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ENVIRONMENTAL SIGNIFICANCE AND IDENTIFICATION OF METAL-CHELATE COMPLEXES USING ION CHROMATOGRAPHY

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The trace analysis of metal-complexes has long been an area of interest for analytical chemists and environmental researchers due to the biological and toxic properties of these compounds. The method for the simultaneous separation of the metal cations and organic and inorganic anions is based on the use of strong chelating anion with high charge. When basic solution contains an excess of strong complexing anion of high charge, such as ethylenediaminetetraacetate (EDTA) or trans-1,2-diamine-cyclohexane-tetraacetic acid (DCTA) ion, most heavy and transition metal ions will occur as anionic complexes. Hence this method provides simultaneous metal and anion separation. The EDTA and DCTA chelating agents exhibit strong complexing power. These aminopolycarboxylic acids can remobilize metals in nature. Because aminopolycarboxylic acids are a potential risk to the environment, it is important to develop an effective analytical technique for their determination. Several factors affect the retention in the separation of the complex anions: complex formation reactions, ion-exchange equilibria and protolysis depending on pH. The aim of this work is the optimization of a simultaneous chromatographic separation and identification of metal ions complexed by the ligand EDTA or DCTA. The method was utilized to separate $CuEDTA^{2-}$, $CuDCTA^{2-}$, $ZnEDTA^{2-}$, $ZnDCTA^{2-}$, $AlEDTA^{-}$, $AlDCTA^{-}$, Cl^{-}, piruvate and maleate anions. An advantage of the developed method is that the same basic pH-range is favourable to the stability of the metal complexes and to the elution.

Keywords: transition metal complexes, EDTA, DCTA ligands, ion exchange chromatography

Introduction

Metal ion speciation and the environment

The presence of transition and heavy metals in the environmental and biological materials justifies the importance of high performance environmental qualitative and quantitative analysis of these species [1]. The transition metals exist in different oxidation states possessing different physical and chemical properties and different toxicity. The main sources of metal-contamination of the environment are the industrial emission, vehicle exhaustion, corrosion processes, households, agriculture, hazardous storage tanks, and waste disposal sites. The presence of inorganic pollutants, especially toxic metal ions, is a serious issue, as metal ions may often be carcinogenic in nature. The identification of pollutants in environmental matrices is a difficult task because of strong interference from other components of the sample. The extended use of palladium in automotive catalytic converters and in the chemical industry has also led to increasing concentrations of this metal in environmental compartments. Platinum group and heavy metals may enter the environment and interact with complexing materials, such as humic substances. Determination of palladium by ion chromatography with ICP-MS detection was developed by M. C. Bruzzoniti et al. [2]

Aluminium plays probably a role in the development of Alzheimer's disease [3]. The route of these toxic metal ions to the human body is through water and other foodstuffs. Therefore, the monitoring of metal ions with different oxidation states in water bodies and foodstuffs is essential and important. Some toxic metal ions are also present in the atmosphere and indirectly affect our health. Some metal ion in oxoanion forms (AsO_4^{3-}, CrO_4^{2-}) are transported across cell membranes.

Copper and zinc within those metals that are essential to life although inherently toxic. The characteristic oxidation forms of copper are: Cu(I) and Cu(II). In case of zinc the most frequent forms are Zn(I) and Zn(II). The change of oxidation state of an element affects the degree of its bio-availability and toxicity. The different oxidation states of a particular metal ion possess different physical and chemical properties. These oxidation states differ in their redox potential, complexation, and hydration properties. Therefore the speciation analysis can differentiate the complexed and free forms of metal ions. Measuring the total concentration of metal ions gives no information about the actual chemical forms it exist, that is important to understand its toxicity and biotransformation. Therefore the speciation of elements can not be omitted.

Aminopolycarboxylic acids as ligands can remobilize heavy and transition metals and their release in nature may cause release of metals into ground water and their uptake by plants. Degradation of chelating agents is controversial. While excessive uptake of heavy metals was viewed as a deterrent for the use of EDTA in agriculture, the same process is now being researched because of the possibility that it could be applied to the phytoremediation of heavy-metal contaminated soils. However, due to the lack of selective analytical techniques, the mechanisms of metal uptake by plants in the presence of EDTA still remain largely speculative. Since aminopolycarboxylic acids are a potential risk to the environment, it is also important to develop a selective analytical technique for their determination.

Chelate chromatography

Chelate chromatography is a special type of ion-chromatography in which chelating agents as eluent additives are employed. The ion chromatography is a suitable speciation technique and it offers reproducible results.

Simultaneous separation of metals and anions is based on the use of a strong complexing anion of high charge [4]. Ethylenediaminetetraacetic acid (EDTA) and trans-1,2-diamincyclohexanetetraacetic acid (DCTA) are excellent chelating agents that are able to form sufficiently stable chelates with different metal ions. The strong complex-forming anions with high charge react with most of the di- and trivalent metal cations and they form complexes with one or two negative charge that makes the simultaneous separation of metal cations, organic and inorganic anions possible.

Several factors affect the chromatographic retention of complex anions. These are the (1) complex formation reactions, (2) the ion-exchange equilibria, and (3) the protolysis that depends on the pH of elution.

Retention models have been developed by Hajos et al. In order to study the retention behavior of metal-complexes in anion exchange chromatography [4, 5]. The theory [5] is based on the generalized ion-exchange-, protonation- and complex-formation equilibria. The unknown ion-exchange equilibrium constants for the sample and the eluent species can be determined from experimental retention data [6] by iterative minimization, using a non-linear regression algorithm. The model was utilized to predict the retention behavior of $CdEDTA^{2-}$, $CoEDTA^{2-}$, $MnEDTA^{2-}$ and $NiEDTA^{2-}$ ions. It was concluded that the chromatographic separation of these species are strongly influenced by the size of ion, the type, concentration, and pH of eluent, and the stability of complex.

Experimental

Instrumentation

A Dionex DX-300 ion chromatograph (Sunnyvale, CA, USA) equipped with a conductivity detector and a Dionex AMMS-I cation micromembrane suppressor was used during the work. The separations were carried out by AS9-HC and AS4A-SC separator columns (250 x 4 mm i.d.) packed with anion-exchangers functionalized with alkyl/alkanol quaternary ammonium ions. The recommended pH-range for the columns was 2–13. All chromatograms were obtained at room temperature at a flow rate of 1.2 mL min^{-1}. The injection volume was 50 µl. The micromembrane suppressor was regenerated with sulphuric acid (0.025 M) at flow rate 3,5 mL min^{-1}.

Reagents and solutions

Eluents were prepared by using analytical grade Na_2CO_3 and $NaHCO_3$ (Fluka, Switzerland). High purity water was obtained by using a Milli-Q system (Millipore, Bedford, MA). The specific resistance of the water was 18.2 MΩ cm^{-1}. Sample solutions of metals, organic and inorganic anions and the chelating agents (EDTA and DCTA) were prepared by dilution of a concentrated stock solution of analytical-grade salts (Fluka). The sample solutions contained chloride salts of metal cations and complex forming ligands. Before analysis, all eluents were treated in an ultrasonic bath for 5 mins in order to remove air.

Basic components and practice of chelate-chromatography

The basic components of chelate chromatography are presented in *Fig. 1*. The delivery system consists of an eluent container (Na_2CO_3, $NaHCO_3$, pH 8–11), liquid transfer lines, eluent and sample selection valves and a pump. The sample components (metal-halogenides, oxoanions) together with complexing agents (EDTA, DCTA) are injected into the separation system via a valve injector. Plastic valves made of chemically inert materials are used. Typical injection volumes are between 10–100 µL. The separator columns are packed with pellicular anion exchanger in order to obtain optimum separation condition for ionic components ($MEDTA^{2-}$, A^-) with an adequately short analysis time. After leaving the separator column, the separated species pass into the conductivity detector. The suppressor-type ion chromatography systems has a unique detection system in which an ion-exchange membrane enhances the sensitivity of analysis. The main function of the suppressor is to chemically reduce the high background conductivity

of the electrolyte used as eluent ($NaOH \rightarrow H_2O$; $Na_2CO_3 \rightarrow H_2CO_3$), and to convert the sample anions into a much more conductive form ($NaCl \rightarrow HCl$). A major advantage of chelate-chromatography – in contrast to other instrumental analysis such as atomic spectroscopy – is its ability to detect different species of anions and cations simultaneously.

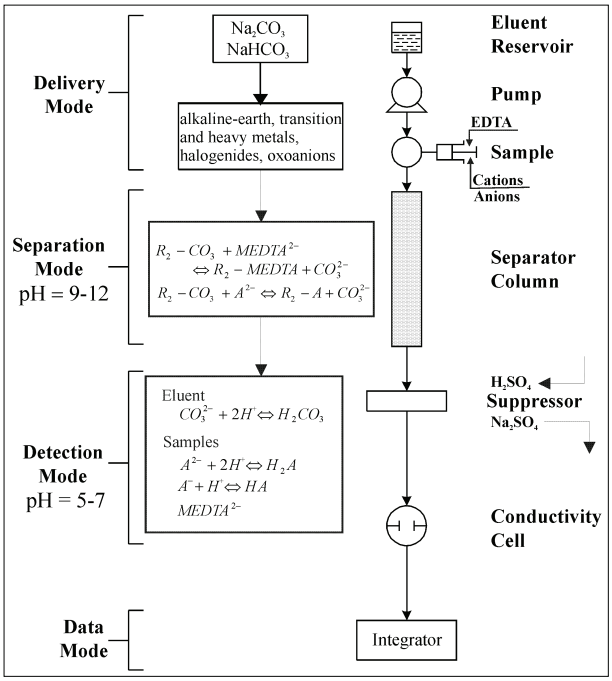

Figure 1: Schematic flow diagram of chelate chromatography

Results and discussion

By adding negatively charged EDTA or DCTA ligand to positively charged metal ions complex anions with negative charge form in the solution according to the following equilibria:

$$EDTA^{4-} + M^{2+} \leftrightarrow [MEDTA]^{2-} \quad (1)$$

$$DCTA^{4-} + M^{2+} \leftrightarrow [MDCTA]^{2-} \quad (2)$$

The conjugate bases of EDTA and DCTA are 6-dentate ligands. In case of complex formation, the 6 donoratoms of the ligand (4 oxygen and 2 nitrogen atoms) are located at octahedron vertices around the central metal ion. The high stability of the metal chelates is due to the fact that the ligand surrounds fully the metal ion and isolates it from molecules of the solvent. The stability of the complexes depends on the pH. When the pH increases, the chelating agents are more and more deprotonated and exhibit their complexing power. During the formation of metal chelates pH-dependent side-reactions occur. At the eluent pH range investigated EDTA and DCTA exist in two forms: HY^{3-} and Y^{4-}.

In this work, carbonate/hydrogencarbonate electrolyte was used as eluent at various concentrations and pHs. The separation system contains three ionic species in the eluent (CO_3^{2-}, HCO_3^- and OH^-) and various forms of organic, inorganic and complex ions in the sample. During elution, the following simultaneous equilibria take place in the separator column:

$$2R\text{-}E + MEDTA^{2-} \leftrightarrow R_2\text{-}MEDTA + 2E^- \quad (3)$$

$$R\text{-}E + MHEDTA^- \leftrightarrow R\text{-}MHEDTA + E^- \quad (4)$$

$$2R\text{-}E + MDCTA^{2-} \leftrightarrow R_2\text{-}MDCTA + 2E^- \quad (5)$$

$$R\text{-}E + MHDCTA^- \leftrightarrow R\text{-}MHDCTA + E^- \quad (6)$$

where:

R – the charged functional group of the ion-exchanger

E – the anion of the eluent.

In the suppressor reaction carbonic acid is formed from the eluent anions:

$$CO_3^{2-} + 2\ H^+ \leftrightarrow H_2CO_3 \quad (7)$$

The retention factors (log k') of the investigated anions at different eluent concentrations and pHs can be seen in *Table 1*. The result shows clearly that the increasing eluent concentration decreases the retention of anions. At the same time, the changing eluent pH affects the sample composition by changing the fractions of differently protonated species.

It can be seen in *Fig. 2* (pH vs. Φ) that at the pH of elution the EDTA can exist in two distinct forms with tri- and four negative charges. It is important to note that the changing pH of the eluent does not affect the order of elution of the metal complexes.

An advantage of the applied method is that the same basic pH-range is favorable to the stability of the metal complexes and also to the elution.

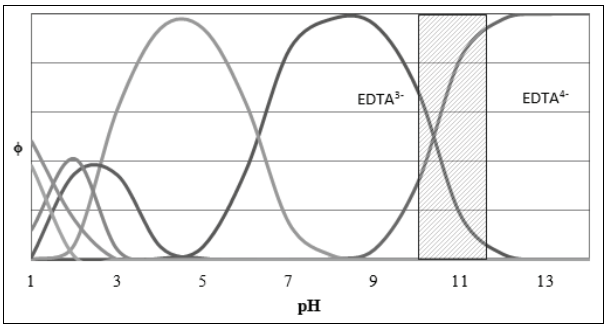

Figure 2: Partial molar fractions of EDTA in the eluent at different pHs. The cross-hatched area represents the pH region of the eluents used

With this separation method, complexes with different ligands and the complexes of different metal ions can be separated in the same run (*Fig. 3-5*). The simultaneous separation of negatively charged metal chelates and carboxylate anions can be performed as well (Fig. 4).

The results indicate that the retention is influenced by the pH of eluent (Table 1). Increasing eluent pH leads to decrease in retention (k') because the predominant form of eluent species is the divalent carbonate above pH 10 that has higher elution strength than the monovalent hydrogen carbonate anion. This important factor has to be considered during the optimization of separation.

Table 1: The effect of pH and eluent concentration on the retention of complexes and ligands

	k'										
C_{elu} [mM]	**5.0**			**6.5**				**8.0**			**9.0**
pH	**10.27**	**10.86**	**11.03**	**9.90**	**10.27**	**10.50**	**10.86**	**9.44**	**10.27**	**10.86**	**11.03**
Cl^-	3.88	3.45	3.60	4.20	3.56	3.42	3.08	5.34	3.53	3.14	2.85
$EDTA^{4-}$	12.00	8.06	9.00	11.89	8.16	6.93	5.95	12.27	6.29	5.38	3.92
$EDTA^{3-}$	26.09	19.38	20.16	25.18	17.78	15.81	14.63	17.29	14.73	12.68	10.58
$DCTA^{4-}$	12.80	9.28	9.81	13.54	10.04	8.73	7.11	n.r.	7.96	6.32	5.07
$DCTA^{3-}$	15.86	11.23	11.93	14.36	12.10	10.55	8.62	21.01	16.96	13.27	10.84
$CuEDTA^{2-}$	30.27	25.98	25.95	22.53	21.67	19.92	18.87	n.r.	17.55	15.86	14.28
$ZnEDTA^{2-}$	22.87	19.30	19.95	n.r.	17.84	16.12	14.54	18.20	14.62	12.77	10.72
$CuDCTA^{2-}$	27.60	19.14	20.84	n.r.	20.41	17.08	14.66	20.22	16.96	13.27	10.65
$ZnDCTA^{2-}$	28.58	20.01	21.31	n.r.	21.45	17.84	15.30	21.90	17.60	13.72	10.50

n.r.: no retention data

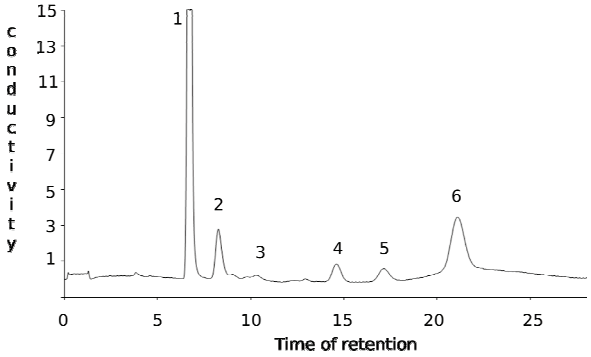

Figure 3: Simultaneous separation of Al and Zn complexes with EDTA and DCTA chelating agents. Peaks: 1. Cl^-, 2. $EDTA^{4-}$, 3. $DCTA^{4-}$, 4. $[AlEDTA]^-$, 5. $[AlDCTA]^-$, 6. $[ZnEDTA]^{2-}$ and $[ZnDCTA]^{2-}$. Eluent: 9.0 mM Na_2CO_3, pH = 11.027. Column: AS9-HC anion exchanger.

Calibration data (*Table 2*) demonstrate that the simultaneous IC analysis of metal-chelate complexes and ligands is sensitive and can be used for quantitation as well.

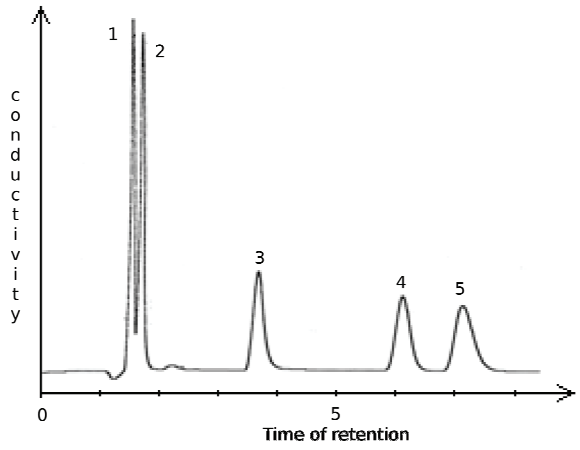

Figure 4: Chromatogram of simultaneous separation of aliphatic carboxyl acids and copper-EDTA complex. Peaks: 1. Piruvate, 2. Cl^-, 3. $EDTA^{4-}$, 4. Maleate, 5. [CuEDTA]. Eluent: 0.7 mM Na_2CO_3 + 1.8 mM $NaHCO_3$; pH=9.66. Column: AS4A-SC.

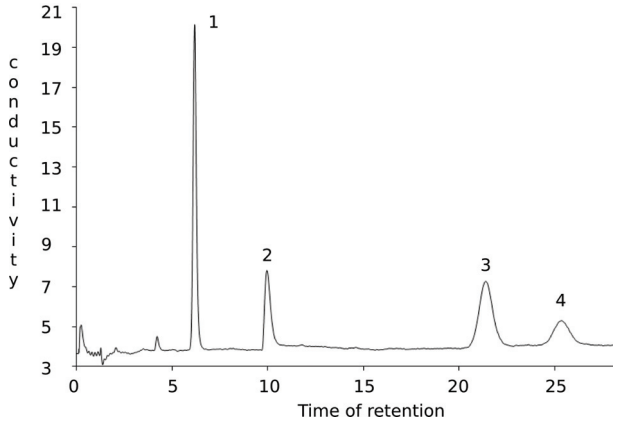

Figure 5: Chromatogram of EDTA-metal complexes. Peaks: 1. Cl^-, 2. $EDTA^{4-}$, 3. [CuEDTA], 4. [ZnEDTA]. Eluent: 8.0 mM Na_2CO_3+ $NaHCO_3$, pH = 10.27. Column: AS9-HC anion exchanger.

Table 2: Calibration data of EDTA and DCTA chelating agents and their complexes.

Sample	Sensitivity (μS sec L mg^{-1})	Linearity (r^2)
$EDTA^{4-}$	3×10^8	0.8645
$EDTA^{3-}$	3×10^8	0.8442
$DCTA^{4-}$	4×10^8	0.8771
$DCTA^{3-}$	2×10^7	0.7558
$[Cu\text{-}EDTA]^{2-}$	10^9	0.9931
$[Cu\text{-}DCTA]^{2-}$	10^9	0.9894

Conclusion

Our experiments verified that the simultaneous analysis of anions and metal cations can be achieved and the change of concentration of components can be detected, the metal complexes and their ligands can be identified. The retention data of chelate-complexes (Cu^{2+}, Zn^{2+}, EDTA, DCTA) were given by the use of anion exchange column packed with pellicular stationary phase, and by the use of carbonate-hydrogencarbonate eluent and suppressed conductivity detection. The effective parameters of the separation were determined considering the composition of eluent.

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REFERENCES

1. I. ALI, H. Y. ABOUL-ENEIN: Instrumental Methods in Metal Ion Speciation, Chromatographic Science Series; Taylor & Francis, 2006, 1–16
2. M. C. BRUZZONITI, S. CAVALLI, A. MANGIA, C. MUCCHINO, C. SARZANINI, E. TARASCO: Ion chromatography with inductively coupled plasma mass spectrometry, a powerful analytical tool for complex matrices. Estimation of Pt and P din environmental samples, Journal of Chromatography A, 997, (2003), 51–63
3. M. C. BRUZZONITI, E. MENTASTI, C. SARZANINI: Simultaneous determination of inorganic anions and metal ions by suppressed ion chromatography, Analytica Chimica Acta 382, (1999), 291–299
4. P. HAJOS, G. REVESZ, O. HORVATH, C. SARZANINI: The simultaneous analysis of metal-EDTA complexes and inorganic anions by suppressed ion chromatography, Journal of Chromatographic Science, 34(6), (1996), 291–299
5. P. HAJOS, G. REVESZ, C. SARZANINI, G. SACCHERO, E. MENTASTI: Retention model for the separation of anionic metal-EDTA complexes in ion chromatography, Journal of Chromatography, 640, (1993), 15–25
6. P. HAJOS, O. HORVATH, G. REVESZ: Advances in Chromatography, 38., Marcel Dekker Inc., New York (1997)