Environ. Sci. Technol. **1998**, *32,* 2043−2050

Complexation of Cobalt by Natural Ligands in Freshwater

JIN QIAN,[†,§] HAN BIN XUE,[*,†]
LAURA SIGG,[‡] AND ACHIM ALBRECHT[‡]
Swiss Federal Institute for Environmental Science and Technology (EAWAG), CH-6047 Kastanienbaum, and CH-8600 Duebendorf, Switzerland

Ligand exchange with dimethylglyoxime (DMG) and differential pulse cathodic stripping voltammetry (DPCSV) was applied to determine free Co(II) ionic concentrations $[Co^{2+}]$ and Co(II) complexation by natural organic ligands in freshwater. $[Co^{2+}]$ and the complexing parameters were calculated on the basis of the thermodynamic equilibria between added and natural ligands. The optimal working conditions were carefully evaluated in synthetic solutions and in natural freshwater samples. The results from samples collected at various sites from the Aare and Rhine Rivers and the Biel and Lucerne Lakes, Switzerland, are presented. The total dissolved Co concentrations are in the range of 0.5−6.5 nM, and $[Co^{2+}]$ is in the range of 0.05−0.5 nM. Organic complexes of Co are predominant in most samples with 80−96% of total dissolved Co. A one-ligand model could well fit the titrations of freshwater samples with Co(II) (using the FITEQL program). The conditional stability constants of the natural ligands were computed to be log K = 9.5−11.6 (pH 8.0 ± 0.1), and the ligand concentrations were [L] = 1.4−7.6 nM. The reliability and applicability of the technique and the implications of the results are discussed.

Introduction

The discharge of various cobalt radionuclides from nuclear reactors to surface water and their accumulation by aquatic organisms and sediments are drawing attention to the cycling and fate of this element in aquatic systems (*1*−*6*). Four nuclear installations are located along the Aare and Rhine Rivers in Switzerland. Some investigations on the transport and deposition of ^{60}Co in surface aquatic systems of that region have been performed (*5*−*7*). For a better understanding of the geochemical cycling and of the biological effects of this element, further studies are needed, especially of the Co speciation in solution.

Cobalt is an essential element that plays a very specific role in vitamin B12 (*8*, *9*). Cobalt is generally a rare element, available only at low concentrations in aquatic systems. With regard to the fate of ^{60}Co in natural waters, it is important to understand whether dissolved Co inputs will be easily bound to particles and retained in sediments or, in contrast, will be strongly complexed in solution and transported with the water phase. Other significant questions concern the pathways by which ^{60}Co may be accumulated in organisms. For all these questions, the speciation of Co with respect to its complexation and to its oxidation state [Co(II) or Co(III)] is crucial.

Few studies on the oxidation state of ^{60}Co (*10*, *11*), on inorganic Co complexation in seawater (*12*), and on the Co complexation in seawater, estuarine waters, and river water (*13*, *14*) are available.

Adsorptive cathodic stripping voltammetry, in which a Co(II)−dioxime complex is accumulated by adsorption onto a hanging mercury drop electrode and the adsorbed species is subsequently reduced, has been successfully used to determine total Co concentrations in industry effluents and seawaters and to study the Co(II) speciation (*13*, *15*−*18*).

We apply here a ligand-exchange method, using dimethylglyoxime (DMG) and measurement by DPCSV (differential pulse cathodic stripping voltammetry) (*13*), to study the Co speciation in lake and river water, assuming that Co(II) is the predominant oxidation state. The optimal working conditions are carefully evaluated in synthetic solutions and in natural freshwater samples. The concentrations of total dissolved and free ionic Co(II) as well as the concentrations and conditional stability constants of the natural ligands are determined in various lake and river waters. Samples were mostly collected in the vicinity of nuclear power plants (Aare and Rhine Rivers, Lake Biel) and compared to other sites (Lake Lucerne) in Switzerland. The results obtained for the stable Co isotope should be applicable to the radioactive ^{60}Co, which is present at much lower concentrations of about 10^{-5} nM, if the isotopic exchange kinetics of ^{60}Co with stable cobalt among various Co complexes are sufficiently fast.

Theory of Ligand Exchange and Data Evaluation. In the ligand-exchange approach used here, DMG (dimethylglyoxime) is added to a water sample to compete for Co with natural ligands (*13*). To evaluate the Co speciation, the concentration of the added dioxime should just be sufficient to form a measurable concentration of CoDMG, but not as high as to complex almost all Co. $[Co^{2+}]$ and the complexation parameters in the system are then obtained by equilibrium calculations.

In the presence of added DMG, the mass balance is

$$\begin{aligned}[Co_T] &= [Co_{nat}] + \sum[CoDMG_i] \\ &= [Co^{2+}] + [Co_{in}] + \sum[CoL_i] + \sum[CoDMG_i] \\ &= \sum[CoL_i] + [Co^{2+}](1 + \alpha_{in} + \alpha_{DMG}) \qquad (1)\end{aligned}$$

where $[Co_T]$, $[Co_{in}]$, and $[CoL_i]$ represent the concentrations of total dissolved, natural inorganic, and organic complexes; $[Co_{nat}]$ is defined as the difference in concentrations of total dissolved and CoDMG complexes ($\sum[CoDMG_i]$) corresponding to natural Co species in the presence of DMG; α_{in} and α_{DMG} represent the inorganic and DMG complexing coefficients, respectively ($\alpha_{in} = \sum\beta_i [L_{in}]^i$, the sum of the products of inorganic ligand concentrations and corresponding stability constants; $\alpha_{DMG} = \beta_{1DMG} [DMG^-] + \beta_{2DMG}[DMG^-]^2$, the sum of the products of the stability constants and the free DMG ligand concentrations).

A calibration must be carried out for each individual water sample by titration of Co. Theoretically, the adsorbed CoDMG concentration is proportional to the peak current (ip), linked via the DPCSV sensitivity (S in A M^{-1}):

$$ip/S = [CoDMG_i] = [Co^{2+}]\alpha_{DMG} \qquad (2)$$

---

[*] Corresponding author e-mail: xue@eawag.ch.

[†] EAWAG, Kastanienbaum.

[§] Present address: Analytical Chemistry Department, Umeå University, 90187 Umeå, Sweden.

[‡] EAWAG, Duebendorf.

S0013-936X(97)01018-3 CCC: $15.00

Practically, the relationship between the [Co_T] and ip in the presence of high concentrations of added Co(II) is used for the calibration. When the natural organic ligands are saturated at higher [Co_T], the concentration of natural organic complexes becomes a constant, and this constant corresponds to total available concentration of the natural organic ligands under the working window, [L_t]:

$$[Co_T] = [L_t] + (ip/S)\ (1 + \alpha_{in} + \alpha_{DMG})/\alpha_{DMG} \qquad (3)$$

Under any given conditions, all values of α should be constant, then

$$\text{let } S^* = S\alpha_{DMG}/(1 + \alpha_{in} + \alpha_{DMG}) \quad (\mathrm{A\ M^{-1}}) \qquad (4)$$

and the labile Co species $[Co]_{lab}$ are then defined as the sum of free ion Co^{2+}, inorganic complexes, and DMG complexes, thus in excess of [L_T]

$$[Co_T] = [L_t] + [Co]_{lab} = [L_t] + ip/S^* \qquad (5)$$

According to eq 5, the slope of the linear part of the titration curve in terms of peak current ip vs [Co_T] at high added Co is simply equal to S^*.

After calibration, [Co^{2+}] and $\sum[CoDMG_i]$ can be calculated from the measured $[Co]_{lab}$ and the known α_{DMG} in a given system:

$$ip/S^* = [Co]_{lab} = [Co^{2+}](1 + \alpha_{in} + \alpha_{DMG}) \qquad (6)$$

[Co_{nat}] is obtained from the difference of total dissolved and DMG complexes at any point of the titration. Therefore, a data set of [Co^{2+}] and corresponding [Co_{nat}] in the titration series of a sample is acquired. A one-ligand model is applied to compute a conditional stability constant K and a ligand concentration [L_t], using FITEQL, a chemical equilibrium-fitting program (*19*). Our data evaluation has shown that the titration data are well fitted with the assumption of one ligand and a 1:1 complex, partially because of the limited Co concentration range (in nM) in the titrations.

The added competing ligand DMG binds a portion of Co in the sample and thereby reduces both [Co_{nat}] and [Co^{2+}]. Thus, to determine the ambient [Co^{2+}] in the original sample, one must be able to correct back to the [Co^{2+}] that was present before the addition of DMG. Using the fitting parameters K and [L_t], one can compute the ambient [Co^{2+}] by interpolation.

The complexing coefficient α_{DMG} is very sensitive to the working conditions and must be strictly calculated at given conditions. In our calculation, the values of $\log \beta_{1DMG}$ (8.17, $I = 0.01$ M) and $\log \beta_{2DMG}$ (16.71, $I = 0.01$ M) were taken from Sillen and Martell (*20*) and corrected for ionic strength using the Güntelberg equation. For the calculation of free DMG concentration, the acidity constant K was taken from Martell and Smith (*21*) and corrected for ionic strength (pK = 10.57, $I = 0.01$ M). Stability constants with major cations are not available from the literature. We have found negligible CaDMG complexation under freshwater conditions (see below). Therefore, the complexes with major cations can be neglected, assuming that Mg has a similar effect as Ca. The competition of other trace metals (Cu, Zn, Ni, etc.) are also neglected in the calculation of free DMG concentration because DMG is added in excess. The value of α_{DMG} is in the range 2−6 in our experiments with optimal [DMG] (2−4 μM, see below) and pH 7.95. Under these conditions the $Co(DMG)_2$ complex is in the range of 63−78%.

The inorganic complexing coefficient α_{in} very much depends on the stability constants of the cobalt(II) carbonate complexes, for which a wide range is found in the literature. With $\log K_{CoCO_3} = 4.4$ and $\log K_{CoHCO_3} = 12.5$ from ref *22*, α_{in} is around 0.3−0.5 in the typical alkalinity range (2−2.5 mM) of our freshwater samples at pH 7.9−8.0. The results shown here were calculated with these low values for α_{in}.

Experimental Section

Determination of the Conditional Stability Constant of DMG β^*_{2DMG}. The conditional stability constant β^*_{2DMG} was experimentally determined in synthetic solutions containing Co^{2+} (10 nM), KNO_3 (0.01 M), HEPES buffer (*N*-(2-hydroxyethyl) piperazine-*N*′-2-ethanesulfonic acid) (0.03 M), DMG (0.1 mM), and Ca^{2+} (1.5 or 2 mM) by ligand competition with EDTA (ethylenediaminetetraacetate) ranging from 0.5 to 80 μM. The procedure is similar to that carried out in UV-irradiated seawater by Zhang et al. (*13*). The CoEDTA complex is not adsorbed at the mercury electrode under these conditions.

The $Co(DMG)_2$ complex is predominant at this high DMG concentration, according to equilibrium calculation. The peak current (ip) is thus directly proportional to the concentration of the adsorbed complex ($CoDMG_2$) via sensitivity (S):

$$ip/S = [CoDMG_2] = [Co^{2+}]\alpha_{DMG} \qquad (7)$$

The sensitivity S does not need to be calibrated here because it is canceled by comparing the concentrations of adsorbed species in the presence of EDTA to the reference in the absence of EDTA.

Defining X as the ratio of ip/S in the presence of EDTA to ip/S in the absence of EDTA, we obtain [for the derivation, see Zhang et al. (*13*)]:

$$\alpha_{DMG} = (X\alpha_{EDTA} + X - 1)/(1 - X) \qquad (8)$$

α_{EDTA} is calculated on the basis of deprotonation and Co complex stability constants of EDTA (10.65, $I = 0.01$ M; 16.31, $I = 0.01$ M) at given conditions, taken from Martell and Smith (*21*); then, an average value of α_{DMG} in the presence of various concentrations of EDTA is obtained.

DPCSV Measurements. All samples measured by DPCSV were buffered to pH 7.95 ± 0.02 by the addition of HEPES, using a stock buffer solution containing 1 M HEPES and 0.6 M NaOH. Standard solutions of Co were prepared by dilution of a 1000 ppm stock atomic spectral standard. Stock solutions of DMG (0.01 M) were prepared in HPLC-grade methanol. The EDTA stock solution was 0.1 M and neutralized with NaOH.

The HEPES buffer is chosen here because of its weak complexing properties (*23*). An enhancement of HEPES on the CSV sensitivity has been observed by Donat and Bruland (*15*). Although the mechanism of this enhancement is not clear (by accelerating or catalyzing the electron transfer), the maximum sensitivity is reached at 0.03 M HEPES. Therefore, 0.03 M HEPES was added to all samples to keep the pH constant and to eliminate differences among samples while enhancing the DPCSV sensitivity.

The titration with Co(II) of a synthetic solution containing KNO_3 (0.01 M), HEPES (0.03 M), and DMG (2 μM−0.1 mM) gave a linear relationship between the peak current and the Co concentration in the range of 0.1−10 nM.

To obtain a set of DPCSV titration data for a sample, a series of subsamples was spiked with different Co concentrations. The 25 mL of a water sample was pipeted into 50-mL high-density polyethylene beakers, and 750 μL of HEPES buffer was added to each beaker to give a final concentration of 0.03 M and pH 7.95 ± 0.02. The DMG solution was added to all beakers to reach the same desired concentration (2−4 μM). Then, the Co solution was added to all beakers but one, giving a concentration range between

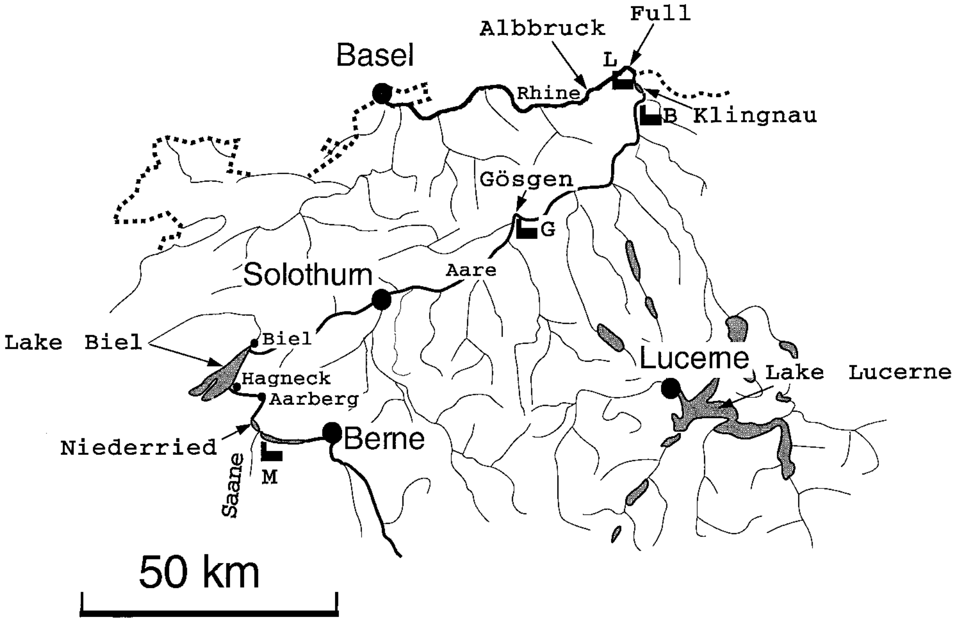

FIGURE 1. Location of the sampling sites.

0.1 and 10 nM in 12 increments. The series was allowed to equilibrate at 22 ± 2 °C overnight.

DPCSV measurements were performed with a HMDE (hanging mercury drop electrode), an Ag/AgCl reference electrode, and a graphite counter electrode held in a Metrohm 647 VA stand combined with a Metrohm 646 VA processor at 22 ± 2 °C. A sample beaker was placed in the measurement cell and deoxygenated with suprapure N_2 for 5 min. A new Hg drop was extruded (after 5 preliminary Hg drops), and the stirrer switched on simultaneously. The potential of −0.6 V was immediately applied to the HMDE, beginning the accumulation by adsorption. After the accumulation time (60 s), the stirrer was turned off, and 15 s later the voltammogram was recorded by scanning in the negative direction in the differential pulse mode. The scanning parameters found to yield the best compromise between sensitivity, resolution, and speed were pulse height of 50 mV and scan rate of 10 mV s^{-1}. The reduction peaks occurred at approximately −1.03 ± 0.02V.

DPCSV precision for labile Co was ±7.8% in a synthetic solution (KNO_3 0.01 M, HEPES 0.03 M, DMG 2 μM) with 0.5 nM Co and ±8.8% in a sample from the Aare River with original 1.4 nM Co, added 2 μM DMG, and 0.03 M HEPES, expressed as the relative standard deviation (RSD) ($n = 6$). Each subsample of any titration series was measured twice by DPCSV, and the average was used to determine $[\mathrm{Co}]_{\mathrm{lab}}$. The detection limit for labile Co depends on the reproducibility of the procedural blank rather than on instrumental noise. A peak measured six times gave a standard deviation of 5.5 pM at an estimated Co concentration of 61.4 pM. This result yields a detection limit (3σ) of 17 pM.

Sampling and Analyses. All sampling and filtering devices, bottles, and membranes (0.45 μm) were washed with 0.01 M HNO_3 and rinsed with bidistilled water before use.

Freshwater samples were taken from the Aare and Rhine Rivers and from Lake Biel (all located near nuclear installations) as well as from Lake Lucerne (Figure 1). Go-Flo sampling bottles (General Oceanics, 5 L) were used to collect samples from different depths in the water columns of the lakes. Samples from rivers were pumped (from about 0.5 m depth, 2−3 m away from the river bank) using a peristaltic pump. Samples were filtered with 0.45-μm Millipore membranes after transport to the laboratory. The filtered samples were stored in the dark at 4 °C until DPCSV determination within 5 days. All Co titrations were carried out in filtered samples.

Total dissolved Co was measured by ICP−MS (Perkin-Elmer SCIEX ELAN 5000) with a detection limit of 0.05 μg L^{-1} ($\approx$0.8 nM) in natural waters. For this purpose, 100 mL of filtrate was immediately acidified to 0.01 M HNO_3.

Dissolved organic C (DOC) was measured by combustion at 680 °C on a Shimadzu TOC 500 instrument. A Metrohm 605 pH meter was used to measure pH. The concentrations of Cl^-, NO_3^-, and SO_4^{2-} were determined by ion-exchange chromatography (Dionex DX 300), and the alkalinity was determined with a Metrohm titroprocessor titrating to pH 4.5 with HCl.

Results and Discussion

Determination of the Conditional Stability Constant of DMG $\beta^*_{2\mathrm{DMG}}$. By competition with EDTA, the complexation coefficient α_{DMG} (at 0.1 mM DMG) was determined as (1.21 ± 0.31) × 10^3 ($n = 6$) at pH 7.80 in the presence of 2.0 mM Ca^{2+} and as (1.26 ± 0.38) × 10^3 ($n = 6$) at pH 7.90 in the presence of 1.5 mM Ca^{2+}.

Because $Co(DMG)_2$ is predominant at 0.1 mM DMG

$$\beta^*_{2\mathrm{DMG}} = \alpha_{\mathrm{DMG}}/[\mathrm{DMG}^-]^2 \qquad (9)$$

Assuming that Ca has no effect, $[\mathrm{DMG}^-]$ is calculated from the protonation of DMG only. The calculated log $\beta^*_{2\mathrm{DMG}}$ is 16.87 ± 0.07 ($n = 6$) at 2 mM Ca^{2+} and pH 7.80 and is 16.61 ± 0.09 ($n = 6$) at 1.5 mM Ca^{2+} and pH 7.90. The values are both in good agreement with the value of 16.71 from Sillen and Martell (*20*) and corrected to 0.01 M ionic strength, which we used for all calculations of α_{DMG} in this study. The agreement indicates that complexation of Ca with DMG can be neglected under freshwater conditions with $[\mathrm{Ca}] < 2$ mM.

Optimal Working Conditions. The dependence of Co sensitivity on the adsorption potential was studied both in synthetic solutions containing 1 nM Co, 0.01 M KNO_3, 2 μM DMG, and 0.03 M HEPES at pH 7.95 and in a water sample from Lake Biel with addition of 1 nM Co(II). The maximum Co sensitivity occurred at adsorption potentials between −0.6 and −0.9 V, as reported by Zhang et al. (*16*). In this study, −0.6 V was chosen as the adsorption potential.

The Co sensitivity was measured as a function of adsorption time at an adsorption potential of −0.6 V in an Aare

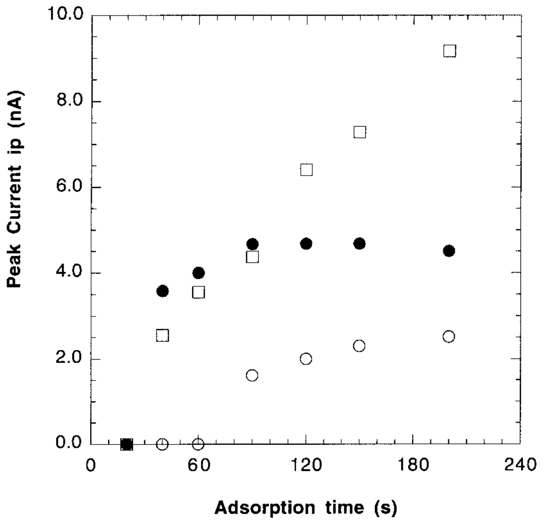

FIGURE 2. DPCSV peak current as a function of adsorptive collection time at a potential of −0.6 V, pH 7.95, in a river sample. The water sample was collected near Klingnau along the Aare River on June 26, 1996, with original dissolved Co 1.2 nM and addition of 1.0 nM Co and 0.5 nM Ni. The symbols are (○) for background without addition of DMG; (●) for Co with addition of 0.1 mM DMG; and (□) for Ni with addition of 0.1 mM DMG.

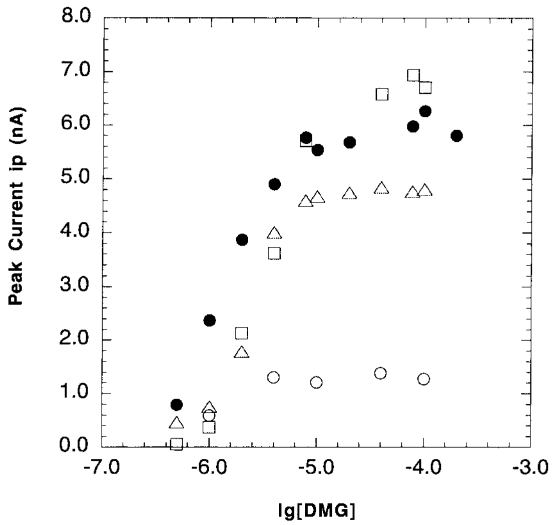

FIGURE 3. Dependence of DPCSV peak current for labile Co on DMG concentration in natural samples. The water samples were collected from the Aare River on July 17, 1996, near Niederried with 1.4 nM original dissolved Co (○) or after addition of 1.0 nM Co(II) (●), and on August 19, 1996, near Hagneck with 1.0 nM original Co and 1.0 nM added Co(II) (△), and from Lake Biel on August 20, 1996, with 1.0 nM original Co and 1.0 nM added Co(II) (□).

River sample. As shown in Figure 2, the peak current increased with adsorption time up to 2 min. On the other hand, the background peak (without DMG in the sample) also increased at longer adsorption times as well as the Ni peak current. The difference between the Co and the background peak current reached a maximum at 1 min adsorption time. The interference occurring with longer adsorption time is probably due to the adsorption of complexes of competing metal ions such as nickel or of natural organic matter (*16*). Therefore, an adsorption time of 60 s was chosen as a compromise between analytical sensitivity and interference minimization.

An optimal concentration range of DMG must be tested prior to determination in unknown natural waters. Therefore, the peak current as a function of DMG concentrations (log[DMG] between −6.3 and −3.4) was examined in the samples from Lake Biel and the Aare River (Figure 3). The tendency is similar for all samples. The peak current increased with added DMG and reached a maximum. Although the rising range of the curve occurs at different DMG concentrations in different samples, the maximum was reached for all samples when $[\mathrm{DMG}] > 10^{-5}$ M. At this maximum, nearly all Co is bound by DMG. Theoretically, an added concentration of DMG should meet the condition that the complexing coefficients of DMG and of the natural organic ligands are of a similar order of magnitude. Practically, the added concentration of DMG should be sufficient to detect $[\mathrm{Co}]_{\mathrm{lab}}$ at the ambient level, but not as high as to bind nearly all Co. This range is just situated around the middle of the rising part of the curves. According to this observation, 2−4 μM DMG is proposed as an optimum concentration for freshwater samples in the present study.

Interferences. Interferences of of Ni, Cu, Cd, Pb, and Zn were examined in the concentration range typically encountered in Swiss lake and river waters (Ni, 10−30 nM; Cu, 10−30 nM; Cd, 0.02−0.5 nM; Pb, 0.1−1 nM; Zn, 10−150 nM, refs *24*−*26* and unpublished data). The formation of DMG complexes with metal ions other than Co should not influence the results because DMG (2−4 μM) was always added in large excess of the concentrations of trace metals (typically <0.1 μM).

Concentrations of 10 nM Ni, 50 nM Cu, 1 nM Cd, 50 nM Pb, and 100 nM Zn did not significantly affect the peak current for labile Co in a synthetic solution with a Co concentration of 0.4 nM as well as in samples from the Aare River at Hagneck and from Lake Biel. Ni forms a complex with DMG that is also adsorbed and reduced at the electrode, like the Co DMG. With a Ni concentration of 50 nM, the peak current of 0.4 nM Co in a synthetic solution decreased to about 80% of its original height. The peaks partially overlap since the Ni reduction peak (at about −0.93 V) was just 100 mV more positive than that of Co (*15, 16*). This interference with Ni was however not observed in our natural water samples.

In few natural samples, even without the addition of DMG, there was a small interference peak at a potential of about −1.03 V that never appeared in the synthetic solutions. No obvious change in the interference peak current was found with Co addition. It seems that the peak is not generated by Co complexes with natural ligands but may originate from the natural organic matter itself. The interfering peak was deducted from the total peak current where it appeared. A shorter adsorption time (60 s) diminished it or even suppressed it in most samples.

$[\mathrm{Co}^{2+}]$ and Complexation in Natural Waters. Typical titration curves with Co in terms of peak current (ip) as a function of $[\mathrm{Co_T}]$ for different water samples are shown in Figure 4. These titration curves have a typical shape, with a concave part at lower $[\mathrm{Co_T}]$ and a linear part at higher $[\mathrm{Co_T}]$. The DPCSV sensitivities S^* for labile Co were extracted from the slopes of the linear portions (see the theoretical part), as shown with the regression lines in Figure 4. The shape of these curves indicates that only a part of the total Co is bound by DMG at lower concentrations and thus that proper competing ligand (DMG) concentrations were used.

The titration data sets of $[\mathrm{Co}^{2+}]$ and $[\mathrm{Co}_{\mathrm{nat}}]$ were used to compute conditional stability constants and concentrations of natural ligands by FITEQL with a one-ligand model (fitting errors <0.1 pK and 0.1 nM [L]). The data could be fitted to a one-ligand model with few exceptions (Table 1). The sensitivity of the results to α_{in} was checked for some of the samples; only minor changes in the calculated log K and $[\mathrm{L_t}]$ were found. The data sets of two Lake Biel samples (2 and 48 m depth, September 1996) could not be converged by FITEQL. Therefore, $[\mathrm{Co}^{2+}]$ for these samples are only estimated by interpolation of the first few points. The

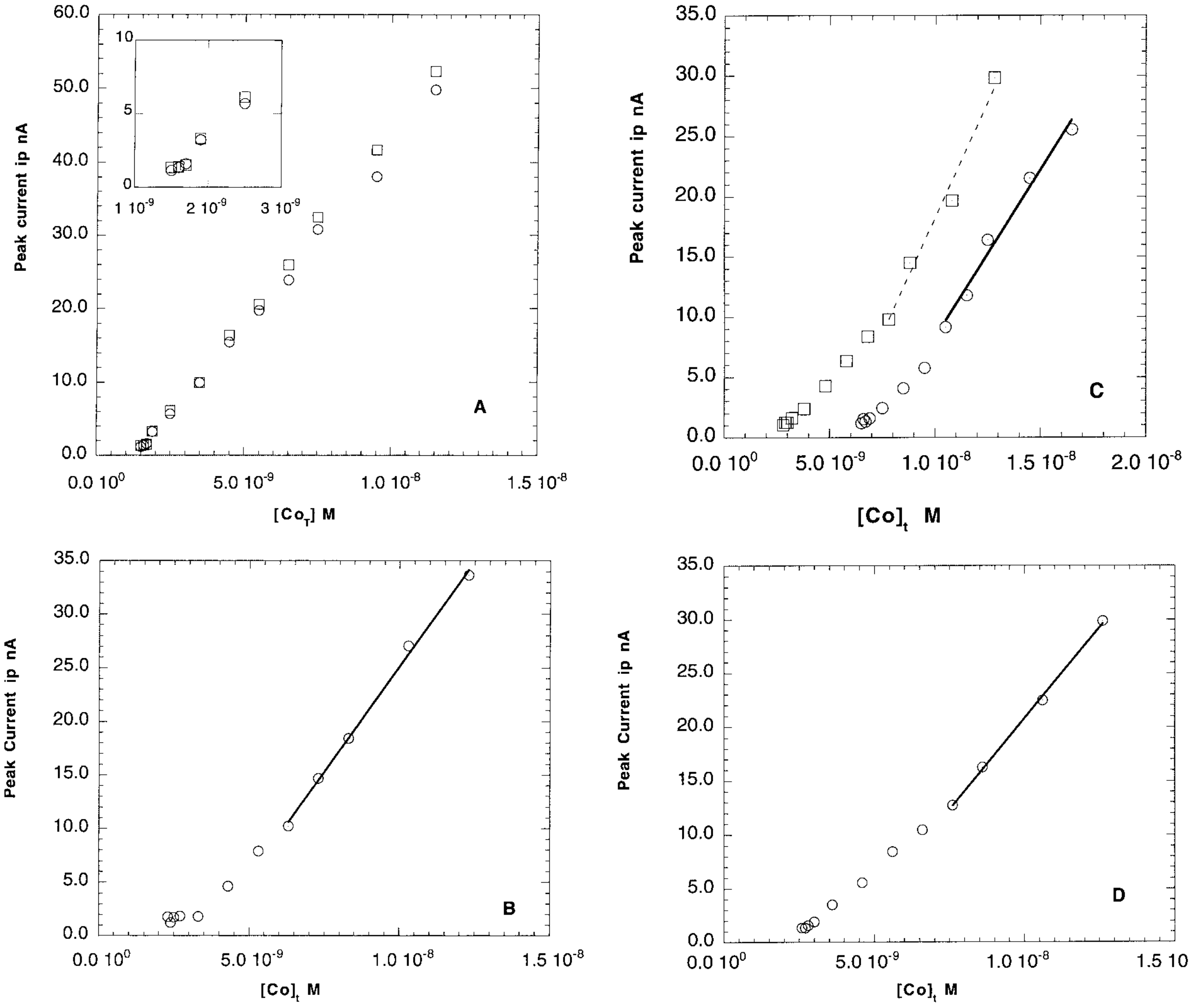

FIGURE 4. Titration curves of natural water samples in the presence of DMG with Co in terms of DPASV peak current as a function of total dissolved [Co$_T$]. (Panel A) Sample from Lake Lucerne (2 m depth, February 12, 1997), measured in the presence of DMG 2 (○) or 2.5 μM (□); the insert depicts an enlargement at the lower concentrations. (Panel B) Lake Biel (25 m depth, January 8, 1997). (Panel C) Aare River, upstream (○) or downstream (□) of the Gösgen nuclear effluent site (December 11, 1996); (Panel D) Rhine River, Albbruck below the nuclear effluent site (December 10, 1996). The regression lines in B−D at higher concentrations are used for calibration of labile Co.

computed parameters fit the data well in most cases, as shown in Figure 5, in which [Co^{2+}] is plotted as a function of [Co$_{nat}$]. Two titration curves are plotted in Figure 5C, that were determined with two different concentrations of DMG in a lake sample. The fitting curves almost overlap, indicating a good reproducibility of [Co^{2+}]. The nearly linear relationship between [Co^{2+}] and [Co$_{nat}$] in Figure 5C indicates that the ligand concentration is lower than the original Co concentration. In samples with ligand concentrations lower than cobalt, the addition of DMG decreased both [Co$_{nat}$] and [Co^{2+}] to lower levels in the first few points of the titrations (Figure 5). Valid complexation parameters could still be extracted by the FITEQL program in these samples.

The concentrations of total dissolved and free ionic cobalt, natural ligand concentrations, and conditional stability constants as obtained from the FITEQL fits and from different sites of several water systems are compiled in Table 1. The total dissolved Co concentrations were in the range of 0.5−6.5 nM and [Co^{2+}] was 0.05−0.5 nM (pCo 9.3−10.4). The organic complexes predominantly contributed to total dissolved Co by 80−96% in most of the samples, with few exceptions (Table 1). Two of the samples with a lower fraction of organic complexes were collected within the plume of dye tracer following the ^{60}Co discharge from a nuclear reactor. Similar conditional stability constants were obtained in most of the samples examined. The average conditional stability constant was log $K = 10.0 \pm 0.1$ (pH $= 8.0 \pm 0.1$, $n = 19$), and the ligand concentrations ranged between 1.8 and 7.6 nM for the samples from the Aare and Rhine Rivers and Lake Biel. In comparison to those samples, stronger ligands with lower concentrations appeared to occur in Lake Lucerne, with log $K = 11.2 \pm 0.3$ and [L$_T$] $= 1.7 \pm 0.4$ nM.

Discussion: Co Complexation in Freshwater. The reliability of the [Co^{2+}] and Co(II) complexation obtained by ligand exchange and DPCSV has to be discussed. A critical point is the possible occurrence of Co(III). From thermodynamic equilibria, it is expected that Co(II) is the predominant oxidation state in natural waters. Co(III) could only occur in strong organic complexes or be associated with particulate matter, especially manganese oxides (*27−29*). Strong Co(III) organic ligands are known to be formed with a number of ligands and to usually exchange slowly (*30*). Co(III) occurs naturally in cyanocobalamine. Co(III) was shown in some cases to occur in anthropogenic Co inputs, for instance downstream of a nuclear reactor (*10, 11*), in which a complexing solution (picolinate) had been used for nuclear reactor decontamination, so that the thermodynamically stable trivalent radiocobalt picolinate complex could be formed and discharged directly to the stream. As far as we know, such strong ligands are not applied in the Swiss nuclear reactors. Because in our study most of the dissolved Co was easily exchanged with DMG and different

TABLE 1. Concentrations of Total Dissolved and Ionic Cobalt and Complexation Parameters in Lake and River Samples[a]

sampling water system	sampling site	sampling date	$[Co_T]$ (nM)	$[Co^{2+}]$ (nM)	pCo	[CoL] (% of $[CO_T]$)	log *K*	[L] (nM)	DOC (mg/L)	remarks
Aare River	Niederried	Jul 17, 1996	1.4	0.12	9.9	91.3	10.1	2.1	3.4	[DMG] = 2 μM
Aare River	Niederried	Jul 17, 1996	1.4	0.09	10.0	93.5	10.5	1.8	3.4	[DMG] = 2.5 μM
Aare River	Hagneck	Aug 19, 1996	1.0	0.43	9.4	56.6	10.2	0.7(?)		within tracer for nuclear effluent
Lake Biel	Lake Biel near Hagneck	Aug 20, 1996	1.0	0.18	9.8	82.3	9.5	2.2		under the tracer
Lake Biel	Lake Biel	Aug 20, 1996	1.0	0.33	9.5	66.7	9.7	1.1(?)		within tracer for nuclear effluent
Lake Biel	Lake Biel	Aug 21, 1996	1.0	0.14	9.8	85.8	9.7	1.9		following tracer but far diffused
Lake Biel	deepest point of lake, 2 m[b]	Sep 3, 1996	1.4	0.46	9.3	67.2			2.8	
Lake Biel	deepest point of lake, 25 m	Sep 3, 1996	1.5	0.12	9.9	91.7	10.3	1.9		
Lake Biel	deepest point of lake, 48 m[b]	Sep 3, 1996	1.5	0.37	9.4	75.6				
Lake Biel	deepest point of lake, 68 m	Sep 3, 1996	1.7	0.12	9.9	92.9	10.4	2.1		
Rhine River	Albbruck	Dec 10, 1996	2.6	0.19	9.7	92.7	10.0	3.7	2.5	following discharge of nuclear effluent
Rhine River	Full	Dec 10, 1996	4.0	0.24	9.6	94.0	10.0	5.4		upstream of the input site of effluent
Aare River	Gösgen down	Dec 11, 1996	2.8	0.13	9.9	95.5	10.0	5.5		following discharge of nuclear effluent
Aare River	Gösgen up	Dec 11, 1996	6.5	0.18	9.7	97.2	10.4	7.6		upstream of the input site of effluent
Aare River	Hagneck	Jan 8, 1997	3.3	0.28	9.6	91.4	10.0	4.2	1.5	
Lake Biel	deepest point of lake, 2 m	Jan 8, 1997	2.5	0.16	9.8	93.5	9.9	4.1	2.5	
Lake Biel	deepest point of lake, 25 m	Jan 8, 1997	2.3	0.14	9.8	93.8	10.0	3.8		
Lake Biel	deepest point of lake, 48 m	Jan 8, 1997	2.5	0.29	9.5	88.4	10.0	2.9		
Lake Biel	deepest point of lake, 68 m	Jan 8, 1997	2.6	0.48	9.3	81.6	9.7	2.9		
Lake Biel	Lake Biel near Hagneck	Jan 8, 1997	2.2	0.24	9.6	88.9	9.7	3.7		same site as on 20 Aug, but without effluent
Lake Biel	outlet of Lake Biel	Jan 8, 1997	2.3	0.32	9.5	86.0	10.0	2.5		
Lake Lucerne	Kreuztrichter 2 m[b]	Jun 24, 1996	<1.0	0.05	10.3	≥ 90			1.2	
Lake Lucerne	Kreuztrichter 2 m	Feb 12, 1997	1.5	0.16	9.8	89.3	11.3	1.4	1.0	[DMG] = 2 μM
Lake Lucerne	Kreuztrichter 2 m	Feb 12, 1997	1.5	0.16	9.8	89.1	11.6	1.4		[DMG] = 2.5 μM
Lake Lucerne	Kreuztrichter 25 m	Feb 12, 1997	1.9	0.07	10.2	96.6	11.1	2.0		[DMG] = 2 μM
Lake Lucerne	Kreuztrichter 25 m	Feb 12, 1997	1.9	0.07	10.2	96.4	10.9	2.2		[DMG] = 2.5 μM

[a] $[Co^{2+}]$, calculated free Co aquo ion concentration; pCo = $-\log [Co^{2+}]$. [CoL], % of total Co bound in organic complexes. log *K*, conditional stability constant obtained by FITEQL fitting, pH = 7.95 ± 0.05. [L], concentration of ligand obtained by FITEQL fitting. [b] These data could not be fitted by FITEQL; the free ionic Co concentrations for these samples were estimated by interpolation of the first few data.

concentrations of DMG gave reproducible results, the contribution of Co(III) was probably small. The independence of added [DMG] (Figure 5C and Table 1) under the optimal conditions verifies the assumptions of the ligand-exchange theory that the other chemical reactions do not significantly decrease the added ligand concentrations, mixed ligand complexes are not formed, and competition between Co and other metals are not significantly affecting Co^{2+} equilibrium. Furthermore, the shift of DPCSV peak potential due to mixed ligand complexes was also not observed.

With regard to the nature of the Co(II) ligands observed in our study, several constraints of the method used have to be taken into account. The range of ligand concentrations and stability constants that may be detected is defined on one hand by the complexing coefficient of DMG (α_{DMG}, see theoretical section) and on the other hand by the concentration range of Co in the titrations (typically 2−20 nM). Only small concentrations of ligands giving complexing coefficients in the range of α_{DMG} can be detected. Furthermore, only filtered samples (<0.45 μm) have been used for speciation determination, so that the role of various colloidal size ligands cannot be distinguished.

Ligands binding Co may therefore include humic and fulvic acids, other more specific organic ligands, and inorganic colloids. Our results indicate that the concentrations of natural organic ligands available for Co complexation are just in the same range as the natural total dissolved cobalt concentrations in most samples and that the total ligand concentrations are correlated with the total dissolved Co concentrations. Humic and fulvic acids, which are certainly present in these waters, would provide a number of complexing groups in large excess of Co(II). Humic or fulvic acids, in the concentration range 1−3 mg C/L, would correspond to about $1-15 \times 10^{-6}$ mol/L of functional groups (*31, 32*). Titration of water samples with Co in the nanomolar range would not saturate these functional groups. Stability constants for Co(II) with humic and fulvic acids reported in the literature appear to be in a lower range than those determined here for unknown ligands (*31−34*). According to calculations using the WHAM database (*32*), only a few percents of the dissolved Co(II) are bound to fulvic and humic acids under representative conditions. Binding of Co to humic and fulvic acids would therefore in our case be restricted to a small number of Co-specific sites in the range of the Co titrations. Colloidal manganese oxides would mostly bind Co after its oxidation to Co(III), which is not likely to react fast enough with DMG.

Our results point thus to the hypothesis that small concentrations of specific Co(II) ligands are present. The relationship between ligand and Co concentrations may indicate that the specific ligands and cobalt originate from the same source. The ligands may be for example degradation products of cobalamine, released by organisms upon degradation, and be therefore very specific for Co or also be porphyrin-type ligands. Nolan et al. (*3*) reported that Co in the form of B12 is quite stable in seawater and that it is

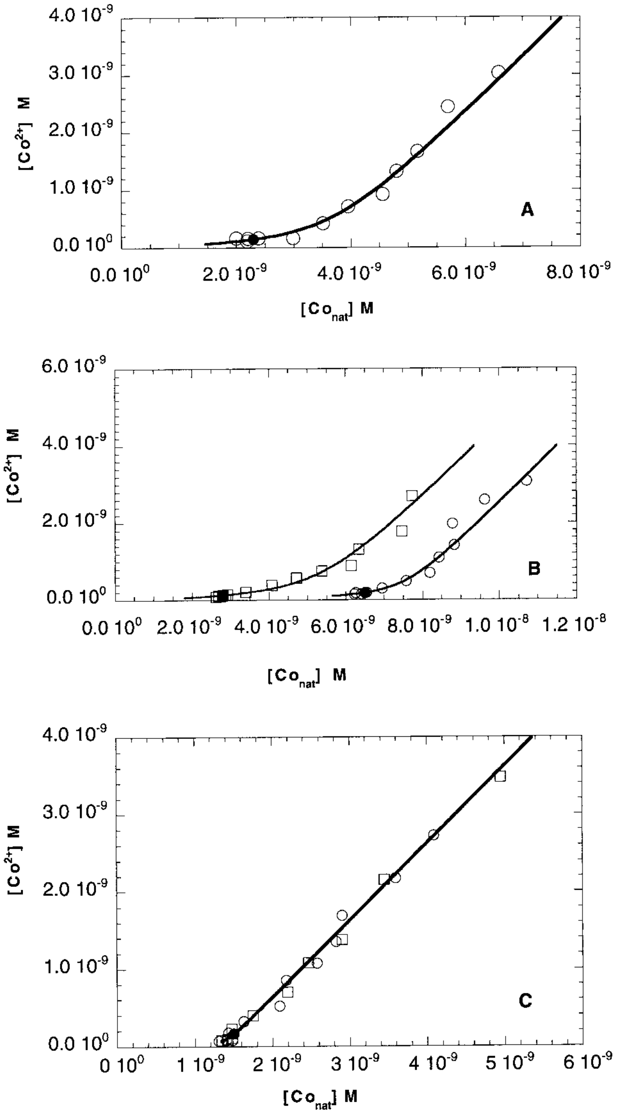

FIGURE 5. Titration curves of natural water samples in terms of [Co^{2+}] as a function of [Co$_{nat}$] ([Co$_T$] − Σ[CoDMG$_i$]). The open points were measured; the solid points indicate ambient [Co$_T$], the curves were calculated with the fitting parameters shown in Table 1. The samples were taken from 25 m depth of Lake Biel on January 8, 1997. (Panel A) From upstream (○) or downstream (□) of the Gösgen effluent site in the Aare River on December 11, 1996. (Panel B) From 2 m depth of Lake Lucerne on February 12, 1997. (Panel C) Determined in the presence of 2.5 μM DMG (○) or 2.0 μM DMG (□).

preferentially taken up, even by phytoplankton. The ligands detected here may play a role for the availability of Co to aquatic organisms.

The total dissolved Co concentrations in the Swiss lake and river waters lie in the same range as those reported for other freshwater systems (*14, 27, 35*−*37*). Only small differences in dissolved Co were observed as a function of depth in Lake Biel, with a small increase with depth in September. In contrast to several other lakes in which Co cycling has been studied (*27, 35, 37*), no oxic/anoxic boundary occurs within the water column of Lake Biel, in which oxygen >4 mg/L is usually present throughout the water column. The cycling of Mn(II)/Mn(IV) is not effective in this lake, and therefore no association of Co with Mn on the basis of redox interactions may be expected.

The available data are not sufficient to compare the Co complexation under different environmental conditions. The effects of discharge from nuclear installations on Co speciation have not been systematically observed and are difficult to evaluate, because their water chemistry changes considerably from discharge to discharge. More samples would be needed to ascertain any effects on the Co complexation.

Only few studies on the Co complexation in freshwater are available for comparison. The fractions of organically complexed Co in our freshwater samples are similar to those in the Scheldt Estuary, which were determined by the same technique (*13*). The conditional stability constants observed here are however 5−6 orders of magnitude smaller than those in the Scheldt Estuary, and the ligand concentrations are somewhat higher. These differences may reflect the presence of much stronger ligands in the Scheldt Estuary, for example, due to the occurrence of anoxic conditions in some parts of the estuary, or may be related to the influence of the Mn redox cycle. We also noticed that Zhang et al. (*13*) worked at a different detection window with higher concentration of added ligands and that they obtained only linear titration curves. In comparison, the curvature found in our titration curves (Figure 4) indicates that we indeed detected reversible Co(II) complexes. In a study on Co complexation in the River Vienne (*14*), retention of complexes by various resins and kinetics of exchange with radiotracers were used. The results indicated that 80−90% of Co was complexed by organic ligands and that these complexes were related to a specific fraction of the organic matter, similarly to the present study.

With regard to the fate of ^{60}Co, this study shows the significance of organic Co complexes. Assuming similar behavior of both radioactive and stable isotopes and allowing sufficient time for equilibration, the results obtained for stable Co should be applicable for radiocobalt. Although we have not yet clearly demonstrated that the speciation of ^{60}Co is the same as that of the more abundant stable Co, it seems likely that ^{60}Co would exchange easily with the ligands observed. These ligands are present in large excess in comparison to ^{60}Co. A large fraction of ^{60}Co is thus also expected to be complexed by organic ligands. The occurrence of strong complexation of Co in solution may thus compete with the adsorption by particles and result in rather low partition coefficients of Co between particles and solution. Typically, partition coefficients between particles and solution for ^{60}Co have been observed in the range $K_D < 1 \times 10^4$ L/kg in the lake and river waters described here (*5*−*7*). In consequence, ^{60}Co may be only weakly retained in the lake sediments and be transported with river water over large distances. The results on Co complexation are of great importance because they are the basis for the evaluation of possible contamination paths by ^{60}Co, for dose rate calculations in the aquatic system, and for accident management.

Acknowledgments

We are grateful to Gerrit Goudsmit, Alfred Lück, Yvo Weidmann, and Daniel Steiner for their help with sampling; to Andrea Ulrich and David Kistler for the determination with ICP−MS; and to Thomas Rüttimann for the determination of inorganic anions and DOC. We would like also to express our acknowledgment to Prof. Xiaoquan Shan, in RCEES, Beijing, for initiating this study in collaboration with EAWAG. A.A. would like to thank for the position support by the Swiss Nuclear Safety Inspectorate (HSK).

Literature Cited

(1) Suzuki, Y.; Nakahara, M.; Nakamura, R.; Ueda. T. *Bull. Jpn. Soc. Sci. Fish.* **1979**, *45*, 1409−1416.
(2) Ueda, T.; Suzuki, Y.; Nakamura, R.; Nakahara, M. *Impacts of radionuclide releases into the marine environment*; IAEA: Vienna, 1981; pp 371−380.
(3) Nolan, C. V.; Fowler, S. W.; Teyssie, J. L. *Mar. Ecol. Prog. Ser.* **1992**, *88*, 105−116.
(4) Mundschenk, H. *J. Environ. Radioact.* **1996**, *30*, 215−232.

(5) Albrecht, A.; Reichert, P.; Beer, J.; Lück, A. *J. Environ. Radioact.* **1995**, *28*, 239−269.
(6) Albrecht, A.; Goudsmit, G.; Qian, J.; Sigg, L.; Xue, H. B.; Kobler, D.; Lück, A.; Weidmann, Y. *EAWAG News* **1997**, *43*, 14−17.
(7) Albrecht, A.; Goudsmit, G.; Zeh, M. *Limnol. Oceanogr.* Submitted for publication.
(8) Bowen, H. J. M. *Trace elements in biochemistry*; Academic Press: London, 1966.
(9) Kendrick, M.; May, M.; Pilshka, M. J.; Robinson, K. D. *Metal in biological systems*; Ellis Horwood: Chichester, 1992; pp 71−79.
(10) Leonard, K. S.; McCubbin, D.; Harvey, B. R. *Sci. Total Environ.* **1993**, *130/131*, 237−251.
(11) Leonard, K. S.; McCubbin, D.; Harvey, B. R. *J. Environ. Radiochem.* **1993**, *20*, 1−21.
(12) Cosovic, B.; Degobbis, D.; Bilinski, H.; Branica, M. *Geochim. Cosmochim. Acta* **1982**, *46*, 151−158.
(13) Zhang, H.; Van Den Berg, C. M. G.; Wollast, R. *Mar. Chem.* **1990**, *28*, 285−300.
(14) Garnier, J.-M.; Pham, M. K.; Ciffroy, P.; Martin, J.-M. *Environ. Sci. Technol.* **1997**, *31*, 1597−1606.
(15) Donat, J. R.; Bruland, K. W. *Anal. Chem.* **1988**, *60*, 240−244.
(16) Zhang, H.; Vire J.-C.; Patriarche, G. J.; Wollast, R. *Anal. Lett.* **1988**, *21*, 1409−1424.
(17) Bobrowski, A. *Talanta* **1994**, *41*, 725−729.
(18) Vega, M.; van den Berg, C. M. G. *Anal. Chem.* **1997**, *69*, 874−881.
(19) Westall, J. C. *FITEQL: A program for the determination of chemical equilibrium constants from experimental data*. Technical Report; Oregon State University: Corvallis, 1982.
(20) Sillen, L. G.; Martell, A. E. *Stability Constants of Metal-Ion Complexes*; Suppl. No. 1, Special Publication 25; The Chemical Society: London, 1971.
(21) Martell, A. E.; Smith, R. M. *Critical stability constants;* Plenum Press: New York, 1974.
(22) Fouillac, C.; Criaud, A. *Geochem. J.* **1984**, *18*, 297.
(23) Good, N. E.; Winget, G. D.; Winter, W.; Connolly, T. N.; Izawa, S.; Raizada, M. M. *Biochemistry* **1966**, *5*, 457−462.
(24) Xue, H. B.; Sigg, L. *Anal. Chim. Acta* **1994**, *284*, 505−515.
(25) Xue, H. B.; Sigg, L. *Anal. Chim. Acta* In press.
(26) Xue, H. B.; Oestreich, A.; Kistler, D.; Sigg, L. *Aquat. Sci.* **1996**, *58*, 69−86.
(27) Lienemann, C.-P.; Taillefert, M.; Perret, D.; Gaillard, J.-F. *Geochim. Cosmochim. Acta* **1997**, *61*, 1437−1446.
(28) Murray, J. W.; Dillard, J. G. *Geochim. Cosmochim. Acta* **1979**, *43*, 781−787.
(29) Moffett, J. W.; Ho, J. *Geochim. Cosmochim. Acta* **1996**, *60*, 3415−3424.
(30) Gerloch, M.; Constable, E. C. *Transition metal chemistry*; VCH: Weinheim, 1994.
(31) Tipping, E.; Hurley, M. A. *Geochim. Cosmochim. Acta* **1992**, *56*, 3627−3641.
(32) Tipping, E. *Comput. Geosci.* **1994**, *20*, 973−1023.
(33) Westall, J. C.; Jones, J. D.; Turner, G. D.; Zachara, J. M. *Environ. Sci. Technol.* **1995**, *29*, 951−959.
(34) Van Loon, L. R.; Granacher, S.; Harduf, H. *Anal. Chim. Acta* **1992**, *268*, 235−246.
(35) Balistrieri, L. S.; Murray, J. W.; Paul, B. *Limnol. Oceanogr.* **1992**, *37*, 529−546.
(36) Quemerais, B.; Lum, K. R.; Lemieux, C. *Aquat. Sci.* **1996**, *58*, 52−68.
(37) Balistrieri, L. S.; Murray, J. W.; Paul, B. *Geochim. Cosmochim. Acta* **1994**, *58*, 3993−4008.

Received for review November 20, 1997. Revised manuscript received April 1, 1998. Accepted April 20, 1998.

ES971018L