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## REVERSED-PHASE HIGH-PERFORMANCE LIQUID CHROMATOGRAPHY OF DIETHYLDITHIOCARBAMATE COMPLEXES USING RADIAL COMPRESSION COLUMNS

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### SUMMARY

The diethyldithiocarbamate complexes of Co(III), Cr(III), Cu(II), Hg(II), Ni(II), Pb(II), Se(IV) and Te(IV) are separable on a Waters Assoc. Radial Pak C<sub>18</sub> column using a mobile phase of methanol–acetonitrile–water (40:35:25), provided the column has been conditioned by prior injection of a concentrated mixture of the complexes. Alternatively, the column can be conditioned by flushing with 0.005 M disodium ethylenediamine tetraacetate (EDTA), with subsequent addition of  $1.25 \cdot 10^{-5}$  M EDTA to the mobile phase. Diethyldithiocarbamate complexes of Pb(II), Cd(II) and Fe(III) gave poor peak shape due to substitution reactions with nickel from stainless steel components of the chromatographic system. The analysis of a synthetic electroplating solution containing salicylsulphonic acid, sulphanyl-amide and six inorganic ions is illustrated.

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### INTRODUCTION

Dithiocarbamates have long been used for the extraction and spectrophotometric determination of many elements. In recent years, dithiocarbamate chelates have been studied by high-performance liquid chromatography (HPLC)<sup>1,2</sup>, with the resultant separations forming the basis of methods for the simultaneous analysis of metals. This approach has been successfully applied to Cd(II), Hg(II), Co(II), Cr(III), Cr(VI), Pb(II), Mn(II), Bi(III), Ni(II), Zn(II), Fe(III) and Se(IV), using both normal-phase<sup>3–8</sup> and reversed-phase chromatography<sup>9–18</sup>.

Within the reversed-phase mode, a variety of column types has been used, including stainless steel<sup>9–16</sup>, glass<sup>18</sup> and microbore<sup>17</sup>, with the most successful separations being achieved on the stainless steel type<sup>10,13</sup>. However, it has been recently pointed out that ligand substitution reactions may occur between the metal parts of the column (particularly the stainless steel end frits) and the metal complexes being chromatographed<sup>18,19</sup>. This effect is most pronounced for the diethyldithiocarbamates of Bi(III), Zn(II), Cd(II) and Pb(II), since these complexes are relatively unstable and may readily undergo an exchange reaction with nickel from the stainless steel components.

This detrimental effect can be minimised by using glass or PTFE-lined columns<sup>18</sup> or by deactivating the metal end frits with an organosilane<sup>19</sup>. The former approach, however, is not completely successful, and a loss of column efficiency is sometimes observed, and while the latter method does appear to prevent ligand exchange, the silanisation procedure is somewhat lengthy and requires dismantling of the column.

In their report describing the abovementioned silanisation method, Shih and Carr<sup>19</sup> observed that Waters Assoc. radial compression columns showed no ligand-exchange effects for Ni(II) and Fe(III), since these columns are largely constructed of non-metallic materials, with the exception of low surface area stainless-steel spreader plates at each end of the column. We have further examined the use of these columns for the separation of metal diethyldithiocarbamate complexes and in this paper we illustrate the separation of eight complexes and also describe the column conditioning procedures necessary to consistently achieve this separation.

## EXPERIMENTAL

### *Instrumentation and reagents*

The liquid chromatograph used consisted of a Waters Assoc. (Milford, MA, U.S.A.) M6000A pump, U6K injector and 440 UV absorbance detector, together with a Houston Instruments Omniscrible recorder. A 5-mm I.D. Waters Assoc. Radial Pak C<sub>18</sub> column with a particle size of 5  $\mu\text{m}$  was used in conjunction with a Waters Assoc. RCM-100 radial compression module. The detector was operated at 254 nm, the mobile phase flow-rate was 2.0 ml/min and the recorder chart speed was 0.5 cm/min.

Analytical grade methanol (May and Baker) was doubly distilled in all-glass apparatus, water was purified using the Millipore Milli-Q system and HPLC grade acetonitrile was purchased from Waters Assoc. All solvents were filtered through 0.45- $\mu\text{m}$  Millipore filters before use. Analytical grade sodium diethyldithiocarbamate was obtained from BDH (London, Great Britain) and all metal salts and other reagents used were of analytical grade. The ion-pairing reagent used in the analysis of the synthetic electroplating solution was tetrabutylammonium phosphate (Ajax).

Individual 0.01 *M* solutions of each metal salt were made up in 0.1 *M* nitric acid, and also a mixture of nine metal ions, all at 0.01 *M* concentration (Mixture A). A dilute mixture solution (Mixture B) was prepared containing the same eight metal ions, but with individual concentrations adjusted so that all peaks in the resulting chromatogram would be on scale (see Fig. 1). The actual concentrations of the components of Mixture B ranged between  $2.7 \cdot 10^{-4}$  *M* and  $2.0 \cdot 10^{-3}$  *M*.

### *Extraction procedure*

An aliquot (usually 5 ml) of the aqueous solution containing metal ions was transferred to a 125-ml separating funnel, to which were added 10 ml of pH 5.78 acetate buffer and an excess of 0.1 *M* sodium diethyldithiocarbamate. The dithiocarbamate complexes so formed were then extracted with four 5-ml portions of chloroform, and the combined organic extracts were back-extracted with 20 ml of Milli-Q water to remove excess ligand. The organic phase was quantitatively filtered through Whatman phase-separation paper into a 25-ml volumetric flask, and the solution was

TABLE I

## CAPACITY FACTORS OF RESOLVED DIETHYLDITHIOCARBAMATE COMPLEXES

Mobile phase 1: methanol–acetonitrile–water (40:35:25); the column was conditioned by injection of a concentrated mixture (mixture B, see Experimental). Mobile phase 2: methanol–acetonitrile–water (40:35:25) containing  $1.25 \cdot 10^{-5}$  M EDTA; the column was conditioned by flushing with 0.005 M EDTA.

Species	Capacity factor	
	Mobile phase 1	Mobile phase 2
Disulfiram	5.5	6.8
Cd(II)	5.9	0
Pb(II)	7.4	0
Ni(II)	10.0	13.0
Co(III)	12.2	16.8
Cr(III)	13.6	19.0
Se(IV)	15.0	20.2
Cu(II)	16.8	23.0
Hg(II)	21.0	29.0
Te(IV)	22.0	30.6

then diluted to the mark with chloroform. A 5-ml aliquot of this solution was evaporated to dryness with a gentle stream of nitrogen and the residue was dissolved in exactly 5 ml of methanol. An aliquot (usually 10  $\mu$ l) of this solution was then injected into the chromatograph.

## RESULTS AND DISCUSSION

*Suitability of complexes for chromatographic analysis*

Studies were performed using chloroform extracts of the diethyldithiocarbamate complexes of the following species: Cd(II), Co(III), Cr(III), Cr(VI), Fe(III), Hg(II), Mn(II), Ni(II), Pb(II), Se(IV), Te(IV), V(V) and Zn(II). The complexes of Fe(III), Mn(II) and V(V) were unstable in chloroform and gave broad, ill-defined peaks when chromatographed. No peak was observed for the Zn(II) complex, however all of the remaining species were suitable for chromatographic analysis, although both Pb(II) and Cd(II) gave asymmetrical peaks. Mobile phases containing binary mixtures of water with methanol or acetonitrile gave rather poor peak shape and inadequate resolution, however ternary solvent mixtures of water, methanol and acetonitrile gave excellent peak shape and resolution. Table I lists the capacity factors for the complexes studied and a typical separation of eight complexes is shown in Fig. 1. Both the number of complexes separated and the quality of the resolution and peak shape in Fig. 1 surpass those achieved in previously published studies.

All chromatograms of individual diethyldithiocarbamate complexes and mixtures of complexes exhibited a major peak which eluted early in the chromatogram. This peak has been observed by other workers and has been attributed to an unidentified disulphide compound produced from the excess ligand used for extraction of the metal ion<sup>14</sup>. We have identified this disulphide as bis(diethylthiocarbamyl)disulphide (disulfiram) which is produced by oxydation of diethyldithiocarbamic acid occurring during the solvent extraction process. This identification was based on comparison of

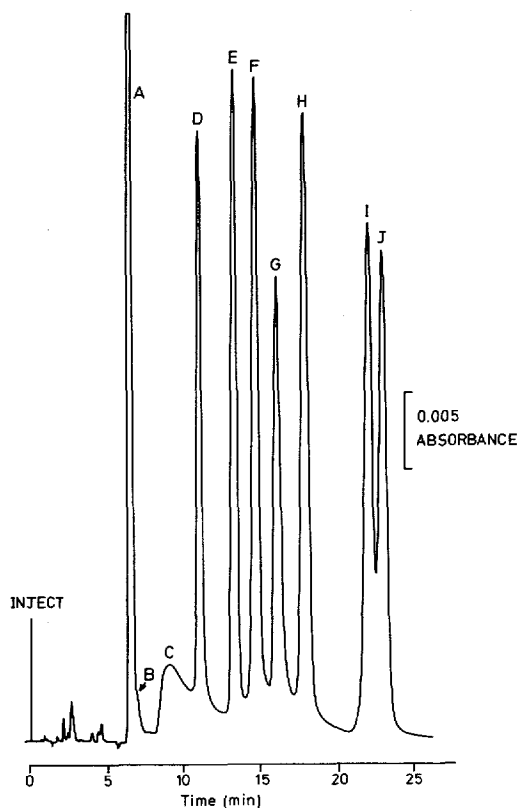


Fig. 1. Separation of a mixture of diethyldithiocarbamate complexes (Mixture B, see Experimental). Conditions: mobile phase methanol-acetonitrile-water (40:35:25) flow-rate 2.0 ml/min, detector wavelength 254 nm; detector sensitivity 0.05 a.u.f.s.; injection volume 10  $\mu$ l; column conditioned by prior injection of Mixture A (see Experimental). A = disulfiram; B = Cd(II), 0.12  $\mu$ g; C = Pb(II), 0.11  $\mu$ g; D = Ni(II), 0.14  $\mu$ g; E = Co(III), 0.07  $\mu$ g; F = Cr(III), 0.11  $\mu$ g; G = Se(IV), 0.32  $\mu$ g; H = Cu(II), 0.18  $\mu$ g; I = Hg(II), 0.51  $\mu$ g; J = Te(IV), 0.17  $\mu$ g.

retention times of the unknown disulphide and an authentic sample of disulfiram. Fig. 1 shows that the disulfiram is well resolved from any of the dithiocarbamate complexes.

It is noteworthy that poor peak shape was observed for Pb(II), Cd(II) and Fe(III), and that no peak was observed for Zn(II), since all of these metals form relatively unstable diethyldithiocarbamate complexes<sup>20</sup> and can be expected to undergo substitution reactions with nickel from the stainless steel in the chromatographic system. It appears that such substitution reactions occur even with the radial compression column, in contrast to the prediction of Shih and Carr<sup>19</sup>, however it was noted that peak shapes were generally superior on this column compared to an equivalent stainless steel column. Indeed, on this latter column, no peak was observed for Cd(II).

#### Column conditioning

In the preliminary investigation of the retention times of the diethyldithiocar-

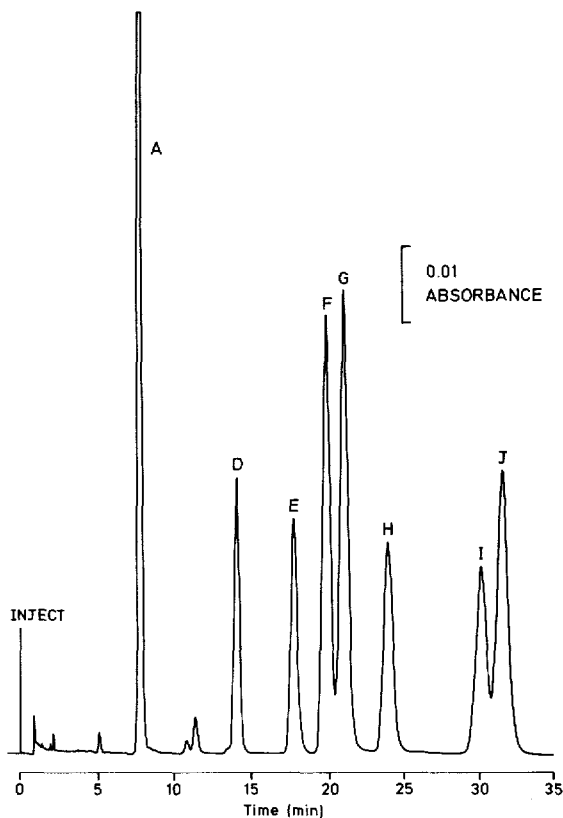


Fig. 2. Separation of a mixture of diethyldithiocarbamate complexes on a column conditioned by flushing with  $0.005\text{ M}$  EDTA. Conditions: mobile phase methanol-acetonitrile-water (40:35:25) containing  $1.25 \cdot 10^{-5}\text{ M}$  EDTA; detector sensitivity  $0.1\text{ a.u.f.s.}$ ; all other conditions and peak identities (including amounts of metals injected) as for Fig. 1.

bamate complexes, concentrated stock solutions of the complexes in chloroform (obtained by extraction of  $0.01\text{ M}$  metal ion solutions) were injected onto the column. A separation of a mixture of complexes was then performed using a concentrated mixture (Mixture A), prior to separation of a dilute mixture (Mixture B), as shown in Fig. 1. When a new column was used for separation of Mixture B, inadequate resolution or even total retention was observed. This indicated that prior injection of a concentrated mixture was necessary for satisfactory resolution of more dilute samples, suggesting that the residual adsorptive sites on the column must first be saturated prior to chromatography of samples containing trace amounts of metal complexes.

We have found that flushing the column with a dilute solution of  $\text{Na}_2\text{EDTA}$  and subsequent addition of EDTA to the mobile phase served as an effective column conditioning procedure. After such treatment, the column gave satisfactory resolution of Mixture B (Fig. 2).

Addition of EDTA to the mobile phase caused the peaks due to the diethyldithiocarbamate complexes of  $\text{Cd(II)}$  and  $\text{Pb(II)}$  to elute at the void volume. This change is presumably due to a ligand exchange reaction between EDTA and the metal com-

TABLE II

DETECTION LIMITS FOR METAL DIETHYLDITHIOCARBAMATE COMPLEXES, EXPRESSED AS THE AMOUNT (OF METAL) REQUIRED TO GIVE AN ABSORBANCE OF 0.0003 USING A 10- $\mu$ l INJECTION

<i>Species</i>	<i>Detection limit</i> ( $\mu$ g)
Cd(II)	0.7
Pb(II)	18.4
Ni(II)	1.0
Co(III)	0.5
Cr(III)	0.5
Se(IV)	1.0
Cu(II)	1.5
Hg(II)	6.1
Te(IV)	1.3

plex which releases the diethyldithiocarbamate ligand. This ligand, due to its ionic nature, is then unretained on the column. For this reason, Cd(II) and Pb(II) were omitted from the mixture used to obtain the chromatogram shown in Fig. 2. The EDTA column conditioning procedure was found to give reproducible peak heights and retention times. Table I shows the capacity factors obtained using this method.

#### *Analytical applications*

To illustrate the analytical utility of the chromatography of diethyldithiocarbamate complexes, simultaneous calibration plots were prepared for the species shown in Fig. 2. Excellent linearity was observed for all complexes for injection amounts covering the sub-microgram range. The detection limits are listed in Table II, and it is expected that these limits could be significantly lowered through the use of more sensitive detection wavelengths and improved solvent extraction and sample enrichment techniques.

An interesting possibility is the application of HPLC to the simultaneous analysis of organic and inorganic species in the same sample. Such an analysis is required for electroplating solutions containing metal ions and organic brightening agents. Typical organic brighteners include aromatic sulphonic acids, sulphonamides and heterocyclic sulphonic acids. Fig. 3 illustrates the separation of a synthetic electroplating solution containing salicylsulphonic acid and sulphanilamide, together with a mixture of six inorganic ions. The acidic nature of the organic components of the mixture necessitated use of a cationic ion-pairing reagent, however this did not alter the resolution or retention of the diethyldithiocarbamate complexes.

#### CONCLUSIONS

Radial compression columns have been shown to be very useful for the separation of up to eight diethyldithiocarbamate complexes, however careful attention must be given to the conditioning of these columns before use. This conditioning may be achieved by injection of concentrated solutions of diethyldithiocarbamate complexes,

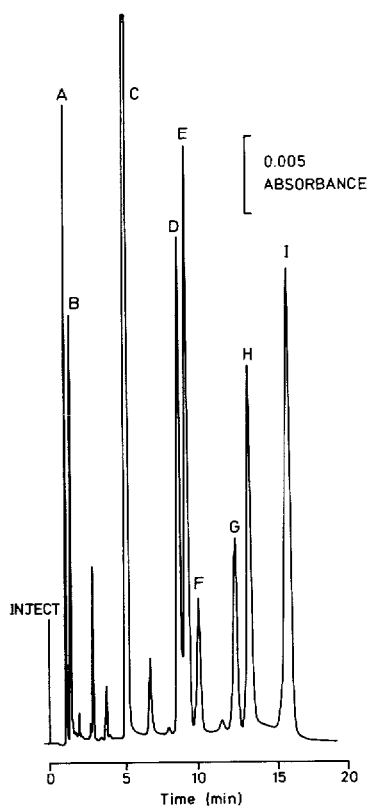


Fig. 3. Separation of a synthetic electroplating solution containing two organic brightening agents and six inorganic ions. Conditions: mobile phase methanol-acetonitrile-water (40:39:21) containing  $1.11 \cdot 10^{-5} M$  EDTA and  $5 \cdot 10^{-3} M$  tetrabutylammonium phosphate; detector sensitivity 0.05 a.u.f.s.; injection volume  $7 \mu\text{l}$ ; other conditions as for Fig. 2. A = salicyl sulphonic acid,  $0.06 \mu\text{g}$ ; B = sulphanilamide,  $0.03 \mu\text{g}$ ; C = disulfiram, D = Ni(III),  $0.10 \mu\text{g}$ ; E = Co(III),  $0.05 \mu\text{g}$ ; F = Cr(III),  $0.04 \mu\text{g}$ ; G = Se(IV),  $0.22 \mu\text{g}$ ; H = Cu(II),  $0.13 \mu\text{g}$ ; I = Hg(II),  $0.36 \mu\text{g}$ .

of by flushing the column with a dilute solution of EDTA, with subsequent addition of EDTA to the mobile phase.

The radial compression column did not significantly reduce the occurrence of substitution reactions between nickel (from the stainless steel in the chromatographic system) and the complexes of Fe(III), Cd(II) and Zn(II), indicating that other stainless-steel components, in addition to the column frits, also contribute to this phenomenon. The simultaneous analysis of organic and inorganic components of a sample has been illustrated using a synthetic electroplating solution containing organic brightening agents and inorganic ions.

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## REFERENCES

- 1 G. Schwedt, *Top. Curr. Chem.*, 85 (1979) 159.
- 2 G. Schwedt, *Chromatographia*, 12 (1979) 613.
- 3 E. B. Edward-Inatimi and J. A. W. Dalziel, *Anal. Proc.*, 17 (1980) 40.
- 4 O. Liška, J. Lehotay, E. Brandšteterová, G. Guiochon and H. Colin, *J. Chromatogr.*, 172 (1979) 384.
- 5 T. W. O'Laughlin and T. P. O'Brien, *Anal. Lett.*, A11 (1978) 829.
- 6 P. C. Uden and I. E. Bigley, *Anal. Chim. Acta*, 94 (1977) 29.
- 7 M. Moriyasu and T. Hashimoto, *Anal. Lett.*, A11 (1978) 593.
- 8 P. Heizmann and K. Ballschmiter, *J. Chromatogr.*, 137 (1977) 153.
- 9 G. Schwedt, *Fresenius Z. Anal. Chem.*, 295 (1979) 382.
- 10 G. Schwedt, *Chromatographia*, 12 (1979) 289.
- 11 G. Schwedt, *Fresenius Z. Anal. Chem.*, 288 (1977) 50.
- 12 P. C. Uden, B. D. Quimby, R. M. Barnes and W. G. Elliott, *Anal. Chim. Acta*, 101 (1978) 99.
- 13 G. Schwedt, *Chromatographia*, 11 (1978) 145.
- 14 T. Tande, J. E. Pettersen and T. Torggrimsen, *Chromatographia*, 13 (1980) 607.
- 15 G. Schwedt and A. Schwartz, *J. Chromatogr.*, 160 (1978) 309.
- 16 A. M. Bond and G. G. Wallace, *Anal. Chem.*, 53 (1981) 1209.
- 17 M. Yamazaki, S. Ichinoki and R. Igarashi, *Bunseki Kagaku (Jpn. Analyst)*, 30 (1981) 40.
- 18 N. Haering and K. Ballschmiter, *Talanta*, 27 (1980) 873.
- 19 Y.-T. Shih and P. W. Carr, *Talanta*, 28 (1981) 411.
- 20 G. D. Thorn and R. A. Ludwig, *The Dithiocarbamates and Related Compounds*, Elsevier, New York, 1962, p. 158.