic surface waters. In sediments this is likely to be the case as well. Typically silver concentrations in sediments are 1 nmole/g sediment. AVS concentrations in freshwater sediments are usually around 5-20 μmole/g sediment (Huerta-Diaz et al., 1993). Thus, molar ratios of silver to sulfide will in most cases be less than one.

Results from Paper I help to address the question of the chemical form of silver in sediments. AVS is the main source of reduced sulfur in the upper layer of sediments and is important for trace metal sequestering. Because silver sulfide is very insoluble compared to iron sulfides ($\log \{K_{sp}(Ag_2S)/ K_{sp}(FeS)\} \sim -33$), the interaction of silver with FeS is likely important in regulating dissolved silver concentrations in sediments. However, the nature of this interaction is unclear. It is not known whether trace metals associated with sediments are present as distinct metal-sulfide precipitates or co-precipitates or sorbed species on the surface of the more abundant iron sulfides.

In Paper I, the reaction of silver with a well-characterized synthetic amorphous FeS phase was investigated. A time series of filtered samples were taken which demonstrated that silver in the presence of the iron sulfide rapidly equilibrates to ultra-trace levels (50 pM). Dissolved silver concentrations agreed with equilibrium calculations based on acanthite solubility, suggesting the formation of distinct silver-sulfide phase rather than a co-precipitate with the more abundant

FeS(s). In these experiments, the molar ratio of Ag/Fe was between 10^{-5} and 10^{-6}. For an ideal solid solution (co-precipitate), silver activity in the solid should therefore be equal to the mole fraction, and the dissolved silver concentration in equilibrium with the solid should be lower by a similar factor (i.e., 10^{-5} to 10^{-6} times lower). Including activity coefficients for a non-ideal solid solution would not compensate for this reduced activity. Since dissolved concentrations agreed with Ag_2S(s) solubility, it appears that silver behaves thermodynamically independently (e.g., Ag_2S(s) sorbed on FeS(s)). This agrees with the findings of Huerta-Diaz et al. (1993) where it was shown that Cd, Co, Cu, Ni, Pb and Zn did not co-precipitate with FeS(s). As well, dissolved silver concentrations were insensitive to initial silver concentrations (Figure 6) and equilibrated to the value predicted by Ag_2S(s) solubility which indicates a discrete solid phase and not a surface complex.

Most metal sulfide minerals nucleate slowly, and instead metastable amorphous metal-sulfides are usually formed under supersaturated conditions. However, silver appears to form a crystalline Ag_2S solid with no intermediate amorphous phase. In a separate experiment, we precipitated silver sulfide under supersaturated conditions and analyzed the product by x-ray diffraction(XRD). Based on the noise to peak ratio, it was estimated that the precipitate was at least 50% acanthite, demonstrating that unlike other metal sulfides, XRD crystalline acanthite forms rapidly (< 1 hour). Therefore it is possible that acanthite could be formed directly from silver reacting with an iron sulfide phase. Studies using X-ray photoelectron spectroscopy (XPS) and scanning electron microscopy (SEM) have shown that Ag_2S forms on the surface of pyrite (Scaini et al., 1995; Buckley, Wouterlood, 1989). Although studies using these techniques can not be performed at environmentally relevant silver concentrations, they provide some support for the possible formation of $Ag_2S(s)$ at lower concentrations.

Earlier it was suggested that silver forms surface complexes with particulate and colloidal bound metal sulfides in WTP effluent and surface waters. Here we are suggesting that silver forms a distinct $Ag_2S(s)$ phase in sediments. The difference between the behaviour of silver in sediments and in the aqueous phase is probably a result of higher silver concentrations in sediments compared to the aqueous phase. Higher silver concentrations in the aqueous phase would likely produce a $Ag_2S(s)$ phase rather than surface complexes.

Other studies have examined silver binding to other phases. For example, Wingert-Runge and Andren (1995) examined silver partitioning to several oxic sediments. They showed it is possible to get log K_d values for bentonite and MnO_2 which agree with the results from oxic lake sediment. Log K_d values measured in the field are typically 5 to 6 in sediments, as well as in overlying oxic waters. However, taking typical sedimentary silver concentrations and the concentration of silver predicted by acanthite solubility (at typical porewater sulfide concentrations) gives a partitioning coefficient (log K_d ~ 5.5) which also falls in the range of measured log K_d values. Since silver is present in sediments at such low concentrations, it is possible that enough sulfide (AVS or pyrite) is present even in oxic sediments for silver to exist as $Ag_2S(s)$ instead of binding to other solid phases. However, the kinetics of reaction between the more abundant phases in sediments and the sulfide phases present in lower abundance may be an important consideration.

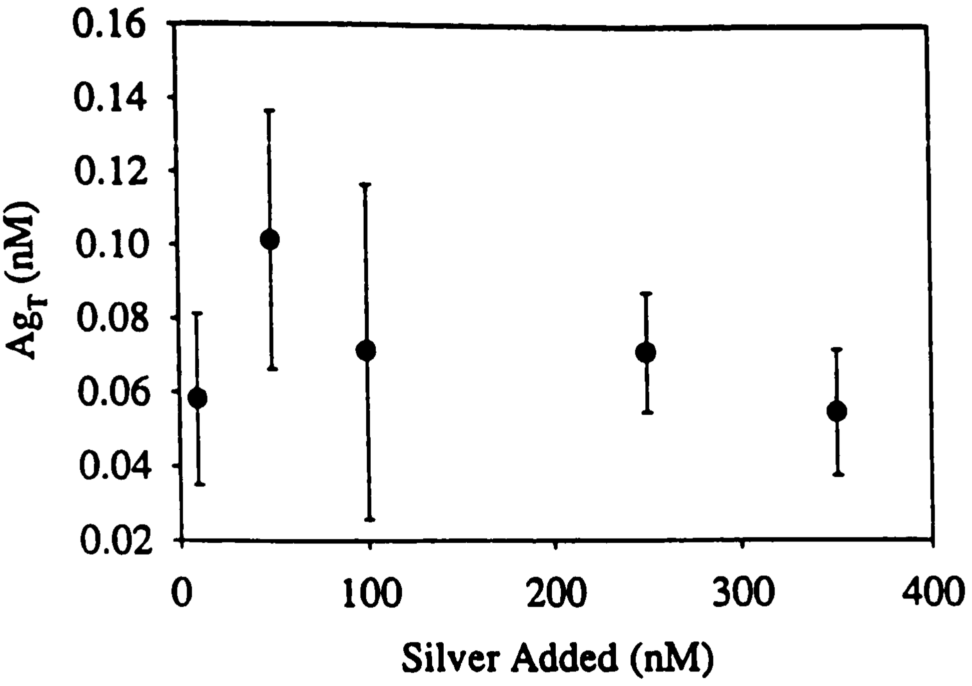

Figure 6. Dissolved silver in the presence of FeS with a 1 hour equilibration time between additions of $AgNO_3$ (10, 50, 100, 250 and 350 nM). Error bars represent ± 1 SD.

Silver Speciation in Porewaters

In the previous section it was suggested that it is reasonable to presume that silver exists in most sediments, even those with only trace amounts of AVS, as a distinct $Ag_2S(s)$ phase. It has been shown that pure metal-sulfide phases (CuS and CdS) are valid for modelling Cu and Cd solubility (Shea, 1985; Framson and Leckie, 1978). To evaluate the validity of using a pure $Ag_2S(s)$ phase for modelling silver solubility, as well as to examine speciation in porewaters and overlying waters, we measured silver, inorganic sulfide and thiols in porewater and overlying water samples. Porewaters were extracted from anoxic sediment cores collected in Hamilton harbour (details are given in Paper III). Sediment cores were collected from three separate locations (A (sandy), B and C (silty)) and segmented into 10 cm sections.

AVS measurements on the sediments from the three locations (for intervals 0-10 cm and 40-50 cm) ranged from 23.3 to 54.4 and 13.6 to 32.8 µmoles/g dry sediment, respectively. The molar ratio of silver to AVS was 0.00001-0.00005 for these sediments.

Silver concentrations were not detectable (<0.01 nM) in the porewaters from sampling location B and C. Silver was also not detectable in the interfacial water samples collected at these locations. However, at location A, low concentrations of silver in the 10 kDa - 0.45 µm fraction (but not the <10 kDa filter fraction) were measured in both the porewater and interfacial water (Figure 7).

Inorganic sulfide concentrations in the 10 kDa - 0.45 µm filter fraction ranged from 350 to 1000 nM and in the <10 kDa filter fraction ranged from 50 to 150 nM. Thiol compounds, 3-mercaptopropionic acid (3-MPA) and mercaptoethanol, were present in the 10-20 cm and 20-30 cm sections from location A. Mean 3-MPA and mercaptoethanol concentrations for these sections were 9.1 and 15.5 and 7.0 and 10.4 nM, respectively.

Using a <10 kDa sulfide concentration of 100 nM (assuming sulfide is this fraction is in the form of HS^-) and a 3-MPA concentration of 15 nM, predicted silver species based on $Ag_2S(s)$ solubility are $10^{-12.5}$ M $AgHS^\circ$ and $10^{-16.5}$ M Ag-3-MPA. Based on this calculation, Ag_T ($\sim[AgHS^\circ]$) should be below the silver detection limit (10^{-11} M). No silver was detectable at location B and C and in the <10 kDa filter fraction of location A, suggesting that dissolved silver could be controlled by $Ag_2S(s)$ solubility. Dissolved silver in equilibrium with $Ag_2S(s)$ can only be detected at higher HS^- concentrations as observed in the laboratory study presented in Paper I. Unfortunately here we can only speculate that dissolved silver concentrations are similar to those predicted by $Ag_2S(s)$ solubility.

Silver was measurable in the colloidal fraction (10 kDa - 0.45 μm) at location A. Silver in this fraction is likely complexed to the colloidal bound sulfide. Presumably silver was measurable as a result of higher sulfide concentrations in the colloidal fraction at location A. Thus, colloidally bound sulfides appear to be responsible for silver present in these sediment porewaters, much the same as was observed in the WTP effluent and surface waters. Colloidally bound sulfide is probably in the form of metal sulfides as discussed previously.

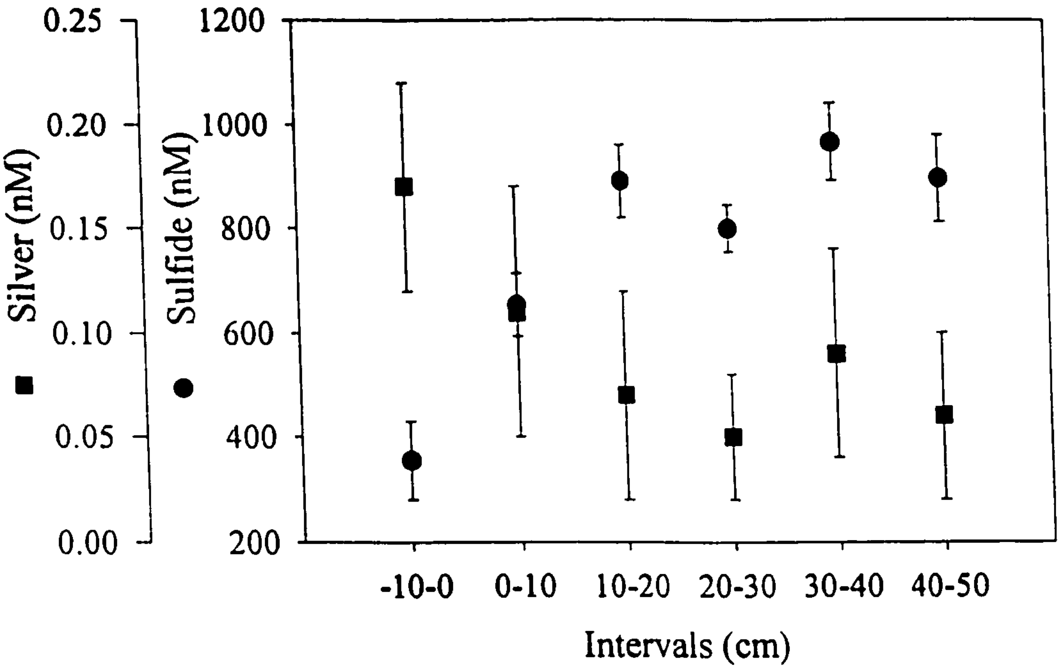

Figure 7. Silver and inorganic sulfide porewater concentrations (10 kDa - 0.45 μm filter fraction) for Hamilton Harbour location A. Error bars represent ± 1 standard error.

4.3 Stability of Silver-Sulfides

The importance of sulfides in natural waters to binding silver(I) leads us to consider the effect of environmental changes, in particular oxidation, on the stability of silver-sulfides. In Paper III, we measured silver and sulfide concentrations in WTP and receiving waters. Silver concentrations in the 10 kDa - 0.45 µm and <10 kDa filter fractions were relatively constant between the WTP and 500 m downstream (which translates to a flow time of approximately 48 hours). Thus, silver appeared to be unaffected by aggregation, sorption and other processes that might affect the distribution of this metal. The constant concentrations of silver in the 10 kDa - 0.45 µm and <10 kDa filter fractions are a result of sulfide concentrations remaining in large excess relative to silver, even 500 m downstream of the WTP.

Sulfide concentrations decreased 50% (>0.45 µm filter fraction) and 40% (10 kDa - 0.45 µm filter fraction) between the WTP and 500 m downstream. This decrease can be attributed to relatively slow oxidation of the sulfide in these fully oxygen-saturated waters. No decrease in sulfide was observed in the <10 kDa filter fraction. Sulfides may be replenished by diffusion of sulfides from the bottom waters, which are higher in sulfides (see Paper III, Table 7 or Figure 4). This would mean that sulfide oxidation rates are probably greater than the 40-50% loss per 48 hours. Although we did not perform any direct studies of sulfide oxidation rates for these waters, we did perform an oxidation study using the CLE/SE method (and have shown that our measurements of silver complexation capacity are related to sulfide concentration). In this study, a <0.45 µm filter fraction collected at the 250 m downstream sampling location showed a 66% decrease in silver complexation capacity after 24 hours and a 88% decrease after 48 hours (Paper IV). These measurements agree reasonably well with the decreases in sulfide concentration observed in Paper III. Slow oxidation of sulfide (days to months) have also been reported by Luther and Tsamakis (1989) and Simpson et al. (1998), and are attributed to complexation of sulfide by metals.

Since photolysis could also affect the stability of silver-sulfides, in a second set of experiments using the CLE/SE technique, we examined the effect of light exposure on silver complexation. We found that complexed silver (silver-sulfides) was unaffected by light, while uncomplexed silver, Ag^+, underwent significant photo-reduction. Thus, complexation by sulfides likely enhances the mobility of silver in the receiving waters downstream of WTP.

5. General Discussion

In the course of this research project it became apparent that colloidally and particulate bound sulfides represent a significant proportion of the sulfide in natural waters (80-100%). Even the majority of sulfide measured in the <10 kDa size fraction appears to be colloidally bound sulfide. To account for the colloidal and particulate bound sulfide, we suggested that this colloidal and particulate sulfide is probably in the form of metal sulfides.

The presence of metal sulfides in marine systems has been the subject of several recent laboratory and field studies. Cutter and Krahforst (1988) and Luther and Tsamakis (1989) reported trace amounts of sulfide in oxic seawater. Luther and Tsamakis also reported that purging of seawater did not remove all the sulfide and postulated that sulfide was stabilized in the oxic seawater through complexation with metals. Dyrssen et al. (1988) and Elliot (1988) have shown that copper (II) complexes are the thermodynamically favoured form of sulfide in oxic seawater. Dyrssen and co-workers showed that 95% of sulfide in oxic seawater should be bound by Cu(II) as dissolved $Cu(S)^{\circ}$ and 5% by Hg(II) as $Hg(S)^{\circ}$. Luther and Ferdelman (1993) have shown that in salt marsh creek waters and Chesapeake Bay waters iron sulfide complexes are important.

Luther and Ferdelman also noted that these iron sulfide complexes are stronger and less labile than iron sulfide complexes generated in the laboratory. They postulated that mixed ligand complexes with organic ligands may help to stabilize the Fe-S bond and make the bond less labile. These authors showed that hydroxyl, carboxyl and nitrogen functional groups could theoretically stabilize the Fe-S bond.

Metal sulfide complexes bound to fulvic and humic matter would account for the silver measured in the colloidal and particulate filter fractions examined in the WTP samples, surface waters and porewaters (Paper III). Complexation of silver by organically bound metal sulfides would also explain the correlation observed by Shafer et al. (1994) between dissolved organic matter (DOC) and silver concentrations in WTP effluents. Metal sulfides may exist as simple moieties, such as $M(SH)^{+}$, as illustrated by the zinc sulfide species in Figure 8a or as more complex metal sulfide clusters as illustrated in Figure 8b. Silver could bind to some or all of the available sulfur(-II) as illustrated in Figure 8c and 8d. Silver might also substitute into the metal sulfide clusters, but this exchange reaction would occur more slowly. Although relatively low molecular weight organic ligands were used in the illustrations shown in Figure 8, these organic ligands could be exchanged for larger macromolecules which would then explain the silver and sulfide measured in the colloidal and particulate filter fractions.

Earlier I discussed the results of the CLE/SE experiments conducted on the Dundas WTP and Desjardin Canal waters which investigated the contribution of organic versus inorganic

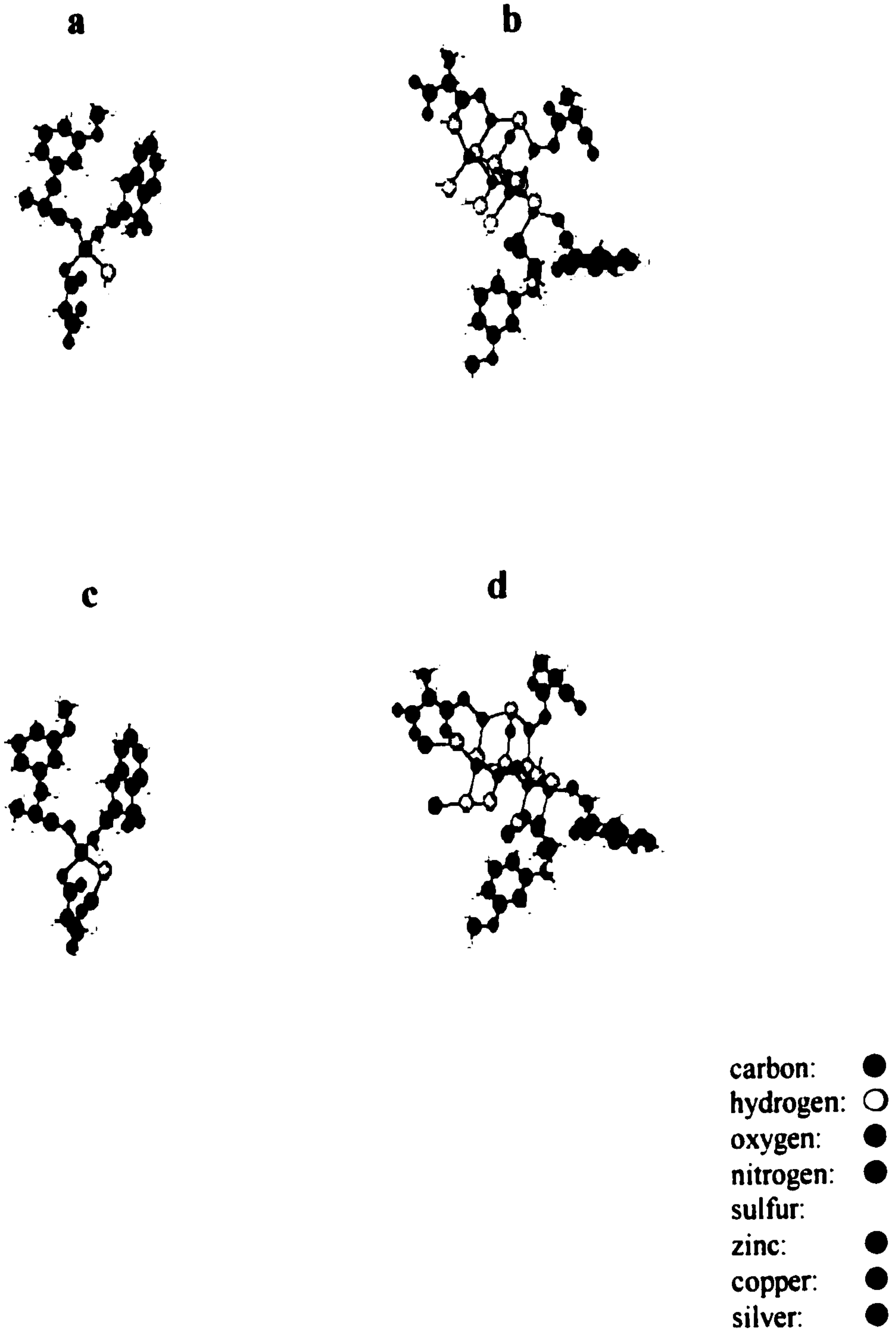

Figure 8. Illustrations of organically complexed (a) zinc sulfide and (b) copper sulfide species and mixed (c) silver-zinc sulfide and (d) silver-copper sulfide species.

ligands to silver complexation. In these experiments we compared UV-irradiated <0.45 μm samples (organics destroyed) to non-irradiated <0.45 μm samples, and showed that there was no difference in silver complexation between these treatments. We concluded from these results that organic ligands (i.e., thiols) were not important to silver complexation. However, these results also tell us that complexation of silver by metal sulfides is unchanged even if the complexing organic ligands are eliminated. This could be further substantiated by the 10 kDa filtration of a set of UV-irradiated samples to determine whether the metal sulfides then appeared as lower molecular weight species (i.e., free from organic complexation).

Rozan and Benoit (1998) also observed sulfides in oxic surface waters at low nanomolar concentrations. They also suggest that the persistence of sulfides in oxic waters is probably aided by the formation of metal sulfide complexes, especially CuS. These authors found metal sulfide complexes were removed to a significant extent by DEAE columns and by filtration using a 3 kDa cut-off filter. Rozan and Benoit also suggest that some metal sulfides are probably present in large organic molecules. Although they also suggest the possibility of amorphous or crystalline metal sulfides making up the colloidal fraction. Rozan and Benoit also showed that sulfides in the river water samples they examined were stable to oxidation. In a laboratory study they showed that sulfides were still measurable after exposure to oxygen for 200 hours. This agrees with our observations that sulfides in natural freshwaters are relatively stable to oxidation compared with what would be expected for HS^- (Papers III and IV).

In the introduction to this thesis I pointed out the concern over possible toxic effects of elevated levels of silver in contaminated waters. I also raised the point that the physcio-chemical form (species) of silver is important to the bioavailability and toxicity of silver to aquatic organisms. The current toxicological literature provides mixed evidence for which silver species are bioavailable and toxic. In the case of fresh and salt water fish, the evidence is quite conclusive that free silver ion, Ag^+, accumulated at the gill is many orders of magnitude more toxic than other species of silver. However, for other aquatic organisms, such as zooplankton and benthic feeders, silver is accumulated through feeding, and it is less clear how silver is accumulated and what silver species are bioavailable.

How does this thesis research contribute to our understanding of silver's bioavailability and toxicity in freshwaters? Firstly, this work demonstrates the importance of sulfides in silver speciation. Sulfides are present in WTPs and effluent receiving waters at levels 100-1000 times greater than silver concentrations. Based on the measured complexation constants and ligand concentrations reported in Paper IV, free silver ion concentrations should be extremely low in these waters (0.01-0.1 picomolar or 0.001-0.01 ng/L), shown by the following calculation:

$$K'_{AgL} = [AgL]/[Ag^+][L^-] = 10^{11\text{-}12}$$

for $[AgL] \approx Ag_T = 0.5$ nM and $[L^-] \approx L_T = 50$ nM,

$$[Ag^+] = (0.5\ \text{nM})/(50\ \text{nM})(10^{11\text{-}12}) = 10^{-13} - 10^{-14}\ \text{M}$$

Total silver concentrations at the Dundas plant outfall (mean of 1.8 nM) were often above the 1.2 nM chronic exposure value suggested by the USEPA (1980) and were also above concentration values shown to adversely affect freshwater zooplankton (fed algae exposed to 0.1 nM $AgNO_3$) and rainbow trout (0.8-1.5 nM $AgNO_3$). However, at the 250 m downstream sampling location, total silver concentrations had dropped to 1.0-1.1 nM as a result of non-conservative processes. Furthermore, the above calculation demonstrates that free silver ion concentrations should be very low in these waters. Thus aquatic organisms sensitive to Ag^+, such as fish, are exposed to concentrations of the toxic species Ag^+ at concentrations well-below where negative effects have been observed (at least five orders of magnitude below the MATC).

In the section on regulating silver, I mentioned several proposals for new aquatic silver criteria. One of these proposals is to base regulatory limits on Ag^+ concentrations. I have demonstrated that Ag^+ concentrations will be very low, since sulfide concentrations are in excess of total silver. The recent measurements of sulfides in other oxic waters systems indicate that this is probably a general phenomenon in fresh waters. Measuring Ag^+ concentrations will be difficult, if not impossible, due to (a) the lack of a sufficiently sensitive analytical method for measuring Ag^+ or (b) a separation method which can separate truly dissolved silver from colloidally bound silver (which could be used in conjunction with more sensitive analytical methods, e.g., ICPMS). However, it may be reasonable to use <10 kDa filtered samples as an indicator of waters needing further investigation.

The other recent proposal for a new aquatic silver criterion is based on the biota ligand model (BLM). In this method, a maximum acceptable concentration is determined for silver bound to the gill of a representative fish species (which is determined in relation to the MATC). By calculating the partitioning of silver between the gill and the solution species, the potential impact on aquatic biota is assessed for a given water body. The BLM method is good in that it allows for competition between biotic ligand sites and ligands in the water and that it is flexible enough to allow for multiple toxic species. However, the two major drawbacks of this method are (a) that it is hard to apply to other aquatic organisms other than fish (b) that it depends heavily on the quality of the input data (i.e., equilibrium constants and ligand concentrations). Using the data collected by Janes and Playle (1995) on silver-gill equilibrium binding constants and number of silver-gill binding sites (log $K_{Ag\text{-}gill}$ = 10.3 and 1.9 nmol binding sites per fish) along with the complexation constants and complexant concentrations determined in Paper IV (log K_{AgL} = 11.5, L_T = 50 nM), I calculated the amount of silver (for Ag_T = 1 nM) bound to the gill. While ignoring the additional protective effect of cations (i.e., Ca-gill and Na-gill), only 0.13% of the gill sites will be bound by silver. This demonstrates that sulfides in natural waters are likely important to keeping silver off the gill. However, measurements of sulfides and their silver binding constants are not routinely available.

For benthic feeding organisms where toxic effects have been observed as a result of exposure to contaminated sediments, other species of silver may be important. The research presented in this thesis demonstrated that silver reacts with AVS sulfide to form $Ag_2S(s)$. Hirsch (1998) has previously shown that silver as $Ag_2S(s)$ is not bioavailable. These findings together support the proposal to base sediment metals criteria on the SEM/AVS ratio (DiToro and Mahony, 1994). As long as the AVS concentration is greater than the silver concentration, no toxic effects should be observed. Although the protective effect of AVS would appear to apply to anoxic sediments only, sulfides may also be an important phase for binding ultra trace metals (i.e., Ag, Au and Hg) in oxic sediments as well. The persistence of sulfides in oxic waters observed in this work and others (Luther and Tsamakis, 1989; Rozan ans Benoit, 1998) suggests that sulfides must be considered. Sulfide measurements must be sufficiently sensitive to determine concentrations at nanogram/g levels.

Gagnon and Fisher (1997) studied the accumulation of cadmium, cobalt and silver from ingested sediment particles. These authors concluded that metals must be in solution in the digestive tract of an organism before assimilation can occur. This would suggest that any silver species entering the gut must disassociate before silver can be bioaccumulated. Silver measured in the Hamilton Harbour porewaters (Paper III) appears to be associated with metal sulfides. An important question which relates to this finding is - what happens to these metal sulfide complexes in the gut environment? This question needs to be addressed before the SEM/AVS proposal (DiToro and Mahoney, 1994) can be adopted as a sediment criterion for silver.

Finally, as this thesis research has demonstrated the importance of S(-II) to silver speciation in a variety of natural waters, I put forward the suggestion that the silver to sulfide ratio could be used as a water quality criterion for silver. The Cline Method represents a sensitive and straightforward method for determining S(-II) in water, and adopting this method would harmonize the water and sediment criteria.

6. Summary

The following statements summarize the major findings gained from this research:

1. At least 85-90% of silver is found in the particulate or colloidal phase for the WTP effluent and the surface waters examined.
2. Silver is complexed by particulate and colloidal bound inorganic sulfide. Inorganic sulfide concentrations are 100-1000 greater than silver concentrations, even is fully oxygen saturated waters.
3. Silver complexation constants for these inorganic sulfides are log $K'_{AgL} = 11\text{-}12$.
4. Other S(-II) complexes ($AgHS^\circ$, Ag-polysulfides and Ag-thiolates) will be present in freshwaters at lower concentrations, well-below the MATC of aquatic organisms.
5. Free silver ion concentrations will be very low ($[Ag^+] \approx 10^{-13}\text{-}10^{-14}$ M).
6. The results of the laboratory and field observations indicated that dissolved silver concentrations in sediments can be modelled based on $Ag_2S(s)$ solubility. However, silver complexed to colloidally bound inorganic sulfides is responsible for higher than predicted dissolved silver in some sediment porewaters.

7. Ideas for Future Research

This thesis research has demonstrated the importance of silver-sulfide complexes to silver speciation in natural waters. The conclusion of this research is that silver in the aqueous phase is likely bound to metal sulfides. To date, little is known about metal sulfide complexes in natural waters. It is unclear which metals are responsible for complexing sulfide, and the chemical form of these complexes is largely unknown.

Fe, Zn and Cu sulfide solution species have begun to be investigated by electrochemical techniques. However, the complexation of metal sulfide moieties by organic macromolecules has not yet been examined. It would be interesting to conduct a series of laboratory studies examining silver complexation with a number of simple metal sulfide species. These could include species already identified in natural waters: $M(SH)^+$, $M_2(SH)^{3+}$ and $M_2S_3^{2-}$ (Luther and Ferdelman, 1993; Rozan and Benoit, 1998). The effect of complexation by organic macromolecules could be assessed through the use of several low molecular weight model compounds. Characterization of these complexes could be performed using both electrochemical techniques and electrospray mass spectrometry.

Another important area of research under investigation by the Environmental Geochemistry Group at McMaster is the effect of oxidation on the mobility of silver from sediments. This work has primarily focused on the oxidation of $Ag_2S(s)$. An extension of this research would be to study the oxidation of Ag-Fe,Zn,Cu-sulfide solution species.

Finally, one of the main impetuses for developing a better understanding of silver speciation is the link between bioavailability and the form the metal. Toxicity tests need to be conducted on a variety of organisms using model Ag-Fe,Zn-sulfide complexes.

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REACTIVITY OF AG$^+$ ION WITH THIOL LIGANDS IN THE PRESENCE OF IRON SULFIDE

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Abstract—Dissolved silver (Ag) in the presence of amorphous iron sulfide (FeS) rapidly equilibrates to ultra-trace levels (~5 ng/L), which agrees with equilibrium calculations based on Ag_2S solubility. This finding, along with the authors' discovery that crystalline Ag_2S (acanthite) precipitates rapidly under supersaturated conditions, suggests that a discrete Ag-S phase is formed. Furthermore, potential interferences (Cl^-, Cu^{2+}, and dissolved organic carbon) show no effect on the Ag-FeS system. Only the presence of organosulfur (thiol) ligands at 10^{-3} to 10^{-4} M result in increased dissolved silver. Agreement between calculations and measured values were good for 1.8×10^{-4} M 3-mercaptopropanoic acid; however, calculations predicted higher dissolved silver than measured values for higher thiol levels, possibly due to Ag-thiol binding to FeS.

Keywords—Silver Iron sulfide Thiols

INTRODUCTION

Recently, silver (Ag) has been found at elevated concentrations in marine waters and freshwaters [1–4]. In addition, Luoma [5] and Crecelius [6] have shown biomagnification among bivalve populations living in waters elevated in silver. However, due to the fact that concentrations of silver in nature are at the picomolal level, at present data are limited on the distribution, reactivity, and fate of silver.

Silver is probably transported in the colloidal phase and is rapidly scavenged by suspended sediment. The partition coefficient between the particulate and aqueous phase of log K_d = 5.4 reflects this effect [7]. Some studies indicate that silver associated with particles is not available to benthic organisms [8]. Therefore, solid–liquid partitioning of silver appears to be important in biomagnification.

Iron sulfides are important for trace metal sequestering in reducing waters [9]. Because silver sulfides are very insoluble compared to iron sulfide (FeS) (solubility product ratios $[K_{sp}](Ag_2S)/K_{sp}(FeS) \sim 10^{-33}$), the interaction of silver with FeS may be important in the removal of silver from the water column. However, the nature of this interaction is unclear. The question of whether trace metals associated with sediments are present as discrete metal sulfide precipitates or sorbed species on surfaces of iron sulfide has not been resolved. Cowan et al. [10] suggested that dissolved silver concentrations are often supersaturated with respect to the formation of a discrete acanthite (Ag_2S) phase. The discrepancy between calculated dissolved silver concentrations and observed values may be attributable to the nature of silver in the solid phase.

Dissolved silver species, not included in equilibrium models, may also account for the difference between measured and estimated values. Silver binds strongly to organosulfur (thiol) ligands, commonly found in reducing environments [11,12]. Shea and MacCrehan [13] showed that complexation of copper by organic thiols accounted for discrepancies between observed copper concentrations in anoxic marine waters and thermodynamic model predictions.

This paper presents results for an experimental investigation of silver interactions with FeS. In this study, we look at the effect of metal complexation, as well as the influence of competition by other trace metals and the potential interference caused by dissolved organic matter, on silver uptake by FeS. Of particular interest is the complexation of silver with thiol ligands such as 3-mercaptopropanoic acid (3-MPA) and cysteine, which are commonly found in reducing environments at micromolal levels. The results are compared to equilibrium calculations.

EQUILIBRIUM CALCULATIONS

The experimental system in this study considers silver reacting with FeS. Experiments are run in an anoxic environment at constant pH and constant total sulfide (S_T). We also examine how this Ag-FeS system is affected by ligand complexation, other metals, and organic matter in the form of fulvic acid. We assume for the calculations that the system is in equilibrium with Ag_2S. Therefore

$$Ag_2S(s) + H^+ \rightarrow 2Ag^+ + HS^- \quad K(Ag_2S) = [Ag^+]^2[HS^-]/[H^+] = 10^{-35.94} \quad (1)$$

and

$$FeS(s) + H^+ \rightarrow Fe^{2+} + HS^- \quad K(FeS) = [Fe^{2+}][HS^-]/[H^+] = 10^{-2.95} \quad (2)$$

We consider the system at a pH of 8.0, where $S_T \sim [HS^-]$, and $[Ag^+]$ and $[Fe^{2+}]$ are fixed by Equations 1 and 2. Equilibrium calculations were performed for all the experiments conducted in this study. (Stability constants used in these calculations are listed in the Appendix.) Figure 1 shows the calculated total dissolved silver (Ag_T) concentrations for an Ag-HS^--H^+-Ag_2S system. The effect on Ag_T of thiol ligand at 10^{-3}, 10^{-4}, and 10^{-5} M concentrations is also shown. The dashed vertical line indicates the expected total sulfide con-

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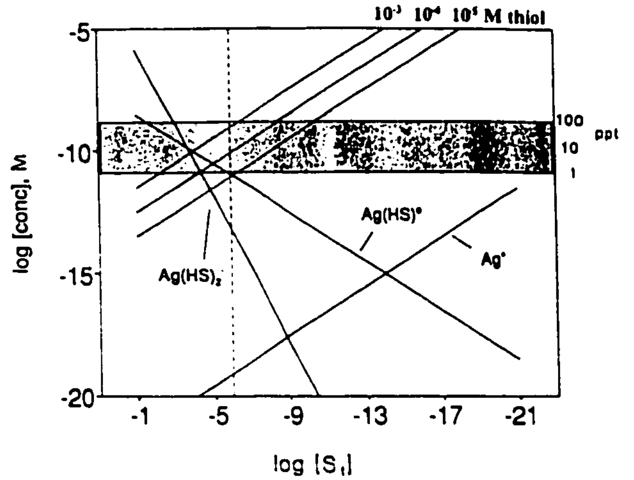

Fig. 1. Silver sulfide solubility as a function of total aqueous sulfide concentration at a pH of 8.0. AgHS° and Ag⁺ define the total soluble silver concentration in the absence of thiol ligands. Total silver (Ag$_T$) is also shown for 10^{-3} to 10^{-5} M thiol. The dotted line represents total sulfur (S$_T$) in equilibrium with FeS, where total iron (Fe$_T$) = S$_T$. The shaded area represents the range of Ag$_T$ found in nature.

centration for a system saturated with respect to FeS, where total iron (Fe$_T$) equals S$_T$. This value is consistent with many reducing environments. Calculations indicate that for S$_T$ = 10^{-6} M, Ag$_T$ is unaffected by Cl$^-$ concentrations ranging from freshwater to seawater levels. For thiol concentrations at $<10^{-9}$ M, chloro complexes will not become important until S$_T$ ~ 10^{-9} M for seawater and S$_T$ ~ 10^{-13} M for freshwater. Calculations also predict that fulvic acid will not affect Ag$_T$ concentrations. Copper(II) will not influence dissolved silver levels because S$_T$ is assumed to be constant, and total copper (Cu$_T$) in equilibrium with CuS(s) is too small to compete with silver for thiol ligands.

METHODS

Preparation of synthetic FeS

Degassed deionized water was prepared by deaeration of Millipore UF 18 MΩ water with N$_2$ (passed through an amalgamated zinc-metavanadate O$_2$ scrubber). A 0.5 M sulfide solution (buffered to pH 10 with Na$_2$CO$_3$) was prepared from Na$_2$ S·9H$_2$O and a 0.5 M Fe(II) solution was prepared from Fe(NH$_4$)$_2$(SO$_4$)$_2$·6H$_2$O. The reaction vessel consisted of a 500-ml three-hole round-bottom flask fitted with an argon in-flow and out-flow, a combination Ag-AgCl pH electrode, and Fe^{2+} and S^{2-} inlets. The S^{2-} and Fe^{2+} solutions were added at 3 ml/min to 300 ml degassed deionized water maintained at 25°C. The precipitate was collected on a 0.45-μm Millipore filter blanketed with argon. Batches of the precipitate were stored at 5°C under argon.

The precipitate was characterized by powder X-ray diffraction (XRD) using Cu K_α radiation. Broad, low-intensity peaks indicated the presence of some mackinawite and XRD-amorphous FeS. The solubility product of the precipitate, determined by dissolution of the precipitate, was approximately 50 to 100% more soluble than the solubility of mackinawite reported by Berner ($K = 10^{-3.64}$, Eqn. 2) [14].

Silver uptake experiments

The reaction vessels consisted of 2.5-L Teflon bottles wrapped in black polyethylene to suppress photochemical activity. Solutions were mixed using a peristaltic pump so that FeS at the bottom of the vessels was not disturbed (Teflon tubing was used except for the silicone manifold pump tubing). At the out-flow side of the pump, a two-way Teflon valve was added to accept a Millipore 0.2-μm in-line disposable polysulfone filter for sampling the solution.

For each experiment, 5.0 g of freshly precipitated FeS was weighed into a Teflon bottle. One liter of 30 nM silver nitrate solution containing the other components (thiols, Cl$^-$, Cu^{2+}, and fulvic acid) was carefully poured into the bottle so as not to disturb the FeS. Solutions were mixed for the duration of the experiment in a glove bag flushed continously with N$_2$. Two 10-ml samples of the solution were taken at 10 min, 1 h, and 2 h. The first sample was filtered through the 0.2-μm in-line filter. The second sample was filtered through the in-line filter, as well as a 10-kDa Millipore polysulfone centrifuge filter. Filtered samples were acidified in a class 100 cleanhood with 50 μl of high-purity 2 N HNO$_3$. The pH of each solution was determined once sampling was complete. Prior to each experimental run, all equipment (bottles, filters, tubing, and other parts) coming in contact with the reaction solution was prepared by soaking in 20% HNO$_3$ (reagent grade) for 3 d and then in 1% HNO$_3$ (trace metal grade) for 3 d. Chemicals used in this study were analytical grade, and included AgNO$_3$ and Cu(NO$_3$)$_2$ (BDH, Toronto, ON, Canada); 3-MPA and DL-cysteine (Sigma, St. Louis, MO, USA); KCl (BDH); and Armadale fulvic acid (Concordia University, Montreal, PQ, Canada) [15].

Analytical methods

Silver and iron were determined using a PE-Sciex Elan-500 ICPMS with ultrasonic nebulizer, adjusted for optimum response as follows: plasma Ar: 12; auxiliary Ar: 2; nebulizer Ar: 1,300 L/min; R.F. power: 1,200 W. Sensitivity was significantly improved by decreasing resolution from 1.0 A to 1.4 A. Samples were analyzed for ^{107}Ag, ^{109}Ag, and ^{54}Fe. Standard addition was used for all analyses. A PlasmaChem certified standard (PlasmaChem Associates, Bradley Beach, NJ, USA) was used to verify spiking solutions prepared in our lab. The three-sigma detection limit for silver in an acidified ionized water was 0.015 nM. Sulfide concentrations for the dissolution of the FeS precipitate were determined by the methylene blue method [16].

RESULTS

Previous work involving amorphous FeS has shown it to be extremely fine grained [17]. Anticipating that this would present a problem in distinguishing truly dissolved silver from colloidal silver, we introduced a method of determining whether colloidal material was present in our filtrate solutions. Iron concentrations were determined for both the 0.2-μm and 10-kDa filtrate solutions. Any differences in the Fe$_T$ concentration could thus be attributed to colloidal FeS. A comparison of the two iron concentrations showed no statistically significant difference between the two filter sizes at the 95% confidence level (using a two-tailed t test). Comparisons between samples taken at 10 min versus 2 h, as well as comparisons between the different experiments, also showed no significant variations in Fe concentrations. Therefore, we conclude that colloidal FeS did not interfere with the determination of dissolved silver concentrations in these experiments.

Also, no difference was found for silver concentrations in the 0.2-μm and 10-kDa filtrate solutions ($p < 0.05$, one-tailed

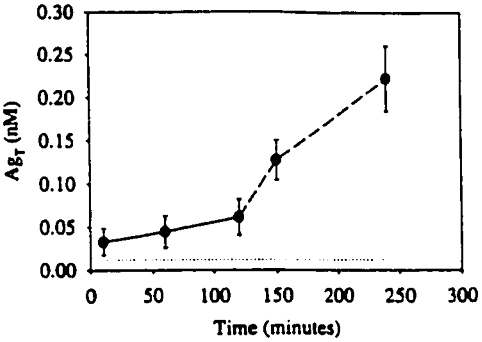

Fig. 2. Dissolved silver in the presence of FeS at pH 8.0 and 25°C. Solid line: dissolved silver in the presence of FeS; dashed line: remobilized silver with 1.8×10^{-1} M 3-mercaptopropanoic acid; dotted line: calculated total silver (Ag_T) for Ag_2S equilibrium. Error bars represent ±1 SD.

t test). As a result, we report Ag_T as the average of the two filtrate concentrations.

Silver interaction with FeS

Points connected by solid lines in Figure 2 show experimental results for uptake of dissolved silver concentrations in the presence of FeS. The removal of dissolved silver to the solid phase is very rapid. Within 10 min, total dissolved silver dropped from an initial 30 nM to 0.05 nM. Samples taken after 1 and 2 h showed no appreciable change, indicating that equilibrium for the Ag-FeS system is achieved quickly. In addition, thermodynamic equilibrium estimates for Ag_T for this system (dotted line, Fig. 2) predicted a similar value to what was found experimentally.

We were sceptical that acanthite would actually form under these conditions, because most metal sulfide minerals nucleate slowly. Instead, metastable amorphous metal sulfides are usually formed under supersaturated conditions. Therefore, we precipitated Ag_2S using the same procedure as used in the FeS precipitations, and immediately analyzed the solid by XRD. Figure 3 is the resulting XRD pattern for the precipitate. It shows the precipitate is at least 50% acanthite. Unlike other sulfides, XRD crystalline acanthite forms rapidly.

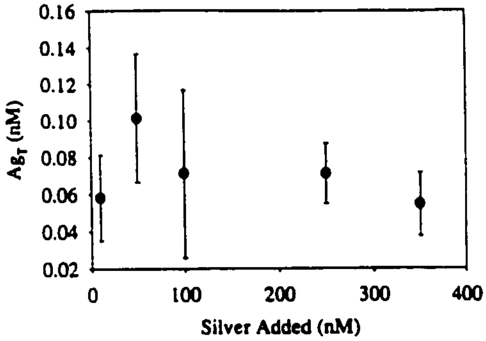

Fig. 4. Dissolved silver in the presence of FeS with a 1-h equilibration time between additions of $AgNO_3$ (10, 50, 100, 250, and 350 nM). Error bars represent ±1 SD.

A set of experiments was performed to study the uptake characteristics of silver in the presence of FeS for initial silver concentrations from 10 to 350 nM. Figure 4 shows that no increase in dissolved silver levels occurred with increasing total silver. The combined results of rapid Ag_2S formation and insensitivity to initial silver concentrations suggest that dissolved silver equilibrates with Ag_2S in the presence of FeS. Under these conditions, total dissolved silver will be at the low picomolal level.

Silver–organosulfur complexation

Experiments were carried out to assess the mobilization of silver in the presence of FeS. We investigated the effect of silver complexation by organosulfur (thiol) compounds. Our calculations show that under reducing conditions (sediment

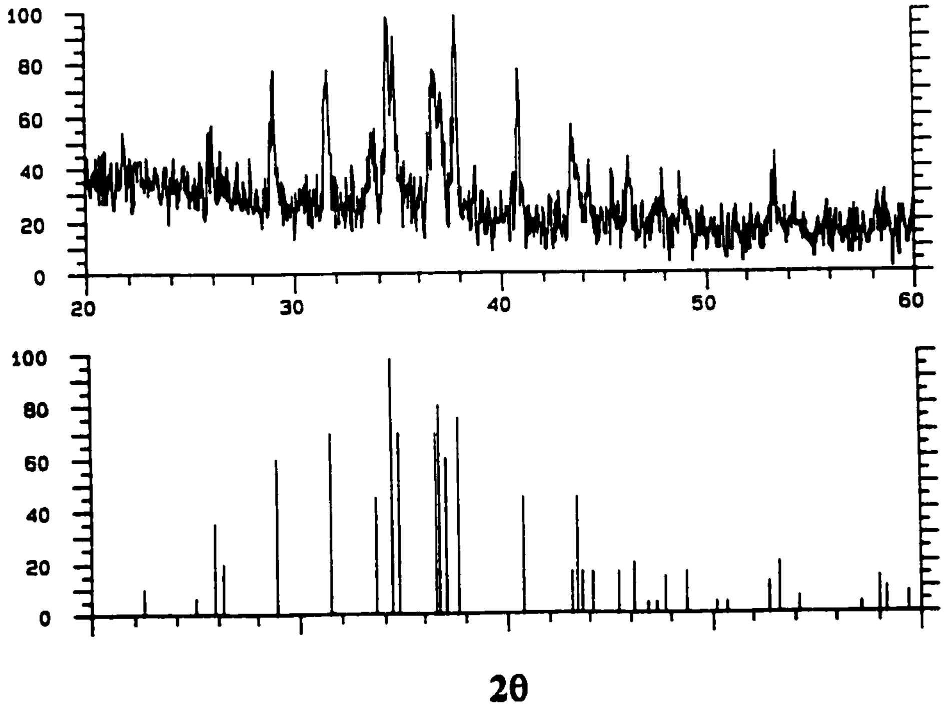

Fig. 3. X-ray diffraction powder (Cu K_α) spectrum for Ag_2S precipitate (upper) with acanthite reference (lower).

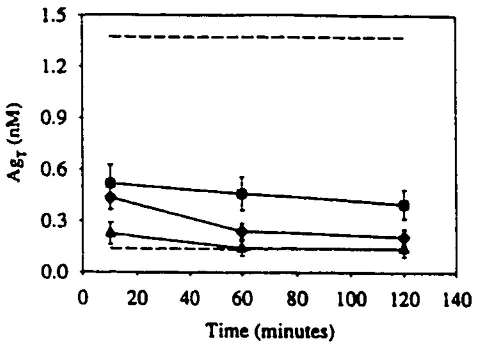

Fig. 5. The effect of thiol ligand on dissolved silver in the presence of FeS. Error bars represent ± 1 SD. —◆—, 1.8 × 10^{-3} M 3-mercaptopropanoic acid (3-MPA); —▲—, 1.8 × 10^{-4} M 3-MPA; —■—, 9.2 × 10^{-4} M cysteine. Dashed lines: calculated total silver (Ag_T) for Ag_2S equilibrium with 1.8 × 10^{-4} and 1.8 × 10^{-3} M 3-MPA.

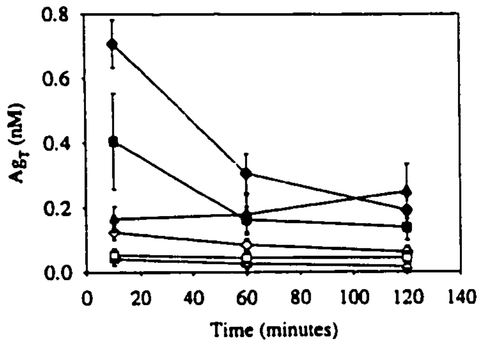

Fig. 6. The effect of fulvic acid, copper, and chloride on dissolved silver in the presence of FeS. Error bars represent ±1 SD. Filled symbols with 1.8 × 10^{-3} M 3-mercaptopropanoic acid, open symbols without. —◆—, 15 mg/L fulvic acid; —▲—, 50 µg/L $Cu(NO_3)_2$; —■—, 10^{-1} M KCl.

interface, high dissolved organic carbon [DOC], and so on), silver complexation with thiol compounds could explain the difference between observed and predicted dissolved silver levels found in many contaminated waters. Our calculations also show that in organically rich systems, including many marine environments, silver will exist predominately as silver thiolate complexes.

We determined dissolved silver in the presence of two thiols, 3-MPA and cysteine. Figure 5 shows experimentally determined dissolved Ag_T compared with calculated values. Reactions occur rapidly as observed in the simple Ag-FeS system. Experimental and equilibrium results compare closely at the lower thiol concentration (1.8 × 10^{-4} M). Equilibrium estimates, however, are two times higher than measured values at higher thiol concentration (1.8 × 10^{-3} M).

We performed experiments where an equilibrated Ag-FeS system was spiked with 3-MPA to investigate silver remobilization. The dashed portion of the plot in Figure 2 shows that silver can be remobilized by thiols. Moreover, because Ag_T is comparable for additions of Ag-thiol to FeS and thiol to Ag-FeS (Fig. 5 vs dashed line in Fig. 2), the order of addition of components (silver and thiol to FeS) does not make a difference in silver solubility, suggesting that this part of the system is reversible.

Secondary factors: Cu^{2+}, Cl^-, and DOC

Equilibrium calculations previously presented suggest a priori that thiol complexation should dominate over Cu^{2+}, Cl^-, and DOC in terms of affecting dissolved Ag_T concentrations. These constituents, however, are common in natural systems and might impose metastable conditions. Silver interactions with FeS were investigated for these three effects on silver mobility in the presence and absence of the 3-MPA thiol ligand. Figure 6 shows these results. After 2 h, in the absence of 3-MPA, none of the three secondary factors (Cl^-, Cu^{2+}, DOC) significantly increased Ag_T ($p < 0.05$, one-tailed t test) above the control condition as determined by the Ag-FeS experiment shown as the solid line in Figure 1. The secondary factors with 3-MPA gave significantly higher results in all cases, and were comparable to what was found for 3-MPA in the absence of the secondary factors.

DISCUSSION

Our experiments demonstrate that trace levels of dissolved silver in the presence of FeS are rapidly transferred to the solid phase. Silver remaining in solution after equilibrating with the solid is at ultra-trace levels (~0.05 nM), agreeing closely with the estimated solubility of acanthite (Ag_2S). This finding, along with the unusually rapid formation of crystalline acanthite, suggests a discrete Ag-S phase.

Thiol concentrations in reducing environments have been measured at the low micromolal level. Equilibrium calculations at $S_T = 10^{-6}$ M (system in equilibrium with FeS) predict that Ag-thiol complexation is about of equal importance to $AgHS^{\circ}$ for this level of thiol. Silver thiolate complexes, however, can be the dominant dissolved silver species at thiol concentrations of 10^{-5} M or higher. These higher thiol concentrations might be expected for systems containing high levels of natural organic matter or for waters in the vicinity of urban centers. The experimental results confirm this prediction. The effect of increasing thiol concentrations on Ag_T, however, was somewhat suppressed relative to predicted values. We propose that this discrepancy is due to Ag-thiol binding to the FeS solid, which is ignored in our calculations.

For $Ag\text{-}HS^-\text{-}Cl^-$ systems, calculations show that it is not until $S_T < 10^{-9}$ M for marine waters ($[Cl^-] = 0.5$ M) and $S_T < 10^{-13}$ M for freshwaters ($[Cl^-] = 10^{-3}$ M) that chloro complexes replace $AgHS^{\circ}$ as the predominant dissolved silver species. Thus, silver chloro complexes should only be important in marine surface waters where total sulfide could be $<10^{-9}$ M. Only trace levels of thiol ligand (10^{-8} M or less) are necessary, however, so that [Ag-thiol] = $[AgHS^{\circ}]$ or $[AgCl_n^{1-n}]$ at $S_T < 10^{-9}$ M. At present, we can only speculate as to the levels and importance of thiol compounds in more oxidizing waters. We are currently developing techniques to investigate the existence of such complexes in natural systems and demonstrate their importance in the speciation of silver.

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Appendix

The following thermochemical data were used for equilibrium calculations.

Sulfide complexes

The solubilities of Ag_2S and CuS were taken from Dyrssen and Kremling [18]. Silver complexation by sulfide has been studied extensively and quite carefully by Schwarzenbach and Widmer [19]. The solubility of FeS was taken from Davison [20] (ionic strength [I] = 0; 25°C).

$$H_2S(aq) = H^+ + HS^- \qquad \log K_{s1} = -7.02$$

$$Ag_2S(s) + H^+ = 2Ag^+ + HS^- \qquad \log K_{s0} = -35.94$$

$$Ag^+ + HS^- = AgHS^\circ \qquad \log K_1 = 14.0$$

$$AgHS^\circ + HS^- = Ag(HS)_2^- \qquad \log K_2 = 3.67$$

$$FeS(s) + H^+ = Fe^{2+} + HS^- \qquad \log K_{s0} = -2.95$$

$$CuS(s) + H^+ = Cu^{2+} + HS^- \qquad \log K_{s0} = -22.3$$

Silver chloro complexes

This information was compiled from Sillen and Martell [21] and Kratohvil et al. [22].

$$Ag^+ + Cl^- = AgCl^\circ \qquad \log K_1 = 3.5$$

$$Ag^+ + 2Cl^- = AgCl_2^- \qquad \log K_2 = 5.4$$

$$Ag^+ + 3Cl^- = AgCl_3^{2-} \qquad \log K_2 = 5.63$$

$$Ag^+ + 4Cl^- = AgCl_4^{3-} \qquad \log K_4 = 5.23$$

Silver and copper thiolate complexation

The constants for silver thiolate complexation (our work) are: 3-mercaptopropionic acid, $\log K_1(AgSR) = 12.0$, $\log K_2(Ag(SR)_2^-) = 14.0$; L-cysteine, $\log K_1(AgSR) = 11.9$, $\log K_2(Ag(SR)_2^-) = 15.2$. The constant for copper(II) thiolate complexation is: for penicillamine (Kuchinskos and Rosen [23]), $\log K_1(CuSR) = 16.5$. Stability constants for copper(II) thiolate complexes are limited. However, calculations with values all the way up to $\log K = 20$ suggest that Cu does not affect Ag-thiol concentrations.

Silver complexation with Armadale fulvic acid

Varshal et al. [24] determined the conditional stability constant for a 1:1 silver–fulvic acid (FA) complex at pH 8.0 ($I = 0.1$). Although the fulvic acid was isolated from the Moscow River, we assume the stability constant will be reasonably similar for the Armadale fulvic acid.

$$Ag^+ + FA = Ag\text{-}FA \qquad \log K = 5.67$$

II

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Potentiometric Determination of Silver Thiolate Formation Constants Using a Ag_2S Electrode

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Abstract. Formation constants for silver thiolates were obtained by titration of the ligand in a constant temperature, ionic strength and pH medium and measuring the potential change at a Ag_2S electrode. A non-linear equation was derived from which the first and second silver formation constants, β_1' and β_2', and the sulfide group acid dissociation constant,.K_a', were determined. An overall estimate of the uncertainty in the derived parameters was obtained using a Monte Carlo approach. The procedure was compared to a previous work on AgHS°. Log β_1', log β_2' and $-\log K_a'$ results were obtained for cysteine (11.9 ± 0.5, 15.2 ± 0.4, 7.8 ± 0.1), glutathione (12.3 ± 0.3, 14.3 ± 0.8, 8.8 ± 0.3) and 3-mercaptopropanoic acid (12.0 ± 0.4, 14.0 ± 0.4, 10.5 ± 0.3) at 20 °C and 0.01 m ionic strength.

Key words: Silver sulfide, thiols, formation constants

1. Introduction

Silver, being a group B element, binds strongly with sulfur with formation constants of 10^{10} or better in many cases (Martell and Smith, 1977). Speciation of silver with sulfur groups in inorganic and organic ligands will affect aqueous silver concentrations (Adams and Kramer, 1998). We have found only a limited number of reliable formation constants for such complexes in aqueous solution (Smith and Martell, 1997; Schwarzenbach and Widmer, 1966).

Silver has a propensity for forming polynuclear complexes with thiol ligands at silver concentrations greater than about 10^{-4} mol L^{-1} (Tunaboylu and Schwarzenbach, 1971). Silver concentrations, however, are at much lower concentrations ($< 10^{-6}$ mol L^{-1}) in natural waters where mononuclear complexes should predominate. Thus the technique for determining stability constants applicable to natural waters must be operative at low total silver concentrations. Tseng and Gutknecht (1975) showed the application of a polycrystalline Ag_2S membrane electrode to the determination of silver thiolate formation constants. Their methodology and data reduction, however, required improvements in order to give more precise formation constant information. We have (a) fabricated a better Ag_2S electrode, (b) developed an experimental procedure for obtaining both silver formation constants and acid dissociation constant, and (c) obtained more rigorous assessment of data by using nonlinear fitting of equations, following Morf et al. (1974).

We determined a formation constant for the AgHS° species to compare to a previous determination (Schwarzenbach and Widmer, 1966). We also report formation constant values for several commonly occurring thiol ligands found in reducing waters: L-cysteine, glutathione and 3-mercaptopropanoic acid (3-MPA) (Shea and MacCrehan, 1988).

2. Theory

The potential measured by a Ag_2S electrode is attributed to the activity of silver Ag^+ in solution, the silver ion activity due to Ag_2S equilibrium, and the silver "leached cation activity", α_D. α_D is due to Frenkel defects in the Ag_2S lattice, impurities in the crystal and chemisorption (Morf, 1981, p. 171 *ff*). Morf concluded that α_D is constant for a well conditioned electrode but may vary with preparation, conditioning and stirring rate and is about $10^{-5.5}$ mol L^{-1} for the Ag_2S electrode.

The Ag_2S electrode potential can be stated as:

$$E_{\mathrm{meas}} = E^0_{\mathrm{Ag}^+} + 2.3\frac{RT}{F}\log(\{\mathrm{Ag}^+\} + \alpha_D) \tag{1a}$$

and when $\{Ag^+\} \gg \alpha_D$ (about 10^{-4} mol L^{-1}),

$$E'_{\mathrm{meas}} = E^0_{\mathrm{Ag}^+} + 2.3\frac{RT}{F}\log\{\mathrm{Ag}^+\} \tag{1b}$$

which reflects the linear portion of the calibration curve. Equation (1b) can be stated in terms of Ag^+ concentration:

$$E'_{\mathrm{meas}} = \left(E^0_{\mathrm{Ag}^+} + 2.3\frac{RT}{F}\log\gamma_{\mathrm{Ag}^+}\right) + 2.3\frac{RT}{F}\log[\mathrm{Ag}^+] \tag{1c}$$

and at constant temperature and ionic strength in linear form:

$$E'_{\mathrm{meas},t,I} = a_0 + a_1\log[\mathrm{Ag}^+] \tag{1d}$$

where $E^0_{Ag^+}$ is the standard state potential, brackets are solution concentrations, braces are activities, γ_{Ag^+} is the activity coefficient for Ag^+ and a_o and a_1 are intercept and slope for a linear equation.

We assume that a ligand, (L^{p-}), forms only mono-nuclear complexes with Ag^+ (AgL_n^{1-np}), since the mononuclear wall should not be surpassed due to the low Ag^+ activity (Stumm and Morgan, 1996, p. 266). The reaction is:

$$\mathrm{Ag}^+ + nL^{p-} = \mathrm{Ag}L_n^{1-np}$$

and the concentration conditional formation constant (β'_n) is given as:

$$[\mathrm{Ag}L_n^{1-np}] = \beta'_n[\mathrm{Ag}^+][L^{p-}]^n. \tag{2}$$

We assume metal complexation occurs only at the sulfhydryl group in these thiol compounds ($\mathrm{H}L^{1-p}$ and L^{p-}), and we can resolve the AgL_1^{1-p} and AgL_2^{1-2p} species.

Mass balance expressions for total Ag, $[\mathrm{Ag}_t]$, and total ligand, $[L_t]$, given the above limitations, are:

$$[\mathrm{Ag}_t] = [\mathrm{Ag}^+] + [\mathrm{Ag}L_1^{1-p}] + [\mathrm{Ag}L_2^{1-2p}]$$

and

$$[L_t] = [L^{p-}] + [\mathrm{H}L^{1-p}] + [\mathrm{Ag}L_1^{1-p}] + 2[\mathrm{Ag}L_2^{1-2p}]$$

For excess ligand compared to silver and for strong Ag-ligand binding, the mass balance expressions simplify to,

$$[L_t] \approx [L_t^{p-}] + [\mathrm{H}L_t^{1-p}] \tag{3a}$$

$$[L_t^{p-}] \approx [L^{p-}] \tag{3b}$$

$$[\mathrm{H}L_t^{1-p}] \approx [\mathrm{H}L^{1-p}] \tag{3c}$$

$$[\mathrm{Ag}_t] \approx [\mathrm{Ag}L_1^{1-p}] + [\mathrm{Ag}L_2^{1-2p}] \tag{3d}$$

For $K'_a = \{\mathrm{H}^+\}[L^p]/[\mathrm{H}L^{1-p}]$, and substituting Equations (3a) and (3b) into Equation (2), we obtain:

$$\begin{aligned}[\mathrm{Ag}L_1^{1-p}] &= \beta'_1\alpha_a[L_t][\mathrm{Ag}^+] \\ [\mathrm{Ag}L_2^{1-2p}] &= \beta'_2\alpha_a[L_t]^2[\mathrm{Ag}^+]\end{aligned} \tag{4}$$

and

$$\alpha_a = \frac{[L_t^{p-}]}{[L_t]} = \frac{K'_a}{\{\mathrm{H}^+\} + K'_a}$$

where $[L_t]$ is the total ligand in solution and $[L_t^{p-}]$ is the total unprotonated ligand (i.e., $[L_t] = [L_t^{p-}] + [\mathrm{H}L_t^{1-p}]$).

For the experimental conditions of $[L_t^{p-}] \gg [\mathrm{Ag}_t]$ and $[\mathrm{Ag}_t] \approx \sum[\mathrm{Ag}L_n^{1-np}]$, the silver ion mass balance simplifies to:

$$\sum[\mathrm{Ag}L_n^{1-np}] = 2[\mathrm{H}S^-] + \alpha_D \tag{5}$$

where $[HS^-]$ is the $[Ag^+]$ due to electrode dissolution (i.e., $Ag_2S + H^+ = 2\,Ag^+ + HS^-$). The solubility product of Ag_2S is:

$$K_{Ag_2S} = \frac{[Ag^+]^2[HS^-]}{\{H^+\}}. \tag{6}$$

Substituting Equations (4) and (6) into Equation (5) and rearranging results in a cubic equation in $[Ag^+]$:

$$[\beta'_1\alpha_a[L_t] + \beta'_2\alpha_a^2[L_t]^2][Ag^+]^3 - \alpha_D[Ag^+]^2 - 2K'_{Ag_2S}\{H^+\} = 0. \tag{7}$$

Parameters K'_a, β'_1 and β'_2 were estimated simultaneously by fitting Equations (1) and (7) to the data using the simplex optimizer in MatLab®. The objective function for non-linear fitting was set to minimize $(\sum(E_{meas} - E_{calc})^2)$.

The intercept, a_0, and slope, a_1, in Equation (1d) and α_D in Equation (7) are estimated from titration of $AgNO_3$. The mean electrode calibration values (a_0, a_1 and α_D) along with E_{meas} and L_t at pH 4, 6 and 8 represents a complete experimental data set to input into the fitting routine, and solve for the formation constants and the sulfhydryl acidity constant.

The leached silver ion activity, α_D, was determined from data when the calibration curve becomes non-linear (e.g. Figure 1). Subtraction of Equation (1b) from (1a), using the definition of a_1, and re-arrangement gives:

$$\alpha_D = [Ag^+]\gamma_{Ag^+}(10^{(E-E)/a_1} - 1)$$

where E and E' are the potentials defined in Equations (1a) and (1b) respectively. The activity coefficient, γ_{Ag^+} is estimated at 0.9 for the experimental conditions (Stumm and Morgan, 1996, p. 109). A plot of $(10^{(E-E')/a_1} - 1)$ versus $(1/([Ag^+]\gamma_{Ag}))$ gives slope α_D and intercept of zero. α_D is obtained from least squares regression of data in the non-linear portion of the calibration curve. The leached silver ion activity, α_D, is assumed constant during the experiment (Morf, 1981, p. 172).

$(-\text{Log } K'_{Ag_2S}) = [HS^-][Ag^+]^2/\{H^+\})$ is estimated as −36.74 from data at 20 °C (Smith and Martell, 1997), and adjusted to an ionic strength of 0.01 mol L^{-1} using the extended Debye–Hückel equation for γ_{Ag+} and γ_{HS^-}. Note that the term , $2K'_{Ag_2S}\{H^+\}$, is small compared to the other two terms in Equation (7). Therefore the variations in the value of K'_{Ag_2S} have negligible influence on the solution of Equation (7).

3. Experimental

The Ag_2S electrode used in this study was fabricated by compressing ultrapure Ag_2S (Johnson Matthey, U.K.) at 30000 psi for 48 hrs. A platinum wire was embedded in one end of the pellet to obtain electrical continuity. The resulting

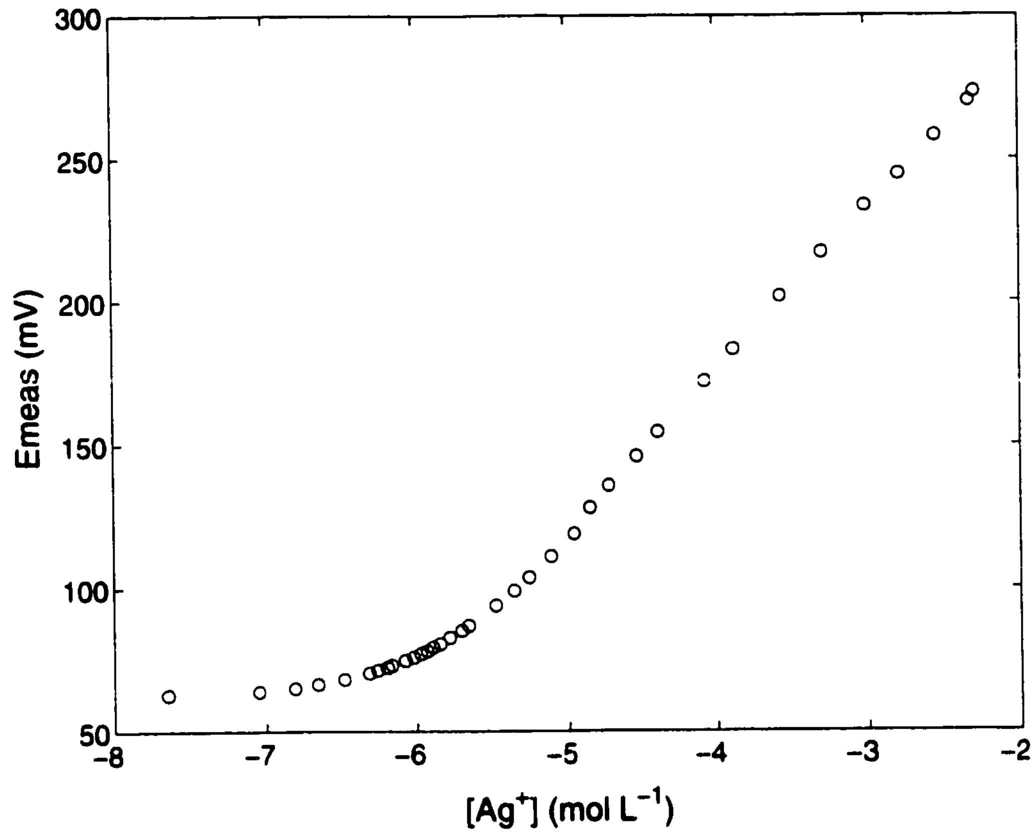

Figure 1. Example of calibration of Ag_2S electrode by titration with $AgNO_3$ at pH of 6, 20 °C and $I = 0.01$. The defect concentration, α_D, as well as a_0 and a_1 are determined from these data.

cylindrical pellet (6 mm ϕ × 6 mm) was mounted on a glass tube using epoxy resin and heat shrink tubing. The contact surface was polished on Microcloth using 0.02 mm alumina as the polishing agent. The electrode was conditioned prior to each experiment with a light polishing and soaking for a minimum of 3 hours in a 10^{-2} mol L^{-1} $AgNO_3$ solution. We obtained a 40% reduction in α_D over other fabrications and conditionings (e.g. Morf, 1981, p 166).

Titrations were carried out in a closed 100 mL glass vessel with three ports in the top for the Ag_2S electrode, pH electrode and titration tubes. The vessel was filled with 50 mL of deionized water and purged using high-purity helium. Ionic strength was set to 0.01 mol L^{-1} with KNO_3. A water-jacket maintained a constant temperature of 20 ± 0.2 °C A dual electrode automated titrator (Tanager Scientific Systems, Ancaster, ON, Canada) was used to obtain pH and E_{meas} data for additions of ligand. pH was held constant within ±0.02 log units using 0.1 mol L^{-1} HNO_3 and NaOH added by the titrator system. A double-junction combination pH electrode (Orion Research Inc., Beverly, MA, USA), filled with KNO_3, was used for pH measurements, and the reference portion was used for both the pH and the Ag_2S electrode. Two buffer solutions (pH 4.00 and 7.00, Anachemia Inc., Montreal, QC, Canada) were used to calibrate the pH electrode. The titrator

Table I. Determination of parameters in Equation (1). Multiple titrations were performed with $AgNO_3$ at pH 4, 6, 8 and 10. $I = 0.01$ mol L^{-1}, $t = 20$ °C.

$a_0 = 405.0 \pm 9.2$ (1 s.d., $n = 8$) mV
$a_1 = 60.8 \pm 4.0$ (1 s.d., $n = 8$) mV
$\alpha_D = 2.29 \times 10^{-6} \pm 1.55 \times 10^{-6}$ (1 s.d., $n = 8$) mol L^{-1}

software was configured so that the E_{meas} (mV) of the solution was recorded when the measured potential changed by less than 0.2 mV over a 5 minute period with no statistical indication of E_{meas} gradient.

Solutions of L-cysteine, glutathione and 3-mercaptopropanoic acid (Sigma Chemical Co., USA) used in titrations were prepared from He-purged, deionized water. A 1 mol L^{-1} solution of HS^- was prepared from $Na_2S{\cdot}9H_2O$, and was standardized using the Cline Method (Cline, 1969).

4. Results and Discussion

The intercept and slope, a_0 and a_1, of Equation (1d) were determined from least squares estimation from multiple titrations with $AgNO_3$. Titrations were performed at constant pH values from 4 to 10 and periodically throughout the study. The calibration of the Ag_2S electrode did not change with pH or during the study. Experiments can not be carried out above a pH of 10, however, since there is a significant E_{meas} shunt due to the formation of silver oxyhydroxides.

Figure 1 is one example of the plot of E_{meas} versus log $[Ag^+]$ for the electrode calibration. Table I gives the overall mean and standard deviation for multiple electrode calibrations of a_0, a_1 and α_D.

We first tested this method by comparing results to previous work by Tunaboylu and Schwarzenbach (1966) for the system: Ag^+—HS^-—H_2O. Titrations were performed at pH 8 and 10. At high HS^- concentrations ($>10^{-3}$ mol L^{-1}), large negative shifts in E_{meas} occurred. This shift suggests that hydrogen bisulfide reacts readily with the membrane surface, resulting in a non-Nerstian response. For this reason, only a formation constant for $AgHS^\circ$ can be reported for a pH 8 of log $\beta = 12.8$ (20 °C, 0.01 mol L^{-1}). However, our value compares favorably with the β value published by Tunaboylu and Schwarzenbach (1966) of log $\beta = 13.3$, (20 °C, 0.1 mol L^{-1}) even though the ionic strengths are different.

L-cysteine (CYS), glutathione (GLU) and 3-mercapto-propanoic acid (3-MPA) were titrated at fixed pHs of 4, 6 and 8 in order to obtain K_a', β_1' and β_2'. Plots of E_{meas} versus log $[L_t]$ for L-cysteine at pH 4, 6 and 8 are shown in Figure 2. Figure 3 compares the fitted function to the experimental data (E_{meas} versus log $[L_t^{p-}]$) for L-cysteine. Titration data for GLU and 3-MPA were fitted following the same procedure. Table II lists the fitted formation constants (β_1' and β_2'), as well

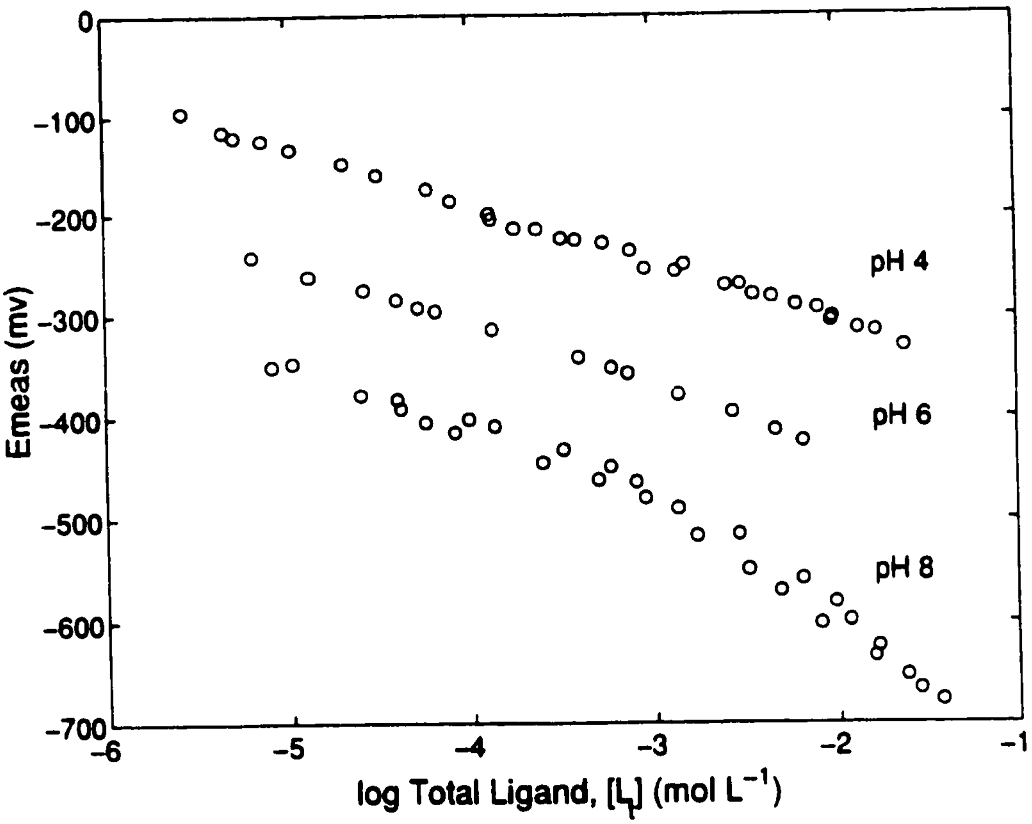

Figure 2. E_{meas} versus log $[L_t]$ for L-cysteine at pH 4, 6 and 8.

Table II. Fitted constants to thiols. Literature values from (Smith and Martell, 1997). $I = 0.01$ mol L^{-1} and $t = 20$ °C, unless otherwise stated.

Thiol	log β_1'	log β_2'	$-\log K_a'$	Literature $-\log K_a'$
L-Cysteine	11.9 ± 0.49	15.2 ± 0.39	7.8 ± 0.11	7.8 (20 °C, 0.01 mol L^{-1})
Glutathione	12.3 ± 0.32	14.3 ± 0.79	8.8 ± 0.31	9.1 (25 °C, 0.16 mol L^{-1})
3-Mercaptopropanoic acid	12.0 ± 0.36	14.0 ± 0.36	10.5 ± 0.25	10.5 (20 °C, 0.01 mol L^{-1})

as the fitted acidity constant (K_a') for these three thiol ligands. Acidity constants obtained by this procedure were compared to other reported values. The acidity constants compared very well for CYS and 3-MPA and GLU, considering that constants at different temperatures and ionic strengths were compared for GLU.

Although CYS, GLU and 3-MPA have multiple protonation sites (i.e., carboxylic acid and amine groups), good agreement between experimental data and the fitted model for all three thiol ligands suggests that for the pH range considered (pH 4–8), complexation of silver occurs with the thiol group (RS$^-$) and is unaffected by changes in the degree of protonation at the other functional sites. The

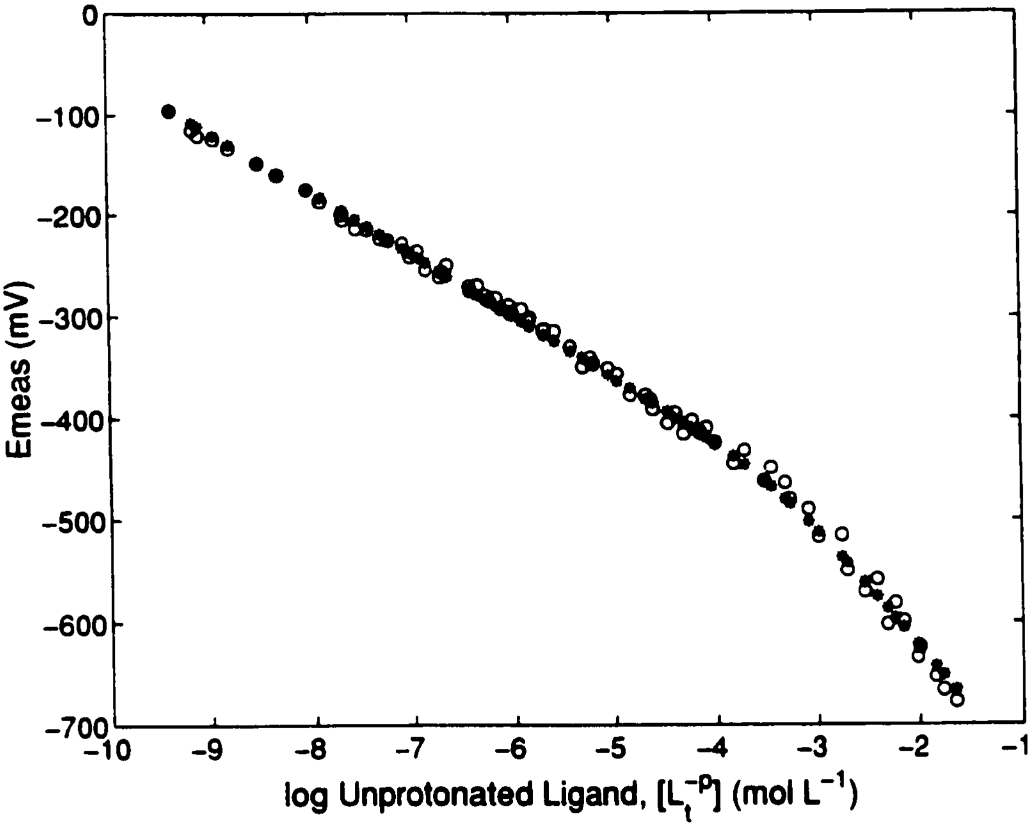

Figure 3. Comparison of fitted data (E_{meas} versus $[L_t^{p-}]$) to experimental points for L-cysteine titration data (Figure 2). ○, experimental; *, calculated.

amine groups present in CYS and GLU do not appear to play a measurable role in silver complexation.

Tseng and Gutnecht (1975) used the same electrode and a similar approach to determine log β values for L-cysteine and 3-mercapto-propanoic acid of 13.5 (0.1 mol L^{-1} NaOH, 20–21 °C) and 14.4 (1.0 mol L^{-1} NaOH, 20-21 °C), respectively. These are about two orders of magnitude larger than what we obtained (Table II). However, the fitting method used by Tseng and Gutnecht to determine β'_1 values from the data places an apparently large and undefined uncertainty on their results.

We used a Monte Carlo approach to estimate the uncertainty in the parameters determined by non-linear fitting of Equations (1) and (7) to the titration data. In nonlinear fitting, exact confidence limits can not be calculated. The Monte Carlo approach is especially useful when used with the simplex method, which does not otherwise provide any estimate of uncertainty in the measured parameters (Motulsky and Ransnas, 1987).

Following the Monte Carlo approach, we calculate a distribution of values for each parameter (β'_1, β'_2 and K'_a). A minimum of one thousand pseudo data sets were created by calculating E_{meas} values from the best fit K'_a, β'_1 and β'_2 estimates

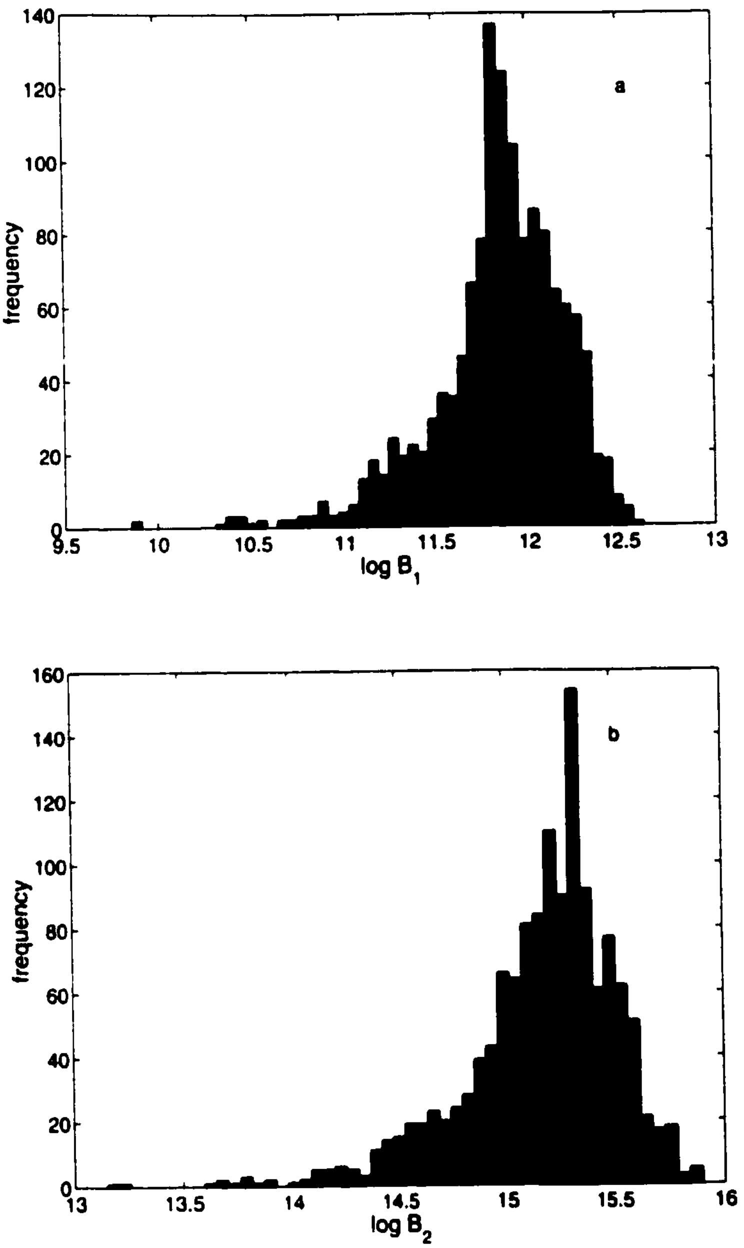

Figure 4a,b.

and mean values of a_0, a_1 and α_D at all pH and ligand concentrations. Random-normal noise with a standard deviation of 5 mV (estimated from repeat titrations) was added to each calculated E_{meas}. The pseudo data sets were fitted following the method outlined above for the original, experimentally determined data set. Different values of a_0, a_1 and α_D, obtained from random-normal sampling using values and standard deviations given in Table I, were used for fitting each of the pseudo data sets. This method provides a distribution of values for each parameter resulting from uncertainty in both the electrode calibration and ligand titrations.

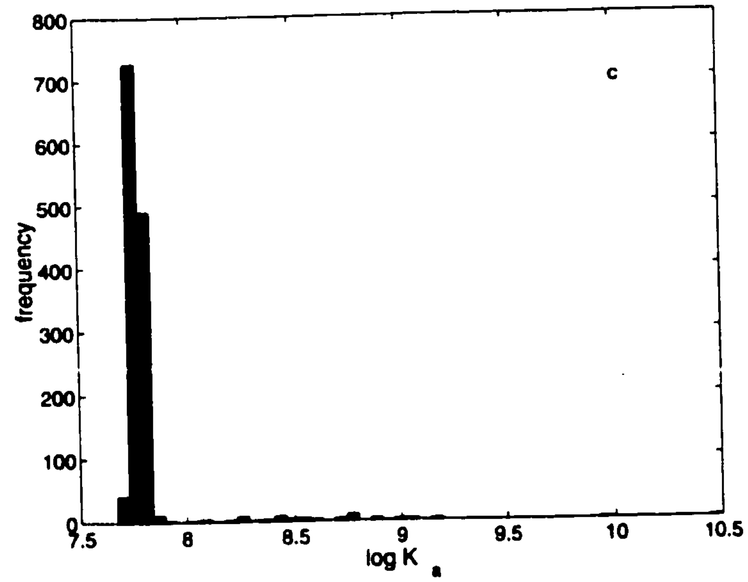

Figure 4. Distribution of fitted parameters for L-cysteine: (a) β'_1, (b) β'_2 and (c) K'_a. Estimates of error of fitting are obtained from these distributions.

Figure 4 shows the distributions of fitted parameters for L-cysteine. The deviation is small for K'_a and is skewed for β'_1 and β'_2. We found that if the standard deviation of α_D was reduced ten-fold while performing Monte Carlo simulations, this skewness was eliminated. Estimates of the uncertainty for K'_a, β'_1 and β'_2 are shown in Table II. In making these estimates, we assumed a log-normal distribution and give one standard deviation about the mean (Table II).

Acknowledgments

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III

Silver Speciation in Wastewater Effluent, Surface Waters and Porewaters

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Running Header: Silver speciation in wastewater effluent, surface waters and porewaters

Abstract

Silver, inorganic sulfide and thiol compounds were measured in municipal wastewater effluent and receiving waters and porewaters from an anoxic lake sediment in order to predict silver speciation in these systems. We found sub-micromolar concentrations of inorganic sulfide even in fully oxic surface water. This inorganic sulfide is likely in the form of colloidal metal sulfides, which have been shown to be stable under oxidizing conditions for periods of several hours. Inorganic sulfide in both the wastewater effluent and receiving waters was found to be 200-300 times in excess of silver concentrations, while inorganic sulfide in the porewaters was 1000-15000 times in excess of silver concentrations. With sulfide in excess of silver, we predict silver-sulfide complexes to dominate silver speciation. Thiols were present at low nanomolar levels in porewaters, but were not detectable (< 1 nM) in wastewater effluent or receiving waters. Thiols do not appear to be important to silver speciation in these freshwater systems. Partitioning of silver into particulate (>0.45 μm), colloidal (10 kDa - 0.45 μm) and dissolved (<10 kDa) size fractions showed that a significant proportion of silver is in the colloidal (30-35%) and dissolved phase (15-20%). Dissolved phase concentrations were relatively constant in the treatment plant effluent and receiving waters, suggesting silver in the <10 kDa size fraction is strongly complexed by ligands that are not significantly affected by aggregation or sorption processes.

Keywords: silver, sulfide, thiols, speciation, wastewater, sediment porewaters

Introduction

Silver is found in unperturbed aquatic systems at ultra-trace levels (5-100 picomolar) [1-5], while silver concentrations near the outfalls of municipal wastewater treatment plants (WTP) tend to be elevated by at least 1-2 orders of magnitude [6]. There is concern that these elevated silver concentrations may impact on aquatic biota.

The bioavailability and toxicity of metals are integrally linked to their chemical form [7,8]. In order to conduct meaningful toxicity experiments, we must, therefore, have knowledge of how silver speciates in the environment. To date, few studies exist which deal with the chemical forms of silver in natural waters. Based on available thermodynamic data, we previously postulated that silver(I) is likely to be complexed by sulfides under almost all natural conditions. Silver(I) forms very strong complexes with S(-II) species. The formation constant for $Ag^{+} + HS^{-} = AgHS^{\circ}$ is log K ~ 13 [9]. Formation constants of similar magnitude have been determined for several organic sulfide compounds (i.e., thiols) [10]. Compounds containing oxygen and nitrogen groups display much weaker silver binding (e.g., EDTA, log K ~ 6) [11].

In marine and estuarine porewaters, sulfide is present in millimolar concentrations [12,13]. Other reduced sulfur species are present at lower concentrations, including a number of thiol compounds [14]. As well, sulfides have been shown to occur at nano- and picomolar concentrations in the open ocean [15,16]. Little equivalent data exists for freshwater. Rozan and Benoit [17] recently measured metal sulfide species at nanomolar concentrations in oxygenated river waters. As well, Wang and Tessier [18] have measured various sulfur species in porewaters from anoxic lake sediment.

Our thermodynamic calculations suggest that silver will be complexed by S(-II), as long as

S(-II) concentrations are greater than silver concentrations [19]. Under this condition, the free silver ion concentration should be negligible, due to the strength of the silver-sulfur(-II) bond.

In this paper, we present the results of a study examining silver, sulfide and thiol concentrations in porewater, surface water and WTP effluent samples. We also present data on the distribution of these species amongst the particulate, colloidal and dissolved phase.

Methods

Surface water and wastewater effluents were sampled throughout the spring and summer of 1997 and 1998. Wastewater effluents were sampled at the Dundas and Burlington (Ontario, Canada) municipal WTPs. Surface water samples were taken at several locations downstream from the Dundas plant in the Desjardin Canal.

Both treatment plants use a tertiary activated sludge treatment process, and add ferric chloride to enhance floc formation and setting in the clarifier tanks. Disinfection is accomplished through chlorination, and treated effluent from the Dundas plant also passes through a sand filter as the final step in the treatment process.

Low-level trace metals techniques were used to avoid contamination and to provide sensitive and reliable analyses. Procedures for the preparation of sampling apparatus, processing of water samples, and analysis by ICP-MS were adapted from protocols used by the Lake Michigan Tributary Monitoring Project (LMTMP) [20]. All surfaces contacting water samples were acid-cleaned in 20% ACS grade HNO_3 acid and 1% high-purity TMA HNO_3 acid (Baker Trace Metal Grade) for a minimum of 3 days. Materials were then rinsed with Milli-Q water (Millipore), and dried in a HEPA filtered environment. Samples for metal, sulfide and thiol analysis were collected in 1L teflon® bottles stored in zip-lock polyethylene bags. Wastewater samples were collected directly into teflon® bottles by personnel wearing arm-length PVC powder-less gloves. Water samples from the Desjardin Canal were collected from a small rubber raft. In all cases, samples were collected with no head-space to limit oxidation. Samples were returned to the laboratory (< 1 h) and placed in a N_2-filled (O_2 scrubbed) glovebag. Aliquots of each sample were transferred to 250 mL teflon® bottles for metal analysis. Sub-samples for metal analysis were then further processed in the HEPA filtered cleanhood, while sub-samples for sulfide and thiol analyses were processed in the N_2-filled glovebag. In each case, sub-samples were filtered using 25 mm Millipore 0.45 μm polysulfone syringe filters along with Millipore 10 kDalton Ultrafree®-15 centrifugal filters. Filters were prepared by rinsing twice with 1% TMA HNO_3 acid and Milli-Q water in the HEPA enclosure.

Sediment core samples were collected in Hamilton Harbour, Lake Ontario, on August 20, 1997 at three stations with coordinates: N4792473.8 E592328.5 (A), N4792258.8 E591758.0 (B) and N4792996.5 E594001.8 (C). Water depth at these locations was between 15 and 20 m. Sediment from Station A was sandy compared to Station B and C which were predominantly silty. Core samples were obtained by pushing 10cm x 50cm polypropylene tubes into a box core collected from a barge. Tubes were back-filled with interfacial water and sealed immediately. Two cores were collected from each of the box cores taken at the three stations. Sediments were extruded from the tubes in 10cm increments using a pneumatic extruder. Sediment sub-samples were placed in 125mL HDPE bottles, cleaned using the trace metals protocol, and back-filled with argon (>99.99%). Sub-samples were centrifuged (IEC, MA, USA) at 3000 rpm (~2000 g) for 20 min. Porewaters were processed for metal, sulfide and thiols analysis as described for the surface water and wastewater effluent samples. No unfiltered samples were analyzed for the porewaters due to its high particulate content.

For metal analysis, 10 mL of the unfiltered, 0.45 μm and 10 kDa filtered sub-samples were stored in HDPE 15 mL centrifuge tubes and acidified with 200 μL Ultrex II HNO_3 acid (J.T. Baker, NJ, USA). Unfiltered sub-samples were double-bagged in zip-lock polyethylene bags and heated at 60°C for 24 hours. Immediately prior to analysis, samples were acidified with an additional 200 μL 2N HNO_3 containing 4 μg/L In as an internal standard. Ultra-trace metals analyses were performed on a PE-Sciex Elan-500 ICP-MS dedicated to low-level samples for periods of two weeks. Since our ICP-MS facility is used for a variety of sample types, we have found it necessary to thoroughly clean the instrument prior to analysis. By following strict protocols, we are able to eliminate contamination and hysteresis problems which commonly affect ultratrace level ICP-MS. Table 1 summarizes ICP-MS operating conditions. Instrumental and procedural blanks and metal standards were placed randomly within a run as part of our quality assurance program. Sample concentrations were determined based on ultrapure calibration standards (PlasmaChem, NJ, USA).

Sulfide concentrations were determined colorimetrically [21]. The colorimetric reagent was made by mixing 2.25 g N,N-dimethyl-p-phenylenediamine oxalate (Sigma) in 660 mL H_2SO_4 and 340 mL Milli-Q and 5.4 g $FeCl_3 \cdot 6H_2O$ (BDH) in 100 mL HCl and 100 mL Milli-Q. 25 mL of the unfiltered, 0.45 μm and 10 kDa filtered sub-samples were added to amber glass bottles, which were sealed with telfon®-coated silicone septa. 2 mL of the colorimetric reagent was added to each bottle and responses were measured one hour later using a Spectronic 20 (Bauch & Lomb) at 670 nm. Calibration standards were made from a Na_2S solution standardized iodimetrically against a thiosulfate standard (Anachemia, Montreal, QC, Canada).

Thiols were determined by reverse phase high performance liquid chromatography (HPLC). Following the method outlined by Mopper and Delmas [22], samples were derivatized using an ortho-phthalaldehyde/2-aminoethanol reagent followed by HPLC and fluorometric detection. A Bio-Rad model 700 gradient module was used as the pumping system. Separation was carried out with a 250 x 4.6mm Vydac (218TP) C18 reverse-phase column containing 5μm packing. A Bio-Sil ODS-10 guard column was placed in front of the analytical column. Detection was accomplished with a Hewlett-Packard 1046A Fluorometric Detector with monochromators set to 350 nm (excitation) and 420 nm (emission). Chromatograms were collected on a Hewlett-Packard 3390A Integrator.

Mobile phases consisted of 0.05 M sodium acetate (pH 5.7) (phase A) and acetonitrile (phase B) degassed with helium (>99.999%). Reagents consisted of 20 mg of ortho-phthalaldehyde (Sigma Chemical) in 1 mL of acetonitrile and 25 μL of 2-aminoethanol (Sigma Chemical) in 1 mL of 0.1 M sodium borate. Both reagents were then diluted 100-fold in the same solvents. Stock solutions of thiol standards were prepared: glutathione, L-cysteine, 3-mercaptopropionic acid, 2-mercaptoethanol, thiolactic acid and ethanethiol. The stock solutions were prepared in 5 mM sodium acetate buffer (pH 4.0) degassed with helium and containing 0.1 mM EDTA. Mixed standards were prepared from stock solutions and diluted to 10-1000 nM with degassed Milli-Q water prior to derivatization. 25 μL of each reagent was added to 1 mL of standard or sample. Solutions (100 μL loop) were injected after precisely 1 min. 0.45 μm and 10 kDa samples were reacted and injected with no further treatment. Separations were performed at 25°C and at a flow rate of 1 mL min^{-1}. The gradient used was as follows: isocratic at 10% B for

1 min; 10% to 50% B in 10 min; isocratic at 50% B for 10 min; isocratic at 100% B for 5 min; 100% to 10% B in 1 min; and 10 min at 10% B.

Unfiltered porewater, wastewater and surface water samples were reacted with a 1 mM solution of $NaBH_4$ to liberate any thiol compounds bound to particulate matter through disulfide linkages. Samples were then filtered, and the pH was adjusted to near neutral with 0.1 N HCl. This method of reducing disulfide linkages was tested on a 1 μM solution of glutathione disulfide, which was fully recovered as glutathione. Other samples were reacted with 1 mM EDTA to liberate thiols bound by metals.

Temperature, pH (Ross combination electrode, Orion), conductivity (Perolin-Bird Archer, Cobourg, ON, Canada) and dissolved oxygen (Model 600, ESD, Newark, DE, USA) were measured at each sampling location. DOC (TOC-DIC) was determined by IR detection of CO_2 following persulfate-UV digestion on a Dorhmann-DC180 automated system (Table 2).

Results and Discussion

Quality Assurance for Silver Data

The instrumental detection limit (IDL) for ICPMS analysis of silver (3σ of 5 blank replicates) ranged from 0.01 to 0.05 nM under conditions outlined in Table 1 (laboratory control blanks consisted of Milli-Q water spiked with the equivalent Ultrex II HNO_3 and In solution used for samples). Field quality assurance for silver measurements included 10% procedural blanks, 25% sample replicates and 10% $AgNO_3$ spiked samples. Procedural blanks, carried to the field and processed concurrently with the samples, were not significantly different from the laboratory control blanks. Individual procedural blanks were always within the 95% confidence interval of the control blank. Precision (RSD) on replicated samples ranged between 7.5 and 15.1% for low-range samples (0.05-0.25 nM) and between 1.6 and 8.1% for higher-range samples (>0.25 nM). Samples from each sampling location were collected, filtered and spiked with $AgNO_3$ to determine recoveries. Spike recoveries are summarized in Table 3.

Quality Assurance for Sulfide and Thiol Measurements

IDL for the sulfide analyses was 1-2 nM (laboratory control blanks consisted of the colorimetric reagent added to degassed Milli-Q). Procedural blanks carried to the field and processed concurrently with the samples were not significantly different from control blanks. Also, field samples with no colorimetric reagent added were not significantly different from control blanks (i.e., indigenous material in the samples did not display absorbence at the measurement wavelength, 670 nm). Precision (RSD) on all sulfide measurements was better than 10%. Recoveries of Na_2S spikes from the three filter fractions were 97±15% (unfiltered), 96±8% (0.45 µm filtered) and 99±7% (10 kDa filtered). Additionally, a 50 nM sulfide solution (pH 8) was filtered through the 0.45 µm and 10 kDa filters. Recoveries were found to be excellent from the 0.45 µm filtration (100±5%), but recoveries from the 10 kDa centrifugal filtration tended to be low and quite variable (74±19%). Even with the special precautions taken in this study, the centrifugation step caused significant losses of sulfide.

IDL for the thiol analyses varied depending on the compound (glutathione and L-cysteine, 25 nM; 3-mercaptopropionic acid, 2-mercaptoethanol, thiolactic acid and ethanethiol, 1-2 nM) (laboratory control blanks consisted of the fluorometric reagent added to degassed Milli-Q). Procedural blanks were not significantly different from control blanks. To determine recovery efficiencies for the thiol analysis, 0.45 μm filtered samples were spiked with 50 nM 3-MPA (recoveries were 95±12%).

Silver in Treatment Plants

Although overall removal efficiencies for silver in municipal WTPs are generally greater than 95% [6] concentrations of silver in WTP effluents are elevated at least 1-2 orders of magnitude relative to background levels. In this study, we focused on the small fraction of silver which is not removed during primary treatment. Silver was measured at three locations in the Dundas plant. Grab samples were taken at the inflow and outflow of the secondary clarifier and at the outflow from the plant after chlorine addition and sand filtration. Silver concentrations measured in the plant are summarized in Table 4.

Particulate-bound (>0.45 μm) silver concentrations in the plant averaged 0.8-1 nM. A small decrease in silver concentrations was observed between the inflow to the secondary clarifier and the outflow from the plant, although, the proportion of particulate silver remained constant (Table 4). The 20% removal observed between the secondary clarifier and the effluent leaving the plant is presumably due to particle settling in the secondary clarifier and entrapment of particulate matter during sand filtration. The proportion of particle-bound silver in the plant outflow averaged 54±5%, which compares favourably with values obtained by Shafer *et al.* (60±16%) who examined silver partitioning in a number of WTPs located in Wisconsin, USA [6].

Colloidally-bound (10 kDa-0.45 μm) silver showed a 30% percent decrease between the secondary clarifier inflow and secondary clarifier outflow, suggesting scavenging by larger particles or aggregation of colloidal particles. No significant change in the colloidally-bound silver was observed between the secondary clarifier outflow and the plant outflow. Colloids containing silver do not appear to undergo further aggregation or sorption after leaving the secondary clarifier and do not appear to be removed by the sand filter.

Ultrafiltration of samples from the three sample locations within the plant showed that dissolved (< 10 kDa) silver concentrations remained constant (Table 4). Dissolved silver does not appear to be affected by aggregation or sorption processes in the final stages of treatment. In a concurrent study, we found high affinity sites for silver in the < 10kDa filter fraction of the secondary clarifier ([23], this volume). Complexation of silver in the <10 kDa filter fraction explains its lack of reactivity towards particulate matter, which have previously been shown to have large partition coefficients, K_d [4,6].

While ultrafiltration studies conducted by Shafer *et al.* [6] found only 2% of total silver passes through the 10 kDa membrane, we found significantly higher proportions of dissolved silver (16-30%) in plant effluent. We found an average 0.28±0.04 nM silver in the <10 kDa filter fraction compared to only 0.03 nM measured by Shafer *et al.* [6]. A single set of samples from the Burlington plant also showed a higher proportion of silver in the < 10 kDa filter fraction (15%). During the April 1998 trip to the Dundas plant, we compared the 10 kDa filtered concentrations determined using the Millipore Ultrafree-15® centrifugal filters and separations carried out on a Millipore cross-flow membrane system. Both methods gave similar results. In a study by Pham and Garnier [24], river water samples were tagged with radioisotopes of several different metals, including silver. The authors found that for the <0.45 μm size fractions, up to 25% of the silver was in the <10 kDa fraction (although some samples contained no <10 kDa silver).

Silver Downstream from the the Dundas Treatment Plant

The fate of silver leaving the Dundas plant was examined by sampling at three locations downstream from the plant in the Desjardin Canal (plant effluent contributes 100% of the baseflow in the Canal). Grab samples, taken a few centrimetres below the water surface, were collected five metres downstream from the submerged plant outflow pipe, midway along the Canal (250 m downstream) and immediately upstream of where the Canal discharges to Cootes Paradise marsh (500 m downstream). At the midway location, bottom water was also collected at a depth of 3 m through 0.25″ O.D. teflon® tubing into a teflon® bottle using a 50cc HDPP syringe outfitted with a two-way valve. Silver concentrations measured downstream of the Dundas plant are summarized in Table 5.

Particulate-bound silver (0.8±0.4 nM) was quite variable at the plant outfall, displaying extremes not captured during sampling trips to the treatment plant. Particulate-bound silver concentrations decreased by approximately 40% between the outfall and the second sampling location 250 m downstream and a further 10% by 500 m downstream (Table 5). Since the effluent from the Dundas plant represents the only major input into the Desjardin Canal, losses of silver in the particulate phase must be due to non-conservative processes, such as particle settling, sorption and uptake by biota. In contrast, concentrations of colloidally-bound (10 kDa - 0.45 µm) and dissolved silver (<10 kDa) remained relatively constant over all three sampling locations. Silver in these filter fractions appears unaffected by aggregation or sorption processes over the length of the Canal (residence time for water in the Canal is approximately 48 hours). As with the <10 kDa filter fraction in the treatment plant, this uniformity in the silver concentration suggests that the silver in these fractions must be associated with strong complexants. Furthermore, the concentrations of these complexants must remain relatively constant in the different filter fractions. Otherwise, changes in the distribution of complexants would be mirrored in the distribution of silver amongst the different filter fractions. As will be shown in the next section, inorganic sulfide concentrations do remain relatively constant over the length of the Canal (Table 7).

Sulfide and Thiols in Wastewater Effluents and Surface Waters

Sulfide concentrations determined by the Methylene Blue colorimetric method include the sulfide species: S^{2-}, HS^- and $H_2S(aq)$ (HS^- is the predominant species at the pH of these waters). However, the addition of the acidic reagent solution to the sample decreases the pH and causes a variety of colloidal metal sulfides to decompose, allowing liberated sulfide to react with the colorimetric reagents. Amorphous FeS reacts quantitatively under these conditions ($FeS \rightarrow Fe + \sum S$), as does ZnS and CuS, although at different rates ($CuS < FeS \sim ZnS$). Polysulfides, S_n^{2-}, also react to yield one equivalent S(II). Thiol compounds, on the other-hand, do not decompose under these conditions and can not be measured by this technique [25].

Measurable sulfide concentrations were present in both the wastewater effluent samples, as well as the fully oxic surface water samples (Table 6 and 7). Particulate sulfide concentrations in the Dundas WTP decreased from the secondary clarifier inflow (270 nM) to the plant outflow (230 nM). There was a dramatic drop between the plant outflow and the outfall in the Canal (145 nM). No change was observed between the outfall and 500 m downstream. Sulfide concentrations in the colloidal phase (10 kDa - 0.45 μm) decreased from 200 nM at the secondary clarifier inflow to 150 nM at the plant outfall and decreased from 130 nM at the outfall to 115 nM

500 m downstream. In contrast, dissolved sulfide (<10 kDa) concentrations remained remarkably constant (60-70 nM). As observed with the distribution of silver amongst the different size fractions, the distribution of inorganic sulfide also remained relatively constant (Tables 6 and 7). Simpson [26] showed that several metal sulfides can be stable under oxic conditions for periods of several hours (CdS, CuS, PbS and ZnS), while FeS and MnS oxidize extremely rapidly under the same conditions. The oxidation rate of free sulfide (HS^-) was found to be slower than that of FeS and MnS, but still fast in comparison to the other metal sulfides examined. Sulfide measured in this study is likely in the form of these more stable metal sulfides. Measurements of copper and zinc concentrations in the Plant and in the Canal indicated that there is always an excess of these metals over measured sulfide concentrations.

Thiol compounds were not measurable in any of the wastewater or surface water samples we examined. Reactions of unfiltered samples with the equivalent of 0.1 mM $NaBH_4$ to liberate thiols bound through disulfide linkages and with the equivalent of 0.1 mM EDTA to liberate thiols bound to metals also did not produce any thiol peaks in the chromatograms. This finding suggests that concentrations of simple thiols of the kinds used as standards in this study are at least 100 times lower than inorganic sulfide concentrations.

Silver, Sulfide and Thiols in Porewaters

Silver concentrations were not detectable (<0.01 nM) in the porewaters from sampling location B and C. Silver was also not detectable in interfacial water samples collected from location B and C. However at location A, low concentrations of silver in the 10 kDa - 0.45 µm fraction (but not the <10 kDa filter fraction) were measured in both the porewater and interfacial water (Figure 1). Sulfide concentrations were higher than measured in the wastewater and surface water samples. Inorganic sulfide concentrations in the 10 kDa - 0.45 μm filter fraction ranged from 350 to 1000 nM and in the < 10 kDa filter fraction ranged from 50 to 150 nM. Two small peaks, corresponding to 3-mercaptopropionic acid and mercaptoethanol, were present in several of the chromatograms from location A. Mean concentrations for the 10-20 cm and 20-30 cm depth intervals were 9.1 and 15.5 and 7.0 and 10.4 nM, respectively. Reactions with $NaBH_4$ and EDTA on unfiltered porewaters did not yield any increase in thiol concentrations. Thiol concentrations in freshwater porewaters are considerably lower than in marine and estuarine porewaters, where thiol concentrations up to 10 μM have been reported [14].

Acid-volatile sulfide (AVS) measurements on sediments from location A (intervals 0-10 cm and 40-50 cm) gave average values of 54.4 and 32.8 μmoles/g, respectively (unpublished work). The relatively large amounts of AVS in these sediments and low silver concentrations in the porewater suggests that silver is effectively scavenged by the sulfides present in the sediment.

Silver Speciation

Our investigation of wastewater effluents, surface waters and porewaters has demonstrated the ubiquity of inorganic sulfide at nano-molar levels even in fully oxic waters. Under these conditions, sulfide concentrations will always be in excess of silver, present at ultra-trace concentrations (<1 nM). This is shown by a plot of total silver versus total inorganic sulfide for all the samples collected during this study (Figure 2). Since silver forms a very strong sulfide complex compared to other metals ($Hg^{2+} > Ag^{+} > Cu^{+} > Pb^{2+} > Zn^{2+}$), it is reasonable to assume that even if sulfide determined by the Methylene Blue method is in the form of metal sulfides, silver should outcompete other metals for the available sulfide. Since sulfide was measurable in all three size fractions we examined, this suggests that silver in these different size fractions is likely in the form of silver-sulfide complexes. However, colloidal metal sulfides are probably entrapped in complex networks of organic material and silver is probably a coprecipitate or surface complex in these complex aggregates. This is evidenced by the correlation between DOC (Table 2), particulate sulfide (Table 6 and 7) and particulate silver (Table 4 and 5). Shafer et al. [6] also observed a correlation between silver and DOC in the effluents of several Wisconsin WTPs. To explain this correlation, the authors suggested that silver was complexed by DOC. However, sulfide to silver ratios of 100-1000 at the Burlington and Dundas WTPs indicate that sulfide is responsible for complexing silver instead.

Our investigation of thiols in wastewater, surface water and porewater suggests that this form of sulfur(-II) is probably not an important species for complexation of metals in freshwaters. Inorganic sulfide concentrations are in too large an excess for thiol compounds to be of importance. However, we have not yet investigated the abundance of sulfhydryl sites in higher molecular weight substances (i.e., humics and fulvics). It should also be noted that thiol compounds, namely glutathione and metal chelating polypeptides (metallothioneins and phytochelatins) likely play an important role in regulating silver and other metal concentrations in living organisms [27]. Although silver sulfide complexes possess large log K values, silver is quite labile between sulfur(-II) species [28,29]. It is possible, therefore, to envision transfer of silver to biota via an exchange between an inorganic sulfide species in the aqueous phase and thiolic sites in the organism.

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Table 1. ICP-MS Operating Conditions

R.F. power (W)	1200
Argon plasma gas (L/min)	12
Argon aux gas (L/min)	2
Argon neb gas (L/min)	1.3
operating vacuum (torr)	$1x10^{-5}$
cones	nickel
nebulizer	Cetac 5100AT ultrasonic
sample uptake (mL/min)	1-1.5
quad mode	peak hop, 200ms dwell, 3 points/peak
acquisition	90s, 5 cycles, 10 replicates
rinse	300s, 2% HNO_3
internal standard	4µg/L In

Table 2. Characteristics of Wastewater, Surface Water and Porewater

	Temp (°C)	pH	Specific conductance (µS/cm)	Oxygen (mg/L) (%sat)	DOC (mg/L)
Secondary clarifier inflow (Dundas)	23.9	7.5	828	8.8 (105)	14.3
Secondary clarifier outflow (Dundas)	24	7.5	838	9 (107)	10.7
Plant outflow (Dundas)	24.2	7.5	835	9.1 (108)	10.4
Plant outfall (Desjardin Canal)	23.2	7.5	856	8.9 (105)	7.1
250 m downstream (Desjardin Canal)	22.5	7.3	795	8.4 (98)	6.5
250 m downstream (bottom water) (Desjardin Canal)	22.5	7.2	812	6.5 (76)	6.8
500 m downstream (Desjardin Canal)	23.4	7.4	742	8.9 (105)	6.1

Data collected on 05/06/97.

Table 3. Spike Recoveries (%)[a]

	Unfiltered	0.45 μm filtered	10 kDa filtered
Dundas WTP	92±9	94±5	102±8
Desjardin Canal	94±11	99±4	105±13
Hamilton Harbour Porewater	97±8	95±10	96±14

a. 0.5 nM $AgNO_3$ spikes

Table 4. Silver Concentrations in the WTP's (nM)

	Particulate (>0.45 μm)		Colloidal (10 kDa - 0.45 μm)		Dissolved (<10 kDa)	
Secondary clarifier inflow (Dundas)	0.95±0.13	1.12/0.80	0.57±0.09	0.69/0.47	0.32±0.07	0.42/0.25
Secondary clarifier outflow (Dundas)	0.79±0.16	0.97/0.57	0.41±0.03	0.44/0.36	0.29±0.06	0.38/0.23
Plant outflow (Dundas)	0.78±0.10	0.89/0.65	0.39±0.01	0.41/0.38	0.28±0.04	0.32/0.23
Plant outflow (Burlington)	1.03	---	0.5	---	0.26	---

mean values ±1 standard deviation; hi/low values; data compiled from 3 sampling trips to the Dundas plant (1997-98).

Table 5. Silver Concentrations Downstream from the Dundas WTP (nM)

	Particulate (>0.45 μm)		Colloidal (10 kDa - 0.45 μm)		Dissolved (<10 kDa)	
Plant Outfall	0.84±0.38	1.54/0.39	0.42±0.04	0.47/0.36	0.25±0.09	0.40/0.14
250 m downstream	0.51±0.06	0.60/0.40	0.42±0.07	0.49/0.32	0.29±0.10	0.45/0.17
250 m downstream (bottom water)	0.42±0.07	0.51/0.34	0.35±0.07	0.44/0.34	0.27±0.09	0.38/0.15
500 m downstream	0.45±0.10	0.56/0.29	0.40±0.04	0.48/0.28	0.31±0.07	0.45/0.25

mean values ±1 standard deviation; hi/low values; data compiled from 5 sampling trips to the Desjardin Canal (1997-98).

Table 6. Inorganic Sulfide Concentrations in the WTP's (nM)

	Particulate (>0.45 μm)		Colloidal (10 kDa - 0.45 μm)		Dissolved (<10 kDa)	
Secondary clarifier inflow (Dundas)	268±36	306/220	195±32	240/165	61±5	65/54
Secondary clarifier outflow (Dundas)	250±47	290/184	170±25	204/144	69±4	75/65
Plant outflow (Dundas)	230±37	260/178	153±20	180/132	70±22	98/45
Plant outflow (Burlington)	240	---	170	---	110	---

mean values ±1 standard deviation; hi/low values; data compiled from 3 sampling trips to the Dundas plant (1997-98).

Table 7. Inorganic Sulfide Concentrations Downstream from the Dundas WTP (nM)

	Particulate (>0.45 μm)		Colloidal (10 kDa - 0.45 μm)		Dissolved (<10 kDa)	
Plant Outfall	145±45	205/102	130±41	205/96	72±17	102/54
250 m downstream	155±59	258/102	121±49	213/84	65±14	90/49
250 m downstream (bottom water)	163±46	243/126	130±45	213/96	76±18	112/64
500 m downstream	137±46	213/90	115±38	184/82	69±20	90/49

mean values ±1 standard deviation; hi/low values; data compiled from 5 sampling trips to the Desjardin Canal (1997-98).

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Figure 1. Silver and inorganic sulfide porewater concentrations (10 kDa - 0.45 μm filter fraction) for Hamilton Harbour location A. Error bars denote ±1 standard error.

Figure 2. Total sulfide versus total silver concentrations for Dundas WTP (squares) and Desjardin Canal (circles), and 10 kDa - 0.45 μm filter fractions for Hamilton Harbour location A (triangles).

Figure 1

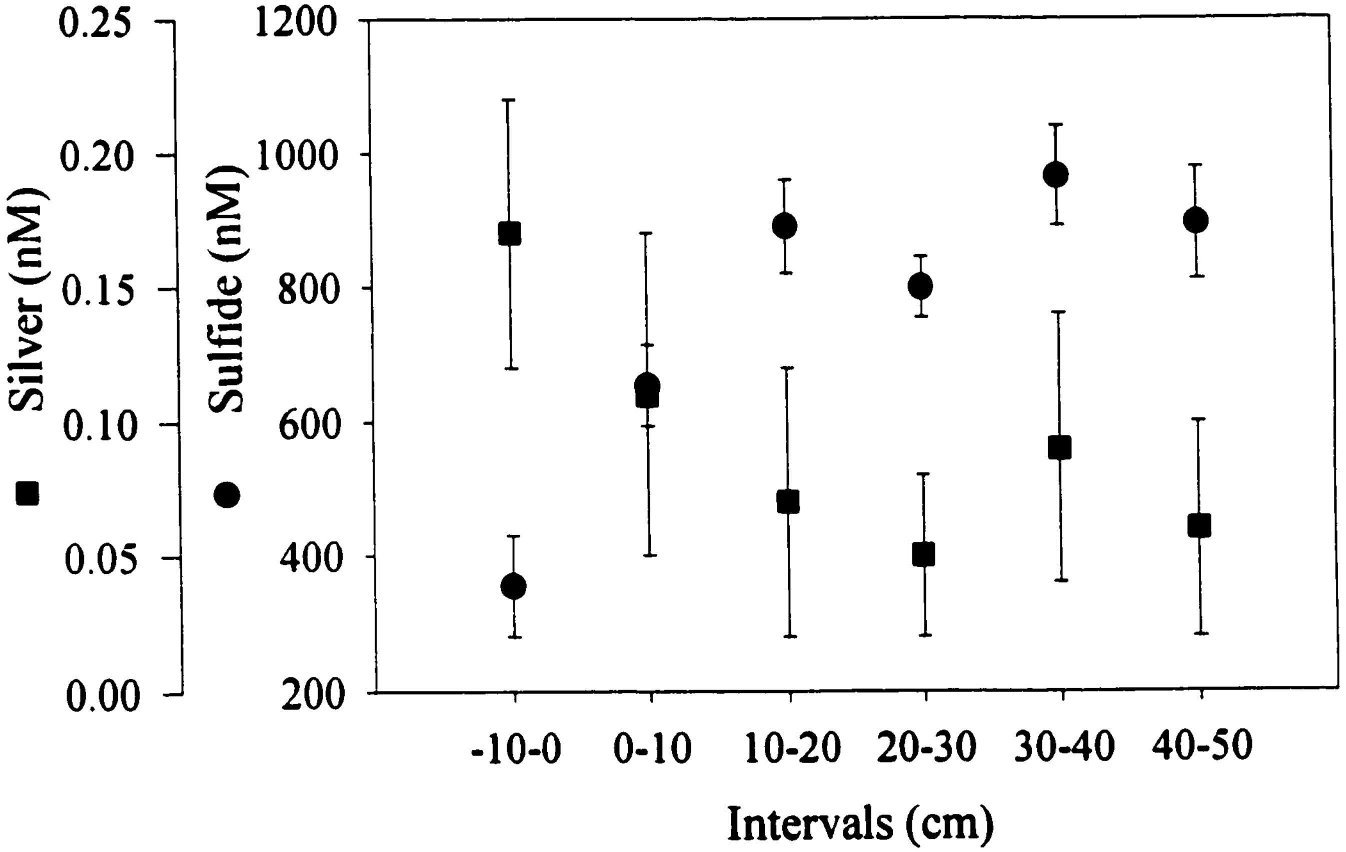
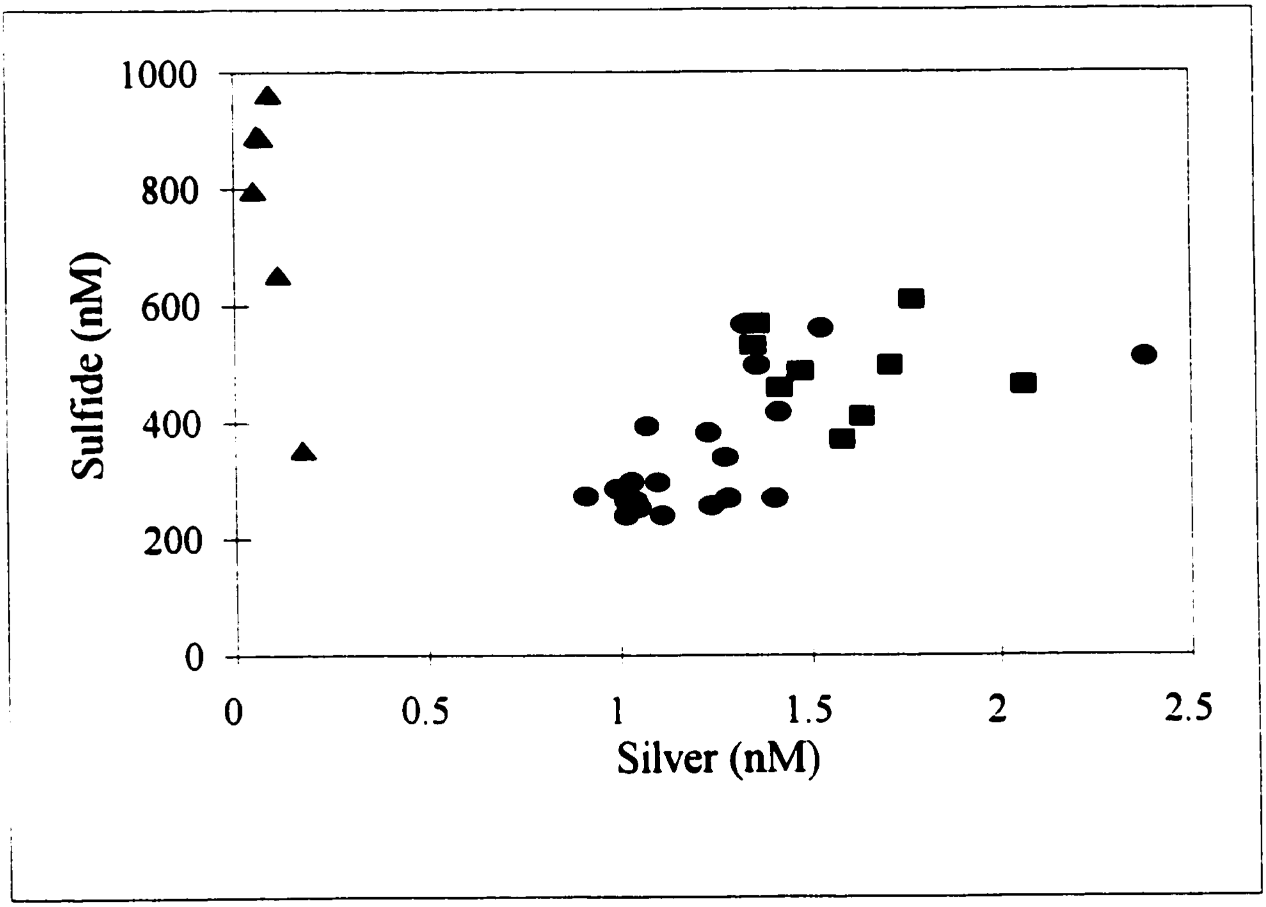

0.25
0.20
0.15
0.10
0.05
0.00
■ Silver (nM)
● Sulfide (nM)
1200
1000
800
600
400
200
-10-0
0-10
10-20
20-30
30-40
40-50
Intervals (cm)

Figure 2



IV

Determination of Silver Speciation in Wastewater and Receiving Waters by Competitive Ligand Equilibration/Solvent Extraction

(Accepted to *Environ. Toxicol. Chem.* February 10, 1999)

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Running Header: Silver speciation in wastewater and receiving waters by competitive ligand equilibration

Abstract

A competitive ligand equilibration/solvent extraction (CLE/SE) technique was used to examine silver complexation in the particulate (> 0.45 μm), colloidal (<0.45 μm) and dissolved (< 10 KDalton) phases of wastewater effluent and receiving waters. Additions of silver at near ambient levels (0.1-10 nM), followed by CLE/SE, allowed the determination of complexation sites with large stability constants (log K_{AgL} = 11-12), responsible for silver binding. Good agreement between these constants and formation constants of well-characterized silver-sulfide complexes suggests that silver in wastewater effluents and receiving waters is likely complexed to sulfur(-II). This is supported by previously collected data on silver and sulfide concentrations in these waters, which showed that inorganic sulfide concentrations are 200-300 times in excess of silver concentrations. Organic ligands appear to be insignificant in comparison to the contribution to silver binding by inorganic ligands. These inorganic ligands probably consist of metal sulfides, which are stable for hours to days in oxic waters. When complexed to these strong affinity ligands, silver(I) is protected from photo-reduction to zero-valent silver.

Keywords: Silver, sulfide, speciation, complexation constants, competitive ligand equilibration, wastewater treatment

Introduction

Speciation involves identification of the various chemical forms of an element within a system. In natural systems, metal species can include the free hydrated ion, inorganic complexes, organic complexes and particulate forms associated with colloids and suspended particles. In marine waters, thermodynamic data for silver(I) complexes suggest that inorganic chloride and sulfide complexes are probably the predominant silver(I) species [1-3]. However, in freshwater, with less chloride, inorganic sulfide and thiols may be important to silver(I) speciation. Previous work, including the preceding paper appearing in this issue, has demonstrated that inorganic sulfides can be persistent even in fully oxic surface waters [4,5]. Colloidal metal sulfides, shown to be stable for periods of hours in oxygenated waters [6], could represent potential complexants for silver in these waters. This would fit with recent studies showing strong partitioning of silver towards the colloidal and particulate fraction [7,8]. Complexation constants for silver complexes with organic ligands containing oxygen and nitrogen functional groups are relatively small ($\log K < 6$) [9]. However, thiol ligands (mercaptans) have similar complexation constants to inorganic sulfides ($\log K \approx 13$) [10,11] and could also be an important complexant for silver(I). Previously, we examined the abundance of low-molecular weight thiol compounds in freshwater (including wastewater effluent and sediment porewaters), and concluded that concentrations of low-molecular weight thiol compounds were insignificant compared to inorganic sulfide concentrations measured in the same waters. However, higher-molecular weight substances, containing thiolic groups, may be present at greater concentrations. Thiolic groups in higher-molecular weight substances would likely be less susceptible to oxidation and therefore available to complex silver.

In order to provide direct evidence of silver complexation in wastewater and receiving waters, we used the competitive ligand equilibration-solvent extraction (CLE/SE) approach developed by Miller and Bruland [3] to determine complexant concentrations and complexation constants for a municipal wastewater treatment plant (WTP), Dundas, Ontario. We evaluated complexation in different size fractions: unfiltered, <0.45 µm filtered and <10 kDa filtered. The findings of this study were assessed in relation to data collected on silver, sulfide and thiol concentrations in these waters [4]. Further studies were conducted on the <0.45 µm size fraction using the CLE/SE technique to examine how exposure to oxygen and light affects silver complexation in these waters. As well, the contribution of inorganic and organic ligands to silver binding was assessed through comparison of UV irradiated and non-irradiated samples.

Theory

Diethyldithiocarbamate (DDC^-) and chloroform are added to the sample in order to establish a competition for silver with the natural ligands. After a new equilibrium is established, the chloroform fraction containing the silver-DDC complex is separated from the aqueous phase. The amount of silver in the chloroform fraction is determined by inductively-coupled plasma mass spectrometry (ICPMS) after back-extraction into nitric acid. Silver complexation constants and natural ligand concentrations can then be estimated from the total silver concentration, $[Ag_T]$, and the silver extracted into the chloroform, $[Ag_{chl}]$.

Competitive ligand equilibrations are conducted on sample aliquots spiked with increasing amounts of silver to generate a titration curve of total silver, $[Ag_T]$, versus silver removed from the aqueous phase, $[Ag^*]$. $[Ag^*]$ normalizes $[Ag_{chl}]$ to the volume of the sample aliquot, allowing direct comparison to the total concentration ($[Ag^{\bullet}] = [Ag_{chl}]V_{chl}/V_{aq}$). The resulting titration plot is curved, concave upward, if strong (relative to DDC^-) natural ligands are present in the sample.

This kind of competitive ligand equilibration approach allows the determination of the total ligand concentrations, $[L'_{T,i}]$, and the conditional complexation constants for their silver complexes, $K'_{AgL,i}$, for all hydrophilic natural ligands which do not extract into the chloroform fraction. This will include organic ligands (e.g., thiols), inorganic ligands (e.g., HS^-), or colloids (e.g., metal sulfides). It is assumed that DDC is selective for silver and undergoes minimal interactions with other metals present in the natural water samples investigated in this study. The competitive ligand equilibration approach is most effective when the strength of the competitive ligand (α_{AgDDC}) matches the strength of the natural ligands (α_{AgL}), where

$$\alpha_{AgDDC} = ([Ag^{\bullet}] + [AgDDC])/[Ag^+] = K_{AgDDC}[DDC^-] \qquad (1)$$

and

$$\alpha_{AgL} = \sum[AgL_i]/[Ag^+] = \sum K'_{AgL,i}[L'_i] \qquad (2)$$

AgL_i are the silver complexes with the natural ligands and $[L'_i]$ are the concentrations of natural ligands not compexed to silver. Miller and Bruland [3] have shown that in general it is possible to estimate $[L'_{T,i}]$ and $K'_{AgL,i}$ if α_{AgDDC} is within an order of magnitude to either side of α_{AgL}. Our previous research has focused on the importance of sulfide ligands to silver speciation in freshwater. Based on measured sulfide concentrations in our samples [4] and previously measured silver-sulfide complexation constants [10], we designed our experiments with an α_{AgDDC} value of approximately 10^4.

In order to estimate $[L'_{T,i}]$ and $K'_{AgL,i}$, $[Ag^+]$ and $\sum[AgL_i]$ have to be calculated for each titration point. To accomplish this, we make use of the slope of the titration curve at high silver addition points, where we assume that the natural ligands capable of competing with the DDC^- are fully titrated. In the absence of natural ligands, the speciation is given by

$$[Ag_T] = [Ag^+] + [AgX_i] + [AgDDC] + [Ag^*] \tag{3a}$$

in which AgX_i are inorganic silver complexes (other than HS^-). This can be written in terms of side reaction coefficients

$$[Ag_T]/[Ag^+] = 1 + \alpha_{Ag'} + \alpha_{AgDDC} + \alpha_{Ag^*} \tag{3b}$$

where

$$\alpha_{Ag'} = [AgX_i]/[Ag^+] \tag{4a}$$

$$\alpha_{Ag^*} = [Ag^*]/[Ag^+] \tag{4b}$$

Because $\alpha_{Ag'}$ is negligible in freshwater, this term was omitted in further equations. Rearranging equation 3b and substituting $[Ag^*]/\alpha_{Ag^*}$ for $[Ag^+]$ gives

$$[Ag^*] = \alpha_{Ag^*}[Ag_T]/[1 + \alpha_{AgDDC} + \alpha_{Ag^*}] \tag{5}$$

The slope of the plot of $[Ag^*]$ versus $[Ag_T]$ at high metal additions where the natural ligands are fully titrated is therefore

$$S = \alpha_{Ag^*}/(1 + \alpha_{AgDDC} + \alpha_{Ag^*}) \tag{6}$$

In the presence of natural ligands, the speciation is given by

$$[Ag_T] = [Ag^+] + [AgDDC] + [Ag^*] + \sum[AgL_i] \tag{7}$$

which can be solved for $\sum[AgL_i]$ using α-coefficients to give

$$\sum[AgL_i] = [Ag_T] - [Ag^*](1 + \alpha_{AgDDC} + \alpha_{Ag^*})/\alpha_{Ag^*} \tag{8a}$$

or

$$\sum[AgL_i] = [Ag_T] - [Ag^*]/S \tag{8b}$$

To solve for $[Ag^+]$, equation 4 is rearranged to get

$$[Ag^+] = [Ag^\bullet]/\alpha_{Ag^\bullet} \qquad (9)$$

and rearranging equation 6 and substituting gives

$$[Ag^+] = [Ag^\bullet](1-S)/S(1 + \alpha_{AgDDC}) \qquad (10)$$

α_{AgDDC} is calculated from equation 1, where K_{AgDDC} was determined independently by a potentiometric titration using a Ag_2S electrode ($\log K_{AgDDC} = 9.1 \pm 0.1$) [10] and $[DDC^-]$ is closely approximated by $[DDC_T]$, since $[Ag_T] << [DDC_T]$. However, this is only valid when there are no significant side reactions with major cations for DDC at the pH of the samples.

In this study, we used nonlinear fitting of one- and two-ligand models (equation 11) to the $\sum[AgL_i]$ and $[Ag^+]$ titration data to estimate $[L'_{T,i}]$ and $K'_{AgL,i}$ values. The non-linear fitting routine was programmed using Matlab®, using the objective function: $\sum([\sum AgL_i]_{exp} - [\sum AgL_i]_{calc})/[\sum AgL_i]_{exp}$. If no improvement in the fit was observed using a two- ligand model, then we conclude that a one ligand model adequately describes the titration data.

$$\sum[AgL_i] = \sum(K'_{AgL,i}L'_{T,i}[Ag^+]/(1 + K'_{AgL,i}[Ag^+])) \qquad (11)$$

Uncertainty in the estimated parameters was determined by conducting Monte Carlo simulations [10]. The Monte Carlo method incorporates both measurement errors and uncertainties associated with model fitting into an overall estimate of the uncertainty in the parameters. Two-thousand pseudo data-sets were generated by adding random-normal errors to $[Ag^\bullet]$ which was first calculated at each original $[Ag_T]$ titration point using the best-fit parameter values (S, K'_{AgL}, and $[L'_T]$). A relative standard deviation of 5-15%, determined from the residuals of the fitted models, was used to generate the random-normal errors. Data-sets were then fitted using values of K_{AgDDC} and S randomly sampled from their gaussian distributions. The standard deviation of 0.1 log units was used for K_{AgDDC} and the standard deviation for S was estimated experimentally. Figures 2b and 2c show examples of distributions of K'_{AgL} and L'_T values generated by this method.

Methods

Samples were collected immediately upstream of the secondary clairifier outflow at the Dundas WTP and at a site 250 m downstream of the plant in the Desjardin Canal into 4 L LDPE bottles using clean sampling and handling techniques. Further details about these samples are given in the preceding paper appearing in this issue [4]. Samples were returned immediately to the laboratory (<1 hour) for processing. 1.2 L of unfiltered sample was decanted into a 2 L teflon® bottle. The remaining 3 L was subjected to sequential cross-flow membrane filtration (Pellicon, Millipore) to produce a 0.45 µm and 10 kDa filtrate (1.2 L each). Prior to filtration, the membranes were thoroughly acid-cleaned with several litres of 1% nitric acid (Baker, TMA grade), followed by several litres of Milli-Q (Millipore) water. Table 1 gives the sampling date, pH and total silver concentration for each sample. Total silver analyses were carried out directly on the filtrates and after acidification and heating at 60°C for 24 hours for unfiltered samples.

Teflon® separatory and reagent bottles were acid-cleaned prior to use. HPLC grade chloroform (Sigma) was further purified by extracting once with 1% Ultrex II nitric acid (Baker, NJ, USA) and twice with Milli-Q water. Diethyldithiocarbamate (DDC^-) solutions were prepared daily by dissolving the sodium salt (Sigma) in deoxygenated Milli-Q water (degassed for 1 hour with high-purity helium). Stock solutions of 3-Mercaptopropionic acid (3-MPA) (Sigma) were prepared in deoxygenated Milli-Q immediately before use. Solutions for the method verification experiments were made by spiking the 3-MPA stock solution into deoxygenated Milli-Q water buffered at pH 8.0 with $NaHCO_3$ (10^{-3} M). Silver stock solutions were prepared in Milli-Q water from the $AgNO_3$ salt (BDH, Toronto, ON, Canada) and stored in the dark.

Titrations were carried out in a darkroom under uv-filtered orange light. 100 mL aliquots of sample were added to the 125 mL separatory funnels, followed by the DDC^- and silver solutions. 10 mL of chloroform was added and the sample was shaken for 1 minute and then allowed to equilibrate for 3 hours, with occasional shaking. Longer equilibration times (up to 24 hours) were tested and produced similar results. The organic phase was drained into a 125 mL separatory funnel and extracted twice with 5 mL of 1.5 M Ultrex II nitric acid. The nitric acid phase was stored for ICPMS analysis in acid-cleaned 15 mL LDPE sample tubes. Silver analyses were carried out using the conditions and procedures outlined in the preceding paper in this issue. Silver calibration standards were made from an ultra-purity standard (PlasmaChem, NJ, USA) in the same concentration of nitric acid as the samples.

The quantity of natural hydrophobic silver complexes in each sample was determined by extracting aliquots without DDC^-, and the amount of silver in the organic fraction was subtracted from $[Ag^+]$. These concentrations were less than 0.01 nM even at high $[Ag_T]$. Procedural blanks averaged 0.041 nM. Blank values were determined by subjecting Milli-Q water to the extraction procedure, and were added to the known silver in the titration calculations. Total silver measured in the samples was also added to the known silver concentrations. The detection limit for silver extracted into the organic phase, Ag_{chl}, was 0.055 nM (based on 3σ for 7 procedural replicates).

Oxidation experiments were carried out on the <0.45 µm filter fraction from 250 m downstream of the plant. Samples were exposed to the air for periods of 24 and 48 hours. The

CLE/SE technique was then used to determine any differences in complexation as a result of oxidation. The effect of light on complexation was examined by exposing silver spiked aliquots of the same <0.45 μm filter fraction to fluorescent light for 12 hours, followed by CLE/SE. To assess the importance of silver complexation by organic ligands compared to inorganic ligands, we irradiated samples for 12 hours by submerging the UV lamp from a Dohrmann DC-180 Carbon Analyzer into the samples. The samples were then titrated with silver and analyzed by CLE/SE.

Results and Discussion

Model Ligand Titrations

In order to verify that the CLE/SE technique could give accurate estimates of the complexation of silver with sulfide ligands, we performed titrations with 3-Mercaptopropionic acid (3-MPA) in deoxygenated Milli-Q water. 3-MPA was chosen because we previously determined its complexation constant with silver by an independent potentiometric titration method using a Ag_2S electrode [10]. We also tested the extractability of the silver-3-MPA complex into chloroform and determined that it partitions strongly towards the aqueous phase. We tested two concentrations of 3-MPA: 1.0e-7 and 1.0e-6 M. For each concentration, two separate titrations were performed using different total silver concentrations. Since no systematic differences were observed between titrations, the data were combined for fitting purposes. The slope, S, from equation 6 was determined by linear regression at high silver additions where the ligand, 3-MPA, was fully titrated. An average slope value of 0.589 (± 0.012) was used in equations 8 and 10 to calculate $\sum[AgL_i]$ and $[Ag^+]$ for each titration point. Figure 1 shows the combined $[Ag_T]$ versus $[Ag^*]$ titration data for 1.0e-7 M. Figure 2a shows a plot of $[Ag^+]$ versus $\sum[AgL_i]$ for 3-MPA (1.0e-7 M). Titration points, where 3-MPA is fully saturated, were removed because of the large uncertainties resulting from the transformation of $[Ag_T]$ versus $[Ag^*]$ to $[Ag^+]$ versus $\sum[AgL_i]$. A one-ligand model was fit to the titration data with a modification to equation 11 to correct for protonation of the thiolic site (equation 12).

$$[AgL] = K'_{AgL}L'_T(K_a/([H^+]+K_a))[Ag^+]/(1 + (K_a/([H^+]+K_a))\,K'_{AgL}[Ag^+]) \qquad (12)$$

Experiments on 3-MPA were conducted at a pH of 8.0, and we used a previously determined value for acidity constant (pK_a) of 10.5 [10].

Fitted parameter values were log $K'_{AgL} = 12.1$ and $[L'_T] = 1.2e\text{-}7$ M. The uncertainties in these parameters, estimated by performing Monte Carlo simulations, are shown in Figures 2b and 2c. The log K'_{AgL} value is 0.1 log units from our previously determined value of 12.0 [10]. Titrations performed with 1.0e-6 M 3-MPA gave good estimates of the log K'_{AgL} and $[L'_T]$ values as well (see Table 2). From our titrations with the model ligand, 3-MPA, it appears that the CLE/SE method can be used to accurately determine log K'_{AgL} and $[L'_T]$ values.

Titrations of Treatment Plant and Receiving Waters

Unfiltered, < 0.45 μm filtered and <10 kDa filtered samples from the outflow of the secondary clairifier at the Dundas WTP and from 250 m downstream of the plant in the Desjardin Canal were examined for silver complexation using the CLE/SE technique. Additions of silver were chosen to adequately characterize complexation at near ambient concentrations (see Table 1). With additions of silver at near ambient concentrations, we can infer the speciation for the original silver in the samples. We found it was only necessary to use one analytical window

($\alpha_{AgDDC} \approx 10^4$), since the titration plots suggested no stronger ligand complexes at low silver additions (characterized by near-zero $[Ag^+]$ values), and we were not interested in weaker ligand complexes which would only be important at well above ambient silver concentrations.

The slope, S, for the natural samples displayed greater variability than for the model ligand titrations (0.57±0.03). Slopes determined for the unfiltered and <0.45 µm filtered samples from the plant were significantly lower than for the other samples and displayed large scatter in the titration data about the regression line ($r^2 = 0.67$). The decreased slopes suggest that these samples contain ligands which were not fully titrated. Alternatively, the high organic content of these samples could produce significant losses of the Ag-DDC complex through sorption. This would produce the lower observed slopes as well. Since the decreased slopes brings into question the reliability of the data for the unfiltered and <0.45 µm filtered WTP samples, we did not include it in any further discussion. Figure 3 shows plots of the log $[Ag^+]$ versus log [AgL] data for the unfiltered (a), < 0.45 µm filtered (b) and <10 kDa filtered (c) samples from 250 m downstream of the plant (04/10/98) and the <10 kDa filtered (d) sample from outflow of the secondary clairifier at the Dundas WTP (04/15/98). Log K'_{AgL} and $[L'_T]$ values for these samples along with samples collected on subsequent sampling dates are given in Table 2. We found no improvement in the overall fit using a two-ligand model (by comparing residual plots and χ^2/N values), and conclude that a one-ligand model adequately describes the complexation in these samples. Although there is no doubt that many different ligands complex silver in these samples, titrations can not be performed with sufficient precision and resolution to differentiate separate ligand classes. Instead, we obtain average complexation constants to describe silver binding in these samples. Figure 4 shows a plot of log K'_{AgL} versus ligand concentration, $[L'_T]$, for the WTP and downstream samples. Log K'_{AgL} values were different for the unfiltered, <0.45 μm filtered and <10kDa filtered fractions from the receiving water samples. Filtration removes a portion of the ligands, which results in a new average log K'_{AgL} value for the remaining ligands. For the 04/10/98 and 05/20/98 samples, average log K'_{AgL} values were lowest for the <0.45 μm filter fraction and highest for the <10 kDa filter fraction. However, in contrast to the samples taken earlier in the season, we found essentially no differences in the log K'_{AgL} values for the 06/10/98 sample. Ligand concentrations, $[L'_T]$, decreased 62-64% between the unfiltered and <0.45 μm filtered samples (for the three sampling dates), and decreased 58%, 76% and 96% between the <0.45 μm filtered and <10 kDa filtered samples. Ligand concentrations in the <10kDa samples from the 250 m downstream location were 16% (April, 1998) and 86% (June, 1998) lower relative to the WTP samples. This decrease may be due to a variety of processes, including aggregation, sorption or degradation. However, the difference between the locations could also be a result of temporal variability in the removal/formation of complexants in the treatment plant since samples at two locations were collected a week apart.

This study demonstrates that there is excess complexation capacity for silver by strong ligands (log K'_{AgL} ~ 11-12). The complexation capacity is 100-1000 times greater than ambient silver concentrations, and free silver ion concentrations should be extremely low under these conditions ($<10^{-13}$ M or 0.01 ng/L). The good agreement between silver complexation constants for the WTP and receiving water samples and complexation constants of known sulfur(-II) species (log K = 11.9-13.0) [9,10,12] suggests that silver in municipal wastewater effluents and receiving waters is complexed to sulfur(-II). Our previous examination of sulfur(-II) speciation in the Dundas WTP and Desjardin Canal [4] found mid- to high-nanomolar concentrations of inorganic sulfide. However, we found no evidence of organic sulfides (i.e., thiols), although the analytical method was limited to the identification of low-molecular weight thiol compounds. To further investigate the contribution of organic versus inorganic ligands to silver complexation, we applied the CLE/SE technique to UV-irradiated samples of the <0.45 μm filter fraction from the 250 m downstream sampling site. Only inorganic ligands should contribute to silver complexation in these UV-irradiated samples, since the organic material should be destroyed. We found only a very small decrease in the ligand concentrations for the UV-irradiated samples (2-4%) compared to non-irradiated samples. This decrease was insignificant when the uncertainties in the estimated ligand concentrations were taken into consideration. As well, log K'_{AgL} values remained the same for UV-irradiated samples compared to non-irradiated samples. From these findings, we conclude that organic ligands (i.e., thiols) do not appear to be important to silver complexation in these waters. Therefore inorganic sulfides are likely responsible for the silver complexation observed by the CLE/SE technique. However, the ligand concentrations determinated by the CLE-SE technique are approximately 70-90% lower than inorganic sulfide concentrations measured by the Methylene Blue colorimetric method [4], which measures $\sum S$ (HS^- and $H_2S(aq)$) and colloidal metal sulfides [13]. This suggests that not all the inorganic sulfide is available to bind with silver. The discrepancy may either be a kinetic effect, as some metal sulfides may not react during the equilibration time used in these experiments (3 hours) or, more likely, colloidal metal sulfide particles react with silver to form surface complexes, with a portion of sulfide remaining unreacted in the interior of the colloidal particle.

Oxidation Experiments

If sulfides are responsible for complexing silver, it is important to determine the stability of these sulfides under the oxic conditions of the WTP and receiving waters. We have previously established that the sample locations in the WTP and Desjardin Canal are oxygen saturated [4]. Under normal flow conditions, travel time between the WTP and the 250 m downstream sample location is approximately 48 hours. Simpson [6] has previously demonstrated that several metal sulfides can be stable under oxic conditions for periods of several hours (CdS, CuS, PbS and ZnS). It is unclear whether sulfides measured in the Canal are transported downstream from the WTP or whether their presence in the Canal is a result of sulfide production in the anoxic zone of the sediments, followed by diffusion of metal sulfides into the water column.

Samples of the <0.45 μm filter fraction from 250 m downstream of the Dundas WTP were exposed to the air for periods of 24 and 48 hours. Figure 5 compares plots of $[Ag^+]$ versus $[AgL]$ data for unexposed, 24 hour exposed and 48 hour exposed samples. Exposure to air for 24 hours resulted in a 66% decrease in the ligand concentrations, $[L'_T]$, over the unexposed samples. The 48 hour exposure resulted in an 88% decrease in $[L'_T]$ over unexposed samples. Log K'_{AgL} and $[L_T]$ values were: 11.0 and 2.18e-8 M (unexposed), 11.4 and 7.48e-9 M (24 hour exposure) and 11.1 and 2.74e-9 M (48 hour exposure).

Our previous study of the Dundas WTP showed that inorganic sulfide concentrations decreased 47% between the secondary clairifier and the 250 m downstream sampling location. This rate of sulfide loss is in keeping with the rate of oxidation for the air exposed samples in this study, which supports the possibility that metal sulfides are transported to the downstream sampling location from the treatment plant. However, further studies are required to elucidate the source of the sulfides to these waters.

Photoreduction

Photo reduction of Ag^+ to zero-valent silver may represent an important process affecting the transport and bioavailability of this metal in the aquatic environment. To examine the effect of light exposure on silver complexation in these waters, <0.45 μm samples from the 250 m downstream location were titrated with different $[Ag_T]$ and exposed to fluorescent laboratory light for a 12 hour period under argon. We found that for samples where $[Ag_T]$ was less than the $[L'_T]$ value determined for this sample, silver extracted by the addition of DDC was not significantly different from samples not exposed to light. However, for samples where $[Ag_T]$ was greater than the $[L'_T]$ value, extracted silver concentrations were lower than expected (see Figure 6). Presumably, the uncomplexed silver undergoes photo reduction to zero-valent silver, which is not extracted into the chloroform phase by the DDC. Silver complexed by the strong ligands observed in this study may be more mobile as a result of this reduced photo-activity.

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Table 1. Sample Locations and Characteristics

Date	pH	$[Ag_T]$ ± 1 s.d. (nM)
Secondary Clairifier Dundas WTP		
04/15/98	7.6	0.25±0.02 (<10 kDa)
06/03/98	7.4	0.22±0.02 (<10kDa)
250 m Downstream from Dundas WTP		
04/10/98	7.5	0.90±0.03 (>0.45 μm), 0.45±0.02 (<0.45 μm), 0.18±0.03 (<10 kDa)
05/20/98	7.3	0.95±0.05 (>0.45 μm), 0.39±0.03 (<0.45 μm), 0.15±0.02 (<10 kDa)
06/10/98	7.4	1.02±0.05 (>0.45 μm), 0.48±0.03 (<0.45 μm), 0.17±0.02 (<10 kDa)

Table 2. Fitted Parameter Values for Titrations[a]

3-Mercaptopropionic Acid

Sample	$\log K'_{AgL}$	$[L'_T]$ (nM)
1e-7 M	12.1±0.1	120±20
1e-6 M	12.2±0.2	980±70

Secondary Clairifier Dundas WTP

Sample	$\log K'_{AgL}$	$[L'_T]$ (nM)
04/15/98	11.8±0.2	8.1±0.5
06/03/98	11.6±0.2	9.0±0.6

250 m Downstream from Dundas WTP

	>0.45 µm		<0.45 µm		<10 kDa	
Sample	log K'_{AgL}	$[L'_T]$ (nM)	log K'_{AgL}	$[L'_T]$ (nM)	log K'_{AgL}	$[L'_T]$ (nM)
04/10/98	11.6±0.2	45±8	11.1±0.1	16±2	11.8±0.1	6.8±0.7
05/20/98	11.5±0.1	56±6	11.3±0.2	21±3	11.5±0.1	5.1±0.6
06/10/98	11.8±0.2	79±9	11.9±0.2	29±4	11.8±0.2	1.3±0.4

a. values reported ± 1 s.d.

Figure 1. Silver titration of 3-Mercaptopropionic acid (3-MPA) (1.0e-7 M) at pH 8. Solid line represents case where added dithiocarbamate out-competes 3-MPA.

Figure 2a. Log $\sum[AgL]$ versus log $[Ag^+]$ for 3-Mercaptopropionic acid (1.0e-7 M) at pH 8 (circles). Fitted one-ligand model (solid line).

Figure 2b. Distribution of log K'_{AgL} values for 3-Mercaptopropionic acid (1.0e-7 M) from Monte Carlo simulations. Best-fit value is indicated by a solid vertical line and one standard deviation about the best-fit value is indicated by dotted vertical lines.

Figure 2c. Distribution of $[L_T]$ values for 3-Mercaptopropionic acid (1.0e-7 M) from Monte Carlo simulations. Best-fit value is indicated by a solid vertical line and one standard deviation about the best-fit value is indicated by dotted vertical lines.

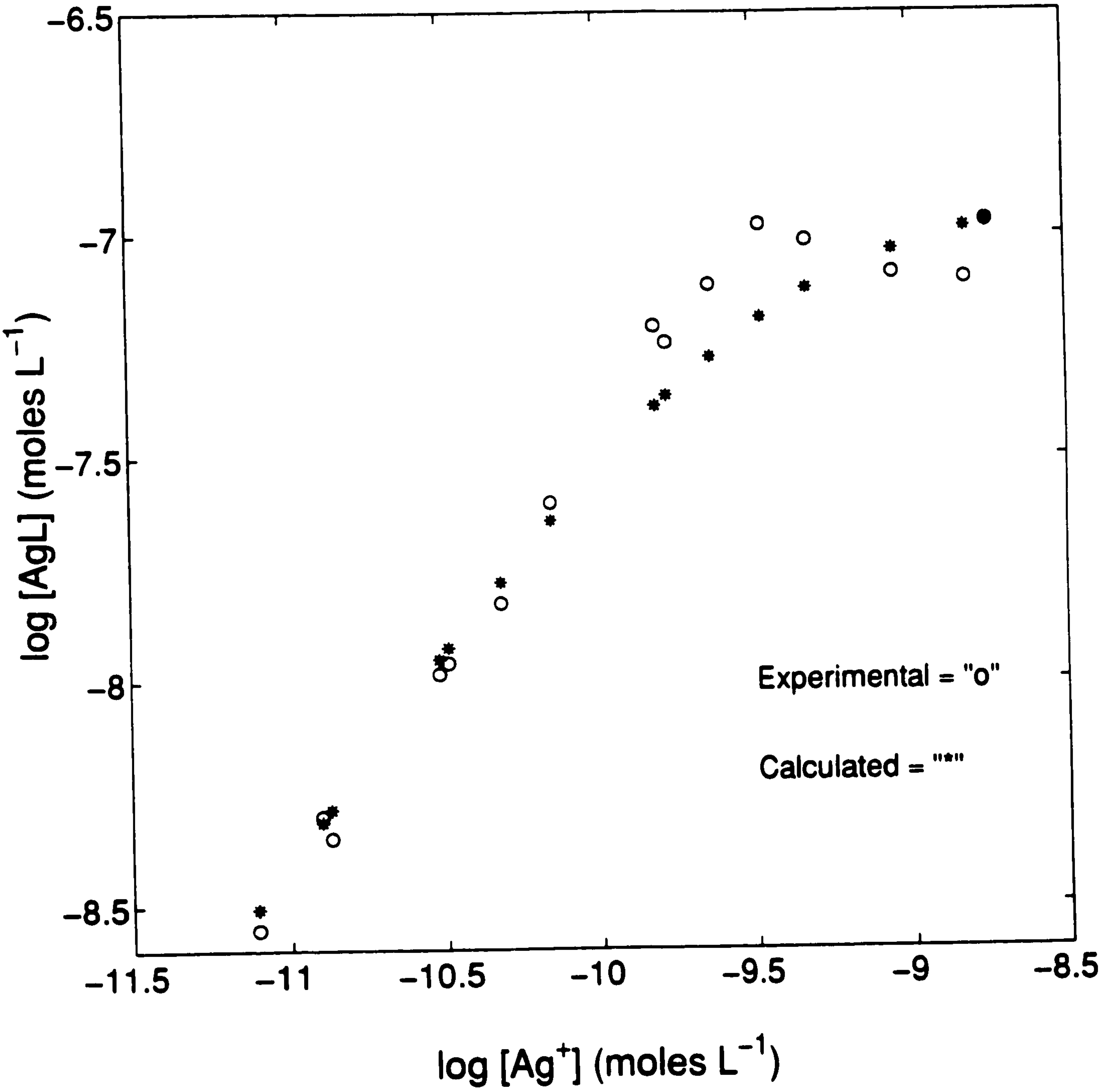
Figure 3. Log $\sum[AgL]$ versus log $[Ag^+]$ data for (a) unfiltered, (b) <0.45 μm filtered, (c) <10 kDa filtered samples from 250 m downstream of the plant (04/10/98) and (d) <10 kDa filtered sample from the outflow of the secondary clairifier at the Dundas WTP (04/15/98).

Figure 4. Silver complexation constants (log K'_{AgL}) and natural ligand concentrations, $[L'_T]$, for (a) unfiltered, (b) <0.45 μm filtered, (c) <10 kDa filtered samples from 250 m downstream of the plant (1- 04/10/98, 2- 05/20/98 and 3- 06/10/98) and (d) <10 kDa filtered sample from the outflow of the secondary clairifier at the Dundas WTP (1- 04/15/98 and 2- 06/03/98).

Figure 5. Log $\sum[AgL]$ versus log $[Ag^+]$ data for the <0.45 μm filter fraction from 250 m downstream of the Dundas plant. (a) Sample unexposed to the air, (b) 24 hour exposure and (c) 48 hour exposure.

Figure 6. Plot of $[Ag^+]$ versus $[Ag_T]/[L'_T]$ for <0.45 μm filter fractions (a) control (stored in the dark for 12 hours) and (b) exposed to light for a period of 12 hours. Error bars show 3 standard errors.

Figure 1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-7
-7.5
-8
-8.5
log [AgL] (moles L^{-1})
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-11
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-9.5
-9
-8.5
log [Ag^{+}] (moles L^{-1})
Experimental = "o"
Calculated = "*"

Figure 2a

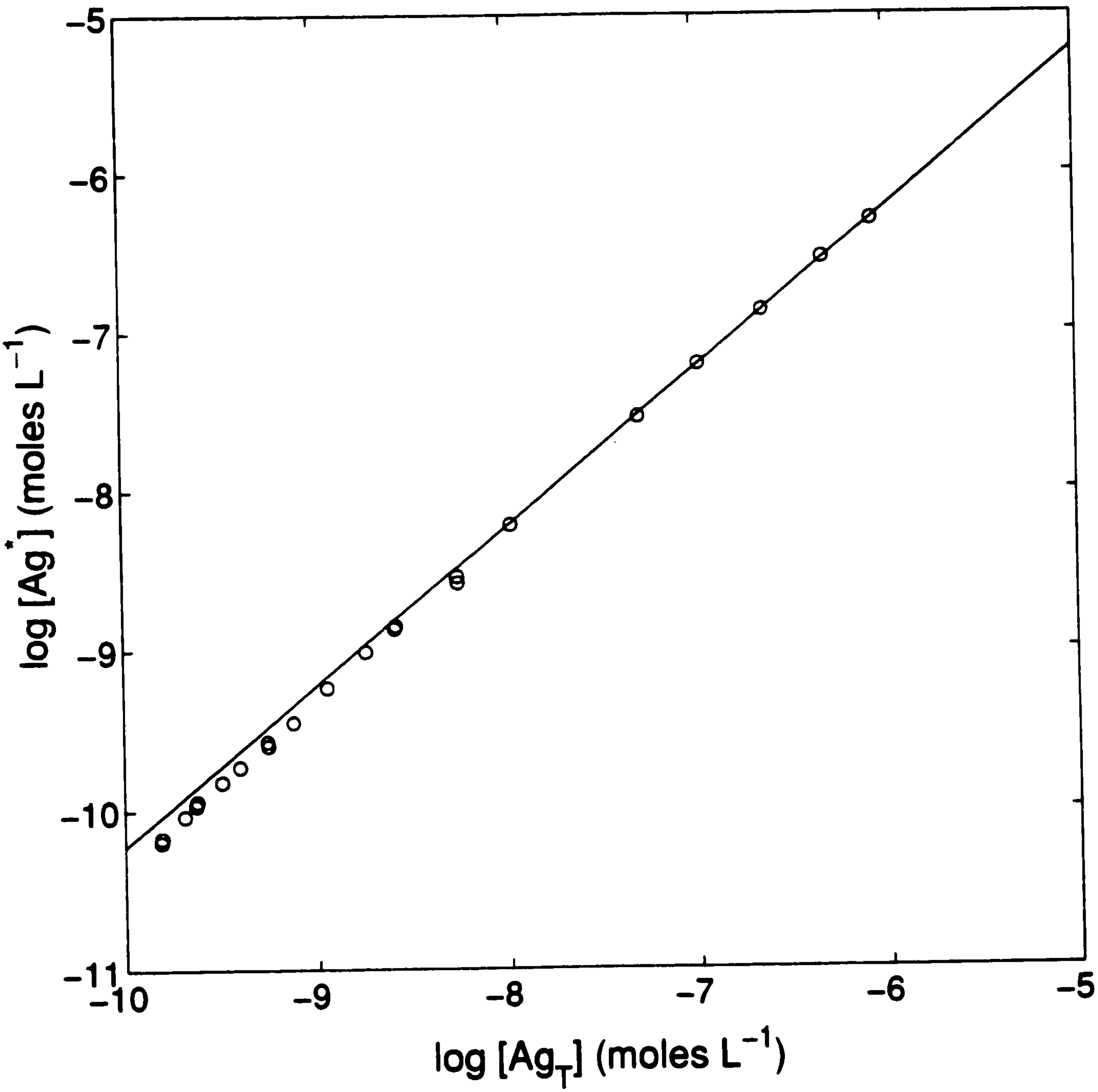
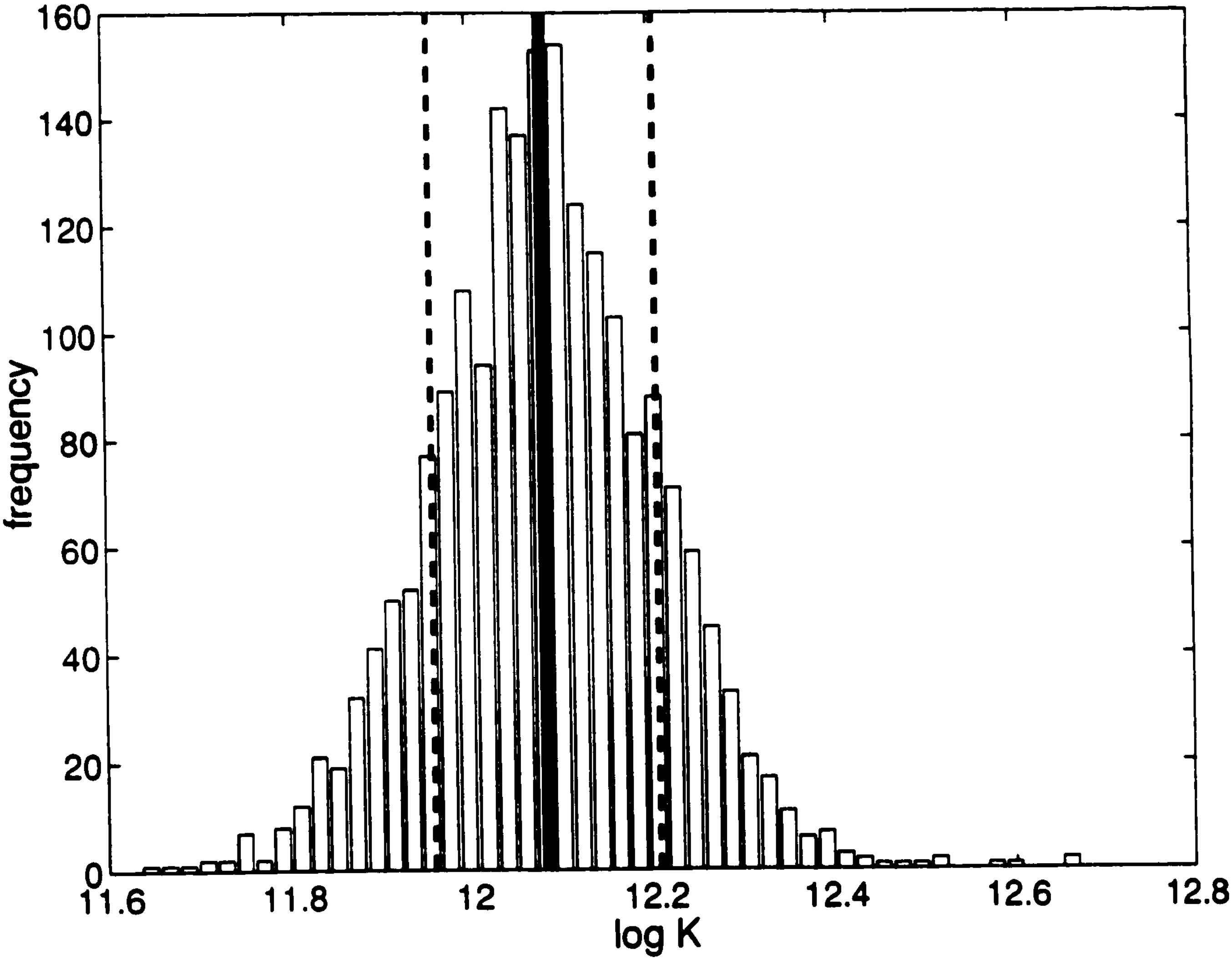


Figure 2b



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140
120
100
80
60
40
20
0
frequency
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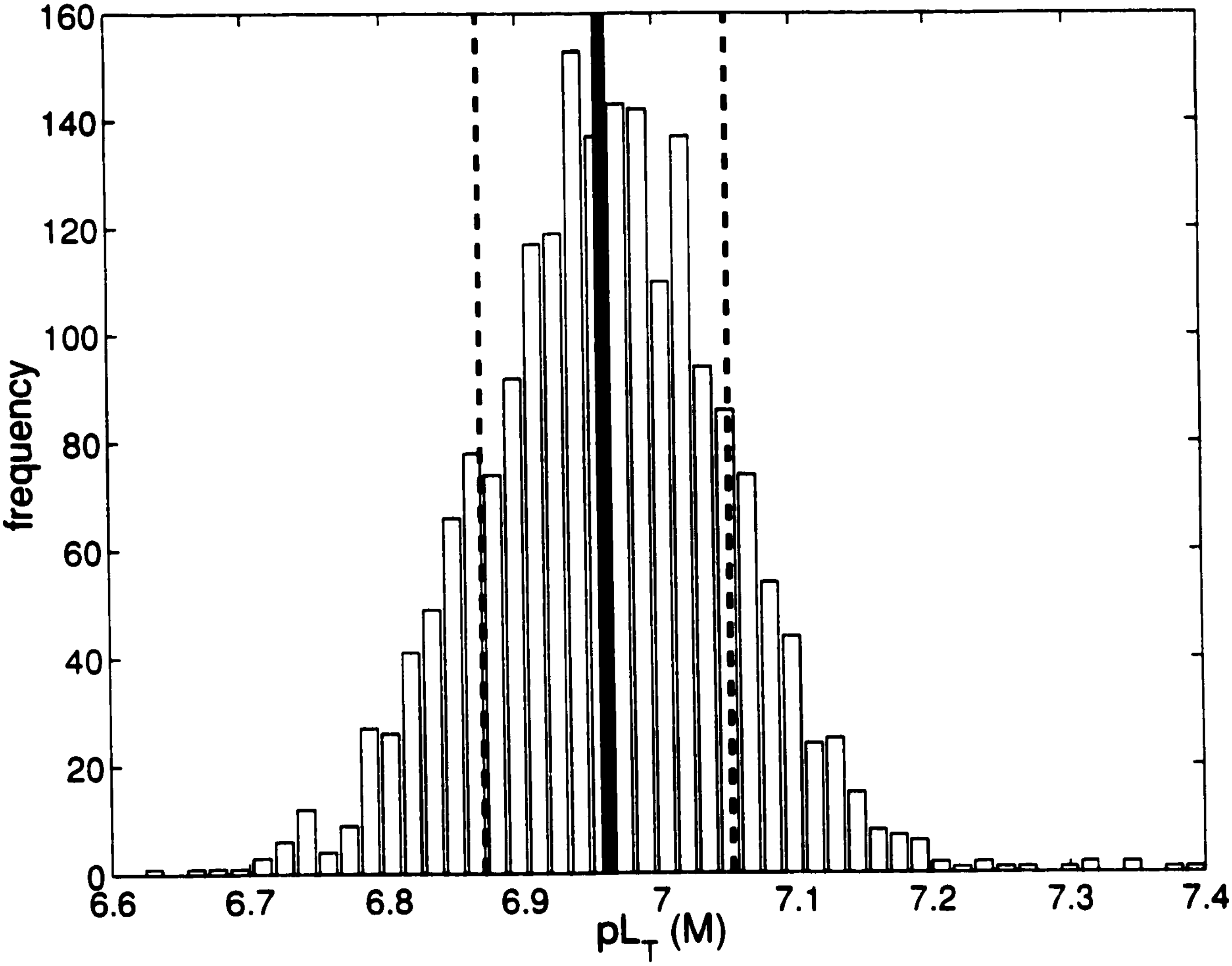
11.8
12
12.2
12.4
12.6
12.8
log K

Figure 2c



160
140
120
100
80
60
40
20
0
frequency
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6.8
6.9
7
7.1
7.2
7.3
7.4
pL_T (M)

Figure 3

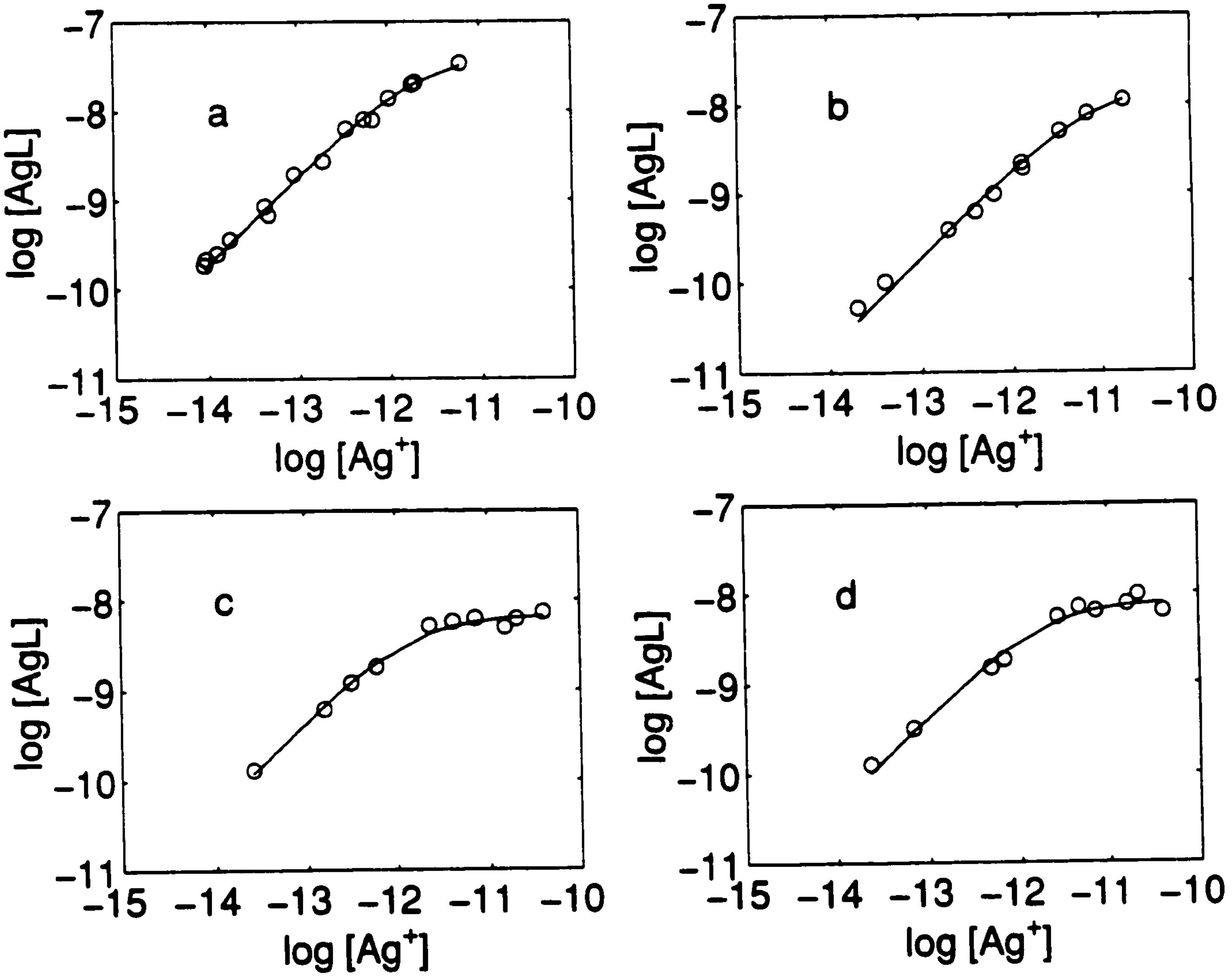
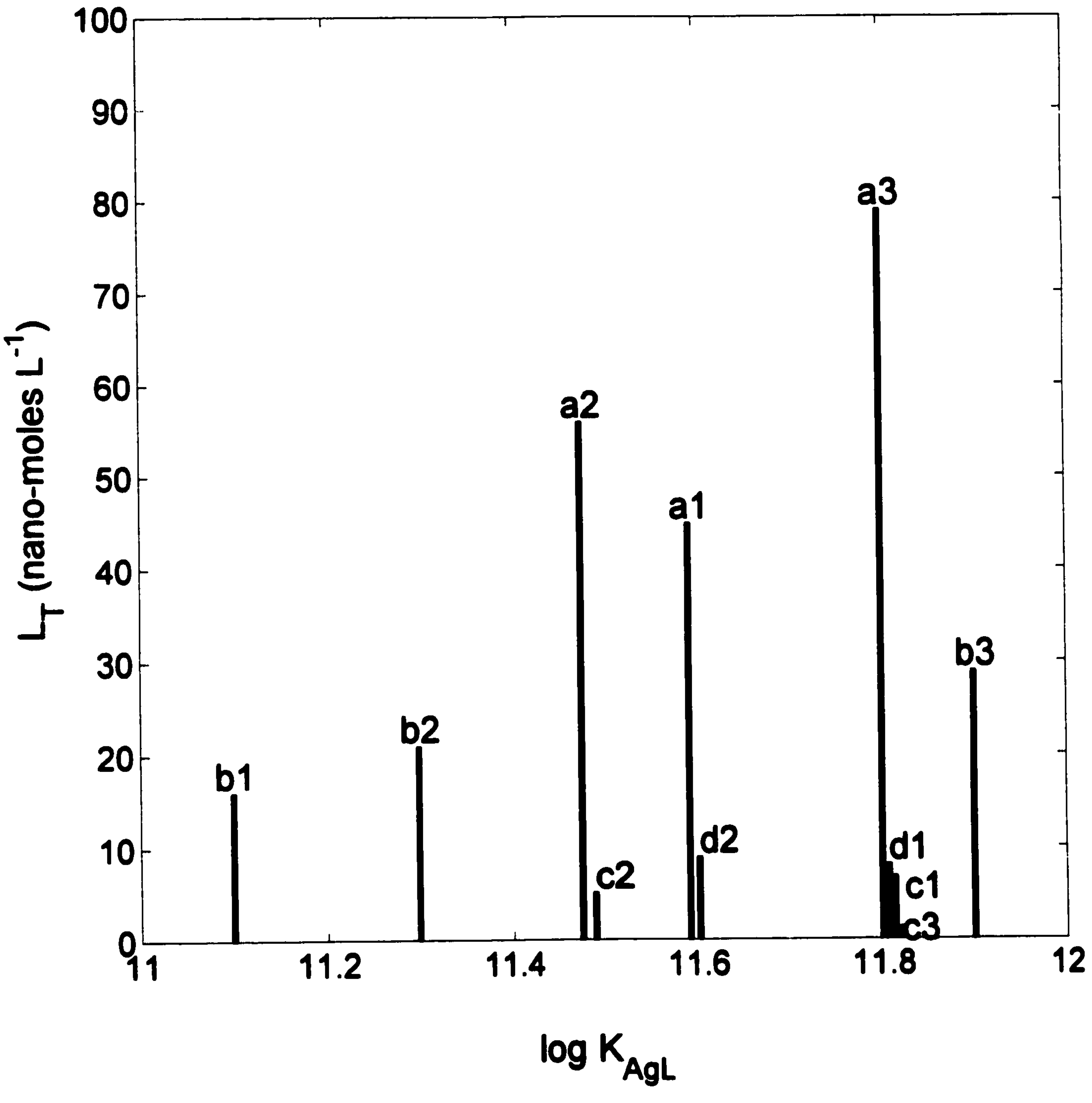


Figure 4



L_T (nano-moles L^{-1})
100
90
80
70
60
50
40
30
20
10
0
11
11.2
11.4
11.6
11.8
12
log K_{AgL}
a3
a2
a1
b3
b2
b1
d2
d1
c2
c1
c3

Figure 5

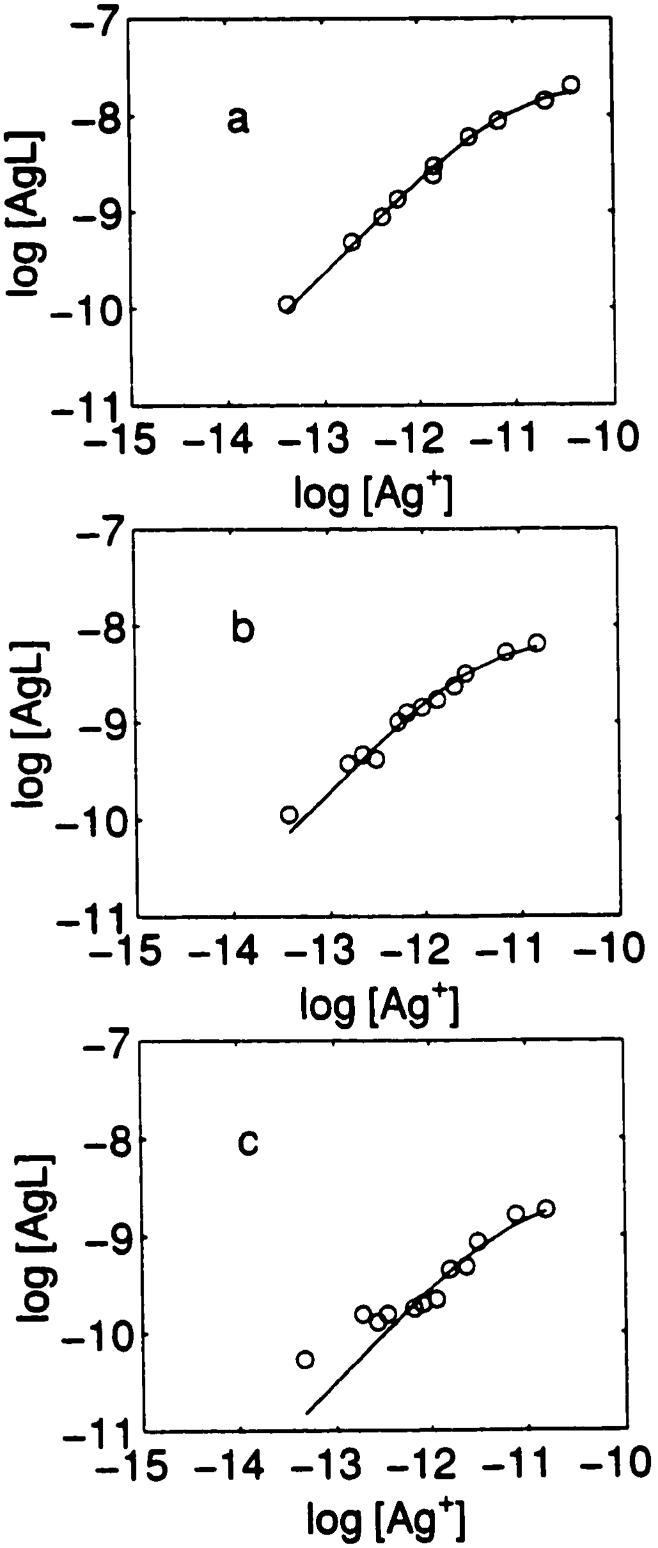
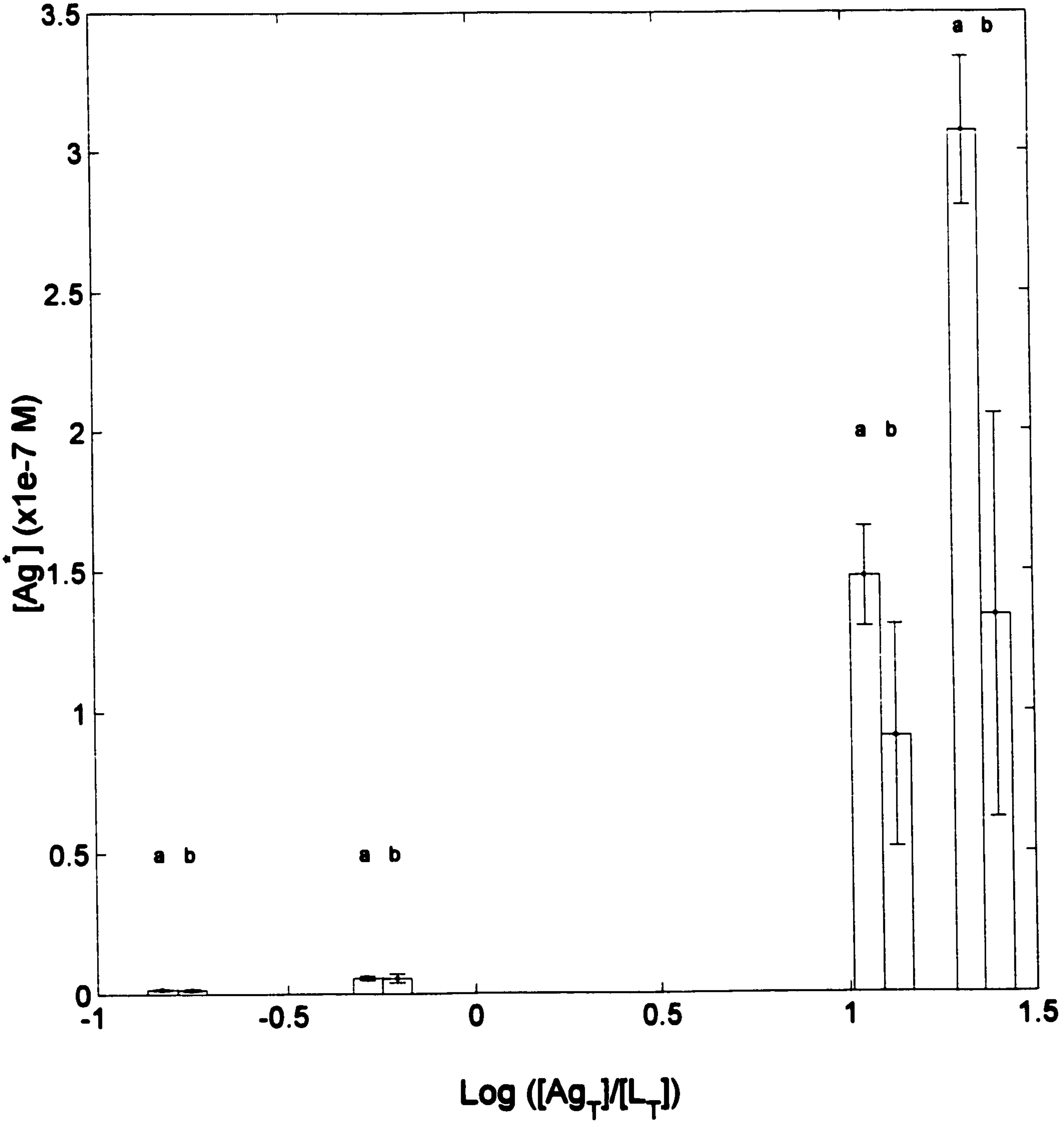


Figure 6



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Annual Review

SILVER AT AN OLD MINING CAMP, COBALT, ONTARIO, CANADA

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Abstract—Aqueous Ag(I) in different size fractions, <5 μm, <0.2 μm, and <10 kDa, was analyzed in a low dissolved organic carbon (DOC) and a high DOC environment. The sediment and tailings, contacting the water, were elevated in silver (10–100 mg/kg) from mining. The area is buffered at pH 6 to 8 by carbonate mineral gangue. Aqueous Ag(I) is present at nanogram per liter levels. Significant amounts of Ag are found in the colloidal (0.2 μm to 10 kDa) and are nondetectable (<0.3 ng/L) or a few nanograms per liter in the <10-kDa fraction. Acid-volatile sulfide (AVS) was found in most (oxic) water samples at the nanomolar level. Ag(I) does not accumulate in plants probably due to the strong binding of Ag(I−) with S(II−). One exception occurs for cattails, when the aqueous AVS concentration is less than the colloidal Ag(I) concentration.

Keywords—Silver   Acid-volatile sulfides   Mining

INTRODUCTION

Silver (Ag) is a trace metal that has widespread uses in society [1,2]. Elevated concentrations (10–100 ng/L) are found in urban surface waters compared with rural areas (<5 ng/L) [3]. Silver(I) (Ag(I)) is concentrated in the solids fractions of the sludge and sediment associated with municipal wastewater treatment plants (WWTPs) [4]. Rozan et al. [5] have also shown that industrial use of Ag during the past 100 years resulted in accumulation of Ag(I) in stream sediment in urban areas.

Silver is usually found in mine tailings and sediment associated with complex ores of copper (Cu), lead (Pb), and zinc (Zn) sulfides. Many of the mines producing Ag have been dormant from 10 to 100 years. Thus, the long-term dissolution and distribution of Ag in sediment and biomass can be determined from studying these mining sites.

Table 1 summarizes the few studies on the water concentrations of Ag(I) at mining locations. High analytical detection limits of most of the studies resulted in "less than" values, which make the results of little use. One paper, however, on a Pb mine in Wales [6], suggests that 0.45 μm filtered aqueous Ag concentrations associated with Ag-laden tailings were <100 ng/L and probably more nearly 10 ng/L. Even the data quoted in this paper may be high, by about 5 ng/L, due to the lack of use of trace metal free acid and the lack of reporting of a blank in their studies.

Many of the mine studies were at locations containing carbonate minerals that resulted in waters with pHs of 7 to 8. Studies in acidic drainage at the Liberty Pit, Nevada, USA [7], found 0.45 μm filtered aqueous Ag concentrations of 21,000 ng/L at a pH of 3.2 and in the Front Range Mineral Belt of Colorado, USA, of ≤200 ng/L at pH of 4.1. However, Ag was not detectable (<100 ng/L) in analyses of pH 2.8 to 3.1 water draining a massive sulfide deposit in Alaska [8]. The large values and large variations may be due to poor analytical detection limits and lack of proper quality control.

The mining camp at Cobalt, Ontario, Canada, was studied to determine the levels of Ag and its migration over time. Mining activities occurred over a span of 100 years, and they have been dormant for the past 15 to 30 years. The objectives of this study were to determine the aqueous concentrations and uptake in plant matter and to relate this information to equilibrium predictions. Tasks included determining the aqueous Ag concentrations in contact with the tailings and stream sediment, the colloidal distribution of Ag in aqueous samples, the accumulation of Ag in rooted vegetation growing on tailings, the association of the Ag to acid-volatile sulfide (AVS) ratio and plant uptake of Ag(I), and the accumulation of Ag in vegetables being grown on modified tailings. The study evaluated an organic sewage outfall system and an inorganic tailings outfall waterway.

Cobalt mining camp

The spectacular occurrences of Ag(0) and Ag(I) were discovered in 1903 at Cobalt, resulting in one of the major Ag mining operations in the world. Silver occurs in a complex ore mineralogy, consisting of arsenides and sulf-arsenides of cobalt (Co), nickel (Ni), and iron (Fe), native Ag and bismuth (Bi), and subordinate amounts of antimonides and sulfides of Pb, Zn and Cu. The gangue is high in calcite with lesser dolomite. There is supergene alteration of the deposit, which develops oxidized Fe-oxy-hydroxides, sulfates, and arsenates [9]. The ubiquitous occurrence of carbonate minerals as gangue results in a pH of 6 to 8 and an alkalinity of >2 mEq/L for waters throughout the area.

1903 to 1933, 1950 to 1960, and 1966 to 1983 represent the three major periods of exploitation of the deposit. Hand picking and other physical methods were used initially. Cyanide and mercury (Hg) amalgamation processes were started in 1911 at Chamber-Ferland and Crosswise Lake (see Fig. 1, study area 1). The later periods emphasized reworking of tailings using flotation and cyanide processing. The last period of exploitation resulted in 1.2 × 10^6 tonnes of tailings being deposited in Cart Lake (see Fig. 1, study area 2). Revegetation studies and a small commercial vegetable garden were also

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Table 1. Summary of concentrations of silver (Ag) at mining locations[a]

Location	Ag(I) concentration	Reference
Ag–lead mines, mid-Wales, UK; aqueous, <0.45 μm filtered aquatic bryophytes	3–200	[6]
Background	0.01–0.06	
Mine areas	0.1–18	
Keno Hill, Yukon, Canada; aqueous, pH 5–7	<1,000 dl	[19]
Front Range, Colorado, USA; aqueous	<200–2,500 dl	[20]
Alaska, mine effluents, USA;		[8]
Massive sulfides, pH 2.8–3.1	<100 dl	
Buffered sulfides, pH ~7	<100 dl	
Robinson District, NV, USA; aqueous, pH 3.2–7.6	21,000	[7]
Yerrington Pit, NV, USA; aqueous, pH 8.5	<10,000 dl	[7]
Getchell Mine, NV, USA; aqueous, pH 5.3–7.7	<5,000	[7]

[a] Water concentrations are in nanograms per liter; sediment and biomass concentrations are in milligrams per kilogram unless otherwise noted. dl = analysis limited by a high detection level.

Table 2. Analysis of silver (Ag) in Cart Lake tailings profile compared with average igneous rock[a]

Soil	Ag
Crust	70
A	17
E	32
B	2
C	13
Average igneous rock	0.1

[a] Concentrations are in milligrams per kilogram [10]. See text for description of soils.

developed at Cart Lake in 1979 and 1996, respectively. Minor reworking of the Chambers-Ferland tailings also occurred during this time period.

Figure 1 is a location map of the Cobalt study area, showing the drainage, the tailings areas, and the two study locations. Location 1 was studied to determine the interaction of Ag(I) in an organic matrix from the sewage outfall for the town of Cobalt with old tailings (Chambers-Ferland tailings of 1907–1930). Location 2 is Cart Lake, containing both older and newer mineral tailings. Cart Lake was nearly filled with tailings during operations from 1966 to 1983. Waters from the site 1 flow over well-vegetated tailings (Sasaginaga Creek) and connect to the outfall of the Cart Lake area at Farr Creek, which also flows through older tailings. Assessment of the Ag(I) geochemistry of water, soil, and plants at sites 1 and 2 and changes downstream are used to indicate ambient water concentrations and aqueous Ag(I) migration. In addition, seepage waters (<35 cm deep) in the tailings as well as surface and bottom waters of Cart Lake pond were sampled and analyzed to estimate near-surface Ag(I) migration. Site 1G acts as a reference for the site 1 samples because it is on older vegetated tailings without sewage flow.

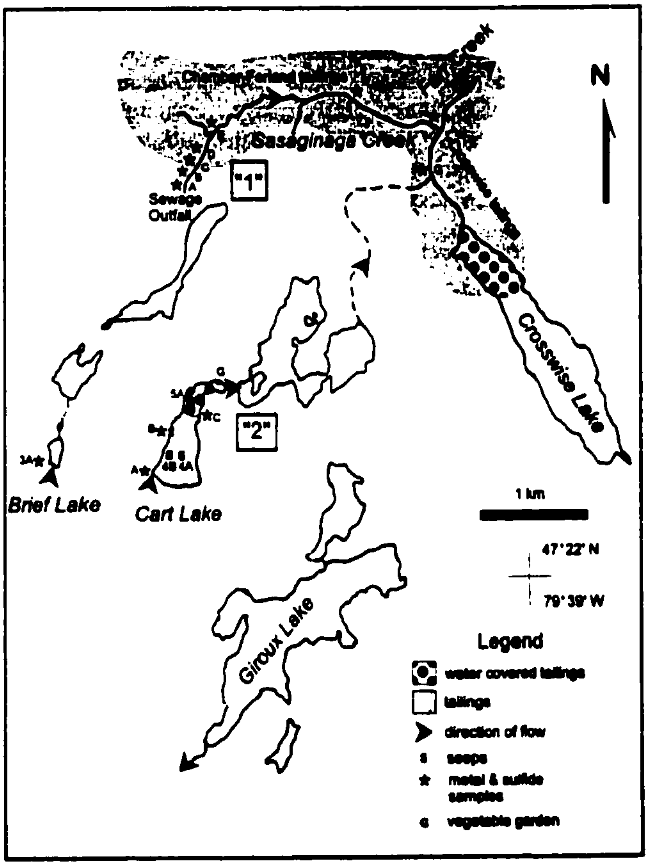

Fig. 1. Location map for Cobalt, Ontario, Canada, showing sites 1 and 2. Sampling locations pertain also to other tables and figures.

Dumaresq [10] studied the tailings at Cart Lake. The tailings are fine- to medium-grained sand size with high porosity and permeability. There are also finely laminated lenses of silt and fine sand. The tailings are desiccated except during rains, and they support only minimal vegetation without topsoil addition. Thus, runoff waters percolate through the upper 30 cm of the tailings and collect along intermittent Cart Lake Creek. Dumaresq described the tailings profile as

crusts:	homogeneous surface sandy porous crusts, about 2 cm thick
E unit:	about 7 cm of light gray, finely laminated silt to clay tailings
B unit:	about 10 cm of reddish brown oxidized tailings
C unit:	unmodified tailings

and in the vegetated area of Cart Lake tailings

A unit: about 7 cm of organic soil (added) above unit E

Table 2 is an analysis of silver in the Cart Lake tailings. Silver is elevated 100 to 1,000 times with respect to average igneous rock. In addition, arsenic (As), Co, Cu, Hg, Pb, and Zn are elevated 10 to hundreds of times in the tailings relative to average igneous rock [10]. There has been mobilization and removal of Ag from unit B because it is depleted about 10-fold compared with the other tailings units. This is true for the other trace metals also. Oxidation of metal sulfides (and formation of the rust) was proposed as the metal mobilization process.

METHODS

Sampling took place on July 5–7, 1996. Sampling occurred during seasonal base flow conditions. Four replicate samples were taken at sites 1C, 1G, 2C, 5A, and 5B spanning a period of 6 h at each location. Duplicate samples were taken on the

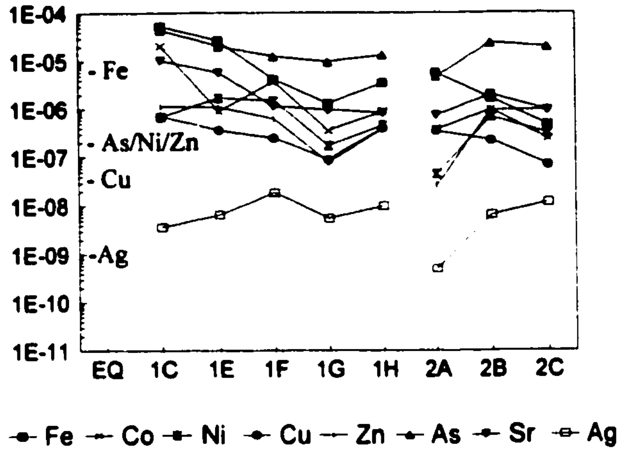

Fig. 2. Concentration of metals other than Ag(I) along sites 1 and 2. EQ refers to Canadian Water Quality Guidelines (http:www.ec.gc.ca/cwqg), mostly for freshwater aquatic life.

first and last days for seepage samples 4A and 4B. Blank and spiked samples were taken at all sample sites.

A reconnaissance study was carried out prior to the intensive sampling and analysis to better plan the program. Figure 2 shows the concentration of selected trace metals at some of sites 1 and 2. The concentrations of metals other than Ag are elevated, often meeting or exceeding environmental quality standards for As, Cu, and Zn. Ag(I), however, was much lower in concentration approaching 10^{-9} mol (100 ng/L) as determined semiquantitatively. This low concentration then precluded the use of ultraclean handling and low-detection techniques [11].

A rigorous sampling and analytical protocol was developed to obtain high-quality data. Soils, vegetation, and water samples were taken and analyzed both in the field laboratory (nearby motel and vehicle) and at the McMaster laboratories.

Water parameters considered were

conventional:	pH, alkalinity, specific conductivity, dissolved oxygen (DO), dissolved organic carbon (DOC), and acid functional group analysis of waters [12]
metal system:	Ag (I), acid-volatile sulfide (AVS)

Soil-vegetation parameters considered were

Ag:	concentrations in embedded soil and different portions of the plant (root, lower stem, upper stem, leaf, and fruit)

Both conventional and ultraclean sampling and analytical techniques were used. Conventional analyses were done at the field site (temperature, pH, specific conductivity, DO, and AVS, the latter within 8 h of collection) and at McMaster laboratories (DOC, alkalinity, and functional group analysis). Different sampling containers were assembled for each task. Blanks were carried to the field, processed, and analyzed along with the samples. Brief Lake (Fig. 1, location 3A) was selected as a background site because it was not within the drainage basin affected by mining.

Low-level ultraclean trace metals techniques were used to avoid contamination and to provide reliable data. Procedures for the preparation of sampling apparatus, processing of water samples, and analysis by inductively coupled plasma-mass spectrometry (ICP-MS) were adapted from protocols used by the University of Wisconsin Lake Michigan Tributary Monitoring Project (LMTMP) [11]. All surfaces contacting water samples were acid-cleaned in 20% ACS grade HNO_3 and 1% high-purity TMA HNO_3 (Baker Ultrex II, Phillipsburg, NJ, USA) over a period of 3 d. Materials were then rinsed with Milli-Q (Millipore, Toronto, ON, Canada) water and dried in a class 100 HEPA filtered laminar flow hood. All filter material was prepared by rinsing with 1% TMA HNO_3 and Milli-Q in the HEPA enclosure. Individual sampling kits containing a 60 cc polypropylene syringe, filters, and polypropylene 50-ml centrifuge tubes for sample storage were double bagged with zip-lock polyethylene bags.

Filter media consisted of Millipore 5-μm Durapore and 0.2-μm polysulfone syringe filters. A Millipore 10-kDa Ultrafree®-15 centrifugal filter was also used. We chose the 5-μm filter to separate out silt and coarse clay particles in the streams. We used the 0.2-μm filter to better isolate the colloidal fraction. We used the 10-kDa fraction, the smallest filter size that does not adsorb Ag(I), to operationally define the soluble portion. All filter media were presoaked in TMA 2% HNO_3 and then washed. Prior to the study, an intensive testing for filter and equipment sorption of $AgNO_3$ was carried out. We detected no removal of ionic Ag(I) at the 10 ng/L level for these filter media. It was impossible, however, to use 2- or 3-kDa filter media due to excessive loss of Ag(I) on the filter material.

Shallow ground water samples (25–35 cm) were obtained at Cart Lake by allowing seepage through a polyvinyl chloride cylinder containing Teflon® netting around an opening. The cylinders were pumped at intervals for 4 d prior to sampling to cleanse the system.

All water sampling was carried out using a hand pump that

Table 3. Operating conditions for the inductively coupled plasma-mass spectrometry

R. F. power (W)	1,200
Argon (Ar) plasma gas (L/min)	12
Ar auxiliary gas (L/min)	2
Nebulizer	Cetac 5100AT ultrasonic
Ar nebulizer gas ((L/min)	1.3
Operating vacuum (torr)	1×10^{-5}
Cones	Nickel
Quad mode	Peak hop, 100-ms dwell, 3 points per peak
Acquisition	90 s, 5 cycles, 10 replicates
Rinse	300 s, 2% TMA HNO_3
Internal standard	4 μg/L internal standard
Field spike	4 μg/L yttrium and thorium

Table 4. Performance data for the clean protocol samples (ng/L)

Instrumental detection limit (1 σ) about 200% variability	0.3
Instrumental determination limit for blanks and standards (3 σ)	1.5
Detection limit with respect to field recoveries	5.0
Relative standard deviation for spiking	
<25 ng/L	15%
25–100 ng/L	10%
>100 ng/L	5%
Relative difference for duplicate samples on different days for 25% of samples: mean ± SD	25 ± 9%
Recovery for yttrium field spike: mean ± SD	104 ± 10%

Table 5. Average results of analyses

Site	Ag (nmol) <10 kDa	Ag (nmol) <0.2 μm	Ag (nmol) <5 μm	AVS nM	pH	T (°C)	Dissolved O_2 mg/L	DIC mg/L	DOC mg/L
1A	0.015	0.17	1.07	570				39.1	16.2
1B	<0.003	0.12	0.22	18	6.84	15.3	8.7	8.7	17.0
1C	0.042	0.21	1.14	485	7.22	13.4	7.5	37.6	16.7
1D	0.036	0.20	2.83	260	7.18	13.5	7.6	26.7	18.7
1E	<0.003	0.65	3.33	310	6.67	19.0	7.9	11.2	3.8
1F	0.025	2.60	2.38	8.5	7.05	18.2	8.2	19.4	5.1
1G	<0.003	0.46	0.35	8.5	7.45	19.1	7.8	22.4	3.2
1H	0.29	1.14	1.00	<1	7.64	19.1	8.2	15.7	4.8
2A	<0.003	0.093	0.01	<1	7.64	18.9	10.2	29.1	9.1
2B	<0.003	0.63	0.43	<1	7.24	19.1	8.5	14.2	10.3
2C	<0.003	1.49	2.08	<1	7.73	19.1	8.5	24.2	10.3
3A	<0.003	0.040	0.067	29	7.19	19.2	6.8	20.4	10.0
4A25-1	0.20	0.16	0.10	230					
4B35-1	0.35	0.85	1.81	<1	6.74	15.6			
4A25-2	0.18	0.16	0.086	240					
4B35-2	0.49	0.56	1.52	<1					
5AS	0.003	1.65	5.35	<1	7.54	23.0			
5AB	0.051	2.17	5.73	<1			11.0		
5BS	<0.003	0.069	0.27	<1	7.69	22.1	9.5		
5BB	<0.003	0.11	0.32	190			9.0		

[a] Refer to Table 4 for expected SD for Ag(I) results at different concentration levels. Ag is expresses here in molar units to compare with acid-volatile sulfide (AVS). O_2 = oxygen; DIC = dissolved inorganic carbon; DOC = dissolved organic carbon.

was connected directly to the sampling syringe by Teflon® tubing. The system was completely isolated from the atmosphere, and sampling equipment was opaque to suppress photochemical reactions.

For field sampling, personnel wore arm-length PVC® powderless gloves. Water samples (about 15 ml, exact amounts determined by difference) were syringe filtered through 5- and 0.2-μm filters. A third sample, syringe filtered through the 0.2-μm filter into a 10-kDa centrifugal filter container, was processed at the end of each day (maximum 8 h). Filtered samples were spiked and acidified in the field with 200-μL double-distilled 2 N HNO_3 containing two exotic metals (yttrium [Y] and thorium [Th]) used to determine analytical recovery on each sample. Samples taken at depth were drawn into a syringe through 1/8" Teflon® tubing.

Sampling and analysis of vegetative matter followed a similar clean procedure. Rooted vegetation was sampled and placed in a clean polyethylene bag after removing a sample of rooted soil into a separate bag. The plant material was later separated and washed first with tap water and finally with Milli-Q deionized water. Separate portions were bagged for laboratory preparations. Laboratory extraction consisted of treating in hot 6 N TMA ultrapure HNO_3 for 30 min. The extract was cooled, filtered, neutralized, and analyzed against appropriate spiked standards by ICP-MS.

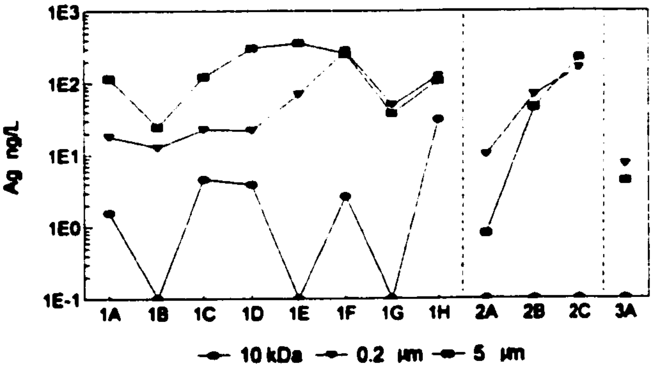

Fig. 3. Ag(I) concentration in different filter fractions along site 1 and 2. Refer to Table 4, duplicates, for estimate of variation.

Acid-volatile sulfide was measured according to Cline [13]. The sample was processed through a 0.45-μm filter in the field, and the derivitization reagent, stored in an opaque container at 4°C, was added immediately after sample collection. All tasks were done without exposure to air and light by using dark bottles and tubing and a closed pumping system. Measurements were made within 8 h after collection and addition of reagent. We previously ascertained that there were no statistically significant changes in results for this delay. Many colloidal sulfides, soluble sulfides, and a portion of polysulfide, but not thiols, are analyzed by this procedure [14].

A PE-Sciex-Elan-500 ICP-MS instrument (Perkin Elmer, Norwalk, CT, USA) was used for trace metal analyses. Table

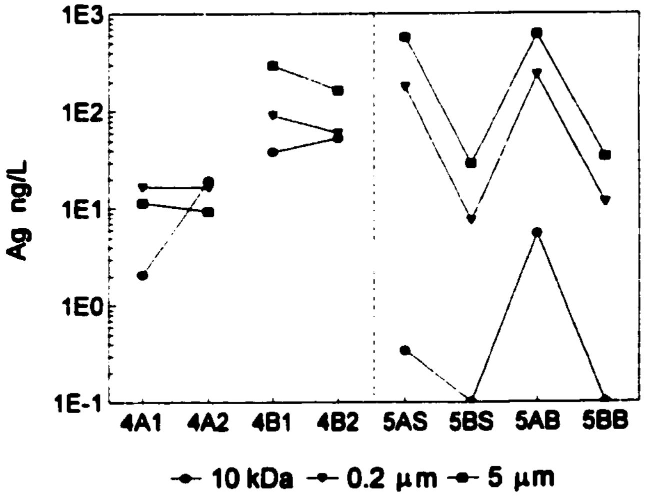

Fig. 4. Ag(I) concentrations in different size fractions in shallow seeps (4) and pond water (5) at Cart Lake Tailings site 2. AS and AB are surface waters at different times, and AB and BB are bottom waters at different times.

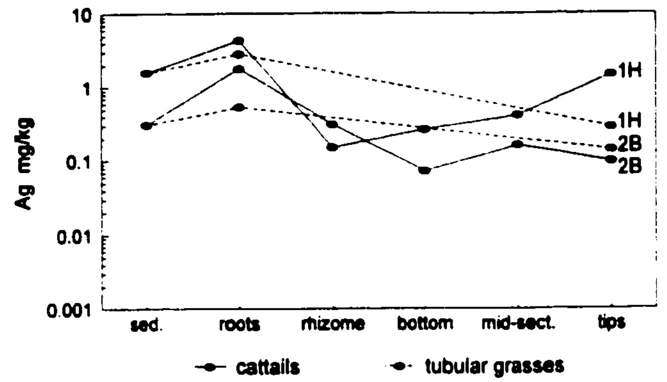

Fig. 5. Ag(I) in sediment, roots, shoots, and tips of cattails. Refer to Table 4, duplicates, for estimate of variation.

3 summarizes the operational conditions for the instrument. Prior to use, the instrument was disassembled, and all components contacting the solution were thoroughly cleaned or replaced with components dedicated to this study. Separate instrument glassware, tubing, and cones were stored in 1 to 2% TMA HNO$_3$ (Baker Ultrex II) prior to analysis. Immediately prior to analysis, 15-ml samples were acidified with 200 μl of TMA 2 N HNO$_3$, containing 4 μg/L In as an internal standard. Both ^{107}Ag and ^{109}Ag were measured against a laboratory internal standard (In) and against field spikes (Y and Th).

RESULTS

Table 4 gives the figures of merit for the quality control and quality assurance portion of the study. Although the instrumental determination limit was defined as 1.5 ng/L using the criterion cited in Table 4, a detection limit of about 0.3 ng/L could be achieved with about ±200% deviation. The relative error, determined by spiking and recovery of samples, varied from 5 to 15% and increased with decreasing Ag(I) concentration. Average replication was poorer (25%) than the spiked recoveries, probably due to other unspecified errors. Overall, the data can be evaluated conservatively with an estimated error of 25%.

Table 5 compiles the data for the study. These data are the first low-level analytical results for Ag and associated parameters at the Cobalt mining camp that were obtained employing a rigorous quality control program. They are significantly lower than those reported for other areas (Table 1).

Figure 3 shows the concentrations of Ag(I) along sites 1 and 2, respectively. The concentrations of Ag(I) are reported for three different size fractions: <5 μm, 0.2 μm, and <10 kDa. The range of concentrations in the different sizes and their medians are

<5μm: 1 to 620, median = 110 ng^{-1}

<0.2 μm: 7.2 to 280, median = 35 ng^{-1}

<10 kDa: <0.3 to 53, median = 1.5 ng L

These ranges may be compared with the control lake site, 3A, with Ag(I) values of 29, 4.3, and <0.3 ng/L for <5-μm, <0.2-μm, and <10-kDa sizes, respectively. Most of the <0.2-kDa Ag(I) is found in the colloidal 0.2-μm to 10-kDa fraction. Often the <10-kDa Ag(I) concentration is below the detection level of 0.3 ng/L. This is particularly true at site 2, where negligible organic colloids and soluble ligands would be found.

Table 6. Concentration ratios of silver for root-soil and leaves-roots along Sasaginaga Creek, elevated in organic matter

Location	Root-soil	Leaves-roots
1B	0.03	NA
1E	0.20	0.11
1H	1.4-3.0	0.09-0.30

Waters draining Cart Lake show a progressive increase in Ag(I) concentrations in the downstream direction for the <0.2- and <5-μm sizes but are not detectable in any of the <10-kDa fraction samples.

The waters draining Sasaginaga Creek are more variable for Ag(I). Ag(I) concentrations in the <5- and <0.2-μm range are in the tens and low hundreds of nanograms per liter, whereas the <10-kDa fraction varies widely and with no trend, from <0.3 to about 5 ng/L, with one site (1H) as high as 31 ng/L. The higher Ag(I) concentration data along study site 1 was related to inflows of waters from mine shafts and contacts with tailings, however. The rapid drop-off in concentration (1F-1G) suggests a rapid removal of Ag(I) in all three size fractions, perhaps due to adsorption on sediment.

Figure 4 shows the shallow seepage (sites 4, 25-35 cm) and pond waters (sites 5) at Cart Lake. Ag(I) concentrations tend to be elevated compared with the two drainages shown in Figure 3. The <10-kDa data are quite variable, ranging from <0.3 to 53 ng/L. Sampling the seepage waters at the two sites at different times also gave highly variable results.

Acid-volatile sulfide was measured at all locations in water samples after 0.45-μm filtration. Although all waters were nearly saturated with respect to atmospheric oxygen, over one half of the samples had measurable AVS in the tens to hundreds nanomolar levels.

Figure 5 shows the change of Ag concentrations (dry weight) from embedded sediment to roots and to tips of tubular grasses and cattails. The tubular plants do not concentrate Ag, showing a systematic decrease from root to tips. This is true for recent grasses (site 2B) and well-established grasses (site 1H). On the other hand, cattails growing on older (100 year) tailings have apparently concentrated Ag in their tips compared with roots, whereas the more recent cattails do not show an increase of Ag in their tips. This is evident in the example for site 1H compared with 2B as shown in Figure 5. The concentration of Ag in the tips of cattails apparently can take a long time to occur.

A study of Ag accumulation in soil, roots, and leaves was done along Sasaginaga Creek, the sewage outlet for Cobalt, to see if added organic matter would change the sediment-plant concentrations of Ag in old tailings derived soil and plants. Samples of aquatic rooted plants and associated sediment were taken at sites 1B, 1E, and 1H. Table 6 tabulates the grass, root, and soil concentration ratios. The <0.2-μm aqueous fraction contained significant Ag(I) (100 ng/L), and the root-associated soils ranged from 0.3 to 4 mg/kg Ag(I). Roots were the major uptake site with root to soil ratios of 3 to 140%. Leaves had only 2% of the soil Ag, with the exception of site 1H, which will be discussed.

A garden plot, about 2 months old and consisting of immature plants, was developed on the northeast shore of Cart Lake. A local red soil with organic matter, about 10 cm deep, was rototilled with Cart Lake tailings. Figure 6 shows the results of analyses for Ag(I) of different market vegetables.

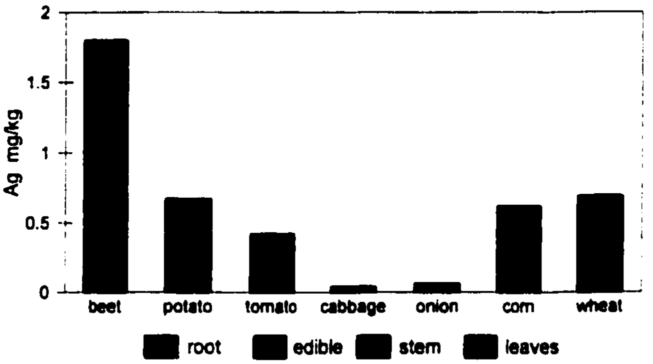

Fig. 6. Silver concentrations (dry weight) for parts of garden vegetable, 2 months old, growing on emended Cart Lake tailings.

At this stage of development, there was no evidence of bioaccumulation of Ag compared with roots levels. Beet, corn, and wheat roots appear to have accumulated more Ag(I) than the other plants.

DISCUSSION

Aqueous Ag(I) concentrations are distributed in different sizes, being more abundant in the "colloidal" fraction, as defined here as between 0.2 μm and 10 kDa. Ag(I) is often not detectable for the <10-kDa fraction. The nature of the colloidal fraction is unknown. Some inferences can be obtained from the review by Bell and Kramer [15]. The increase in concentration of colloidal and <10-kDa Ag(I) at site 1, a water system elevated in organic matter, compared with site 2, suggests that Ag(I) is bound as a thiolate or sorbed on the surfaces of colloidal organic matter. At present, there are no means of defining the Ag(I) association better. Furthermore, the bioavailability of the Ag(I) of this colloidal fraction is not known. Indirect evidence from the Ag(I) concentrations in grasses and cattails, growing on the tailings for many years, suggests that Ag(I) accumulates only in the roots of this vegetation. There are similar indications of concentrations only in roots of market garden plants, and the preliminary results for Ag(I) accumulation here are similar to those found by Hirsch [16], site S-3A.

The bioavailability of Ag(I) and other metals has been hypothesized to be related to a mole ratio of metals to AVS [17]. First, it is most interesting that AVS was detected in many of the <0.45-μm filtered water samples, although all of the samples had significant DO and would be considered aerobic. Figure 7 compares <0.2-μm Ag(I) with <0.45-μm AVS. In most cases, moles of AVS exceed moles of Ag(I). Because Ag(I) would replace other metals [except Hg(II), gold, and Cu(I)] due to stronger binding to S(II-) [17], we propose that as long as AVS (mole) > Ag(I) (mole), Ag(I) should not accumulate in the plant material. There are four cases where there is excess Ag compared with AVS. One is found in the shallow groundwater sample (4B35), two at the "inorganic" Cart Lake location (5AS and 2B), and one at location 1H. Location 1H also had an anomalously high <10-kDa Ag(I) water concentration (Fig. 3). In addition, site 1H is the only location to exhibit concentration of Ag(I) in the tips of cattails (Fig. 5). Although the data presented here do not contradict the metal–AVS concept, there are some questions to be resolved. As mentioned earlier, the exact form of the colloidal and "soluble" (e.g., <10-kDa) Ag(I) is unknown. We estimate the amount of Ag(I) that would be in "solution," assuming an equilibrium with Ag_2S, assuming the ambient pHs of the "inorganic" site 2, and assuming the major species is $AgHS^o$ [18]. We find that the predicted total soluble Ag is ≤ 0.1 ng L^{-1} $AgHS^0$. Similarly, the elevated <10-kDa Ag(I) concentration at site 1H can be interpreted as not being in equilibrium with Ag_2S, i.e., AVS<Ag(I). The elevated values along site 1 in the 10 to 100 ng L levels cannot be explained by equilibrium with Ag_2S, although there is an excess of AVS compared with Ag(I). There is the possibility that the <10-kDa Ag(I) includes some colloidal material or that there are other stronger Ag(I) species such as thiolates or polysulfides [15]. Neither the polysulfide S(O) nor the thiolates would be measured by AVS technique [14]. They may form significant Ag(I) complexes.

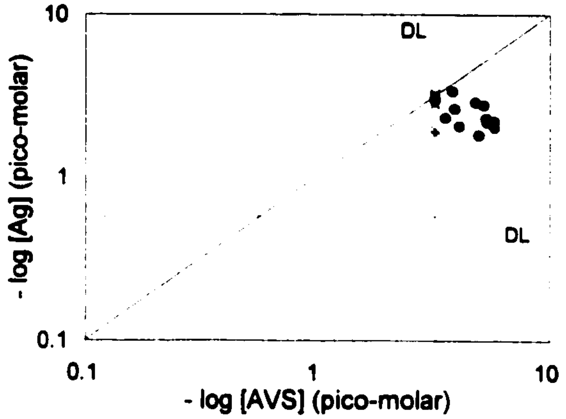

Fig. 7. Comparison of acid-volatile sulfide (AVS) with Ag(I) in the <0.45-μm and <0.2-μm fractions, respectively. DL is detection limit, and the inclined line represents equal moles of Ag(I) and AVS.

Although we cannot explain all of the Ag(I) data within known equilibrium systems, Ag(I) is strongly bound to the solid phase and is at low nanogram per liter levels in the apparent soluble phase. Perhaps, more importantly, the majority of operationally defined soluble (usually <0.45 μm) Ag(I) occurs in the colloidal phase.

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