

Trace Molybdenum Determination in Drinking Waters by USN-ICP-OES After Solid Phase Extraction on Ethyl Vinyl Acetate Turnings-Packed Minicolumn

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INTRODUCTION

Molybdenum (Mo) is an essential trace element for plants, animals, and humans, and is the only metal of the second transition series which is of major biological significance (1). In animals, it is an indispensable co-factor for several enzymes, such as xanthine oxidase and sulphite oxidase (2, 3). In plants, although present at low levels, Mo is an essential micronutrient and is involved in the biochemical processes related to fixing the N₂ of the atmosphere by bacteria. Nevertheless, it is harmful to plants, animals, and humans at high concentrations.

Although Mo is essential for animals, it becomes toxic at high levels (4, 5). It must be emphasized that studies pertaining to the role of Mo in man or the environment have often been hampered by the lack of sufficiently sensitive analytical methods for determining trace Mo levels. For this reason, it is often difficult to determine whether the symptoms attributed to Mo deficiency or excess are due to biological variations or simply to experimental error (6). A better understanding of the role of Mo in humans, plants, and animals as well as in the environment depends on improving the sensitivity and accuracy of the analytical methods involved (7).

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ABSTRACT

This study presents the development of a novel on-line pre-concentration method for inorganic molybdenum by flow injection solid-phase extraction with detection by inductively coupled plasma optical emission spectrometry and ultrasonic nebulization (SPE-USN-ICP-OES). This method employs a minicolumn filled with ethyl vinyl acetate turnings as the adsorbent material. This system was applied to the on-line pre-concentration and determination of molybdenum in water samples with good selectivity and reproducibility; molybdenum was retained on the minicolumn without further complexation.

The time required for the pre-concentration of a 20-mL sample, injection/data acquisition, and conditioning was about 5.0 minutes, resulting in a sample throughput of 12 samples per hour. A 300-fold total enhancement factor for this sample volume was obtained with respect to the molybdenum determination by ICP-OES without pre-concentration.

The precision for six replicate measurements of a solution containing 20 µg L⁻¹ Mo was 3.5% relative standard deviation, calculated at peak height. The limit of detection (3σ) was 0.04 µg L⁻¹.

This method was successfully applied to trace molybdenum determination in drinking water samples and in a certified reference material (Metals in Natural Waters, NIST CRM 1643e).

One of the major routes of incorporation of Mo is through water intake or uptake. For the analysis of water samples, inductively coupled plasma mass spectrometry (ICP-MS) (8, 9) has the analytical capability to determine Mo at trace and ultra-trace levels because of its high sensitivity and selectivity, and it also offers high sample throughput. However, the cost of the instrumentation is not affordable for many laboratories. Trace Mo determination in water samples has been successfully carried out by atomic absorption spectrometry with electrothermal atomization (ETAAS) (10) and inductively coupled plasma optical emission spectrometry (ICP-OES) (11). The drawback of these two techniques is that the Mo concentration in well water, tap water (12), seawater samples (13), etc., is too low to be directly determined. Pre-concentration is an effective means for extending the detection limits of the ICP-OES technique. However, when practiced manually in the batch mode, the operations are usually too laborious and time-consuming.

Pre-concentration and the determination of trace Mo has been proposed by using N-benzoyl-N-phenylhydroxylamine (14) and α-benzoin oxime (15) as complexing reagents in a liquid-liquid extraction system. Ammonium pyrrolidinedithiocarbamate (APDC) and activated carbon has also been used for the pre-concentration and determination of Mo in silicates by flame AAS (FAAS) (16). Giacomelli et al. (17) determined Mo in steels by ETAAS after

complexation with ammonium salt of dithiophosphoric acid O,O-diethyl ester and adsorption onto activated carbon. Molybdenum determination in natural water samples by ETAAS after enrichment using pyrocatechol violet has also been proposed (18). Molybdenum has been determined by ICP-MS (19) and ICP-OES (20) after complexation using oxine. Thiocyanate was used for the preconcentration and determination of Mo in steel (21). Calmagite and activated carbon has been used for the separation and preconcentration of trace Mo in water samples prior to the determination by ICP-OES (13). In addition, in recent years there have been reports (22-29) utilizing the solid-liquid separation and preconcentration of trace elements using microcrystalline naphthalene loaded with organic reagents.

Alternatively, microcolumns packed with ethyl vinyl acetate (EVA) turnings have been successfully employed for the on-line preconcentration of trace elements. The beneficial use of this novel form of EVA as a sorbent material for As, Se, and Te on-line preconcentration and determination by atomic absorption spectrometry was actually demonstrated by our research group (30,31).

The aim of this work was to develop a sensitive and selective flow injection (FI) preconcentration method for the determination of inorganic Mo in natural waters by on-line SPE, assisted by an EVA turnings-packed microcolumn coupled to ICP-OES with ultrasonic nebulization (USN). Because of the very good chemical resistance and the unlimited lifetime of the EVA material, the column does not need any regeneration or repacking and remains unaltered after the preconcentration or elution step. Moreover, Mo can be retained onto the microcolumn without using a complexing reagent. To the best of our

knowledge, it is the first time that EVA turnings are used as sorbent material for Mo preconcentration. The proposed method was optimized and applied to the selective determination of Mo in natural water samples and a certified reference material.

EXPERIMENTAL

Instrumentation

All measurements were performed with a Model ICP 2070 sequential ICP spectrometer (Baird, Bedford, MA, USA). The 1-m Czerny-Turner monochromator had a holographic grating with 1800 mm⁻¹ grooves. A U-5000 AT ultrasonic nebulizer (CETAC Technologies, Omaha, NE, USA), involving a desolvation system, was used.

The ICP-OES and ultrasonic nebulization (USN) conditions are listed in Table I. Minipuls™ 3 peristaltic pumps (Gilson, Villiers-Le-Bel, France) were used. Sample injection was achieved using a Rheodyne® (Cotati, CA, USA), Model 50, four-way rotary valve. The minicolumn (85 mm long, 4.0 mm i.d.) used as the EVA turnings holder was prepared using cross-linked ethyl vinyl acetate

(EVA) Microline tubing. Tygon® pump tubing (Ismatec, Cole-Parmer Instrument Company, Niles, IL, USA) was employed to propel the sample, reagent, and eluent. The FI system used is shown in Figure 1. The 202.030-nm Mo spectral line was used and the FI system measurements were expressed as peak height emission, which was corrected against the reagent blank.

Reagents

A molybdenum stock solution (1000 mg L⁻¹) was prepared by dissolving 920.3 mg (NH₄)₆Mo₇O₂₄ · 4H₂O to a final volume of 500 mL with water and a few drops of concentrated nitric acid (Fluka).

Ultra-pure water (18 MΩ cm⁻¹) was obtained from an EASY pure RF (Barnstedt, Dubuque, IA, USA).

All other solvents and reagents were of analytical reagent grade or better, and the presence of Mo was not detected in the working range.

Column Preparation

Ethyl vinyl acetate (EVA) is a flexible (rubbery), transparent polymeric material with good low temperature flexibility, good chemical resistance (to acids, alkalis, and

TABLE I
ICP-OES and Ultrasonic Nebulizer Instrumental Parameters

ICP-OES Conditions	
RF Generator Power Plasma	0.8 kW
Frequency of RF Generator	40.68 MHz
Gas Flow Rate	8.5 L min ⁻¹
Auxiliary Gas Flow Rate	1.0 L min ⁻¹
Observation Height (above load coil)	15 mm
Analytical Line for Mo	202.030 nm
Ultrasonic Nebulizer Conditions	
Heating Temperature	140.0 °C
Condenser Temperature	4.0 °C
Carrier Gas Flow Rate	1.0 L min ⁻¹

change under reproducible conditions.

Sample Preparation

Water samples were filtered through 0.45- μm pore size membrane filters immediately after sampling, and the pH was adjusted to pH 7.0 with hydrochloric acid or sodium hydroxide solution, then stored at 4 °C in Nalgene® polyethylene bottles (Nalge Nunc International, Rochester, NY, USA). All glass materials used were washed with 10% (v/v) HNO_3 and ultrapure water before use.

Preconcentration Step

A schematic of the preconcentration manifold is shown in Figure 1. At the beginning of the preconcentration/determination cycles, the minicolumn was conditioned for preconcentration first with 10 mL of HCl solution (1.0 mol L^{-1}) and finally washed with ultrapure water until a neutral pH was achieved (valve V1 in position B).

Loading

Molybdenum solutions were loaded on the EVA minicolumn at a flow rate of 5.0 mL min^{-1} with valve V1 in position S and valve V2 in load position (a), while the HCl solution was passing throughout the manifold directly to the USN and later to the plasma (4 minutes).

Injection

After the loading time, the injection valve V2 was switched on the injection position (b) and the retained metal was quantitatively eluted with the HCl solution at 1.8 mL min^{-1} directly into the USN (0.5 minutes).

Conditioning

After Mo determination was completed, injection valve (V2) was switched back to the loading position, and ultrapure water was passed (by the tubing lines) through the minicolumn (V1 in position B) in order to eliminate the

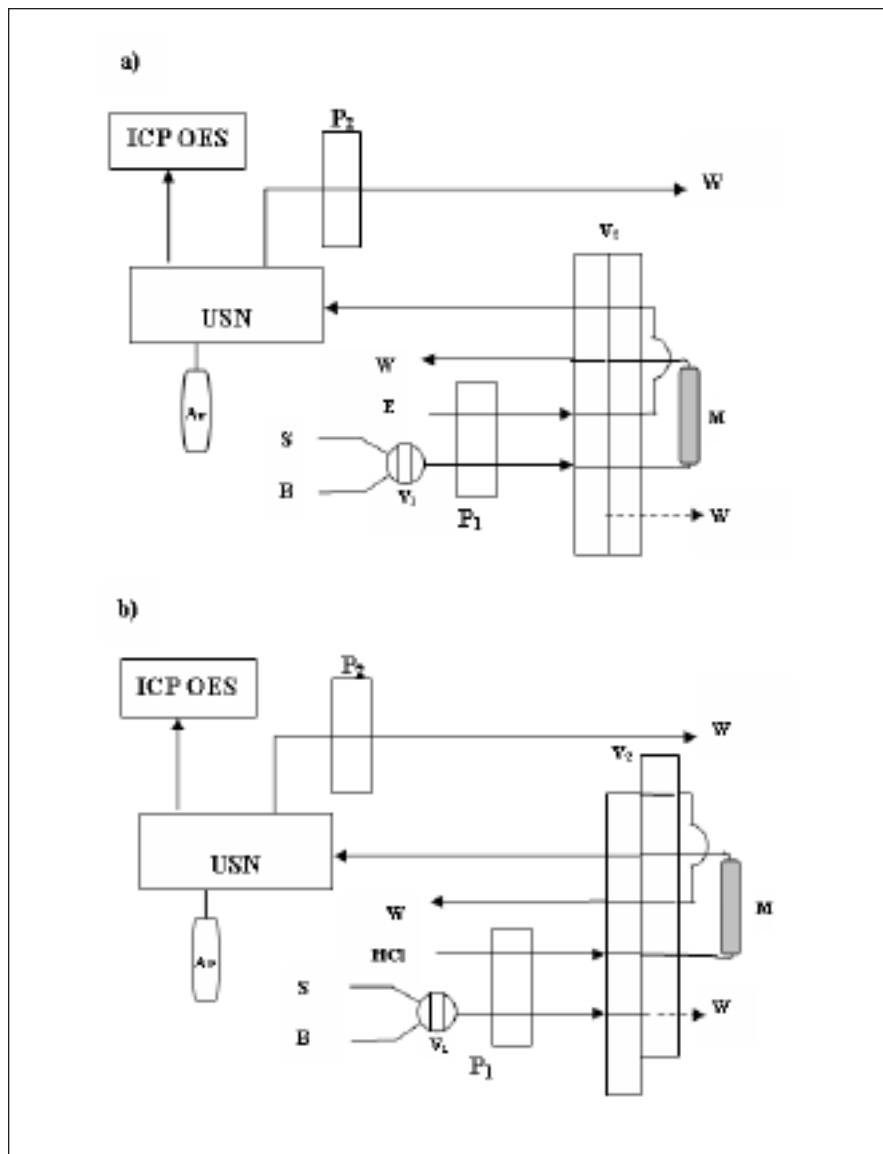


Fig. 1. Schematic diagram of the instrumental setup. B, conditioning line; S, sampling line; W, waste; V₁, two-way rotary valve, V₂, load injection valve (a, load position; b, injection position); M, conical minicolumn; USN, ultrasonic nebulizer; Ar, argon gas supply either for plasma or for USN.

alcohols) and high friction coefficient. This material was employed to construct the preconcentration minicolumn as follows:

The minicolumn was prepared by placing 100 mg of EVA turnings (small particles of spiralled shape) into an empty cylindrical EVA tubing using the dry packing method. To avoid loss of EVA particles when

the sample solution passes through the minicolumn, a small amount of quartz wool was placed on both ends of the minicolumn. The column was then connected to a peristaltic pump with PTFE tubing to form the preconcentration system. This method was used for around 600 preconcentration cycles, and the retention capacity did not

remaining acid (0.5 minutes).

The emission measurements (peak height) were proportional to the Mo concentration in the sample and were used for all measurements.

The operating conditions were established and the determination was carried out.

RESULTS AND DISCUSSION

Effect of pH on Molybdenum Retention

The pH value plays an important role in the retention phenomenon of different ions on the EVA turnings. In order to evaluate the effect of pH, a series of sample solutions were adjusted to a pH range of 1.0–12.0 with HCl or sodium hydroxide and processed according

to the recommended procedure. The effect of pH on molybdenum retention is shown in Figure 2. It can be seen that quantitative adsorption (close to 100%) was achieved in the pH range between 5.0 and 7.0.

Accordingly, a pH of 6.0 was selected for further experiments. All these studies were carried out with synthetic Mo solutions with a concentration of $20 \mu\text{g L}^{-1}$.

Effect of Sample Loading Flow Rate on Molybdenum Retention

The sample flow rate through the microcolumn is one of the steps that controls the analysis time. In this study it was verified that with flow rates up to 5.0 mL min^{-1} there was no effect on analyte recovery, which under optimum conditions was close to 100% (Figure 3). At higher flow rates, the recovery showed an obvious decrease with a diminishing contact time between the analyte and the adsorbent material.

Effect of Eluent

A satisfactory eluent should effectively elute the analyte in a discrete volume in order to obtain the best analyte recovery. Different eluents were tested at a flow rate of 1.8 mL min^{-1} through the column in order to evaluate and compare the analyte recovery (95% confidence interval $n = 6$). Best results were obtained with HCl and, consequently, this acid was employed as the eluent. In this way, different concentrations were tested and we verified that 1.0 mol L^{-1} was the minimum concentration necessary to obtain the best response. At lower HCl concentrations, a lower sensitivity was obtained due to an increase in the dispersion effect.

The elution flow rate is a variable which is often optimized care-

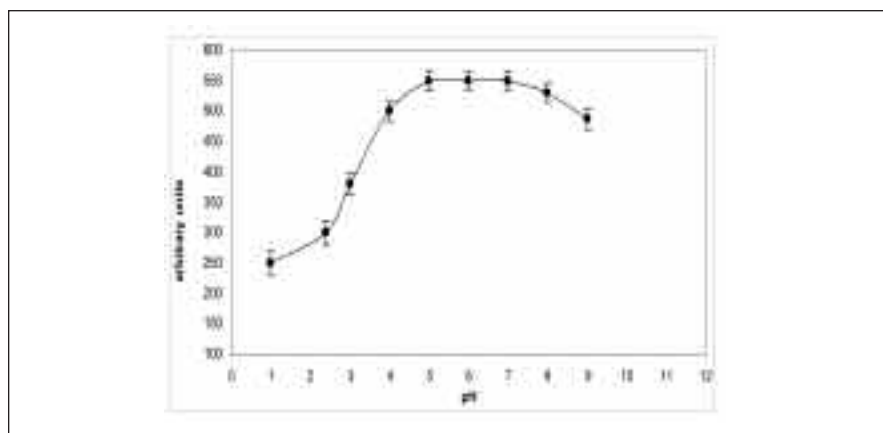


Fig. 2. Analytical signal (expressed as % relative response) as a function of the sample pH. Mo concentration: $5.0 \mu\text{g L}^{-1}$; Sample flow rate: 5.0 mL min^{-1} , HCl concentration: 1.0 mol L^{-1} .

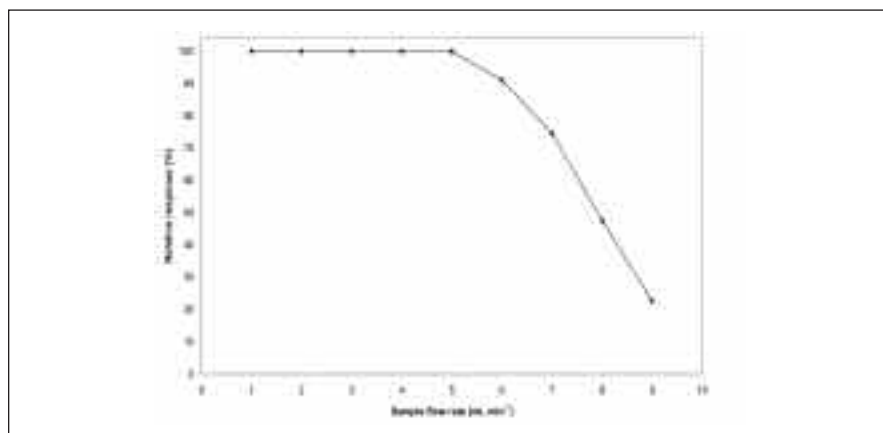


Fig. 3. Analytical signal (expressed as % relative response) as a function of the sample load flow rate. Mo concentration: $5.0 \mu\text{g L}^{-1}$; pH: 6.0; HCl concentration: 1.5 mol L^{-1} .

fully. In this case, this variable was subjected to the optimal intake flow rate of the USN which was 1.8 mL min^{-1} . Accordingly, a flow rate of 1.8 mL min^{-1} was selected as the fixed elution flow rate.

Column Reuse

The stability and regeneration of the column were investigated. Due to the high chemical and physical resistance of the EVA, the column was stable up to at least 500 pre-concentration cycles, and neither column deterioration nor a decrease in Mo recovery was observed.

Interference Studies

The effects of common coexisting ions in various drinking waters and a certified reference material NIST CRM 1643e (National Institute of Technology, Gaithersburg, MD, USA) on the adsorption of Mo on the EVA turnings minicolumn were investigated. In these experiments, solutions of $20 \mu\text{g L}^{-1}$ of Mo(VI) containing the added interfering ions were treated according to the recommended procedure. The tolerance limits of the coexisting ions, defined as the largest amount of ion that produces a recovery of Mo less than 90% were evaluated. The results showed that Cu^{2+} , Zn^{2+} , Cd^{2+} , Ni^{2+} , Co^{2+} , Cr^{3+} , Mn^{2+} , and Al^{3+} could be tolerated up to at least $2500 \mu\text{g L}^{-1}$; Fe^{2+} and Fe^{3+} could be tolerated up to at least $5000 \mu\text{g L}^{-1}$. Commonly encountered matrix components, such as alkaline and alkaline earth elements, are not retained on the minicolumn.

Figures of Merit

The time required for the pre-concentration of 20.0 mL of sample (5.0 mL min^{-1}), acquiring injection/data (0.5 min at 1.8 mL min^{-1}), and conditioning was about 5.0 minutes, resulting in a sample throughput of 12 samples per hour. A 300-fold total enhancement factor (10 for

the minicolumn and 30 for the USN) for a sample volume of 20.0 mL was obtained with respect to the Mo determination by ICP-OES without preconcentration.

The relative standard deviation (RSD) for six replicate measurements of a solution containing $20 \mu\text{g L}^{-1}$ Mo was 3.5%. The limit of detection (LOD), calculated as the amount of Mo required to yield a net peak equal to three times the standard deviation of the background signal (3σ), was $0.04 \mu\text{g L}^{-1}$. The calibration curve was linear from levels close to the LOD up to $500 \mu\text{g L}^{-1}$, with a correlation coefficient of 0.9989.

Recovery Study and Application to Real Samples and Reference Materials

In order to evaluate the Mo recovery of this method, 200 mL of

a natural drinking water sample was collected in our laboratory and divided into 10 portions of 20 mL. The proposed method was applied to six portions and the average quantity of the Mo obtained was taken as the base value. Then, increasing quantities of Mo(VI) were added to the other aliquots of the sample and Mo was determined by the same method. The recoveries were in the range of 98.3–100.0% (see Table II).

Additionally, the accuracy of the proposed method was evaluated by determining Mo in the certified reference material NIST CRM 1643e with a molybdenum content of $121.4 \pm 1.3 \mu\text{g L}^{-1}$ and a density equal to 1.016 g mL^{-1} at 22°C .

TABLE II
Concentration of Molybdenum in Drinking Water Samples and NIST CRM 1643e (95% confidence interval; n = 6)

Sample	Mo(VI) Conc. Base ($\mu\text{g L}^{-1}$)	Mo(VI) Conc. Added ($\mu\text{g L}^{-1}$)	Mo(VI) Conc. Found ($\mu\text{g L}^{-1}$)	Recovery ^c (%)
A (Drinking Water - 1st week) ^a	1.23 ± 0.08	0.00	1.23 ± 0.08	—
A	1.23	0.30	1.53 ± 0.09	100.0
A	1.23	0.60	1.82 ± 0.10	98.3
A	1.23	0.90	2.12 ± 0.10	98.8
A	1.23	1.20	2.42 ± 0.09	99.1
B (Drinking Water - 2nd week) ^a	0.97 ± 0.08	0.00	0.97 ± 0.07	—
C (Drinking Water - 3rd week) ^a	0.88 ± 0.08	0.00	0.88 ± 0.08	—
D (Drinking Water - 4th week) ^a	1.11 ± 0.10	0.00	1.11 ± 0.09	—
E (NIST CRM 1643e) ^b	120.50 ± 1.20	0.00	120.50 ± 1.20	—

^a Drinking water collected in our laboratory.

^b Certified Value = $121.4 \pm 1.3 \mu\text{g L}^{-1}$

^c Recovery (%) = [(found-base)/added] x 100

CONCLUSION

On the basis of the results of this study, the ethyl vinyl acetate (EVA) turnings have shown to be a suitable substrate for the preconcentration of inorganic molybdenum.

The on-line coupling of solid phase extraction with ultrasonic nebulization (SPE-USN) and inductively coupled plasma optical emission spectrometry (ICP-OES) for Mo determination in water samples increases the speed of preconcentration and the analysis process, reduces sample consumption and contamination risks, and improves sensitivity and selectivity. As an additional advantage for total Mo determination, this metal can easily be retained without using a complexing reagent. The preconcentration system was able to determine Mo in drinking waters at sub- $\mu\text{g L}^{-1}$ levels with recoveries close to 100%.

To the best of our knowledge, this is the first report where EVA was used as a substrate in an on-line preconcentration system for molybdenum determination.

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