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A new method for the quantification of different redox-species of molybdenum (V and VI) in seawater

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ABSTRACT

A new method for the direct determination of reduced and oxidized Mo species (Mo (V) and Mo (VI)) in seawater was developed and used for the first time. The method includes the complexation of Mo (V) with tartrate, solid phase extraction of the Mo (V)–tartrate complex by a XAD 7HP resin, followed by elution with acidic acetone. In this study, the eluted Mo (V) was quantified by graphite furnace atomic absorption spectrometry. The detection limit of this protocol was on the order of 0.2 nM. The analytical precision was 10% of ~10 nM. This method was successfully applied to the determination of Mo (V) and Mo (VI) in surface and bottom waters at the head of Peconic River Estuary. Total Mo (Mo (V) + Mo (VI)) ranged from 100–120 nM in most bottom saline waters, and 2.5–15 nM for surface fresher waters. Concentrations of Mo (V) in these environments ranged from 0 nM to ~15 nM, accounting for 0%–15% of the total dissolved Mo pool. The time series experiments showed that the Mo speciation changed within 1 h after the water collection, and therefore it is strongly suggested that speciation analysis be carried out within the first 15 min. However, since these are the first Mo speciation data in concentration ranges typical of normal marine and coastal waters, additional research may be required to optimize the methodology and further explore Mo cycling mechanisms.

1. Introduction

Molybdenum (Mo) is the most abundant transition trace metal in the ocean, and displays conservative behavior with depth (about 107 nM) (Collier, 1985; Morris, 1975), although slight depletion in Mo concentrations are observed in surface waters (e.g., Yamazaki and Gohda, 1990; Adelson et al., 2001; Tuit and Ravizza, 2003). In contrast to open ocean waters, a sharp depletion in Mo levels occurs in the deep waters of the Black Sea and Cariaco Basin (Bertine, 1972; Emerson and Huested, 1991). Reduction of Mo is expected to occur in those waters, although the actual amount of reduced Mo has never been reported. Molybdenum (VI) is generally assumed to be the dominant oxidation state of Mo in oxic natural waters, while Mo (V) is thermodynamically stable in reducing waters (Bertine, 1972; Brookins, 1988).

Molybdenum is an essential trace element for plants, animals and microorganisms (e.g., Bortels, 1930), required for different metalloenzymes that execute key transformations in the biogeochemical cycles of nitrogen, sulfur and carbon (e.g., Mendel 2005). For example, Mo is part of several cofactors required for nitrogenase and nitrate reductase (e.g., Fogg and Wolfe, 1954; Fogg, 1962; Mendel, 2005), which catalyze the reduction of N_2 and nitrate to bioavailable N in the ocean (e.g., Fogg and Wolfe, 1954; Mendel, 2005). In fact, more than 60 enzymes have been identified that contain Mo (Spriro, 1985; Stiefel and Cramer, 1985; Kramer et al., 1987; Smith et al., 1988; Palmer and Reedijk, 1991; Lippard et al., 1994; Hille, 1996; Stiefel, 1997; Kisker et al., 1997).

The lack of Mo may limit the growth of phytoplankton (e.g., Howarth and Cole, 1985; Ter Steeg et al., 1986; Wallen and Cartier, 2008). Bacteria and algae can acquire Mo as molybdate using different surface transporters, siderophores and the sulfate and phosphate uptake systems (Heuwinkel et al., 1992; Marschner, 1995; Liamas et al., 2000; Pau and Lawson, 2002; Liermann et al., 2005; Fitzpatrick et al., 2008). However, Mendel (2005) suggests that molybdate is not biologically active until Mo is incorporated into different enzymes. Electron paramagnetic resonance has confirmed the existence of Mo (V) in biological samples (e.g., Godfrey et al., 1984; Kaul et al., 1985; Sears et al., 1995; Canne et al., 1997; Raitsimring et al., 2008), suggesting that this reduced form of Mo may be more bioavailable in aquatic environments (e.g., Howarth and Cole, 1985; Howarth et al., 1988). Therefore, as reported for other trace metals, the bioavailability of Mo is probably more dependent on the relative abundance of different chemical species than on the total Mo concentration.

Although many studies have reported the levels of total Mo concentrations in different aquatic systems (e.g., Collier, 1985; Emerson

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and Huested, 1991; Windom and Niencheski, 2003; Dalai et al., 2005; Dellwig et al., 2007; Chappaz et al., 2008), none of them have reported the concentrations of the different redox species of this element in those environments. In the limited work published on Mo speciation (e.g., Sugio et al., 1988; Ahmed and Haque, 2002), Mo (V) concentrations were on the order of several mM, orders of magnitude higher than the total concentrations found in natural waters (5 nM in average world rivers and 107 nM in seawater, Martin and Meybeck, 1979; Collier, 1985). These methods are only suitable for analyzing samples with high concentrations of Mo such as mine effluents (Sugio et al., 1988), but are inapplicable for quantifying the extremely low levels of Mo (V) expected in marine systems.

In this study, a method based on solid-phase extraction was developed to measure the Mo speciation (V and VI) in seawater. The analytical protocol was thoroughly investigated, and used to examine surface and bottom waters at the head of the Peconic River Estuary in Long Island, New York.

2. Materials and methods

2.1. Method description

The analytical protocol established for separating Mo (V) and Mo (VI) in natural waters is shown in Fig. 1. In this protocol, a 60 ml water sample was collected and the pH adjusted to 7.0 with a phosphate buffer (pH = 7.0, 0.25 M KH_2PO_4 and 0.25 M Na_2HPO_4) and a small amount of perchloric acid when the seawater pH was higher than 8.0. The reason perchloric acid was used instead of nitric was because the latter oxidized the reduced Mo (results not shown). The effect of other acids, such as HCl, on the chemical speciation of Mo was not evaluated in this study. The Mo (V) in the pH-adjusted sample, and not Mo (VI), was complexed with tartrate (1.0 ml of 10% tartrate solution) and extracted with 2.0 g of pretreated XAD-7HP resin packed in a poly-prep column at a flow rate of 2 ml/min until dry (Fig. 1) to ensure the complete adsorption of Mo (V) onto the resin. Because this resin is selective for the Mo (V)–tartrate complex, the water sample was collected after passing through the resin for the subsequent analysis of Mo (VI). Mo (V)–tartrate was then eluted with 40 ml (8 elutions of 5 ml each) 1 N acidic acetone (10% of concentrated nitric acid in acetone, v/v) to completely elute Mo (V). These eluents containing Mo (V) were dried on a hot plate, redissolved with 5 ml 0.1 N nitric acid and quantified by GF-AAS.

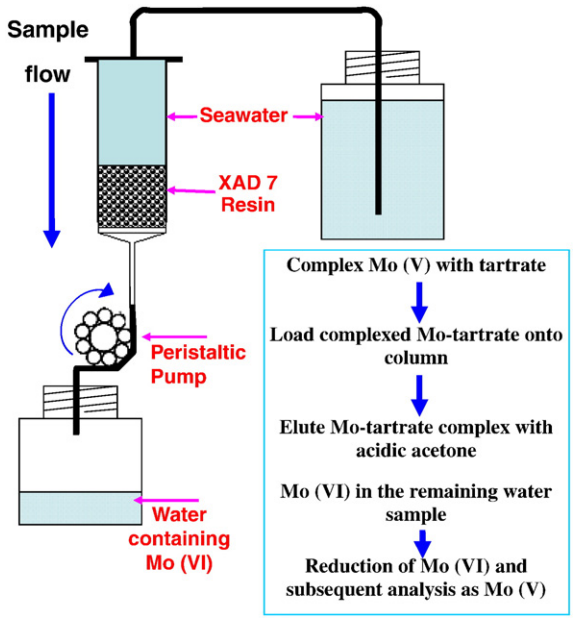

Fig. 1. Schematic of the analytical protocol for separating Mo (V) from Mo (VI) in natural waters.

The eluents containing Mo (VI) were reduced to Mo (V) by adding the reducing agent (0.5 ml 10% (w/v) $SnCl_2$ in 0.1 N HCl), and buffering to pH 7, and analyzed using the protocol described for Mo (V). The total Mo concentration was obtained by adding both Mo (V) and Mo (VI). The detection limit of this protocol was on the order of 0.2 nM for both species of Mo. The analytical precision was ~10% for samples with 10 nM Mo.

2.2. Preparation of standard solutions for Mo speciation recovery experiments

Mo (VI) stock solutions were prepared by dissolving 184.0 mg ammonium molybdate tetrahydrate in 100 ml Milli-Q water, and subsequently standardized by GF-AAS. Mo (V) stock solutions were prepared by dissolving ~30 mg molybdenum (V) chloride in 100 ml 0.1 N perchloric acid (due to the fact that Mo (V) is easily oxidized when exposed to the atmosphere and reacts with water, caution should be taken when making this solution). Standard solutions of Mo (VI) and Mo (V) were prepared by diluting the stock solution in synthetic seawater (degassed for at least 0.5 h with nitrogen gas) to obtain the following Mo species concentrations: 100 nM Mo (VI) only; 100 nM Mo (V) only; and 100 nM of both Mo (V) and Mo (VI). It should be noted that, in contrast to previous studies where several mM of Mo were used (e.g., Sugio et al., 1992), the 100 nM level used in this analytical protocol reflects the natural concentrations of Mo found in seawater.

Tartrate solution was prepared by dissolving 10 g potassium sodium tartrate tetrahydrate in 100 ml Milli-Q water. The phosphate buffer was prepared by dissolving potassium dehydrogenate phosphate and disodium hydrogen phosphate in Milli-Q water to a final concentration of 0.25 M for each solute. The buffer solution was adjusted to pH 7 with 10 N NaOH. All of the chemicals used were analytical reagent grade or the highest purity available (e.g., HPLC-grade absolute acetone).

2.3. The use of tartrate as a ligand for Mo (V)

Mo (V) is not thermodynamically favored under oxic conditions. However, Malthouse et al. (1980, 1981) and Yamanouchi et al. (1977) reported the existence of Mo (V) in biological samples as a dominant species. The exact form of Mo (V) in natural waters is not well understood. Bertine (1972) suggested that Mo (V) may exist in natural waters as Mo_3O_8 (a mixture of Mo (V) and Mo (VI)). However, Mo_3O_8 is usually crystalline (Titley and Anthony, 1961). Szilágyi (1967) suggested that MoO_2^+ occurred when Mo (VI) was reduced by humic acids. The structure of this Mo (V) compound was confirmed by measuring the distance of the Mo–O complex (Spivak and Dori, 1975; Stiefel, 1987). In addition, Mo (V) has a strong tendency to dimerise (Bard et al., 1985). Therefore, as previously reported (e.g., Yamanouchi et al., 1977; Coughlan, 1980; Modec and Brenčič, 2002; 2004; Modec, 2008), Mo (V) most likely exists as $Mo_2O_4^{2+}$ in natural environments.

$Mo_2O_4^{2+}$ can be easily complexed with many organic ligands (e.g., EDTA and oxalate; Bard et al., 1985; Modec and Brenčič, 2004). Such ligands usually yield either polymeric materials or discrete entities (Modec and Brenčič, 2002, 2004). During the development of this analytical protocol, we found that Mo (VI) also complexed with oxalate and EDTA, although not as strongly as Mo (V) (results not shown). Therefore, Mo (V) could not be completely separated from Mo (VI). Compared to oxalate, the dicarboxylic acid tartrate has fewer functional groups (Modec and Brenčič, 2002, 2004) suitable only for the bonding of Mo (V). Ahmed and Haque (2002) further reported that tartrate can form complexes only with Mo (V) and not Mo (VI). Therefore, tartrate was used as the complexing ligand to selectively extract Mo (V) from the water samples.

2.4. XAD resin preparation

By yielding higher enrichment factors, greater efficiency and handling simplicity (Soylak et al., 1997; 2001), solid phase extractions (SPE) have some advantages over other techniques used to isolate Mo species at high concentrations (e.g., spectrophotometric and colorimetric techniques). Furthermore, the XAD resin used in this SPE analytical protocol is of high purity and has good sorption properties including porosity, durability, uniform pore distribution, and high surface area (e.g., Soylak et al., 1997, 2001; Tewari and Singh, 2001). Such amberlite XAD-copolymers have been widely used for the preconcentration of trace metal ions (e.g., King and Fritz, 1985; Soylak et al., 1997; Tunceli and Türker, 2004; Soylak et al., 2001). In both the previous and the present applications, the trace metal ions were first converted into metal chelates or inorganic complexes, then absorbed on the XAD resin, and finally eluted with different eluting agents (e.g., Soylak et al., 2001). The amberlite XAD-7HP resin has recently been used for the isolation of specific organic complexes from aqueous solutions (Graetz and Volk, 1983; Miyata et al., 1996; Chang et al., 2001; Goslan et al., 2002; Ken'Ichi, 2005; Kim et al., 2006; Sasaki et al., 2007). As a nonionic aliphatic acrylic polymer, amberlite XAD-7HP has proved to be a useful adsorbent for some organic compounds such as ester, ketones or aliphatic molecules (e.g., Kim et al., 2006; Lai et al., 2005; Shimizu and Li, 2005). The XAD-7HP resin was therefore used to isolate Mo (V)–tartrate complexes from dissolved Mo (VI) in seawater for this study.

The XAD-7HP resin was first preconditioned and purified to eliminate trace metal ions and other contaminants using the method of Soylak et al. (2001). In this method, the XAD-7HP resin was washed with methanol, Milli-Q water, 1 N nitric acid in acetone, Milli-Q water, 1 N NaOH, Milli-Q water, and finally an acetone and phosphate buffer. A preconcentration system was set up using a peristaltic pump with rotor heads (Cole-Parmer, Masterflex, Model 7553-12, 1–100 rpm), poly-prep columns and Teflon tubing that extended from the head of the columns into the bottom of 60 ml polycarbonate bottle reservoirs (Fig. 1).

2.5. Graphite Furnace Atomic Absorption Spectrometry

Graphite Furnace Atomic Absorption Spectrometry (GFAAS) offers the necessary sensitivity for Mo measurements (Johnson et al., 1973; Morrice et al., 1989). In this study, Mo concentration was determined by GF-AAS using a Perkin-Elmer spectrometer (AAnalyst-800) with a longitudinal Zeeman-effect background correction system, a transversely-heated graphite atomizer (THGA), and an auto-sampler (Perkin-Elmer, AS-800). The hollow cathode lamp (Perkin-Elmer, PN: N305-0186) for Mo was operated at 15 mA, with a slit of 0.7 nm and a 313.3 nm wavelength. The volume injected into the graphite tube was 20 µl for samples and calibration solutions, and high-purity argon (UHD grade) was used as the purge gas. The thermal program used in the GF-AAS was the following (Table 1): drying at 130 °C for 30 s, pyrolyzing at 1500 °C for 20 s, atomizing at 2450 °C for 5 s, and cleaning at 2500 °C for 5 s. The gas flow rate was set to 250 ml/min.

2.6. Stability of Mo (V)

In order to establish whether changes in Mo speciation occur during the solid-phase extraction, the stability of Mo (V) at pH = 7.0 was determined. For this objective, a bottom water sample (temperature = 22 °C, salinity = 14, pH = ~7.5, and dissolved oxygen (DO) = ~10 µM) was collected at the head of the Peconic River Estuary. The water samples (250 ml, two replicates) were immediately adjusted to pH = 7.0 with perchloric acid, and incubated in the lab at room temperature (20 °C). Mo (V) was then determined at different time intervals (up to 4 h after collection) following the method described above.

The stability of naturally occurring Mo (V) was also monitored to determine if biological activity occurring inside the sample bottles affected Mo speciation during the handling and operation by conducting experiments under different temperatures. Water samples (1 l) were collected from the head of the Peconic River Estuary (same hydrological condition as above) and incubated at 20 °C and at 0 °C (two replicates). Mo (V) concentrations were measured according to the protocol described above at different time intervals over 24 h.

2.7. Determination of Mo speciation in natural water samples

Surface and bottom water samples were collected for Mo speciation measurements at the head of the Peconic River Estuary where conditions are strongly affected by sewage effluents, tidal intrusion, and frequent algal blooms (e.g., Breuer et al., 1999; Gobler et al., 2005, 2008). Bottom water samples were collected weekly from June to October 2006, and surface water samples were collected in September 2007 during the summer anoxia at the upper Peconic River and the head of the Peconic River Estuary (Breuer et al., 1999). Bottom water samples were collected with a peristaltic pump through Teflon tubing extended on a plastic pole and lowered to the depth of 0.2 m above the bottom. Surface water samples were collected from 0.2 m below the surface. Dissolved samples for Mo speciation were obtained by filtration through a trace metal-clean, polypropylene capsule filter (0.2 µm). All sampling materials used in this study were prepared using trace metal-clean techniques (e.g., Flegal et al., 1991). Note to the editor: Trace-metal clean techniques are established protocols for this type of studies. Plastics are metal-free but not trace metal clean. Trace metal clean protocols use extensive acid-washes of metal free materials.

Water samples for Mo (V) and Mo (VI) speciation were immediately processed on site for Mo (V), and analyzed in the laboratory within a week for Mo (VI). The eluents were stored at −10 °C in a freezer until analyzed by GF-AAS. Salinity, pH (NBS), temperature and dissolved oxygen in all the water samples were also determined using analytical probes and standard protocols.

3. Results and discussion

3.1. Recovery experiments in synthetic seawater

In order to establish the accuracy of the Mo speciation technique, a series of recovery experiments were conducted with spiked Mo (V) and Mo (VI) concentrations typical of oceanic waters (Fig. 2). The average recovery of the Mo species was 101% for Mo (V) and 93% for Mo (VI) when added together, and 94% (Mo (V)) and 108% (Mo (VI)) when added alone (Fig. 2). These are excellent recoveries suggesting that Mo (V) ions could be analytically isolated from Mo (VI). Naturally occurring Mo (V) species are expected to be more stable than the free Mo (V) ions due to complexation with organic ligands, and most of these natural complexes should also be retained on the exchange column.

Since there is no certified reference material for Mo species available yet, the currently available certified reference seawater, CASS-4, was analyzed for Mo (V) and total Mo. CASS-4 has a certified total Mo concentration of 91.5 ± 9.0 nM. The total Mo concentration

Table 1
Optimized graphite furnace temperature program for the determination of molybdenum.

Step	Temperature (°C)	Ramp time (s)	Hold time (s)	Gas flow (ml/min)
Dry 1	110	1	30	250
Dry 2	130	15	30	250
Pyrolysis	1500	10	20	250
Atomization	2450	0	5	0
Cleanout	2500	1	5	250

obtained using the speciation method was 86.5 ± 3.1 nM; no Mo (V) was detected in the SRM. However, the CASS-4 seawater was collected in 1988 from coastal waters off Halifax, Canada and acidified for more than 18 years (pH ~ 1.6) (NRC, 1999). It is reported that MoO_4^{2-} is subject to successive protonation up to the formation of the possible cationic species, $MoO_2(OH)(H_2O)_3^+$ under acidic conditions at Mo concentrations of less than 10^{-4} M (e.g., Cruywangen and Rohwer, 1975; Pettersson et al., 1986; Tytko et al., 1985). The dominance of Mo (VI) in the acidified SRM, therefore, is likely due to the formation of the cationic species.

Spike experiments were also conducted using natural seawater collected from the Atlantic Ocean about 5 miles off Southampton, NY. The speciation protocol showed that the water sample contained only Mo (VI) with a concentration of 117 nM (Fig. 3). After spiking the water sample with 117 nM of only Mo (V), 84% of the added Mo (V) (101 nM) was recovered (Fig. 3). The amount of Mo (VI) present in the sample also increased slightly to 124 nM, consistent with the fact that free Mo (V) ions are easily oxidized to Mo (VI) (oxidation half-time of ~30 min; Sugio et al., 1992).

3.2. Stability of Mo (V)

The short-term stability study showed that the levels of Mo (V) in the water sample collected in the Peconics changed rapidly after collection with a pH adjusted to 7.0 (Fig. 4). The results suggest that at a pH of 7.0 the reduced form of Mo varied with time in excess of the possible measurement uncertainty. Mo (V) was initially about 2.5 nM and almost doubled within an hour, and afterwards was followed by a slight decline that suggests a complex system with multiple processes operating. These results indicate that the speciation analysis has to be carried out within the first 15 min of the water collection to remain within measurement uncertainty (based on a linear extrapolation of 1 and 0.5 h concentrations to time 0). In contrast to Mo (V), the Mo (VI) concentration first declined and then increased slightly (Fig. 4). An inverse relation between Mo (VI) and Mo (V) suggests a dynamic behavior of Mo species, although it is still not entirely clear what mechanisms are responsible for that trend. Furthermore, comparable results were obtained in the different temperature incubation experiments with initial Mo (V) of ~2.5 nM (0° and 20 °C) (Fig. 5). Mo (V) similarly varied with time showing an inverse relationship with Mo (VI), suggesting that redox reactions occurred rapidly and with possibly undergoing multiple processes. From our results, where temperature dependent biological effects were minimized, whatever is acting as the reductants present in the water (e.g., humic substances, Szilágyi, 1967; Bertine, 1972) may be critical for Mo speciation. Regardless of the mechanism of reaction, the separation of Mo (V) and Mo (VI) should be conducted on site as soon as possible.

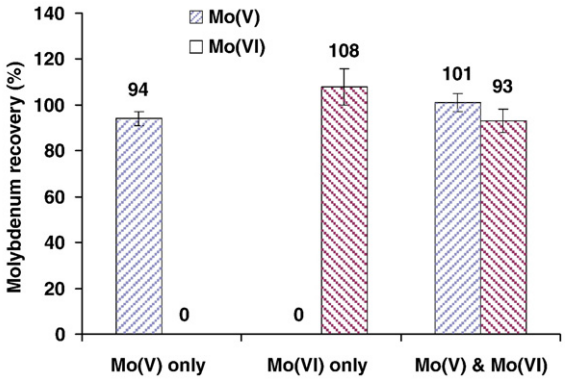

Fig. 2. Molybdenum recoveries of Mo (V) only, Mo (VI) only, and both Mo (V) and (VI) in synthetic seawater (All additions = 100 nM; Error bars indicate the standard deviation, $n = 4$).

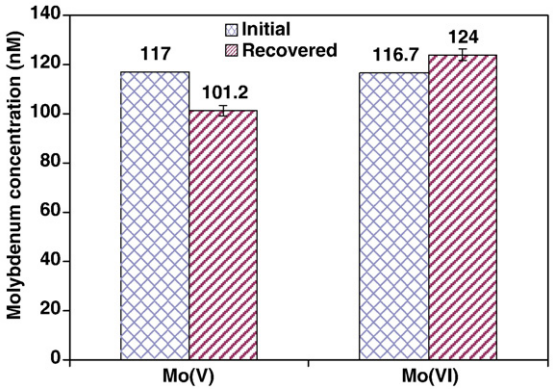

Fig. 3. Recovery of Mo (V) in spiked seawater samples (The water sample was collected in the Atlantic Ocean, ~5 miles offshore of Southampton, NY; The initial Mo is essentially all Mo (VI) with a concentration of 116.7 nM as the initial bar Mo (VI); The initial bar Mo (V) represents the Mo (V) added to the seawater samples. The other bars are the recoveries of the spikes).

3.3. Mo (V) and Mo (VI) concentrations in estuarine waters

The new method was first applied to measure Mo speciation in surface and bottom waters collected at the head of the Peconic River Estuary during the summer of 2006 when water column anoxia occurred. The speciation of both Mo (V) and Mo (VI) were investigated in bottom waters (DO = 0–10 μM, which are considered suboxic (<10 μM O_2) or anoxic (<1 μM O_2) as defined by Tyson and Pearson (1991), as well as in surface oxic waters (DO = 150–180 μM).

Consistent with other reports (e.g., Prange and Kremling, 1985; Collier, 1985; Martin and Meyback, 1979), total dissolved Mo (as Mo (V) + Mo (VI)) at the head of the Peconic River Estuary was relatively high in most bottom saline water (100–120 nM) but was low in the surface water (2.5–15 nM) (Fig. 6). The lowest levels of total dissolved Mo (2.5–4.0 nM) were measured in the oxygenated surface fresh-waters in the upper Peconic River (salinity: 0–5). These Mo concentrations are consistent with the values reported for lake waters in Eastern Canada (0.1–3.4 nM, Chappaz et al., 2008), the Chao Phraya

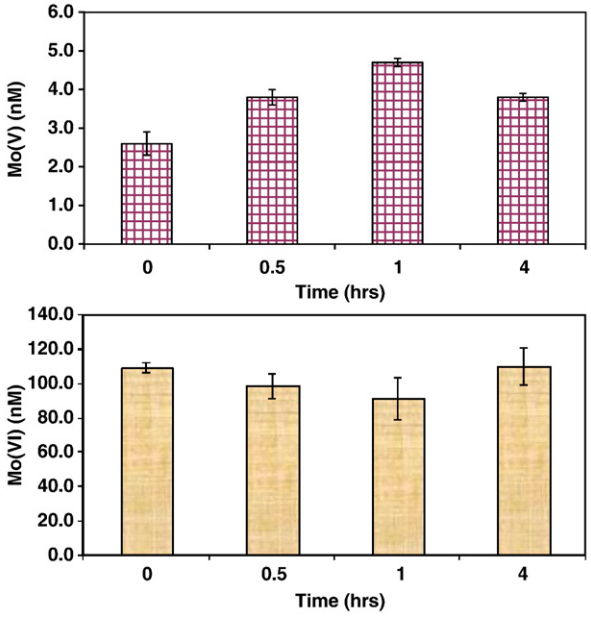

Fig. 4. Changes in Mo (V and VI) concentrations with time after adjusting the sample pH to 7.0 (Error bars are standard deviation, $n = 2$. The samples were bottom waters collected at the head of Peconic River Estuary in September 2007 with dissolved oxygen levels ~10 μM and pH = 7.5).

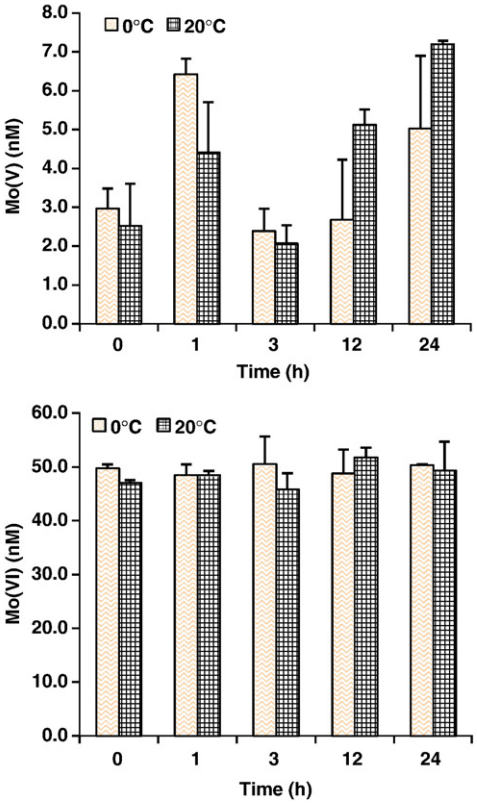

Fig. 5. Changes in Mo (V and VI) concentrations in water samples incubated at 0 and 20 °C for up to 24 h (Error bars are standard deviation, $n=2$; The samples were bottom waters collected at the head of Peconic River Estuary with DO levels of ~10 μM and pH = ~7.5).

River (3.2–4.9 nM, Dalai et al., 2005), and the average world river water (5.0 nM, Martin and Meyback, 1979). The levels of total Mo in surface low salinity water in the Peconics were also lower (total dissolved Mo: 10–15 nM; salinity: ~20) than those measured in the bottom anoxic or suboxic saline waters (Total dissolved Mo: 100–120 nM; Salinity: 15–27) (Figs. 6 and 7). Total Mo measured in the bottom anoxic or suboxic waters showed a non-conservative excess with respect to conservative mixing (Fig. 6). In addition to freshwater dilution (e.g., rain input, sewage effluents, and river runoff), low levels of total dissolved Mo in surface waters may be attributed to large inputs of river-borne particles and sewage effluents, which scavenged Mo (e.g., Brumsack and Gieskes, 1983; Yamazaki and Gohda, 1990; Falkowski, 1983; Paerl et al., 1987; Tuit and Ravizza, 2003), strong phytoplankton and bacterial uptake, and associated adsorption with organic matter from algal blooms during the study period (Breuer et al., 1999; Gobler et al., 2005, 2008). On the other hand, in anoxic or suboxic waters, desorption of Mo from river-borne or suspended materials (Jones, 1974; Dalai et al., 2005; Morford et al., 2005) and diffusion from reducing sediments (e.g., Dellwig et al., 2007) may contribute to the non-conservative excess of Mo.

Generally pH may be strongly influenced by biological activities, such as photosynthesis and remineralization in water column, which, along with O_2, likely affects Mo cycling. In our study, total dissolved Mo concentrations showed an inverse relationship with water pH and a direct relationship with O_2 (Fig. 7). In general, total dissolved Mo decreased from 120 nM in low O_2, bottom saline water with lower pH (~7.0) to less than 20 nM in surface freshwater with higher pH (~7.5) and O_2 (Fig. 7). Between surface water pHs of 7.5 and 8.3, the levels of dissolved Mo were low and less variable (<20 nM). Mo (V) was also low in Peconics, ranging from 0 to 15 nM, accounting for as much as 15% of the total dissolved Mo in some samples (Fig. 8). Mo (V) concentrations were higher in waters with low pH, and lower in

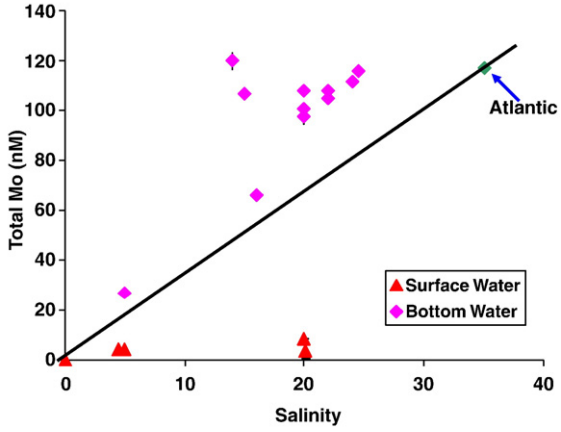

Fig. 6. Total dissolved Mo concentrations (as Mo (V) + Mo (VI)) versus salinity at the head of Peconic River Estuary (The Atlantic seawater sample was collected from ~5 miles offshore of Southampton, NY).

waters with high pH (Fig. 8). In general, Mo (V) is present over a wide range of concentrations in waters of low pH (from 7.5 to 7.0) and low O_2, ranging from 0 to 15 nM.

In surface waters, the levels of Mo (V) were also very low (<2 nM) and apparently independent of water pH during the study, although the low Mo concentrations compromise any strong conclusion regarding pH dependence. Because total Mo concentrations were low in surface waters of relatively low salinity, Mo (V) accounted for a high proportion of the total dissolved Mo (as high as 30% in some samples).

The source of the Mo (V) species is uncertain, although it could be the result of reduction of Mo (VI) in the water column due to the existence of multiple reductants (e.g., Fe^{2+}, S^0 and S^{2-}) (e.g., O'Sullivan et al., 1991; Lloyd, 2003) and/or diffusion from underlying reducing sediments.

4. Conclusion

A new method for the direct determination of reduced and oxidized forms of Mo in seawater was developed. The protocol is based on the complexation of Mo (V) with tartrate under neutral conditions, extraction of this complex with an XAD 7HP resin, elution with acidic acetone, and quantification by GFAAS. The detection limit of this SPE protocol was on the order of 0.2 nM for both species. The analytical precision was ~10%. The method was successfully applied for the determination of Mo (V) and Mo (VI) in surface and bottom waters at the head of the Peconic River Estuary. Consistent with

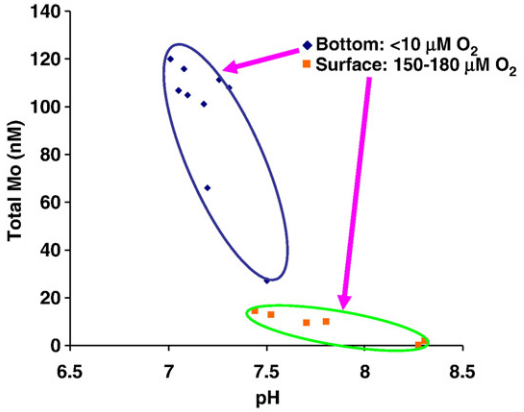

Fig. 7. Total Mo concentrations of Mo (as Mo (V) + Mo (VI)) versus water pH in surface and bottom waters collected at the head of Peconic River Estuary.

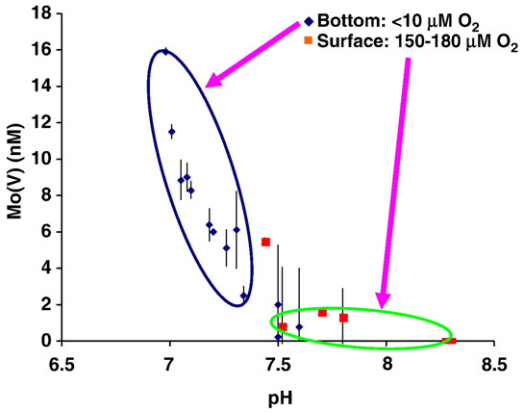

Fig. 8. Mo (V) concentrations versus water pH in surface and bottom waters at the head of Peconic River Estuary.

thermodynamic calculations, the existence of Mo (V) was confirmed in natural waters. Concentrations of Mo (V) ranged from 0 nM to ~15 nM, accounting for 0%–15% of the total dissolved Mo in most waters in the Peconic River estuary. Total Mo (Mo (V) + Mo (VI)) ranged from 100–120 nM with a non-conservative excess in most bottom suboxic or anoxic waters with respect to conservative mixing, and only 2.5–15 nM in low salinity waters in the estuary during the study period. The results also showed that Mo (V) contributed to this excess. Although the source of this reduced Mo is still unknown, benthic remobilization from reducing sediments and reduction of Mo (VI) are the most probable sources.

The results also showed that Mo concentrations and speciation change dynamically in natural waters in response to redox conditions, suggesting that multiple processes are occurring. The method developed here should be an important aid in elucidating the processes involved and serve as a basis for future optimization.

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