National Library of Canada
Canadian Theses Service
Ottawa, Canada
K1A 0N4

Bibliothèque nationale du Canada
Service des thèses canadiennes

NOTICE

The quality of this microform is heavily dependent upon the quality of the original thesis submitted for microfilming. Every effort has been made to ensure the highest quality of reproduction possible.

If pages are missing, contact the university which granted the degree.

Some pages may have indistinct print especially if the original pages were typed with a poor typewriter ribbon or if the university sent us an inferior photocopy.

Previously copyrighted materials (journal articles, published tests, etc.) are not filmed.

Reproduction in full or in part of this microform is governed by the Canadian Copyright Act, R.S.C. 1970, c. C-30.

AVIS

La qualité de cette microforme dépend grandement de la qualité de la thèse soumise au microfilmage. Nous avons tout fait pour assurer une qualité supérieure de reproduction.

S'il manque des pages, veuillez communiquer avec l'université qui a conféré le grade.

La qualité d'impression de certaines pages peut laisser à désirer, surtout si les pages originales ont été dactylographiées à l'aide d'un ruban usé ou si l'université nous a fait parvenir une photocopie de qualité inférieure.

Les documents qui font déjà l'objet d'un droit d'auteur (articles de revue, tests publiés, etc.) ne sont pas microfilmés.

La reproduction, même partielle, de cette microforme est soumise à la Loi canadienne sur le droit d'auteur, SRC 1970, c. C-30.

NL-339 (r. 88/04)

Canadä

Gradient Columns in the GC Analysis of Metal Diethyldithiocarbamates

A. Sadek Elgawahergui

A Thesis

in

The Department

of

Chemistry

Presented in Partial Fulfillment of the Requirements
for the Degree of Master of Science at
Concordia University
Montréal, Québec, Canada

June 1987

ISBN 0-315-41615-7

ABSTRACT

GRADIENT COLUMNS IN THE G C ANALYSIS OF METAL DIETHYLDITHIOCARBAMATES

A. Sadek Elgawahergui

Novel gas chromatographic columns were prepared, characterized, and used in the separation of diethyldithiocarbamates $M(DDC)_n$ of Zn, Cd, Cu, Ni, Pb, Hg, Co, Cr, and Mo. Solute retention, column efficiency, and resolution of $M(DDC)_n$ were studied on these gradient columns. Separation of complex $M(DDC)_n$ mixture including up to nine metals was achievable by using 3-2-1% QF-1 or (1-2-3%) QF-1 + 1% OV-225 gradient columns.

All gradient columns studied were found to be superior to conventional columns in terms of efficiency, reduced analysis times, column bleeding, resolution, and column capacity. The relative standard deviation for retention times and resolution data was generally below 5%. Linearity was obtained in most cases when calibrating plots were prepared for the various $M(DDC)_n$.

The gradient columns studied were characterized by the Retention Index System.

ACKNOWLEGEMENTS

The author wishes to express his sincere thanks to Dr. R.H. Zienius for supervising this project and for his continuous assistance and valuable suggestions.

Special thanks to my family for their continuous support and understanding throughout this project.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

1.0 PURPOSE OF THIS THESIS

The main objective of this thesis was to prepare novel gradient gas chromatographic columns with higher resolving characteristic than those of the conventional columns, to characterize such columns with respect to retention indices, and to use these columns to accomplish the separation of metal diethyldithiocarbamates of Zn, Cd, Cu, Ni, Pb, Cr, Hg, Co,and Mo.

1.1 INTRODUCTION

1.1.1 Gradient Columns in Gas Chromatography

Ever since James & Martin (1) introduced gas-liquid chromatography in 1952 the search has continued to improve column technology to attain faster and better separations. The main objective of a chromatographic method is to separate components in a mixture. If a separation cannot be achieved by isothermal chromatography then some critical parameter may be varied to enhance the resolution and minimize the analysis time. As an example, temperature programming has provided a simple method for the analysis of wide-boiling point mixtures in a minimum of time with enhanced resolution. Flow programming, which offers these features as well, can be utilized for the separation of thermally sensitive solutes and with stationary phases exhibiting high bleed at temperatures approaching their maximum temperature limit.

Another approach for improving resolution has involved the development of gradient columns by Porath(2). He described a step-gradient adsorption column constructed of six sections where each successive section down the column contained charcoal deactivated with stearic acid to a lesser extent than the preceeding section. Satisfactory results were claimed for the analysis of pea-root exudate.

Purnell (3) suggested that a gas chromatographic column in which the capacity factor, k', of the solute was varied within the column might exhibit capabilities similar to those offered by programmed temperature gas chromatography.

Meloan et al (4,5) obtained excellent agreement between theoretical predictions and experimental results for multi-stage, gradient loaded squalane columns prepared by linearly or exponentially decreasing the percent loading of the the stationary phase, squalane, from the beginning to the end of the column. Improved column efficiency and resolution of solutes of low or intermediate k' was observed on a linearly decreasing stationary phase gradient column. The overall k' was reduced to 50% of its value on a conventional column having a liquid loading equivalent to that at the start of the gradient column. Utilizing exponential gradient columns, superior resolution and efficiency and shorter retention times were obtained than was possible with either conventional, uniformly loaded column or linear gradient columns of the same average liquid loading.

In studying columns possessing a linear variation in k' from the beginning to the end of the column they made use of solute retention, column efficiency and resolution factors for characterization. For solute retention they derived the following equation:

$$k' = k'_o \left(\frac{(3a^5 - 5a^3 + 2)}{[5(a^2 - 1)(a^3 - 1)]} \right) \qquad \text{EQ-1}$$

Where;

k'_o = the initial capacity factor.

a = p_i/p_o the ratio of inlet to outlet pressure.

This equation predicts that the capacity factor on the gradient column is a function of the initial capacity factor, k'_o, and the inlet/outlet pressure ratio, a. As this ratio increases, the solute retention on the more heavily loaded end of the column must have a disproportionately large effect. Hildebrand et al (6) observed this to be the case using a column consisting of two stages in series, the simplest example of a gradient column. Equation (1) provided some theoretical basis for interpreting their observations.

Meloan et al (4) derived an equation for the resolution, R, of two closely spaced peaks on gradient columns:

$$R = \sqrt{N}\left(\frac{\alpha-1}{\alpha}\right)\left(1-\frac{\ln(1+k'_{o2})}{k'_{o2}}\right) \quad \text{EQ-2-}$$

Where;

c = is the selectivity factor

k'_{o2} = is the initial capacity factor for the second peak.

They offered an explanation for the better resolution that was obtained and in a shorter time on a gradient column having a decreasing load. Physically, as the components of the mixture move down the column their resolution as well as their relative velocities increase as they move onto ever more lightly loaded regions of column packing. It was predicted that a gradient column with decreased loading would accept at least a two fold increase in sample size and feed volume compared to a regular column. Meloan et al also recommended the use of gradient columns rather than temperature programming in cases where severe column bleeding results at increased temperatures and where working with sensitive solutes.

Bunting and Meloan (5) found that the resolution obtained with exponential gradient columns was superior to that of linear gradient columns or of conventional columns containing the same average loading of stationary phase. Typical of the columns they studied were those prepared in a 25-stage stepwise approximation of a continuous gradient using squalane coated on Chromosorb P 100 cm long.

Merritt et al (7) studied the chromatographic performance of columns comprised of segments with different liquid phase loading with respect to efficiency, resolution and solute capacity factors. Their aim was to decrease the retention and analysis time of solutes that would normally be strongly retained on conventional columns. They constructed columns with segments consisting of 15-5-3-2% Carbowax 20m coated on 80/100 mesh Chromosorb W and 10-5-3-1% SP 2100 on 100/120 mesh Supelcoport and studied their behaviour as a function of flow direction. They reported that advantages with respect to separation or analysis time could be obtained by a judicious choice of flow direction. It was also reported that using the high-to-low percent loading configuration may provide increased sample capacity and improved separation whereas the reverse configuration, under the same experimental conditions, offers reduced retention times and greater column efficiency. An explanation was given for the behavior of the segmented columns based on the volumetric flow rate of the carrier gas. They also noted decreased liquid phase bleeding for gradient columns. In this connection, Giddings (8) studied the vapor pressure of stationary phases on GC supports and concluded that at loadings lower than 2% the vapor pressure dropped sharply, resulting in decreased bleed effect and longer column life. They explained this phenomena as being due to the strong attraction between the support and the stationary phase.

Duty (9) studied gradient loaded preparative columns. A comparison was made between a linearly gradient column and a conventional column, each 10 ft long. Plate number, skewing and resolution were calculated at several flow rates and sample sizes, using toluene and benzene as solutes. The stationary phase used was Silicon Fluid DC710, at a column temperature of 130°C. It was concluded that distinct advantages were obtained with the gradient column, advantages being greatest for sample sizes of more than 0.1ml. Little advantage was gained in increasing the flow rate of the carrier gas from 100ml/min to 300 ml/min.

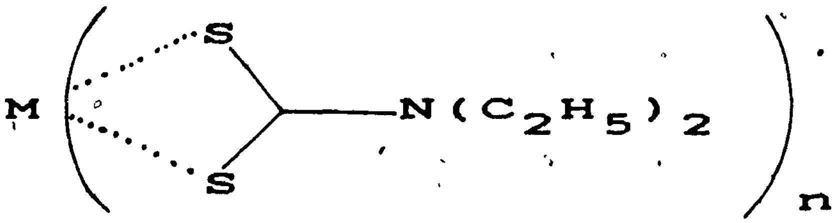
Given the above advantages reported for gradient columns by workers who restricted their experiments to model compound mixtures (Purnell (3), Locke and Meloan (4), Bunting and Meloan (5), Barry and Merritt (7) and Duty (9)), it was decided to attempt to apply this technique to the separation of metal diethyldithiocarbamate compounds which are not all that successfully separated with conventional columns. It was also expected that this ~~project~~ would aid in further defining the scope and the usefulness of gradient columns.

2.0 GAS CHROMATOGRAPHY OF METAL DIETHYLDITHIOCARBAMATES

The ligand, diethyldithiocarbamate has been used for metal ion derivatization prior to GC analysis because of its stability, volatility (10,11), ease of formation (12,13), and its large usage in the field of analytical chemistry (14-16). It has also been employed in sample clean-up and pre-concentration steps prior to metal determination by atomic absorption (17,18) and spectrophotometry (19).

D'Ascenzo and Wendlandt (10,11) established that metal diethyldithiocarbamates (DDC) (Fig 1) are thermally stable and volatile. They reported the thermal properties of

FIG. 1 STRUCTURE OF METAL DIETHYLDITHIOCARBAMATE.



M = Metal ion

n=Oxidation state of metal

Cd(II), Zn(II), Hg(II) AND Co(III) $(DDC)_n$.

Cardwell et al (12) carried out the thermal analysis of DDC 's of Ni(II), Cu(II), Pd(II), Zn(II), Cd(II); Hg(II), Pb(II), Pt(II), Ag(I), Fe(III) and Co(III). They reported that Ni, Zn, Fe and Co showed complete volatility under the conditions used. In GC analyses, all the complexes studied, except for those of Fe(III) and Ag(I), could be eluted successfully at column temperatures in the range of 220 - 245°C and with injector and detector temperature of 250°C. They reported separation of Zn, Cd, Pb and in another run of Zn, Ni, Pd, and Pt, on 5% OV 101 on Chromosorb W AW-DMCS. They reported column efficiencies of up to 1000 theoretical plates with stainless steel tubing columns (1m x 2.1mm, ID). They mentioned that DDC was the first chelate system found to be suitable for the gas chromatographic elution of Cd and Hg. They confirmed that the order of elution was in close agreement with the volatilities of the complexes as determined by thermal analysis.

Ahmad and Aziz (13) detected dimeric structures for Zn(II)DDC between 230 - 250°C and Cd(II)DDC between 230-260°C in the vapor phase. They established that the electron capture detection limits for Pb, Zn, Ni, Hg and Cd were 2-6 ng using a column of 3% SE 30 on Varaport 30 (100/120 mesh).

Radecki et al (20) determined Zn, Cu, and Ni (after conversion to diethyldithiocarbamates) in natural marine bottom sediments using a column packed with 5% OV-101, and, in another publication they reported determination of these three metals in sea sand and sea mud using a column of 5% OV-101 and 5% QF-1 on Gas Chrom Q at 240°C.

Liangbin et al (21) reported the separation and the determination of traces of Co(III) diethyldithiocarbamate using a column packed with 5% OV-101, and tetratriacontane as an internal standard. The limit of detection for Co was 0.01 ng. This method was used to determine Co in vitamin B_{12} and in a Co porphyrin complex.

Jiming et al (22) studied the volatility, thermal stability and gas chromatography of diethyldithiocarbamates of: n=(I) Ag, Au, Ti, and

n=(II), Cd, Cu, Hg, MoO_2, Ni, Pb, Pt and Zn, and

n=(III), As, Bi, Co, Fe, In, Sb, and n=(IV) Se, Te, Sn.

Differential thermal analysis and thermogravimetric analysis were carried out as well as GC.

Riekkola et al (23) reported the separation of Se(II), Zn(II), Cd(II), Pb(II), Pd(II), As(III), Co(III), and Rh(III) as their diethyldithiocarbamates with glass capillary columns coated with OV-101.

Neeb (24) summarized most of the GC work done with metal diethyldithiocarbamates and also published some work done with WGOT and SCOT capillary columns in a comparative study.

Carvajal and Zienius (25) reported highly efficient separation of mixture of up to five metal diethyldithiocarbamates, in a mixture containing Ni(II) or Hg(II), Cu(II) or Pb(II), Zn(II), Cd(II), and Co(III) using a 5% OV-101 and 5% QF-1 mixed stationary phase on Chromosorb W column using temperature programming 210°C to 250°C at 10°C/min. They extracted these metals at trace levels from synthetic sea water. Low ppb detection levels were reported. They also proposed theoretical principles to account both for the nature of the GC separation and for degradation problems encountered and overcome. It was reported that the peaks for Cu(II) and Pb(II) and for Hg(II) and Ni(II) DDC interfered with one another.

No work has been reported to date on successful separation of a mixture of more than five metal diethyldithiocarbamates (DDC), on packed GC columns.

3.0 General Features of the Column in GC

The column is the principal part of the GC system upon which the success or failure of a particular sample mixture separation depends. In gas-liquid chromatography both capillary and packed columns exist. The latter type is the one used in this project. A packed column consists of:

(i) Glass or metal tubing

(ii) packing material which is a solid support coated with a stationary phase.

3.1 SOLID SUPPORT

The primary function of the support material is to provide a surface onto which the stationary phase is deposited uniformly. The choice and treatment of the solid support is very important. Supports made from Diatomite (26, 27), are most generally used, and are called diatomaceous earth, diatomaceous silica or Kieselguhr. Such supports are not ready for GC work when first prepared. The surface needs to be deactivated. To reduce the surface activity acid washing which removes some residual (Al and Fe) oxides, and silanization (described by Schupp (26)) which eliminates silanol groups are practiced.

Two of the best grades of support available were used in this project. Chromosorb WHP (CW-HP), (which is acid washed and DMCS treated), and Gas Chrom Q (GC-Q), (which is acid, base, and DMCS treated).

3.2 STATIONARY PHASE

The presence of the liquid phase is fundamental for the sample mixture partitioning process to occur. Each sample component must interact differently with the stationary phase, under the operating conditions in order to achieve separation. If the difference in chromatographic properties of the sample components is small, then maximum operating efficiency is required for a satisfactory separation to be achieved. The general requirements for a liquid phase are as follows (28, 29):

i) Low volatility, between 0..01 to 0.1 mm at operating temperature, for low "bleed", reasonable column life, and minimal effect on the detector.

ii) Thermal stability, which can be undermined by catalytic influence of the support as the temperature increases.

iii) Chemically inert towards the solutes of interest at operating column temperature

iv) Upper temperature limit should be relatively high compared to the intended operating temperature.

v) Must be liquid at column operating temperature. Some stationary phases are solids at room temperature. If the column temperature is below the melting point of the liquid phase , then the sample components' solubility in the phase is reduced drastically, which causes column overloading, poor separation, short retention times and distorted peaks. Hawkes and Mooney (29) have pointed out that the viscosity of the liquid phase is important in column performance. The liquid phase mass transfer coefficient (C_1 in the Van Deemter equation) is proportional to the viscosity and the square of the film thickness. Therefore, when the viscosity of the liquid phase is high a low column efficiency is expected. This phenomenon will be especially evident for heavily loaded columns owing to the d_f^2 factor (in the Van Deemter equation).

3.2.1 POLY(DIMETHYLSILOXANES) PDMS

The PDMS are the most widely used GC liquid phases, mainly because of their good thermal stability and narrow molecular weight distribution (29). Their general structure is shown in Fig 2. Two of this class of phases, SE 30 and OV-101, were used in this project. They generally separate organic compounds according to their boiling points, but higher molecular weight aromatic and aliphatic compounds appear to be separated by other mechanisms as well. For example, retention time and boiling point patterns are not consistent on OV-101 (30). One of the advantages of PDMS liquid phases is that diffusion and mass transfer of low molecular weight n-alkanes was found to be faster in higher molecular weight gums such as SE 30 than in lower molecular weight oils such as SF 96 (30). Faster diffusion means more efficient partitioning and better separation efficiency.

3.2.2 POLY(METHYLPHENYLSILOXANES)

Substitution of methyl group in PDMS by a phenyl group results in increased chromatographic selectivity (31,34) The polarity of the liquid phases increase with increasing phenyl content. The general structure is shown in Fig 3. Examples of such phases are OV-3 and OV-17.

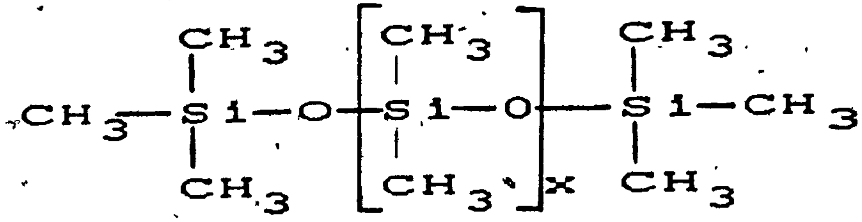
3.2.3 POLY(METHYL-TRIFLUOROPROPYLSILOXANES)

This type of liquid phase structure (eg QF-1), is shown in Fig 4. These materials show unique selectivity because the (CF_3) group has a high dipole moment and a strong electron acceptor ability (31), which results in an unusual retention order. For example, carbonyls and nitro groups are reported to elute after alcohols and esters (32,33) due to the ability of the CF_3 groups to interact with free electron pairs of the oxygen in the carbonyl and nitro groups. This selectivity is observed at 15% liquid phase loading (31). This order is reversed, however, at a loading of 6%. Using QF-1, 2-alkanones can be made to elute after 1- alkanols of the same carbon number even though the latter have higher boiling points (31).

3.2.4 POLY(CYANOALKYLSILOXANES)

The general structure is illustrated in Fig 5. The presence of the polar cyano group, a strong electron attracting group with a high dipole moment, results in greater retention of hydrocarbons bearing pi-electrons, such as olefin (31), aromatic and carbonyls. The most polar of such liquid phase are OV-275 and OV-225. These liquid

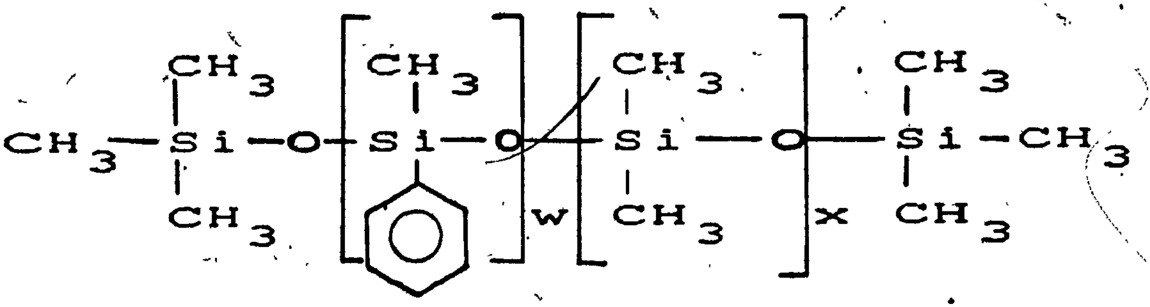
FIG. 2 STRUCTURE OF POLY(DIMETHYLSILOXANES)



LIQUID PHASE		X	TEMPERATURE RANGE
OV-101	OIL	100	20 - 350°C
SE-30	GUM	100	50 - 350°C
OV-1	GUM	100	100 - 350°C

WHERE ; X = MONOMER PERCENT

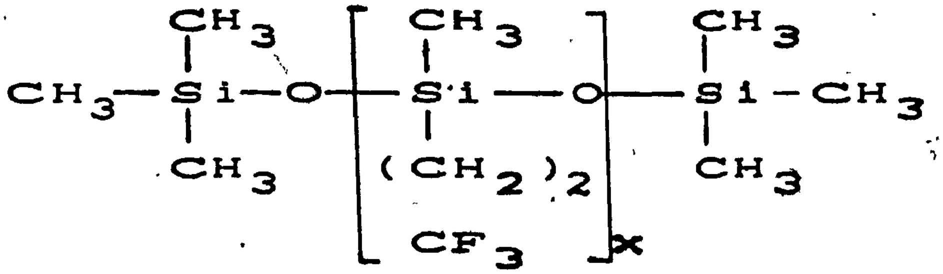
FIG. 3 STRUCTURE OF POLY(METHYLPHENYLSILOXANES)



LIQUID PHASE	% PHENYL	X	W	TEMPERATURE RANGE
OV-3	10	80	20	20 - 350°C
OV-7	20	60	40	20 - 350°C
OV-17	50	-	100	20 - 350°C

WHERE ; U AND W = MONOMER PERCENT

FIG. 4 STRUCTURE OF POLY(METHYL-TRIFLUOROPROPYLSILOXANES)

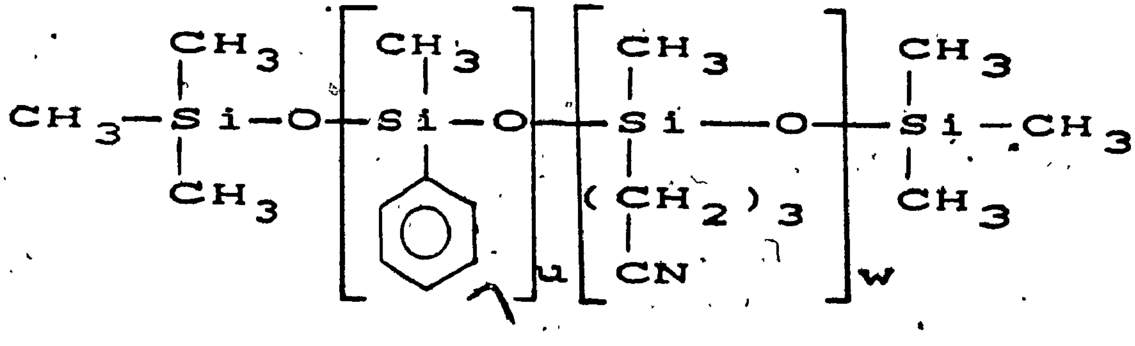


LIQUID PHASE	X	VISCOSITY(CST)	TEMPERATURE RANGE
OV-210	100	10,000	20 - 275°C
QF-1	100	*	20 - 250°C

* AVAILABLE IN VISCOSITY GRADES OF: 300,100,10,000 cst

WHERE ; X = MONOMER PERCENT

FIG. 5 STRUCTURE OF POLY(CYANOALKYLSILOXANES)



LIQUID PHASE	U	W	TEMPERATURE RANGE
OV-225	50	50	20 - 250°C

WHERE ; U AND W = MONOMER PERCENT

phases were used both alone and mixed with another liquid phase in this research project.

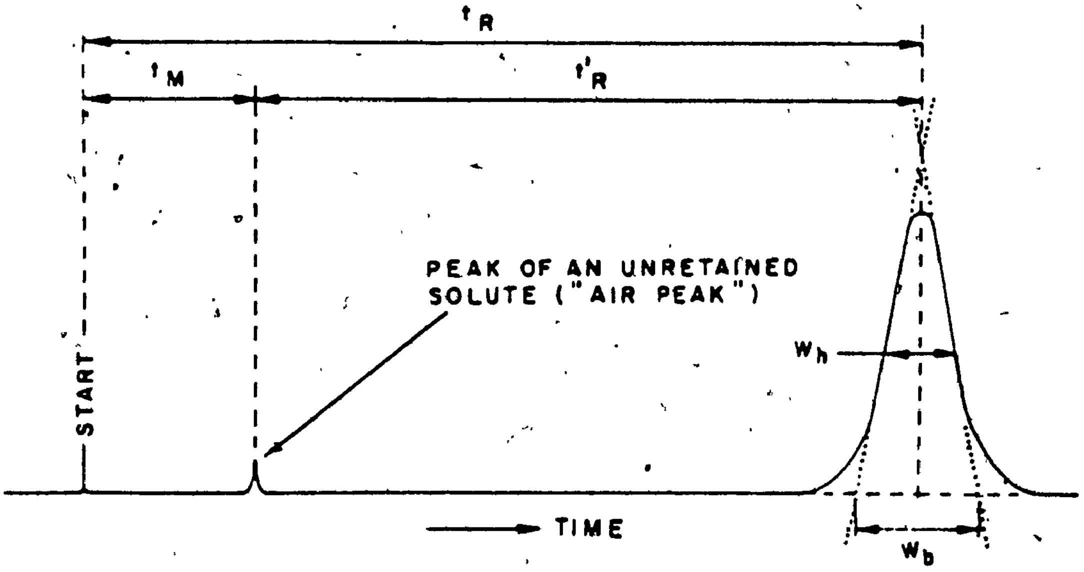
4.0 EVALUATION OF COLUMN PERFORMANCE

The main objective in gas chromatography is to separate components in a mixture. When all the operational conditions are optimum the best separation is achieved, and column efficiency is maximized. There are several parameters which can be measured to evaluate the performance of a column. A good gas chromatographic column is considered to have high separation power, high speed of separation, and high capacity (35). These factors are related and when any one is improved it is usually at the expense of another.

4.1 COLUMN EFFICIENCY

Giddings (36) emphasized the importance of evaluating a column in terms of the number of theoretical plates it contains. To measure the column efficiency in terms of the number of theoretical plates, N, from the chromatogram as shown in Fig 6 the following equations are used (36):

FIG. 6 DETERMINATION OF THEORETICAL PLATES



$$N = 16\left(\frac{t_R}{W}\right)^2 \qquad \text{EQ-3}$$

Where;

t_R = Retention time of sample

W = peak width at peak base

Also,

$$N = 5.55\left(\frac{t_R}{W_{0.5}}\right)^2 \qquad \text{EQ-4}$$

Where;

$W_{0.5}$ = Peak width at half height

It can be seen from EQ-3 and EQ-4 that the narrower the peak, the more efficient is the column.

The Height Equivalent to a Theoretical Plate or HETP is another measure of column efficiency, and it is obtained as follows:

$$H = \frac{L}{N} \qquad \text{EQ-5}$$

Where;

L = Column length in mm

N = Number of theoretical plates

The calculation of HETP allows comparisons between columns of different lengths.

The efficiency of a column is related to column variables by the Van Deemter equation (37).

$$HETP = A + \frac{B}{u} + C \cdot u \qquad \text{EQ-6}$$

It accounts for the mechanisms affecting column performance, and these are:

a - Multipath effect or eddy diffusion (A term)

b - Longitudinal molecular diffusion (B term)

c - Resistance to mass transfer in gas and liquid phases (C term)

u the linear gas velocity (or flow rate) through the chromatographic column. The linear gas velocity is given by:

$$u = \frac{\text{length of column, cm}}{\text{retention time of air, sec}}$$

If HETP is plotted against u one obtains a hyperbola with a minimum HETP, see Fig 7. The minimum corresponds to the flow rate (u optimum) at which the column is operating most efficiently. However, owing to the compressibility of the carrier gas, u is not constant over the entire length of the column. Hence only a small section can operate at maximum efficiency.

The expanded versions of the Van Deemter equation are well documented and reviewed in the literature (27,35, 38, 39).

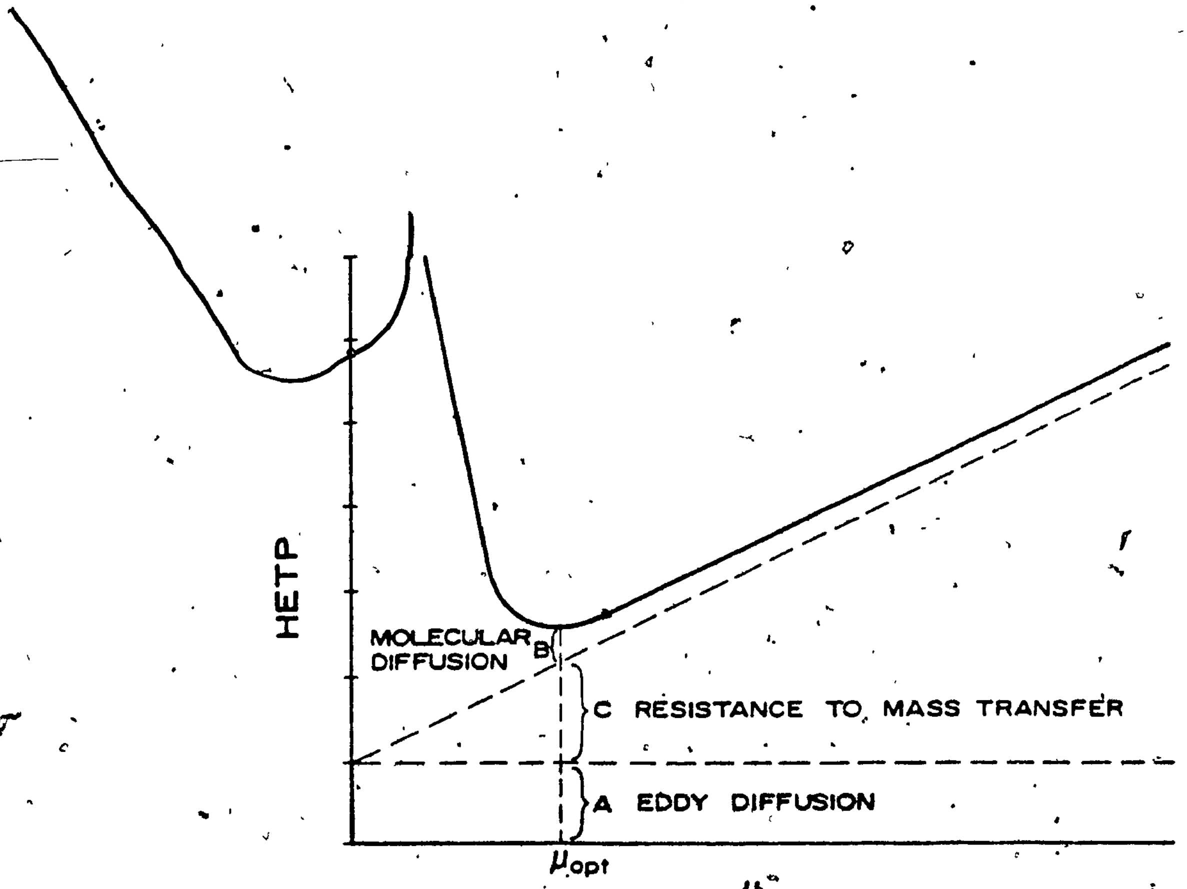
4.2 CAPACITY FACTOR, k'

Column efficiency depends on the capacity factor (k'). The smaller the capacity factor the harder it is to achieve the separation.

The capacity factor k' can be calculated by using the following relationship:

$$k' = \frac{t_R - t_m}{t_m} \qquad \text{EQ-7}$$

FIG. 7 PLOT OF HETP VS GAS VELOCITY (39)



Where, t_m = is the gas hold up time (retention time of a non retained component such as air injected together with sample)

Capacity of a column can also be arrived at by the consideration of the partition coefficient, K_r:

$$K_r = \frac{C_1}{C_M} = B\left(\frac{W_1}{W_M}\right) \qquad \text{EQ-8}$$

Where;

C_1 = concentration of solute in the liquid phase, g/ml

C_m = concentration of solute in the mobile phase, g/ml

W_1 = weight of solute in the liquid phase, g

W_m = weight of solute in the mobile phase, g

B = phase ratio = volume of the gas phase to volume of the liquid phase.

The amount of sample that can be injected into a column without overloading depend on the liquid phase loading which in turn influences the phase ratio. A large liquid loading lowers the phase ratio and diminishes the partition coefficient.

4.3 RESOLUTION (R_s)

The degree of separation between two adjacent peaks (35) is given by the resolution factor (R_s) Fig 8;

$$R_s = \frac{2\left(t_{R_2} - t_{R_1}\right)}{W_1 + W_2} = \frac{2\,\Delta t}{W_1 + W_2} \qquad \text{EQ-9}$$

If R = 1.0, the resolution of two equal-area peaks is approximately 98% complete. If R = 1.5, baseline separation is obtained (40). If R_s is greater than 2.0, an additional (R_s - 1) peak should be resolvable between the two peaks in question (41).

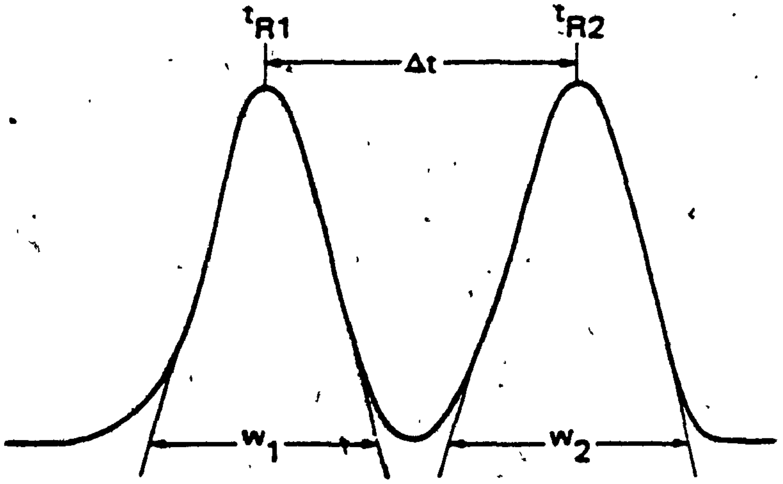
The relationship between R_s, k', and N is as follows:

$$R_s = \sqrt{\frac{N}{4}}\left(\frac{\alpha - 1}{\alpha}\right)\left(\frac{k'_2}{1 + k'_2}\right) \qquad \text{EQ-10}$$

Where α is the selectivity factor defined as:

$$\alpha = \frac{t'_{R_2}}{t'_{R_1}} \qquad \text{EQ-11}$$

FIG. 8 DEGREE OF SEPARATION BETWEEN TWO ADJACENT PEAKS.



and N and k'_2 refer to the peak eluted second.

4.4 QUANTITATIVE ANALYSIS

The size of a chromatographic peak is proportional to the amount of material contributing to that peak and can be measured in a number of ways (35). Two basic concepts can be used, namely:

- Peak height measurement. However, peak shape and height are liable to be changed by variations in operating conditions such as temperature, and carrier gas flow.
- Peak area measurement, which may be accomplished in various ways.

An electronic integrator with a relative standard deviation of 0.44%, was used in this project for peak height and area measurements (39).

5.0 CHARACTERIZATION OF COLUMNS BY RETENTION INDICES

Since the introduction of gas chromatography, numerous authors and committees have studied gas chromatographic data with a view towards its classification to make possible comparisons between laboratories. Various proposals led to the adoption of solute retention times relative to those of standard compounds to catalogue data (42), i.e

$$r_{x/s} = \frac{t'_{R(x)}}{t'_{R(s)}} \qquad \text{EQ-12}$$

where $t'_{R(x)}$ is the retention time of the substance of interest and $t'_{R(s)}$ is that of the standard. The basic shortcoming here was the inability of workers to agree on a single standard. Therefore, published data refer to widely different standards. To overcome this difficulty, Kovats proposed the retention index system (43). The retention behavior of the substance of interest is expressed relative to that of a series of closely related standard paraffins. The retention indices of the normal paraffins are arbitrarily set as 100 times the number of carbon atoms. The calculation of the retention index is accomplished by the following equation:

$$I_s = 100\left\{\frac{\log X_s - \log X_n}{\log X_{n+1} - \log X_n}\right\} + 100n \qquad \text{EQ-13}$$

Where;

I_s = Retention index of sample

X_s = Retention time of sample

X_n = Retention time of n-Paraffin peak preceding the sample peak

X_{n+1} = Retention time of the n-Paraffin peak following the sample peak

n = Number of carbon atoms in the n-Paraffin eluted prior to the compound

Retention index data is used both for the identification of unknown peaks and characterization of liquid phases. The difference in retention indices for a compound on various liquid phases is a characteristic of the polarity of the phases. This has led several researchers to propose the characterization of liquid phases using a multi component test mixture, referred to as the "Polarity Test Mixture" (44, 45).

Numerous modifications of the retention index equation have been proposed for a variety of applications. These are thoroughly reviewed in the literature (46). The overall polarity and selectivity of a column can be determined by the method proposed by McReynolds (47) which is based on theoretical considerations suggested by Rohrschneider (48,49). The basic principle of the Rohrschneider/ McReynolds system is the additivity of intermolecular forces as evaluated from the differences in retention index values for a series of test probes measured on the column to be characterized and on a squalane column used as a non-polar reference phase (50, 51). The chosen set of test probes adequately responds to the principal molecular interactions responsible for retention in gas chromatography: i.e, dispersion, orientation, induction, and donor-acceptor complexation (including hydrogen bonding). Rohschneider suggested the use of benzene, ethanol, methylethylketone, nitromethane, and pyridine as test probes. McRenoylds recommended ten probes (Table 1) including Rohrschneider's five except that butanol, 2-pentanone and nitropropane were substituted for the lower molecular weight homologs used by Rohrschneider. To characterize a column, McReynolds constant are determined using the following equation:

$$\Delta I = I_s - I_{sq} \qquad \text{EQ-14}$$

Where;

I_s = Retention Index of sample on the phase being tested

I_{sq} = Retention Index of sample on squalane

The calculation is repeated for each probe. Liquid phase loadings of 10 - 20% and column temperatures of 100°C to 120°C are commonly used. The Kovats and McReynolds retention index systems were used in this project to characterize gradient loaded columns.

TABLE 1

INTERACTIONS CHARACTERIZED BY McREYNOLDS' PROBES (48)

TEST SUBSTANCE	INTERACTIONS MEAUSRED	CHARACTERISTIC SUBSTANCE GROUP
BENZENE	PRIMARILY DISPERSION WITH SOME WEAK PROTON ACCEPTOR PROPERTIES.	AROMATICS, OLEFINS
BUTANOL	ORIENTATION PROPERTIES WITH BOTH PROTON DONOR AND PROTON ACCEPTOR CAPABILITIES.	ALCOHOLS, NITRILES, ACIDS
2-PENTANONE	ORIENTATION PROPERTIES WITH PROTON ACCEPTOR BUT NOT PROTON DONOR CAPABILITIES	KETONES, ETHERS, ALDEHYDES, ESTERS, EPOXIDES, DIMETHYLAMINO DERIVATIVES
NITORPROPANE	DIPOLE ORIENTATION PROPERTIES	NITRO AND NITRILE DERIVATIVES
PYRIDINE	WEAK DIPOLE ORIENTATION WITH STRONG PROTON ACCEPTOR CAPABILITIES. PROTON DONOR PROPERTIES ARE ABSENT	AROMATIC BASES
2-METHYL-2PENTANOL		BRANCHED CHAIN COMPOUNDS, PARTICULARLY ALCOHOLS
IODOBUTANE		HALOGENATED COMPOUNDS
2-OCTYNE		
1,4-DIOXANE	ORIENTATION PROPERTIES WITH PROTON ACCEPTOR BUT NOT PROTON DONOR CAPABILITIES.	
CIS-HYDRINDANE		

6. EXPERIMENTAL

6.1 PREPARATION OF METAL DIETHYLDITHIOCARBAMATES

The diethyldithiocarbamates of Zn, Ni, Cu, Co, Mn, Pb, Hg, Cd, Cr, and Fe were prepared by Carvajal's general procedure, 21. The metal salts used, sulfate or nitrate, were obtained from J.T. Baker Chemical Co., Phillipsburg, New Jersey, USA, or Fisher Scientific Company, Montreal, Canada. A few modifications were introduced in the method; for example, purification of the sodium diethyl dithiocarbamate reagent (certified, A.C.S. Fisher Scientific Co.) by extraction with a chloroform - acetone mixture (1:1 V/V), as recommended by Radecki (52). The water used for dissolving the metal salts and the sodium diethyldithiocarbamate was deionized and purified with a reverse osmosis, millipore water purification system. The isolated complexes were purified by recrystallization from chloroform - ethanol 1:1.

6.2 PREPARATION OF COLUMN PACKINGS

The method utilized in this project to prepare packings was the rotary evaporation method as outlined by Supina (27, 35) and McNair (39).

The percentage of stationary phase was calculated as follows (38, 53):

$$\% \text{ LOADING} = \frac{\text{wt. of stationary phase} \times 100}{\text{wt. of solid support}}$$

The supports used to prepare the packings are listed in Table 2. The stationary phases and supports were obtained from Chromatographic Specialities Inc. Brockville, Canada, and Applied Science Laboratories, State College, PA., USA.

TABLE 2

SUPPORTS USED

SUPPORT	MESH SIZE	SOURCES
Chromosorb W-HP (CW-HP)	80/100 & 100/120	CHROMATOGR. SPEC. INC.
Gas Chrom Q (GC-Q)	80/100	APPLIED SCIENCE LABS

6.2.1 PROCEDURE FOR COATING SUPPORTS USING THE ROTARY EVAPORATOR

The typical procedure is illustrated for the case of the packing made to contain 3% QF-1 on CW-HP 80/100 mesh.

All glassware was cleaned thoroughly, before use, with soap and water, rinsed with distilled water and finally dried with acetone and left in a hot air oven overnight.

A 3.05 g portion of support was weighed into a beaker. Any aggregated portions were excluded. The support was dried overnight at 120°C and then cooled in a desiccator. QF-1 0.092 g, was weighed out and disssolved in 100 ml of degassed ethyl acetate.

The rotary evaporator flask was washed with distilled water, then with 25 ml of acetone, and finally with 25 ml of ethyl acetate. It was dried with acetone and left in the oven at 120°C for one hour. The prepared stationary phase solution was then transferred to the rotary flask. The dry support was added slowly to the solution, with slight swirling. The rotary flask was rotated at 3-5 rpm and purged with pure nitrogen. Vacuum was applied, and the flask was immersed in a water bath at 60°C. Care was taken to assure that no tumbling would occur. At near dryness, rotation was reduced to about 1-2 rpm to reduce friction and thereby minimize support breakage.

After removal of solvent was complete, the rotary flask was stoppered with filter paper and put in a vacuum oven at 60°C for 3 hours. The free flowing packing was weighed. The final weight was compared to the sum of the original weights of stationary phase and support. The deviation in weights before and after was always within 0.1 to 0.15%.

For mixed phases, such as QF-1 + OV-225, the same procedure was used since ethyl acetate was a common solvent for both stationary phases. In situations where the solvent for one stationary phase was not suitable for the other, coating was carried out in two stages, the stationary phases being coated on the support individually one after the other.

6.2.2 TREATMENT OF COLUMN TUBING PRIOR TO PACKING

Although glass tubing is the best choice for analysis of most compounds, it must be chemically pre-treated to reduce surface activity since glass contains the same detrimental silanol groups which are found on diatomite surfaces. The recommended method of treatment is washing with a solution of 5% dimethyldichlorosilane in toluene, which provides deactivation up to 350°C to 400°C (21, 38). This procedure was followed in this project.

To remove any residual materials, the tubing was washed with deionized water, then with acetone, followed by hexane and finally dried with pure dry nitrogen gas.

The tubing was then filled with Sylon - CT, from Supelco Inc.,Oakville, Canada, which is a 5% dimethyldichlorosilane in toluene solution, and allowed to stand for two minutes. The tubing was then emptied, rinsed with toluene and finally with methanol. Dry nitrogen was then blown through the column after which it was kept stoppered at both ends until it was filled with packing. This was carried out whenever a length of tubing was to be filled with packing regardless of whether it was new or previously used.

6.2.3 PROCEDURE FOR LOADING PACKING INTO TUBING

The glass columns used in this project were 60cm x 2 mm I.D, U - shaped, or 180 cm x 2 mm I.D, coiled shaped. For filling purposes the tubing was clamped onto a stand. A minimum size silanized glass wool plug was inserted in one end of the tubing, and a vacuum source was connected at this end. A small stainless steel funnel was inserted in the other end. The prepared packing was poured gently into the funnel, and a slight vacuum was applied, gently enough to prevent breakage of the support. The tubing was gently vibrated during packing. After The tubing was completely packed a silanized glass wool plug was inserted in the inlet end of the tubing.

Columns prepared in this way were temperature conditioned before use. When not in use they were stored with the ends tightly capped.

6.2.4 COLUMN CONDITIONING

All columns were pre-conditioned at 240°C with nitrogen passing through at a flow rate of 30 ml/min for 16 hours. Columns were never removed hot from the oven. The carrier gas flow was not discontinued until columns were at ambient temperature.

6.3 PROCEDURE FOR PACKING A GRADIENT COLUMN

Depending on the number of gradient steps planned, the tubing was marked off into appropriate number of equal lengths. Different sections were filled with packing containing the same stationary phase in different amounts, or, different stationary phases. The latter combination packings OV-101 and QF-1 are listed in Table 3.

For combinations experiments intended to evaluate the effect of gradient direction, duplicate columns were packed such that in one the OV-101 was at the injector and QF-1 at the detector, while in the other, QF-1 was at the injector, and OV-101 at the detector.

6.4 CARRIER GAS

The carrier gas used was nitrogen (Canadian Liquid Air, Montreal, Canada), ultra pure grade (certified 99.99%) passed through a combination molecular sieve and oxygen trap (Chromatographic Specialities Inc, Brockville, Canada), which was replaced after installation of every sixth carrier gas cylinder.

6.5 GC EXPERIMENTAL CONDITIONS

All experimental work was performed on a Hewlett-Packard Gas Chromatgraph Model HP 5880A, equipped with a dual flame ionization detector, Model HP 19301, and electronic carrier gas flow/pressure controller, Model HP 19306A. Sample injections were carried out either manually with a 10 ul Hamilton Microsyringe, Model 701N, accuracy of ± 1%, or, with an Automatic Sample Injector, Model HP 7671A, which was equipped with an automatic transport system, Model HP 19324B. The automatic transport system could be

TABLE 3

GC COMBINATION PACKING COLUMNS

% LENGTH CONTAINING 2.5% OV-101	% LENGTH CONTAINING 2.5% QF-1
100	0
75	25
66.7	33.3
50	50
33.3	66.7
25	75
0	100

TABLE 4

GAS CHROMATOGRAPHIC EXPERIMENTAL CONDITIONS

PARAMETER	RANGE		
	HIGH	OPTIMUM	LOW
NITROGEN FLOW RATE, ml/min	60	30	20
HYDROGEN FLOW RATE, ml/min	30	30	30
AIR FLOW RATE, ml/min	400	400	400
INJECTOR-DETECTOR TEMP. C^{o}	250	250	250
CHART SPEED, cm/min	1	1	0.5
ATTENUATION	2 TO 10	2 TO 3	2 TO 4
SAMPLE VOLUME, ul	4	0.8	0.7

programmed to automatically move to either of two preselected injection port locations. The accuracy and reproducibility was better than ± 1%.

Chromatograms were displayed on a terminal level four Model HP 19308D with alphanumeric keyboard, basic programming and single column compensation (SCC) systems (Appendix II), The chart speed used was 1 cm/min, or 0.5 cm/min.

Experimental conditions are shown in Table 4. It should be noted that the parameters listed under "optimum" were generally used. The column temperature was varied depending on the analysis being performed. The hydrogen and air flow rates were set according to Hewlet-Packard recommendations for optimum performance (54).

6.6 MELTING POINTS DETERMINATION

Melting points were determined using a Fisher-Johns hot-stage apparatus (55) (Appendix I, Table 45).

6.7 IR SPECTRA

The IR spectra of each metal chelate was obtained as a solid dispersion, (approximately 1% w/w), in dry KBr using a mini-press as described by Silverstein (56). The instrument used was A Perkin - Elmer-710A Infrared Spectrophotometer (fast scan speed and normal slit). All spectra were obtained were in the range of 600 to 4000 cm^{-1} (Appendix I, Fig 53 to 57).

6.8 PREPARATION OF STANDARD SOLUTIONS

The solvent used to dissolve each metal chelate was pesticide grade chloroform (Fisher Scientific). The following concentrations were prepared:

- 0.4 mg/ml for resolution and efficiency experiments.
- 0.1 to 1 mg/ml for calibration curve preparation.
- 0.04, 0.004, and 0.0004 mg/ml for determination of limits of detection.

6.9 SOLUTES USED FOR COLUMN CHARACTERIZATION

The following compounds were used to characterize the gradient columns in terms of retention indices : a McReynold's kit obtained from PolyScience Corporation (Niles, Illinois, USA), consisting of: benzene, n-butanol, 1,4 dioxane, 1-nitorpropane, pyridine, 2-methyl-2-pentanol, 1-iodobutane, 2-octyne, 2-pentanone, and cis-hydrindane; three hydrocarbon mixtures consisting of

i) Mixture # 1: C_5, C_6, C_7, C_8, C_9

ii) Mixture # 2: C_8, C_9, C_{10}, C_{11}, C_{12}

iii) Mixture # 3: C_{10}, C_{12}, C_{14}, C_{16}

One microliter of each of these reference standards was injected in tests carried out.

7.0 DISCUSSION

7.1 PRELIMINARY EXPERIMENTS WITH MIXED STATIONARY PHASE OV-101 AND QF-1 COLUMNS:

The aim was to find a stationary phase for use in the gradient column mode which would be able to separate more metal chelates in a single run than what has been reported in the literature to date. The following columns were selected for preliminary experimentation:

1 - 2.5% OV 101 + 2.5% QF-1 on CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing)

2 - 5% OV 101 + 5% QF-1 on CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing)

These columns were reported previously (25, 52) to be succsseful in the separation of up to five metal chelates, namely: Zn(II), Cd(II), Cu(II), Ni(II) and Co(III) diethyl -dithiocarbamates.

7.1.1 2.5% OV-101 + 2.5% QF-1 ON CW-HP, 80/100 mesh, (60 cm X 2 mm I.D, glass tubing), (Table 5, Figs 9 & 10)

The results obtained using this column are given in Table 5 for mixtures of : Zn(II), Cd(II), Cu(II), and Ni(II) DDC. These results were compared with previously reported data (25). Both the column efficiency and the resolution were reproduced with some improvement. The chromatograms obtained are shown in Fig 9.

In a mixture of nine DDC, consisting of Zn(II), MoO_2(II), Cd(II), Cu(II), Pb(II), Hg(II), Ni(II), Cr(III), and Co(III) $(DDC)_n$, only five peaks were observed as is shown in Fig 10, which was in agreement with results reported previously (25), at a carrier gas flow of 60 ml/min, and column temperature of 200°C. Better resolution was obtained when were changed the latter parameters to 220°C and 30 ml/min.

7.1.2 5% OV-101 + 5% QF-1 ON CW-HP, 80/100 mesh, (60 cm X 2 mm I.D, glass tubing), (Tables 6 & 7, Figs 11 & 12)

The results obtained with this column are listed

TABLE 5

COMPARISON OF RESULTS USING
2.5% OV-101 + 2.5% QF-1 ON CW-HP OR ON GC-Q,
80/100 MESH, 200C^{o}, AND 60 ML/MIN

	RETENTION-DATA			
METAL DDC	REPORTED BY CARVAJAL USING GAS CHROM Q (25)		EXPERIMENTALLY OBTAINED ON CHROM WHP	
	RT	N	RT	N
Zn	4.0	256	3.4	232
Cd	5.5	484	4.9	392
Cu	6.5	433	6.1	341
Ni	8.5	514	8.7	541

	RESOLUTION	
	REPORTED (25)	OBTAINED
Zn-Cd	1.5	1.6
Zn-Cu	2.2	2.3
Zn-Ni	3.6	4.4
Cd-Cu	0.9	0.9
Cd-Ni	2.4	3.0
Cu-Ni	1.5	1.9

Fig. 9 SEPARATION OF FOUR M(DDC)$_n$ ON 2.5% OV-101 + 2.5% QF-1 ON CW-HP, 100/120 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 200°C, AND 60 ml/min.

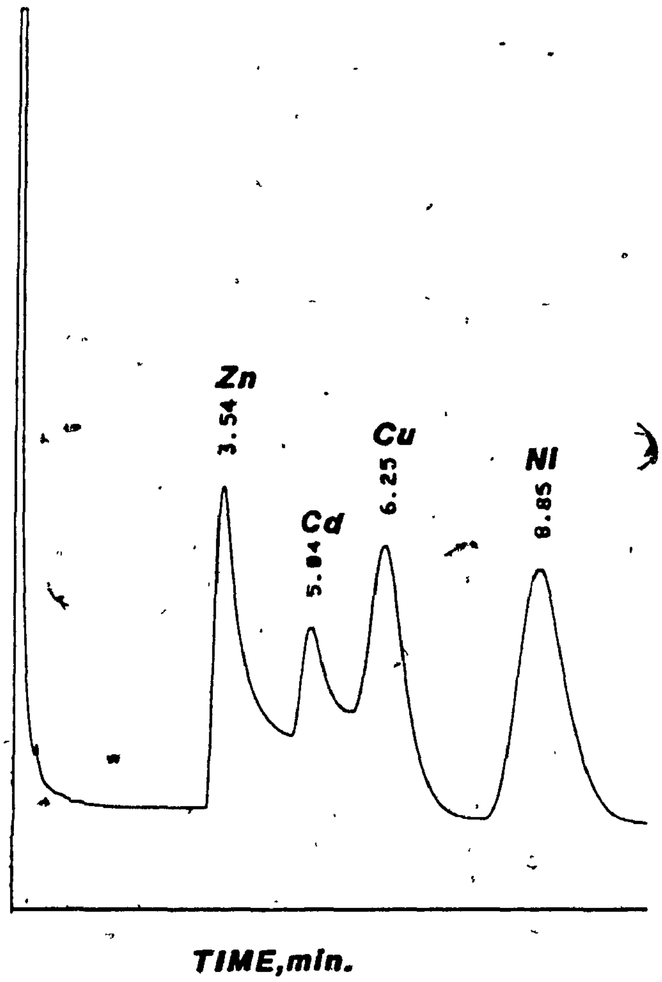
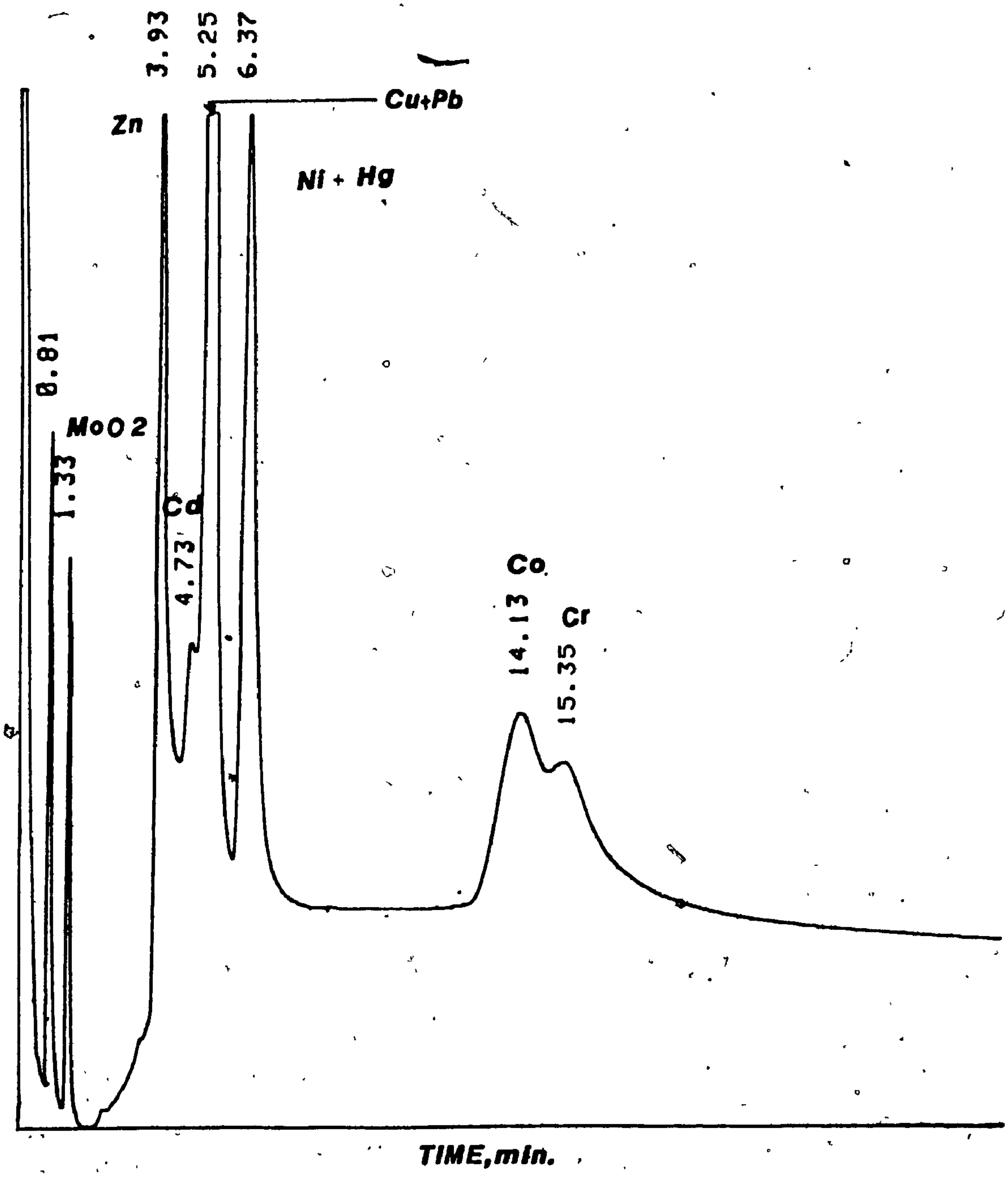


FIG. 10 SEPARATION OF NINE $M(DDC)_n$ ON 2.5% OV-101 + 2.5% QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 200°C FOR 5 MINS, THEN PROGRAMMED TO 240°C AT 10°C/min, NITROGEN FLOW RATE OF 30 ml/min.



in Tables 6 & 7 and Figs 11 & 12. For the separation of the mixture of: Zn(II), Cd(II), Cu(II), and Ni(II) DDC, the column showed higher efficiency and resolution in comparison to what has been reported (25), Table 6, Fig 11. In Fig 11, the chromatograms obtained are shown, under both temperature programming and isothermal conditions The resolution obtained under isothermal conditions was satisfactory and all four metals were separated.

To further test the column's capability, a complex mixture of nine metal $(DDC)_n$, Mo, Zn, Cd, Cu, Pb, Hg, Ni, Cr and Co, was injected. Only five large peaks were obtained at 240°C (Table 7 & Fig 12). The resolution was inferior to that in the previous run .The Cd and Pb peaks overlapped as well as Cu and Hg peaks. The resolution was very poor for Co and Cr. However, considering Zn, Cd, Cu and Ni only, the resolution was still better than that reported in the literature (25, 52), using the same temperature but lower flow rate. This was probably due, mainly, to the more efficient stationary phase coating method employed to prepare columns, and perhaps due to differences in the instrumentation available.

TABLE 6

COMPARISON OF RESULTS USING (5% OV-101 + 5% QF-1) ON CW-HP, 80/100 MESH, (60 cm x 2 mm ID, GLASS TUBING), AT 210°C FOR 10 MINS, THEN PROGRAMMED TO 250°C AT 4°C/min AND AT 30ml/min FLOW RATE

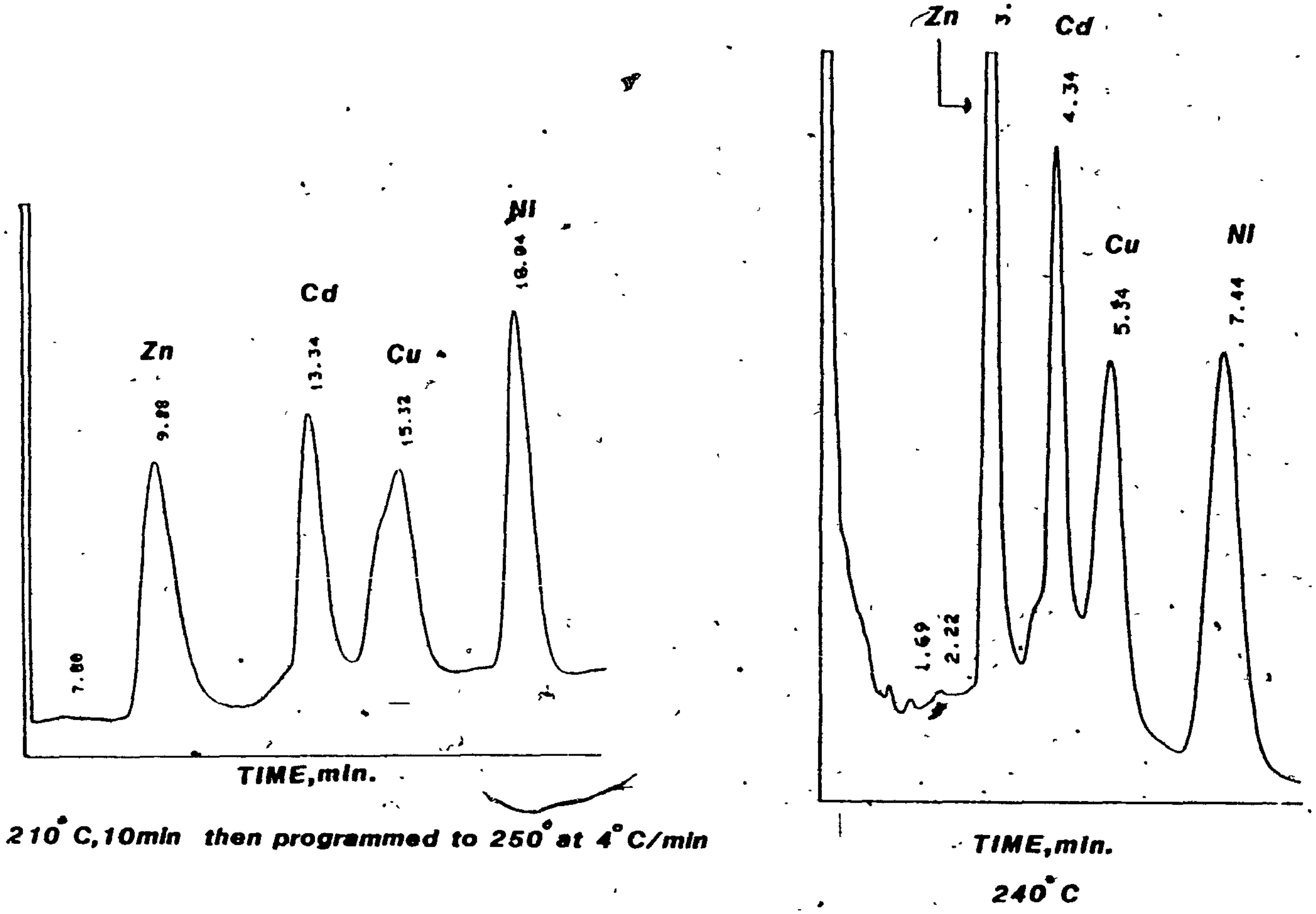
METAL DDC	RETENTION DATA REPORTED(25)*		OBTAINED**	
	RT	N	RT	N
Zn	5.0	237	9.8	650
Cd	7.2	324	13.3	1977
Cu	9.2	339	15.3	1379
Ni	13.3	676	18.0	5207

	RESOLUTION REPORTED(25)	OBTAINED
Zn-Cd	1.5	2.5
Zn-Cu	2.5	3.4
Zn-Ni	4.8	6.4
Cd-Cu	1.1	1.4
Cd-Ni	3.2	4.3
Cu-Ni	1.9	2.1

* Flow rate at 60ml/min and programmed at 10°C/min to 250°C

** Due to the tailing encountered during temperature programming, the flow rate used was 30ml/min and single column compensation was used in order to facilitate area measurements.

FIG. 11 SEPARATION OF FOUR M(DDC)$_n$ ON 5% OV-101 + 5% QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), 30 ml/min, AT 240°C, AND 210°c for 10 mins, THEN PROGRAMMED TO 250°C AT 4°C/min.



210°C,10min then programmed to 250° at 4°C/min

240°C

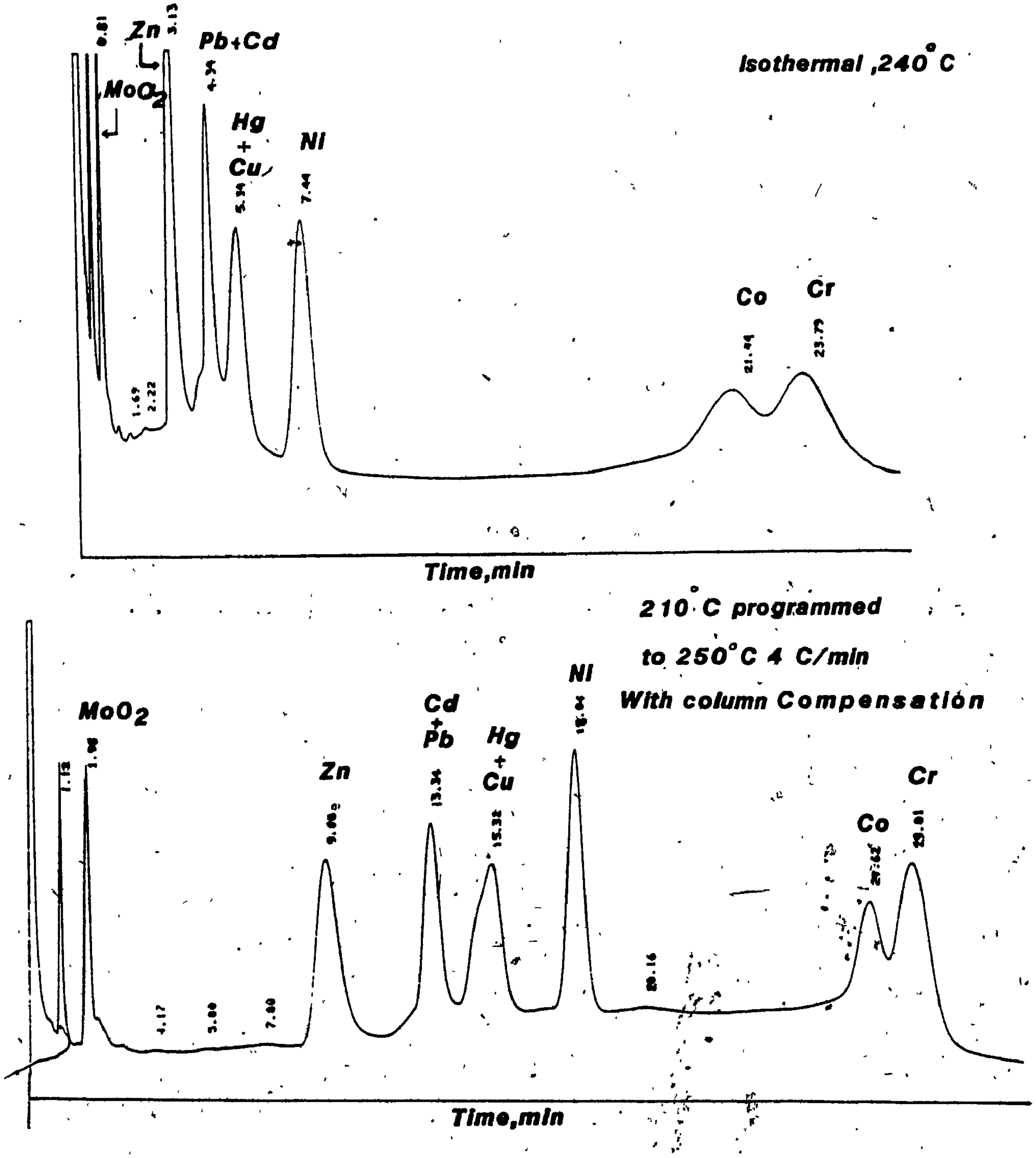
TABLE 7

SEPARATION OF A COMPLEX MIXTURE OF NINE METAL DDC ON 5% OV 101 + 5% QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm ID, GLASS TUBING), AT 240°C, 30 ml/min

METAL DDC	RETENTION DATA RT(MIN)	N
Mo	0.8	66
Zn	3.1	245
Cd	4.3	178
Pb	4.3	178
Cu	5.3	250
Hg	5.3	250
Ni	7.4	394
Co	21.4	438
Cr	23.8	471

	RESOLUTION
Mo-Zn	3.9
Zn-Cd	1.1
Cd-Pb	0.0
Cu-Cd	0.8
Cu-Hg	0.0
Cu-Ni	1.5
Co-Cr	0.3

FIG. 12 SEPARATION OF A NINE $M(DDC)_n$ MIXTURE ON 5% OV-101 + 5% QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), 30 ml/min AT 240°C, AND AT 210°C FOR 10 MINS, THEN TO 250°C AT 4°C/min.



7.1.3 CONCLUSIONS BASED ON THE PRELIMINARY RESULTS:

1 - Both columns offered excellent separation of Zn(II), Cd(II), Cu(II), Ni(II) and Co(III)(DDC), as had already been reported in the literature (25).

2 - Both columns failed to resolve more than five $M(DDC)_n$ per run. Various temperature programming profiles failed to increase the number of chelates which could be separated. In addition, excessive column bleeding was observed during the programming runs.

3 - Since OV-101 and QF-1, at the loadings reported, did not appear to be efficient enough for separation of more than five $M(DDC)_n$ at a time, they would not be adequate for analysis of complex metal mixtures such as those found in marine environments. Therefore, more studies were required to find more efficient stationary phases or more appropriate loadings. It was decided to use the results in Fig 12 as a reference against which to judge separations on other phases.

7.2 OV-101 / QF-1 COMBINED IN VARIOUS PROPORTIONS IN SEQUENCE

(Table 8, Fig 13)

Since a mixed 2.5% OV-101 + 2.5% QF-1 column was successful in achieving very good resolution of five metal chelates, it was decided to prepare combination columns having different proportions of these two stationary phases, in sequence, from 100% OV-101/ 0% QF-1 to 0% OV-101 /100% QF-1, to establish the effect of relative proportions on the resolution that could be achieved. Three metal DDC chelates were chosen as test samples, namely, $Zn(DDC)_2$, $Cd(DDC)_2$ and $Ni(DDC)_2$ for this study because of their excellent thermal stability at 200°C (11, 25, 52).

The results obtained for this series of columns are listed in Table 8, and the regression lines plotted for the resolution factors, obtained in both directions of the carrier flow, and the percent column composition are shown in Fig 13.

The resolution of the Zn-Cd pair decreased with decrease in fraction of OV-101 relative to QF-1. In addition, the retention times for both Zn and Cd varied, but not drastically, with change in relative proportions of the two phases, on the other hand, the retention time of $Ni(DDC)_2$ increased with increasing proportion of QF-1. The resolution

7

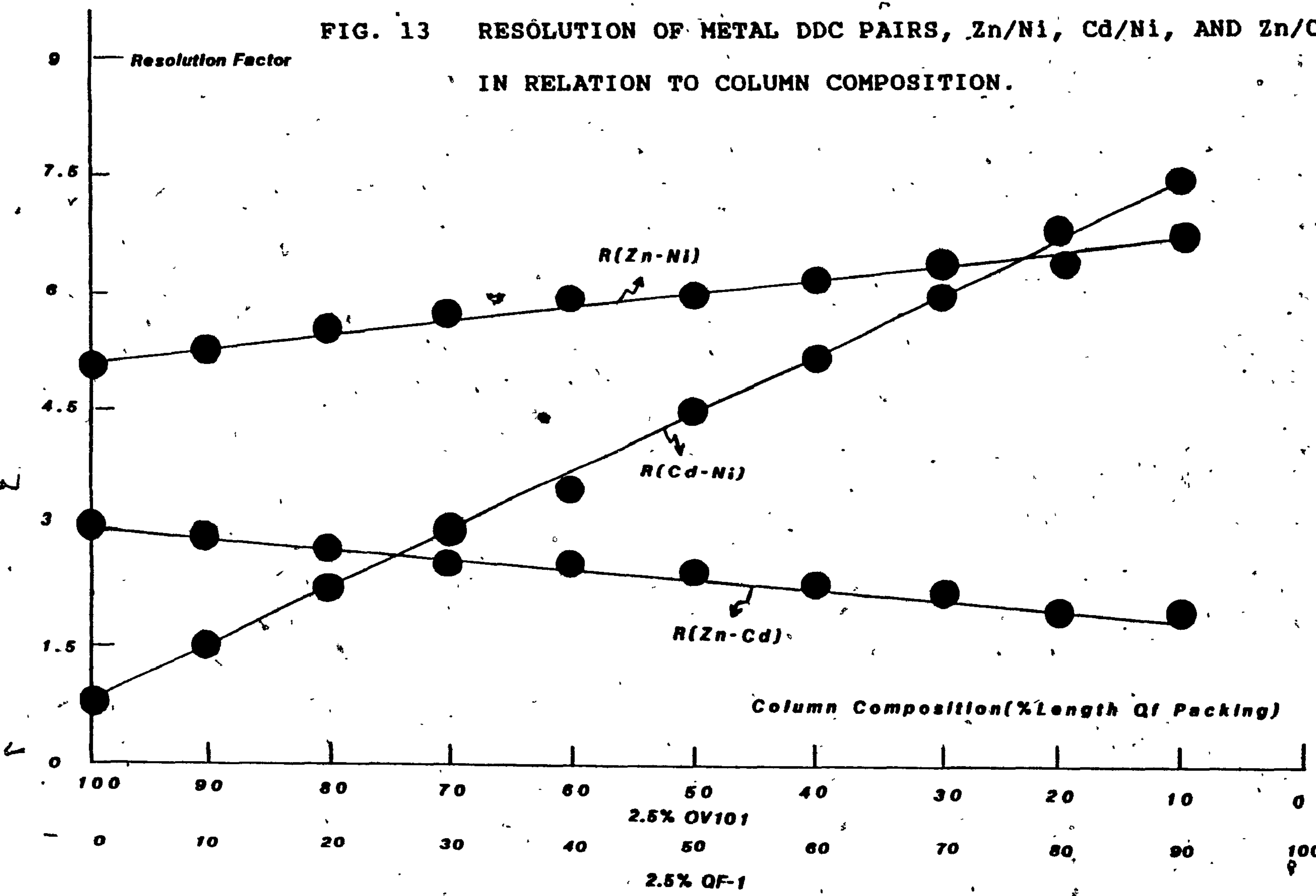
TABLE 8

RESOLUTION AND RETENTION TIMES ON OV-101/QF-1

COMBINATION COLUMNS AT 200°C

COLUMN 2 ft x 1/8 in O.D % LENGTH		DIRECTION OF CARRIER FLOW	RESOLUTION			RETENTION TIME (MIN)		
2.5% OV-101	2.5% QF-1		Zn-Cd	Cd-Ni	Zn-Ni	$Zn(DDC)_2$	$Cd(DDC)_2$	$Ni(DDC)_2$
100	0	→	3.0	1.7	5.1	4.7	7.7	9.9
75	25	→	2.7	2.8	5.3	5.6	8.8	13.4
		←	2.1	3.2	5.5	5.4	8.2	13.8
66.7	33.3	→	2.8	3.1	5.7	5.1	7.9	12.6
		←	2.6	3.3	5.4	5.2	7.9	13.3
50	50	→	2.3	3.7	6.1	5.3	8.2	14.3
		←	2.3	3.9	6.3	5.5	8.5	15.7
33.3	66.7	→	2.2	4.3	6.6	4.7	7.4	14.2
		←	1.9	3.9	5.3	4.8	6.8	14.4
25	75	→	2.2	4.6	6.5	5.8	8.2	17.5
		←	1.9	4.6	6.5	5.1	7.3	16.0
0	100	→	1.8	5.2	6.9	4.9	6.7	16.6

FIG. 13 RESOLUTION OF METAL DDC PAIRS, Zn/Ni, Cd/Ni, AND Zn/Cd, IN RELATION TO COLUMN COMPOSITION.



of Cd and Ni also increased, also, with increasing QF-1 content.

7.2.1 CONCLUSION

From the study of these OV-101/ QF-1 segment columns it was possible to :

1 - conclude that there is a strong affinity of Ni towards QF-1, its retention time increasing with the proportion of QF-1 used.

2 - conclude that there is a logical relationship between resolution (and to the same extent retention time) and proportions of OV-101 and QF-1 used.

3- predict the potential of QF-1 gradient columns to separate metal DDC that elute after Ni, and of OV-101 for the metal DDC which elute before Ni.

7.3 EXPERIMENTS WITH GRADIENT COLUMNS

The approach adopted in the study of gradient columns is outlined below:

(1) - Metal DDC used for preliminary column testing

were selected on the basis of their thermal stability (13, 20, 25); i.e Zn(II), Cd(II), Cu(II) and Ni(II) DDC.

(2) - Whenever a gradient column gave excellent separation of the above four chelates it was evaluated futher by attempted separation of a more complex mixture of metal DDC.

(3) - Temperature programming studies were carried out to obtain the optimum resolution for the maximum number of metal DDC in a mixture on the more promising columns.

(4) - For every gradient column, the following points were considered:

a - Efficiency (number of theoretical plates, N)

b - Retention times, with a view towards obtaining reduced analysis times.

c - Extent of column bleeding at high temperatures.

d - Possible degradation of the metal DDC.

e - Degree of improvement of resolution over that obtained with conventional columns and and that shown in Fig 12 using an OV-101/ QF-1) combination.

7.3.1 GRADIENT COLUMNS WITH OV-101 STATIONARY PHASE

7.3.1.1 1-2-3% OV-101 ON CW-HP, 100/120 mesh, (60 cm X 2 mm I.D, glass tubing), (Table 9, Fig 14 to 17)

The results achieved with this gradient column compared to those on conventional 1%, 2%, and 3% loaded columns at 220°C are shown in Table 9. The resolution on the 1-2-3% gradient column was superior to that on the individual 1%, 2% or 3% columns. The chromatograms on these columns are shown in Fig 14 . None of the four columns, however, could separate the Cd-Cu DDC in the mixture of the four metal DDC injected together.

A mixture of eight metal $(DDC)_n$ was injected into both the 2% OV-101 (the average gradient loading) and the 1-2-3% OV-101 columns using temperature programming from 200°C to 240°C, at 4°C/min, and then constant column temperature 240°C for ten minutes. Peak tailing and column bleeding were observed on the 2% conventional column but not on the 1-2-3% gradient column, Figs 15 & 16, even though the two columns had been prepared and conditioned at the same time in identical fashion. This advantage of gradient columns has also been reported by Berry et al (7).

Some tests were carried out injecting various metal DDC individually to check for possible degradation. $Pb(DDC)_2$

TABLE 9

COMPARATIVE RESULTS OBTAINED ON A 1-2-3% OV-101 GRADIENT COLUMN AND ON 1%, 2% AND 3% CONVETIONAL COLUMNS, AT 220°C.

	RETENTION TIME, min*			
METAL DDC	1%	2%	3%	1-2-3%
Zn	0.9	1.7	1.7	1.6
Cd	1.3	2.6	2.7	2.5
Cu	1.4	2,7	2.8	2.7
Ni	1.7	3.3	3.4	3.2
	RESOLUTION*			
Zn-Cd	1.2	1.2	0.9	2.2
Zn-Ni	1.7	1.9	1.5	3.7
Cd-Ni	0.8	0.7	0.7	0.9
Cd-Cu	0.0	0.0	0.0	0.0

* RSD% <±1%

FIG. 14 COMPARISON OF CHROMATOGGRAMS OBTAINED ON A 1-2-3% OV-101 GRADIENT COLUMN, AND ON 1%, 2%, AND 3% OV-101 CONVENTIONAL COLUMNS, AT 220°C, AND 30ml/min.

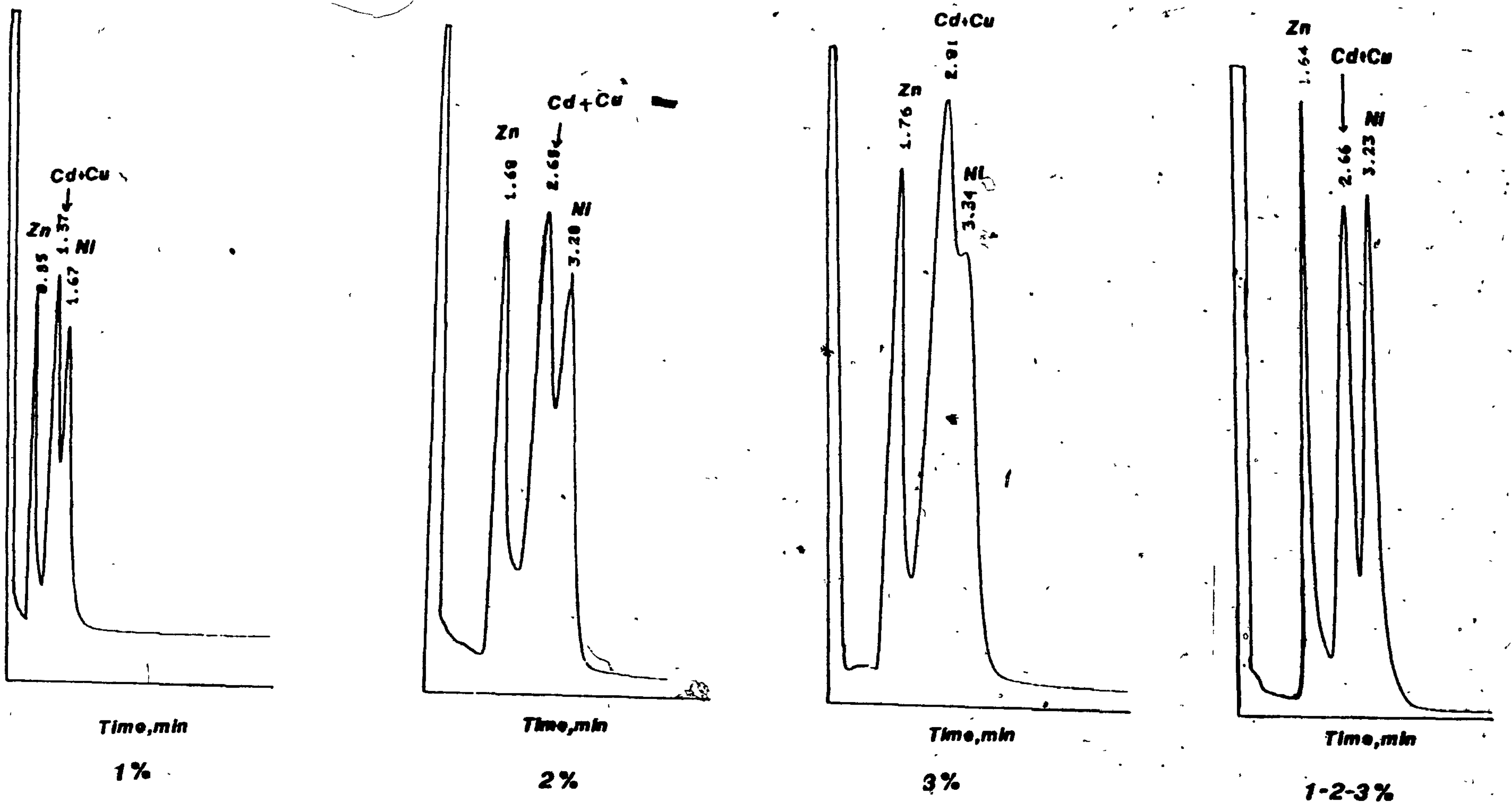


TABLE 39

CHARACTERIZATION OF A 1-2-3% OV-225 GRADIENT COLUMN, AND A 2% OV-225 CONVENTIONAL COLUMN ON CW-HP 80/100 MESH, (180 cm x 2 mm I.D, GLASS TUBING) USING KOVATS RETENTION INDICES

SOLUTE	2%		1-2-3%	
		RSD% ±		RSD% ±
BENZENE	762	1.0	766	0.7
N-BUTANOL	925	0.4	931	0.7
1-4 DIOXANE	886	0.5	911	0.2
PYRIDINE	990	0.2	991	0.2
2-METHYL2-PENTANOL	931	0.4	945	0.3
1-IODOBUTANE	973	0.2	978	0.2
2-PENTANONE	881	0.5	905	0.3
CIS-HYDRINDANE	1032	0.4	1045	0.3

column showed higher chromatographic selectivity when with the 2% column.

8.3.5 1-2-3% OV-210 ON CW-HP, 80/100 mesh, (180 cm x 2 mm I.D, glass tubing) (Table 40)

Shown in Table 40 are the RI data obtained on this column. The RI values for butanol, and 2-methyl-2-pentanol were slightly higher on the gradient column compared to the 2% column. The RI for benzene on this column was similar to that obtained on 1-2-3% QF-1, but lower values were observed than on 1-2-3% QF-1 for the remaining chemical probes, thus indicating that QF-1 has more chromatographic selectivity than does OV-210.

8.3.6 1-2-3% EACH OF QF-1-OV-101 ON CW-HP, 80/100 mesh, (180 cm x 2 mm I.D, glass tubing), (Table 41)

The RI values obtained for this two-phase gradient column are shown in table 41. Generally the RI values on the gradient column were similar but slightly lower than on

TABLE 40

CHARACTERIZATION OF A 1-2-3 % OV-210 GRADIENT COLUMN, AND OF A 2% OV-210 CONVENTIONAL COLUMN ON CW-HP, 80/100 MESH, (180 cm x 2 mm I.D, GLASS TUBING) USING KOVATS RETENTION INDICES

SOLUTE	2%		1-2-3%	
		RSD% ±		RSD% ±
BENZENE	687	1.0	673	1.0
N-BUTANOL	805	0.5	813	0.8
1,4 DIOXANE	835	0.7	831	0.4
PYRIDINE	909	0.3	895	0.3
2-METHYL-2PENTANOL	851	0.6	854	0.6
1-IODOBUTANE	904	0.2	874	0.3
2-PENTANONE	892	0.3	885	0.3
CIS-HYDRINDANE	961	0.2	956	0.3

TABLE 41

CHARACTERIZATION OF A 1-2-3% EACH OF OV-101-QF-1 GRADIENT COLUMN, AND OF A 2% EACH OF OV-101-QF-1 CONVENTIOANL COLUMN ON CW-HP, 80/100 MESH, (180 cm x 2 mm I.D, GLASS TUBING) USING KOVATS RETENTION INDICES

SOLUTE	2%		1-2-3%	
		RSD% ±		RSD% ±
BENZENE	655	0.7	653	0.6
N-BUTANOL	692	0.6	681	0.3
1,4 DIOXANE	731	0.5	732	0.4
PYRIDINE	789	0.4	787	0.7
2-METHYL 2-PENTANOL	760	0.2	758	0.5
1-IODOBUTANE	829	0.2	829	0.2
2-PENTANONE	741	0.5	739	0.4
CIS-HYDRINDANE	964	0.1	966	0.04

the 2% column indicating higher selectivity on the 2% column.

8.3.7 (1-2-3%) QF-1 + 1% OV-225 ON CW-HP, 80/100 mesh, (180 cm x 2 mm, I.D, glass tubing), (Table 42)

This column combined the stationary phases of the two most selective columns. Shown in Table 42 are the RI data obtained on this column compared to the 2% column. The RI values were generally lower, by approximately 10 RI units on the gradient column. The chromatographic selectivity of this column for benzene and pyridine was between that on 1-2-3% QF-1 and 1-2-3% OV-225 .

8.4 General conclusions on characterization of gradient columns by Retention Indices

1. It has been demonstrated that the retention index system can differentiate between polarity gradient and conventional columns and can serve to classify gradient loaded columns.
2. The variable percent loaded gradient columns differed in behavior, to modest extents when compared with

TABLE 42

CHARACTERIZATION OF A (1-2-3%) QF-1 + 1% OV225 GRADIENT COLUMN, AND OF A 2% QF-1 + 1% OV-225 CONVENTIONAL COLUMN, ON CW-HP, 80/100 MESH, (180 cm x 2 mm I.D, GLASS TUBING) USING KOVATS RETENTION INDICES

SOLUTE	2% QF-1 + 1% OV225		(1-2-3%) QF-1+ 1% OV225	
		RSD% ±		RSD% ±
BENZENE	722	0.7	712	0.8
N-BUTANOL	856	0.6	848	0.8
1,4 DIOXANE	876	0.6	863	0.7
PYRIDINE	952	0.3	945	0.4
2-METHYL-2PENTANOL	891	0.3	882	0.7
1-IODOBUTANE	932	0.2	930	0.4
2-PENTANONE	892	0.1	888	0.7
CIS-HYDRINDANE	1028	0.2	1018	0.1

conventional columns, sometimes showing more selectivity.

3. The two best columns found for the separation of a complex M(DDC)$_n$ mixture were coated with QF-1 stationary phase. The obtained RI values for the probes representing molecular orientation properties, such as butanol, dioxane, iodobutane and pentanone, were higher for the gradient column than the conventional. This was in agreement with the fact that the separation of the M(DDC)$_n$ complex mixture on the 3-2-1% QF-1 column was more successful than on the 2% QF-1 column. This finding, which is mainly due to differences in electronegativity of each metal, is also in agreement with the reported different dipole moments of M(DDC)$_n$ (65).

9.0 Detection limits

Tests were carried out with the following columns to establish the lower detection limits for metal $(DDC)_n$, using the Flame Ionization detector.

1. 1.0-1.5-2.0-2.5-3.0% QF-1 on CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing).
2. 3-2-1% QF-1 on GC-Q, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing).

The results obtained are summarized in Table 43.

The detection limits depended on the percent by weight metal content of each chelate. This is because the FID responds only to the organic part of the metal chelate and not to the metal itself. For example, as shown in Table 44, the best detection limit was for $Ni(DDC)_2$, and the poorest for $Pb(DDC)_2$. The determination of trace metals in marine sediments and sea water is usually carried out using either atomic absorption (AA) (66), or emission spectroscopy such as the versatile inductively coupled plasma emission (ICP), with good detection limits (67). For example, the reported AA (67) detection limits for Ni, Zn, and Pb were 0.1, 0.01 and 0.05 ppm respectively and for ICP were 2,1 and 2 ppb respectively. In comparison the detection limits obtained (Table 43), in this project are within the limits of these spectroscopic methods.

TABLE 43

LOWER LIMITS OF DETECTION# FOR $M(DDC)_n$ USING FID

METAL DDC	COLUMN TEMPERATURE C°	DDC (ng)	METAL(ng)
$Zn(DDC)_2$*	190	4.0	0.72
$Cd(DDC)_2$*	190	1.6	0.44
$Cu(DDC)_2$*	210	1.6	0.28
$Ni(DDC)_2$**	210	0.4	60 pg
$Pb(DDC)_2$*	210	16.0	6.60
$Co(DDC)_3$*	230	4.0	0.47
$Cr(DDC)_3$*	230	4.0	0.42
$Hg(DDC)_2$*	210	10.0	4.04

* 3-2-1% QF-1 on GC-Q, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing).

** 1.0-1.5-2.0-2.5-3.0% QF-1 on CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing).

\# FID DETECTION LIMITS FOR A SIGNAL/NOISE RATIO = 3.

TABLE 44

PERCENT METAL IN METAL DIETHYLDITHIOCARBAMATES

COMPOUND	MOLECULAR WEIGHT	% METAL
$Zn(DDC)_2$	361.91	18.06
$Cu(DDC)_2$	360.08	17.65
$Hg(DDC)_2$	497.13	40.35
$Co(DDC)_3$	503.62	11.70
$Ni(DDC)_2$	355.25	16.53
$Pb(DDC)_2$	503.73	41.13
$Cd(DDC)_2$	408.94	27.49
$Cr(DDC)_3$	496.00	10.48

10.0 CONCLUSIONS

It was found that gradient loaded columns were superior to conventional columns with respect to the following points:

1. **EFFICIENCY (NUMBER OF THEORETICAL PLATES):**

   The gradient columns were found to be more efficient than conventional columns in the case of all the columns studied.

2. **REDUCED ANALYSIS TIME:**

   On the gradient columns, the $M(DDC)_n$ eluted faster than on the conventional columns tested for comparison. Therefore, the total analysis time was reduced using gradient columns.

3. **LESS COLUMN BLEEDING:**

   It was observed that with most of the gradient columns studied, stationary phase bleeding was less than on conventional columns when programmed temperature analyses were carried out.

4. **RESOLUTION:**

   The resolution obtained on most of the gradient columns studied was superior to that on conventional columns having stationary phase loading in the concentration range included in the gradient columns. Fine tunning of gradient columns provided separations which could had not been achieved previously. For example, the separation of the Cd/Pb DDC pair was successful on the 1.0-1.5-2.0-2.5-3.0% QF-1 gradient column.

5. **HIGHER COLUMN CAPACITY:**

It was shown that by reversing the gradient loading ~~so that~~ the largest loading is at the beginning of the column, i.e 6-4-2%, rather than 2-4-6%, sample capacity, was increased.

In a mixture of nine metal $(DDC)_n$, eight were separated. $Pb(DDC)_2$ was still a problem and resolution of the Cr/Co pair was only 50%.

The best separations obtained for the mixture of nine metal$(DDC)_n$, were on the 3-2-1% QF-1, and on the 1.0-1.5-2.0-2.5-3.0% QF-1 gradient columns. Separation of the Pb/Cu and Hg/Ni pairs were successful on the 1-2-3% QF-1 + 1% OV-225, and on the 1.0-1.5-2.0-2.5-3.0% QF-1 gradient columns, when they were present in a mixture of seven metal $(DDC)_n$. The separation of these two pairs was not achieved on the 5% OV-101 + 5%QF-1 column developed by Carvajal (25).

OV-225 gradient columns were found to be unsuitable for the separation of metals eluting after the $Ni(DDC)_2$, such as $Co(DDC)_3$ and $Cr(DDC)_3$, due to their very long retention times.

For some metals such, as Hg and Pb, degradation was observed on OV-101, QF-1, and OV-210 at temperatures above 200°C. After several injections, however, a single peak was obtainable for each.

Characterization of the gradient columns using the Retention Index System was shown feasible.

The reproducibility of retention times and resolution was generally good, with relative standard deviations usually below 5% for gradient columns.

The FID detection limits were in the order of 0.40 to 16 ng for the $M(DDC)_n$ on QF-1 gradient columns. These detection limits are as good as what has been reported for ICP and AA. The best gradient column developed in this project would accordingly be appicable to marine bottom sediments.

11.0 Suggestions for further research

Although synthetic chelates were used in this project it should be feasible to extend the use of gradient columns to real soil and marine water analysis. With the proper gradient columns it should be possible to separate metal chelates from sample matrix interferences.

Another suggestion for further research would be a study of quantitative recovery of metals from a complex nine metal mixture, and to determine accuracy and precision using FID.

Exponential gradient loaded columns can be evaluated for trace metal analysis a as means of further fine tunning of columns.

The study of the effect of conditioning temperature and prolonged operation at elevated temperatures on the migration of stationary phases used in these gradient columns could be an interesting project.

Finally, it would be interesting to study the apparently variable retention behavior of Pb in complex mixtures of metal chelates, possibly by using Mass Spectrometry for the identification of eluted peaks.

REFERENCES

1. A.T. James and A.J.P. Martin, Biochem. J., 50 (1952) 679.

2. Porath, J. Arkiv Kemi 7, (1954) 535; C.A. 49, 7437C (1955).

3. Purnell, H. "Gas Chromatography", Wiley, New York, (1963), p. 388.

4. D.C. Locke and C.E. Meloan. Anal. Chem., 36, (1964) 2234.

5. T. Bunting and C.E. Meloan. Anal. Chem., 42 (1970) 586.

6. G.P. Hildebrand and, Reilly C.N., Anal. Chem., 36 (1964) 47.

7. E.F. Barry, K.P. Li, and C. Merritt, J. Chromtogr Sci, 20 (1982) 357.

8. Hawkes S.J. and J.C. Giddings, Anal. chem., 36 (1964) 2229.

9. Duty R.C., J. Gas Chromtogr., 6 (1968) 193.

10. G. D'Ascenzo and W.W. Wendlandt, J. Thermal Anal. 1 (1969) 423.

11. G. D'Ascenzo and W.W. Wendlandt, J. Inorg. Nucl. Chem., 32 (1970) 2431.

12. T.J. Cardwell, D.J. Desarra, and P. Uden, Anal. Chim. Acta., 85 (1976) 415.

13. M. Ahmad and A. Aziz, J. Chromatogr., 152 (1978) 542.

14. M. Delepine, Bull. Soc. Chim . Fr., 3 (1908) 652.

15. E.E. Reid, Organic Chemistry of Bivalent Sulfur, Vol. 4, Chemical Publishing Co., N.Y. (1962) p. 110.

16. G.D. Thorn and C.A. Ludwig, The Dithiocarbamates and Related Compounds, Elsevier, N.Y. (1962) p. 45.

17. K. W. Bruland, R. P. Franks, G. A. Knauer and J. H. Martin, Anal. Chim. Acta, 105 (1979) 233.

18. J. D. Kinrade and J. C. Van Loon, Anal. Chem., 46 (1974) 1894.

19. T. Takada, Y. Okabe and K. Nakano Bull. Chem. Soc. Jpn, 54 (1981) 3589.

20. A. Radecki, and J. Halkiewicz, J. Chromatgr., 187 (1980) 363.

21. L. Liangbin, L. Jinchum, C. Jieke, Fenxi Huaxue, 12 (1984) 439, C.A. 101(8): 65161b (1984).

22. H. Jiming, L. Liangbin, L. Jinchum, C. Jieke and X. Youlan, Wuhan Daxue Xuebao, Ziran Kexveban, 4 (1982) 47, C.A. 99 (6): 47042Z (1983).

23. M.L. Riekkola, O. Makitie, M. Sundberg, Kem-Kemi, 6 (10) (1979) 525; C.A. 92(6); 51296h (1979).

24. R. Neeb, Pure & Appl. Chem., 54 (1982) 847.

25. N.J. Carvajal and R.H. Zienius, J. Chromatogr., 355 (1986) 107.

26. O.E. Schupp III, "Techniques of Organic Chemistry", Vol XIII, E.S. Perry and A. Weissberger, Edits., Interscience, New York, (1968), p. 173.

27. W.A. Supina, " The Packed Column in Gas Chromatography", Supelco Inc., Bellefonte, Pensylvania (1974), p. 22.

28. Dane S.B., J. Chromatogr. Sci., 7 (1969) 389.

29. S.J. Hawkes and E.F. Mooney, Anal. Chem., 36 (1964) 1473.

30. T.G. Southern, A. Lacchelli, D. Cuthiell, and M.L. Selucky, Anal. Chem. 57 (1985) 303.

31. J.A. Yancey, J. Chromatogr. Sci., 23 (1985) 161.

32. J.K. Haken, J. Chromatogr., 141 (1977) 247.

33. Ibid, 300 (1984) 1.

34. A.E. Coleman, J. Chromatogr. Sci., 11 (1973) 198.

35. R. L. Grob, " Modern Practice of Gas Chromatography", John Wiley, New York (1977) 611-615.

36. J.C. Giddings, J. Gas Chromatogr., 2 (1964) 167.

37. Van Deemter, J.J., Zuyderweg, F.J. and K. Linkenberg, A. Chem. Eng. Sci, 5 (1956) 271.

38. J, Tranchant, Edit. "Practical Manual of Gas Chromatography", Elsevier Publishing Company, New York, 1969 p. 24.

39. H.M. MacNair, E.J. Bonelli, "Basic Gas Chromatography", Varian Aerograph, Berkeley, California (1968) p. 64.

40. H. W. Moody, J. Chem. Educ., 59 (1982) 291

41. L.S. Ettre, Chromatographia, 8 (1975) 291.

42. L.S. Ettre, Chromatographia, 6(1973) 490.

43. E. Kovats, Helv. Chim. Acta, 41 (1958) 1915.

44. W. Averill in "Gas Chromatography", N. Brenner, J.E. and M.D Weiss ed., Academic Press, New York, (1964), p. 1-6.

45. L.S. Ettre, J. Gas Chromatogr., 1 (1963) 36.

46. M. V. Budahegyi, and J.M. Takacs, J. Chroamtogr., 289 (1983) 213.

47. W.O. McReynolds, J. Chromatogr. Sci., 8 (1970) 685.

48. L. Rohrschneider, J. Chromatogr. Sci., 11 (1973) 160.

49. W. R. Supina and L. R. Rose, J, Chromatogr. Sci, 8 (1970) 214.

50. L.S. Ettre, Chromatographia, 7 (1974) 261.

51. J.K. Haken, J. Chromatogr. Sci., 12 (1974) 113.

52. A. Radecki and J. Halkiewicz, J. Grzybowski, and H. Lamparczyk, J. Chromtogr., 151, (1978) 259.

53. J.R. Conder and C.L. Young, "Physicochemical Measurement by Gas Chromatography", John Wiley & Sons, New york, (1979) p. 32.

54. Hewlett-Packard Application Note 228-14, "Precision of the HP 5880A Chromatographic System Achieved for a Complex Sample".

55. D.J. Pasto and C.R. Johnson, "Organic Structure Determination", Prentice-Hall Inc, N.J., (1969) p. 62.

56. Robert Silverstein, G. Bassler, T.C. Morrill, "Spectrometric Identification of Organic compounds", John Wiley & Sons, N.Y., Third Edition, 1974 p. 82.

57. G.S. Zhdanov, Z.V. Zvonkova and N.V. Rannen, Kristallografiua, 1 (1956) 514; C.A., 51 (1957) 4793e.

58. J.A. Yancey, J. Chromatogr. Sci., 24 (1968) 117.

59. N. Kabayashi and T. Fujisawa, Bull. Chem. Soc. of Japan, 49 (1976) 2780.

60. A. Tavalaris and R. Neeb, Fresensius Z. Anal. Chem., 292 (1978) 199.

61. J. Masaryk, J. Krupcik, J. Garaj and M. Kosik J. Chromatogr., 115 (1975) 256.

62. F.W. Moore and M.L. Larson, Inorg. Chem., 6 (1967) 998,

63. L. Malatesta, Gazz. Chim. Ital. 69 (1939) 762.

64. H. Malissa and H. Kolbe-Rode, Talanta, 8 (1961) 841; C.A. 56 (1962) 9538d.

65. Caucauals D, Inorg. Chem. vol 11, (1970) 234

66. Robinson J.W., Atomic Absorption Spectroscopy, 2nd ed., Marcel Dekker, N.Y. 1975, p. 56.

67. Robinson J.W., "Undergraduate Instrumental Analysis ", Marcel Dekker, N.Y. 1987, p. 246.

APPENDIX I

TABLE 45

MELTING POINTS OF M(DDC)$_n$

M(DDC)$_n$	MELTING POINT, C^o		
	OBTAINED	REPORTED	REFERENCE
$MoO_2(DDC)_2$	145 - 146	133 - 133.5	62
$Zn(DDC)_2$	178 - 179	225, 178-179	60, 61
$Cd(DDC)_2$	250 - 251	254, 252-253	60, 61
$Pb(DDC)_2$	210 - 211	208, 209-210	60, 59
$Cu(DDC)_2$	201 - 202	196, 201-202	60, 61
$Hg(DDC)_2$	128 - 130	127 - 130	
		139 - 141	60, 57
$Ni(DDC)_2$	234 - 236	235 - 236	60, 61
$Cr(DDC)_3$	252 - 253	250	63, 64
$Co(DDC)_3$	260 - 263	263 - 264	61, 57

FIG. 53 IR SPECTRA OF SODIUM DIETHYLDITHIOCARBAMATE AND $MoO_2(DDC)_2$ IN DRY KBR.

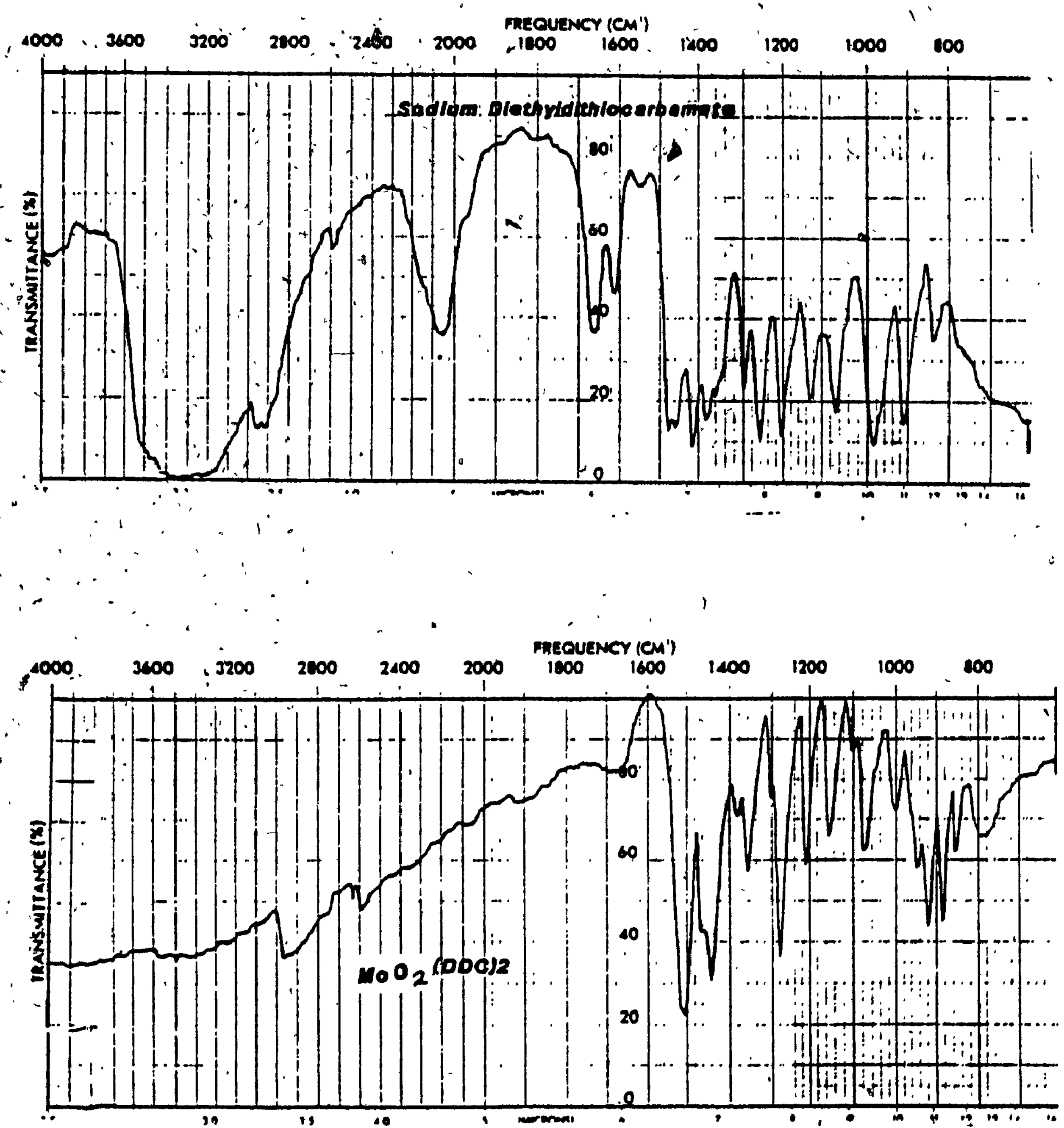


FIG. 54 IR SPECTRA OF $Co(DDC)_3$ AND $Cr(DDC)_3$ IN DRY KBr.

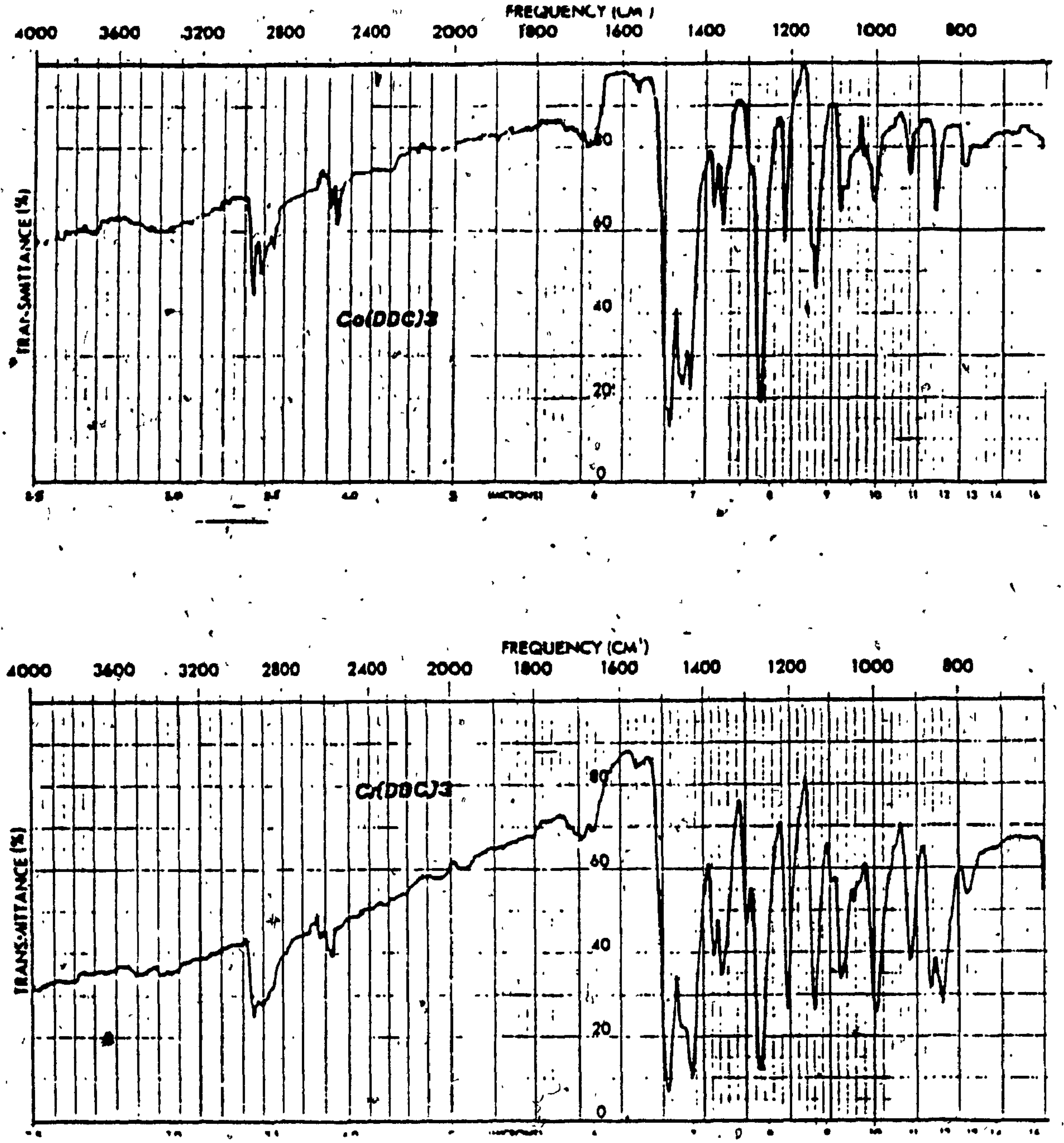


FIG. 55 IR SPECTRA OF $Hg(DDC)_2$ AND $Pb(DDC)_2$ IN DRY KBr.

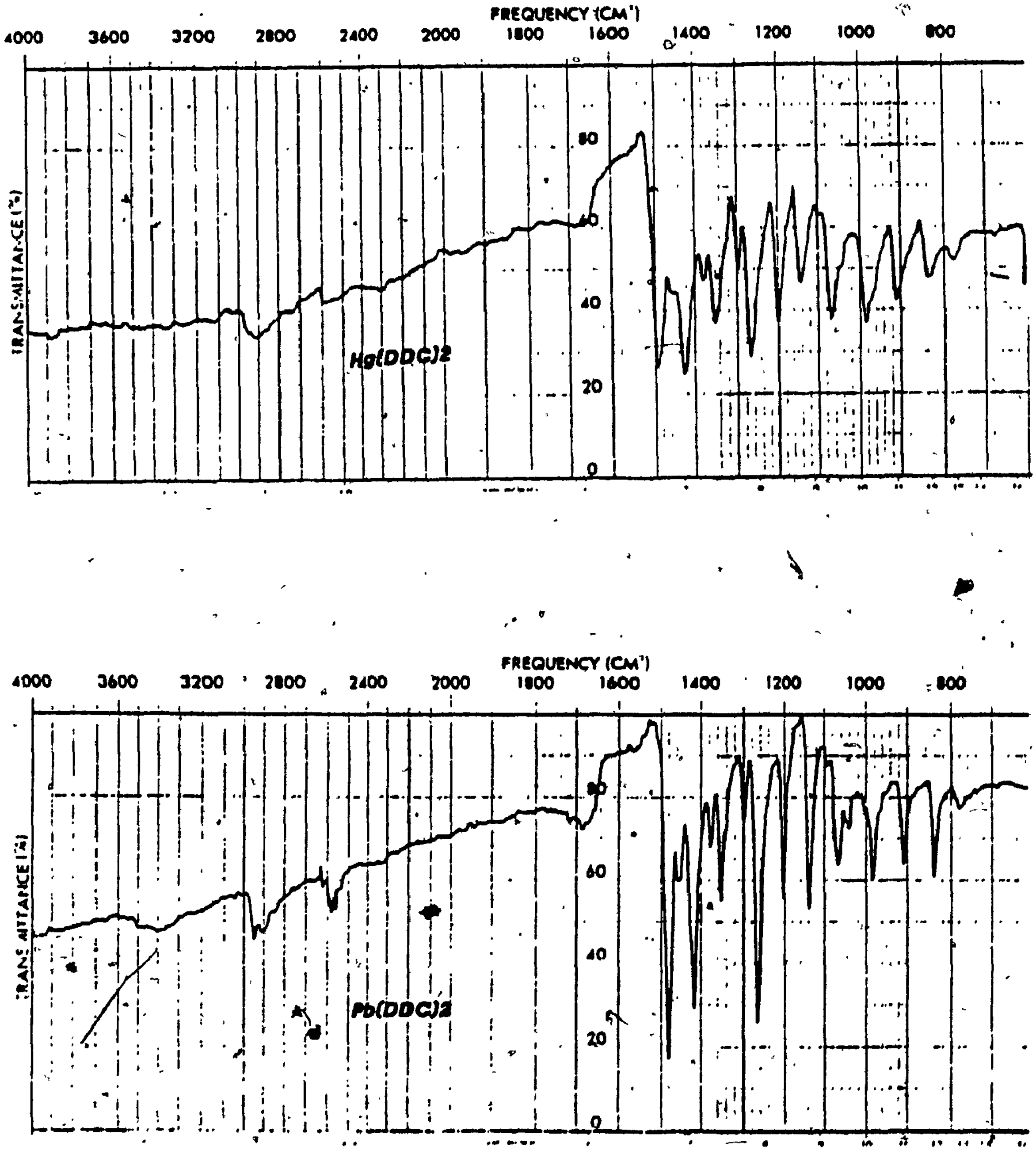
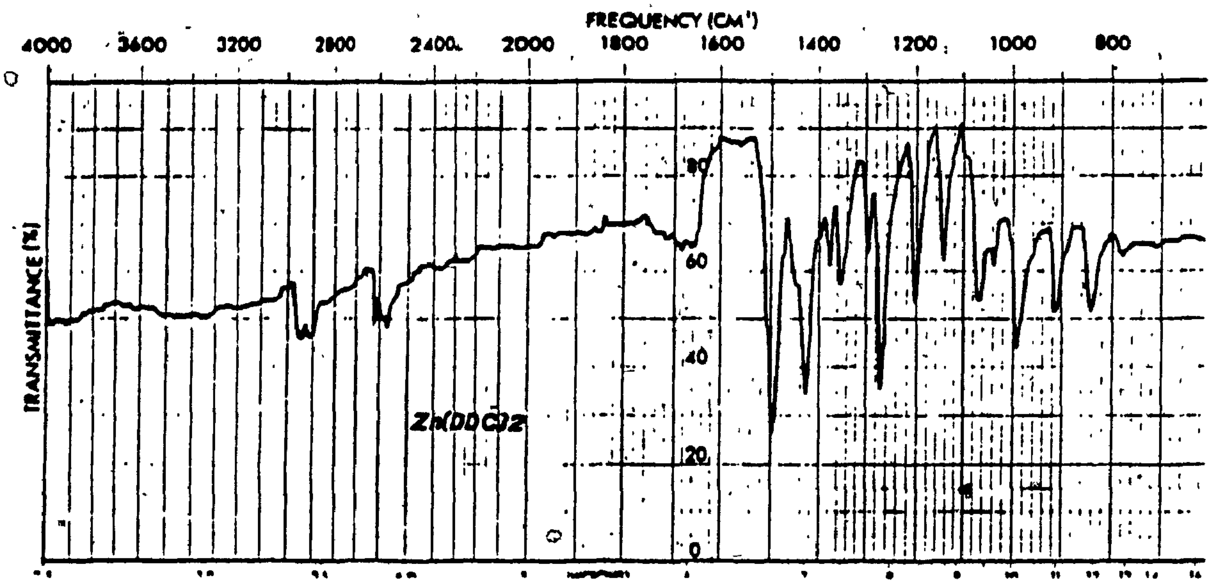


FIG. 56 IR SPECTRA OF $Zn(DDC)_2$ AND $Cd(DDC)_2$ IN DRY KBr.



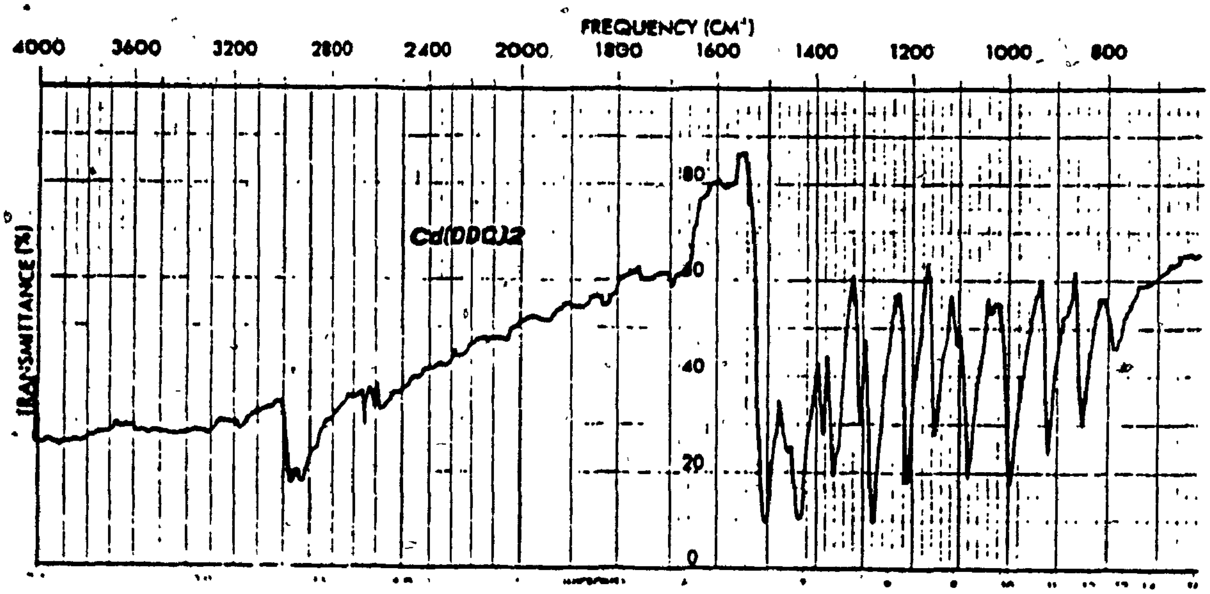


FIG. 57 IR SPECTRA OF $Cu(DDC)_2$ AND $Ni(DDC)_2$ IN DRY KBr.

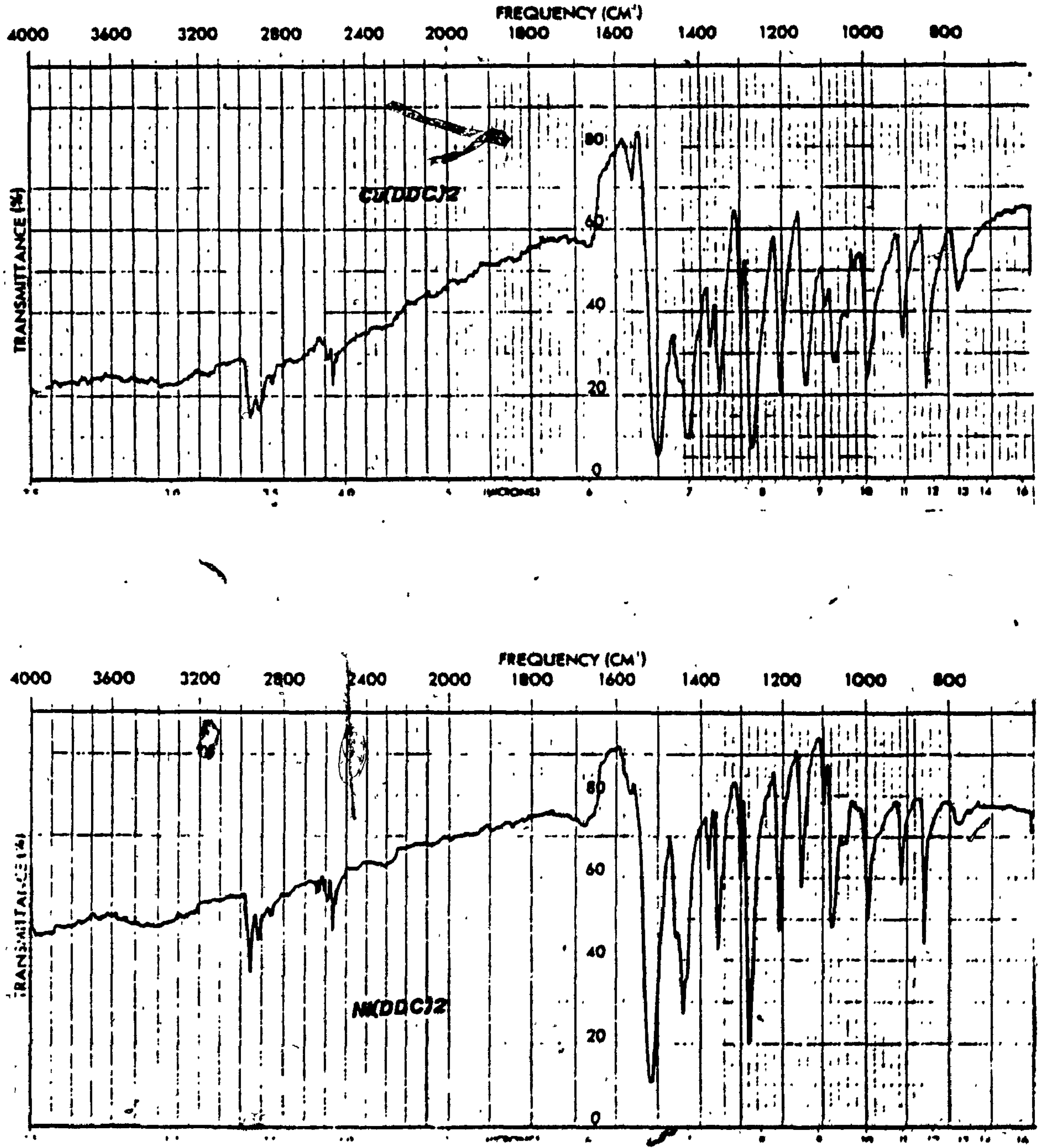


TABLE 46

CALIBRATION CURVES OBTAINED FOR $M(DDC)_n$ ON (1-2-3%) QF-1 ON CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing), at 200C°

CONCENTRATION: 0.1-1.0 ug/ul, N_2 FLOW RATE: 25 ml/min

METAL DDC	RETENTION TIME (min)	SLOPE (cm/ug)	REGRESSION ANALYSIS CORRELATION COEFFICIENT	REGRESSION ANALYSIS Y INTERCEPT (cm)
$Zn(DDC)_2$	4.4	33.6	1.00	- 2.40
$Pb(DDC)_2$	6.0	12.2	0.99	0.22
$Cd(DDC)_2$	7.0	11.0	0.99	- 1.01
$Ni(DDC)_2$	14.8	11.4	0.99	0.05
$Co(DDC)_3$	40.4	2.7	1.00	0.02

FIG. 58 CALIBRATION CURVES FOR $Zn(DDC)_2$ AND $Pb(DDC)_2$ ON 1-2-3% QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 200°C, AND 25 ml/min.

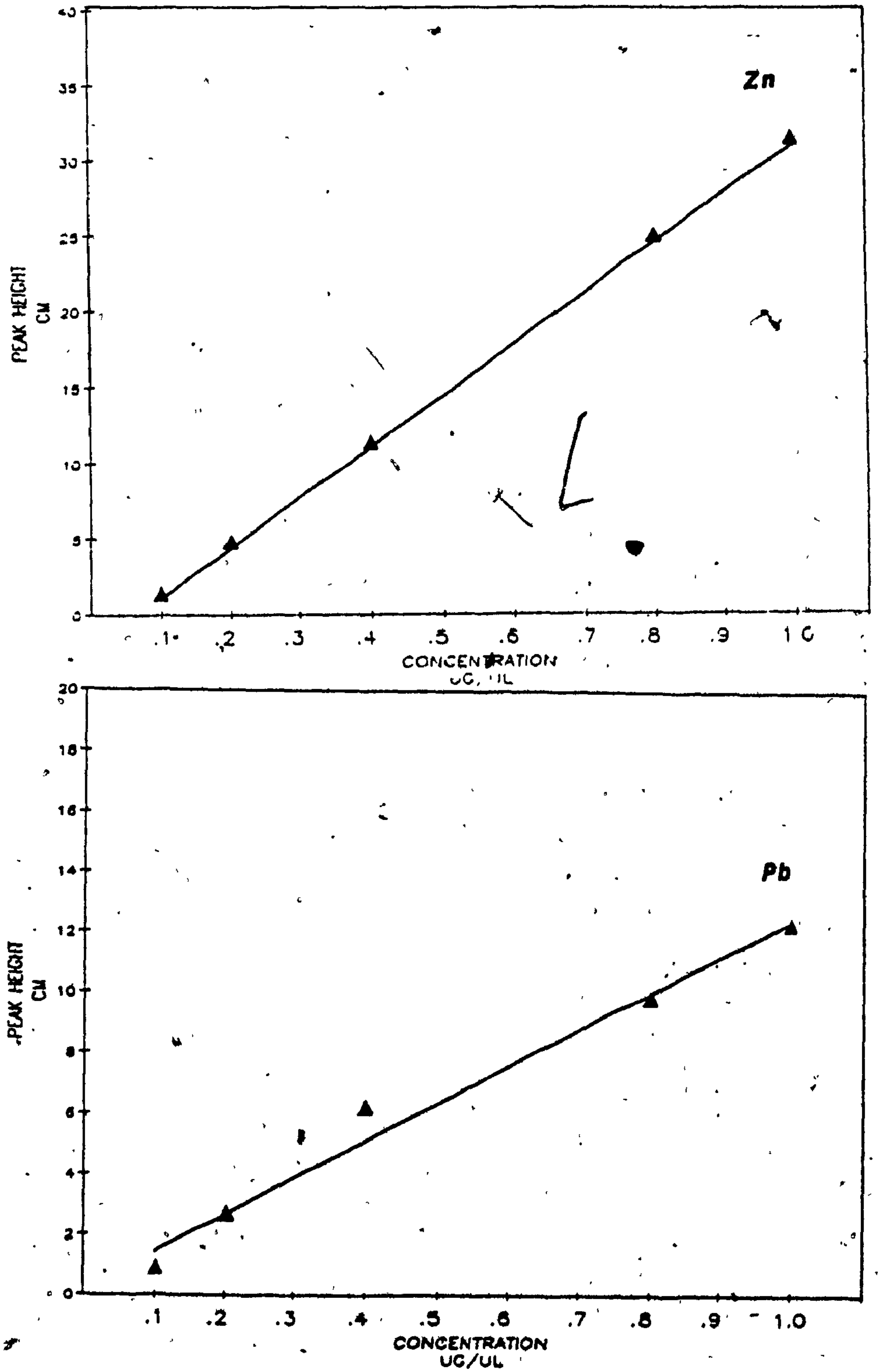


FIG. 59 CALIBRATION CURVES FOR Cd(DDC)$_2$ AND Ni(DDC)$_2$ ON 1-2-3% QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 200°C, AND 25 ml/min.

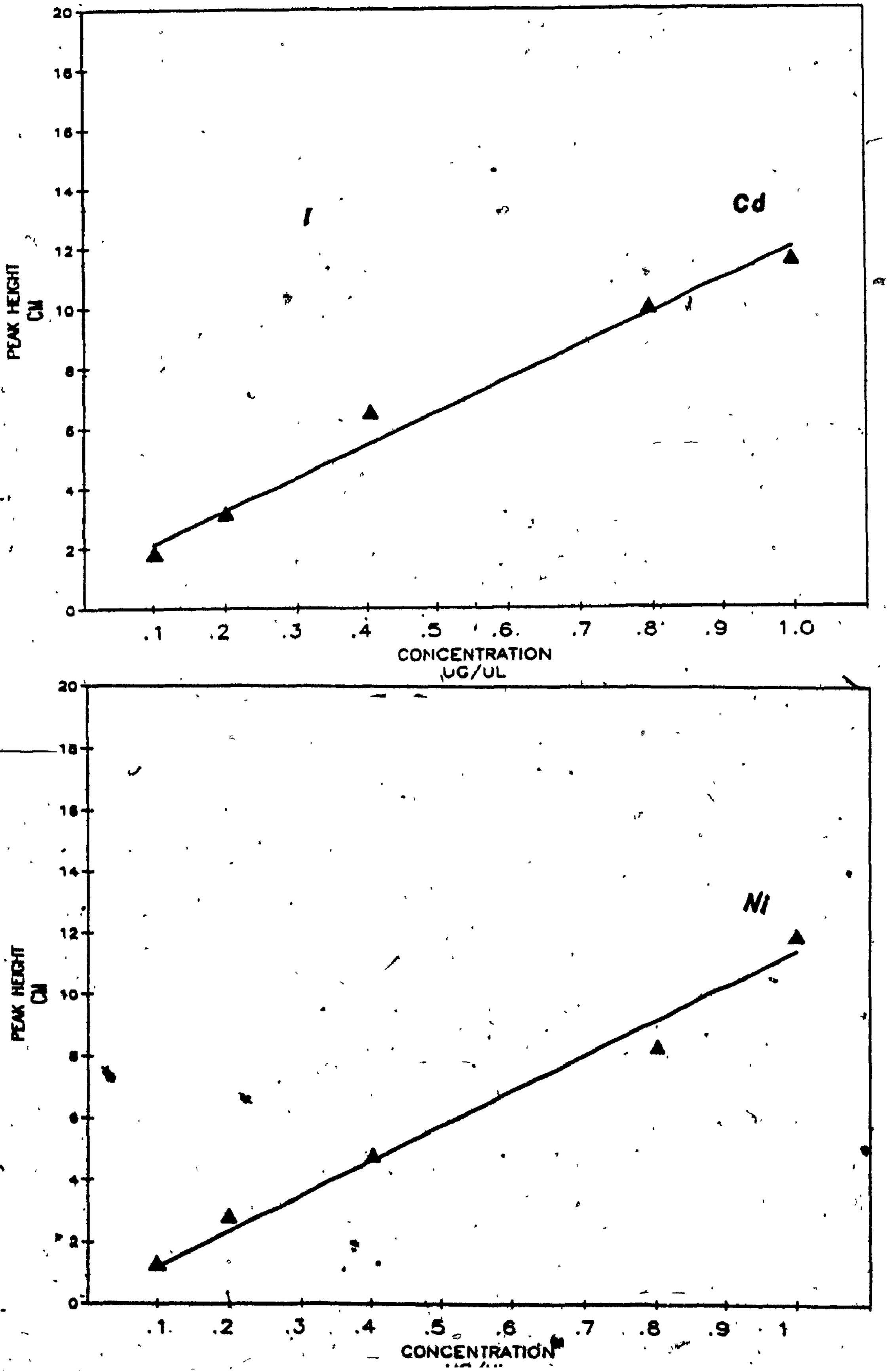


FIG. 60 CALIBRATION CURVE FOR $Co(DDC)_3$ ON 1-2-3% QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 200°C, AND 25 ml/min.

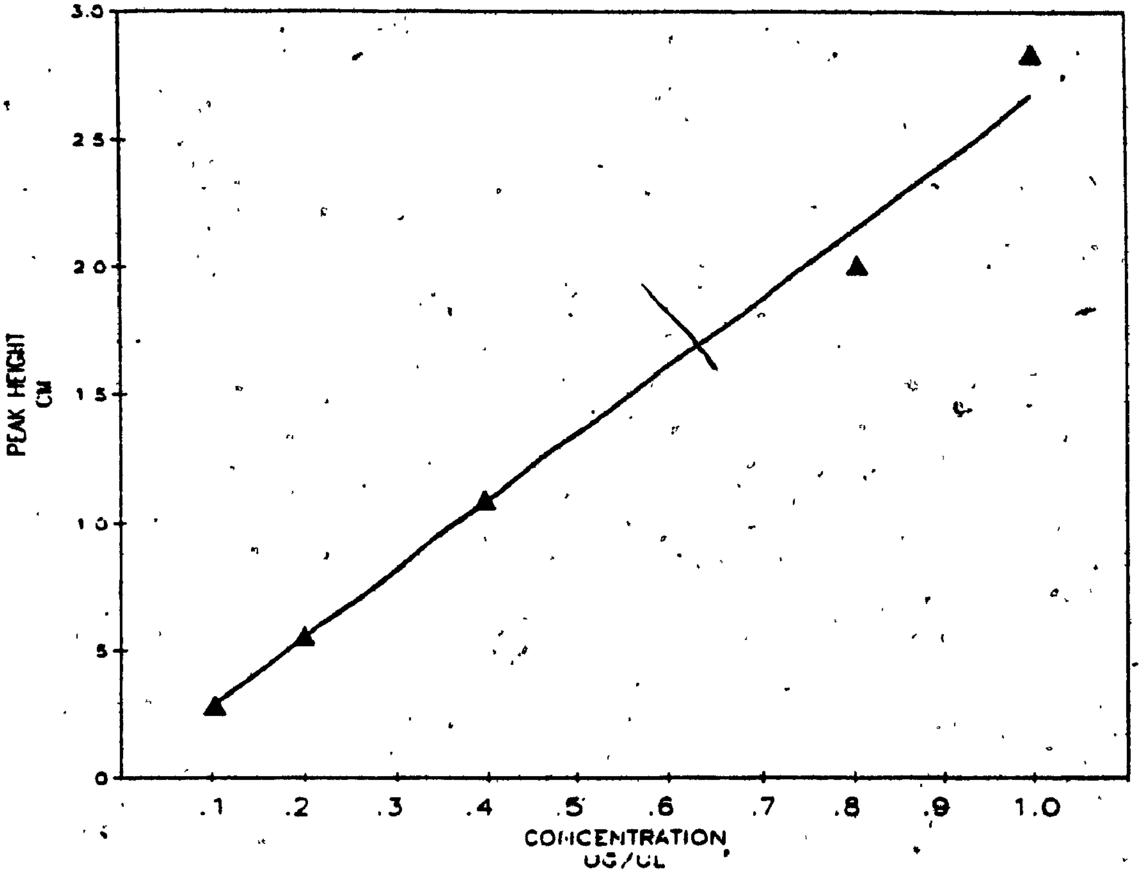


TABLE 47

CALIBRATION CURVES OBTAINED FOR $M(DDC)_n$ ON
(1.0-1.5-2.0-2.5-3.0%) QF-1 ON CW-HP, 80/100 mesh,
(60 cm x 2 mm I.D, glass tubing), AT 200°C,
CONCENTRATION: 0.1-1.0 ug/ul, N_2 FLOW RATE: 25 ml/min

METAL DDC	RETENTION TIME (min)	SLOPE (cm/ug)	REGRESSION ANALYSIS	
			CORRELATION COEFFICIENT	Y INTERCEPT (cm)
$Zn(DDC)_2$	3.9	15.6	1.00	- 1.32
$Pb(DDC)_2$	5.4	11.7	1.00	- 0.75
$Cd(DDC)_2$	5.8	10.9	0.98	0.50
$Ni(DDC)_2$	13.2	11.2	1.00	0.06
$Co(DDC)_3$	35.8	9.9	1.00	0.09

FIG. 61 CALIBRATION CURVES FOR $Zn(DDC)_2$ AND $Pb(DDC)_2$ ON 1.0-1.5-2.0-2.5-3.0% QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 200°C, AND 25 ml/min.

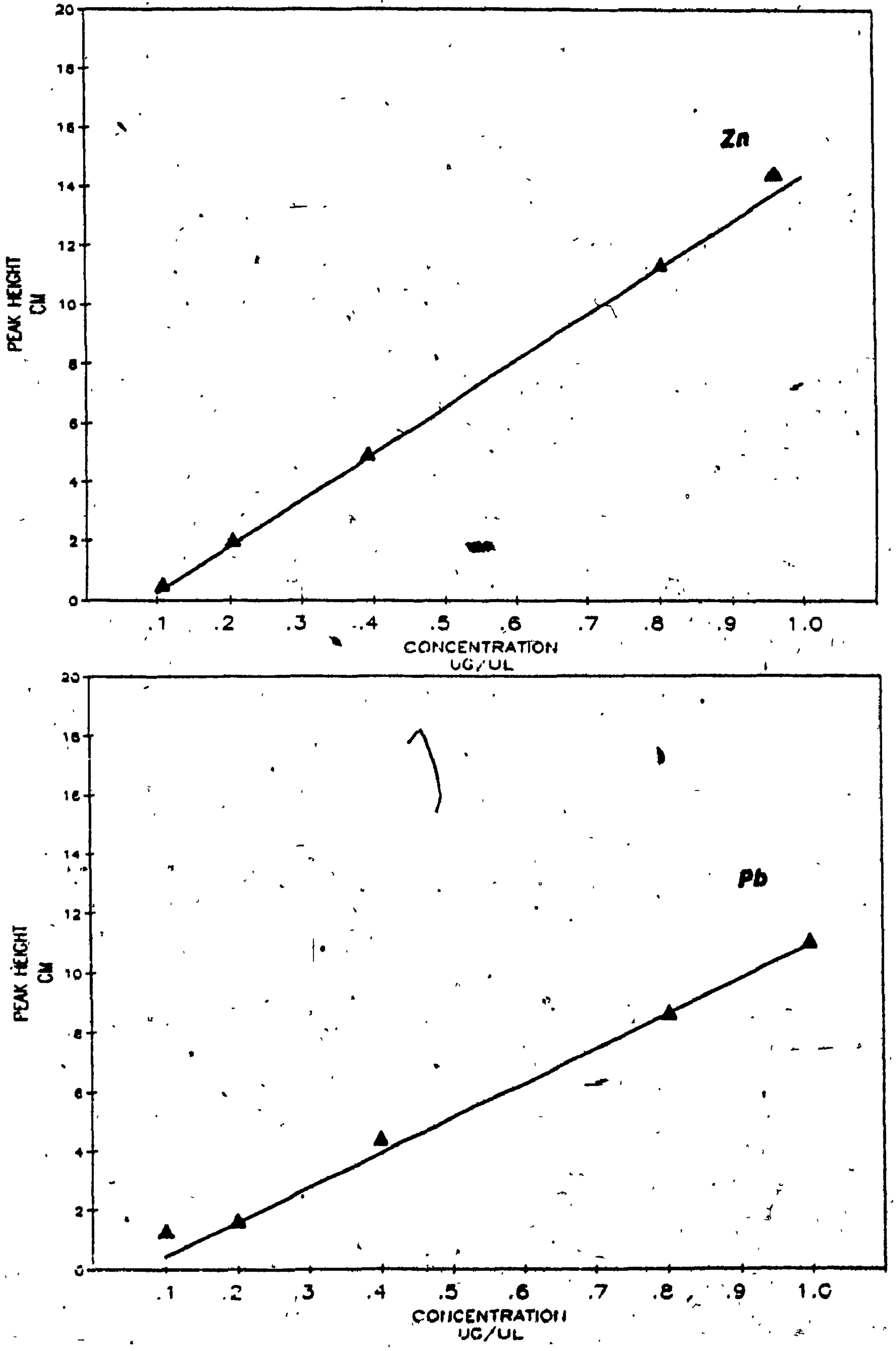


FIG. 62 CALIBRATION CURVES FOR Cd(DDC)$_2$ AND Ni(DDC)$_2$ ON 1.0-1.5-2.0-2.5-3.0% QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 200°C, AND 25 ml/min.

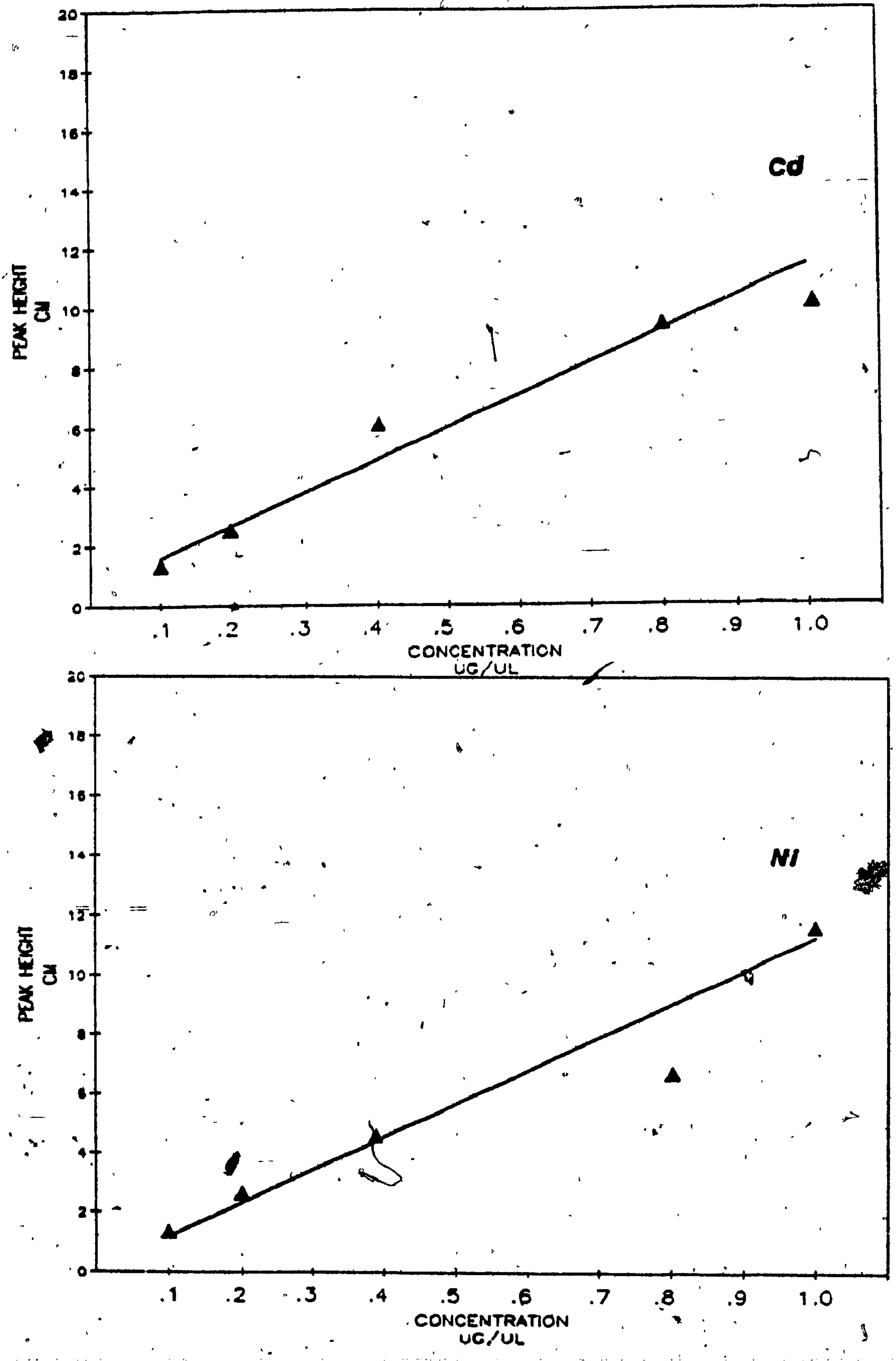


FIG. 63 CALIBRATION CURVE FOR $Co(DDC)_3$ ON 1.0-1.5-2.0-2.5-3.0% QF-1 ON CW-HP, 80/100 MESH, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 200°C, AND 25 ml/min.

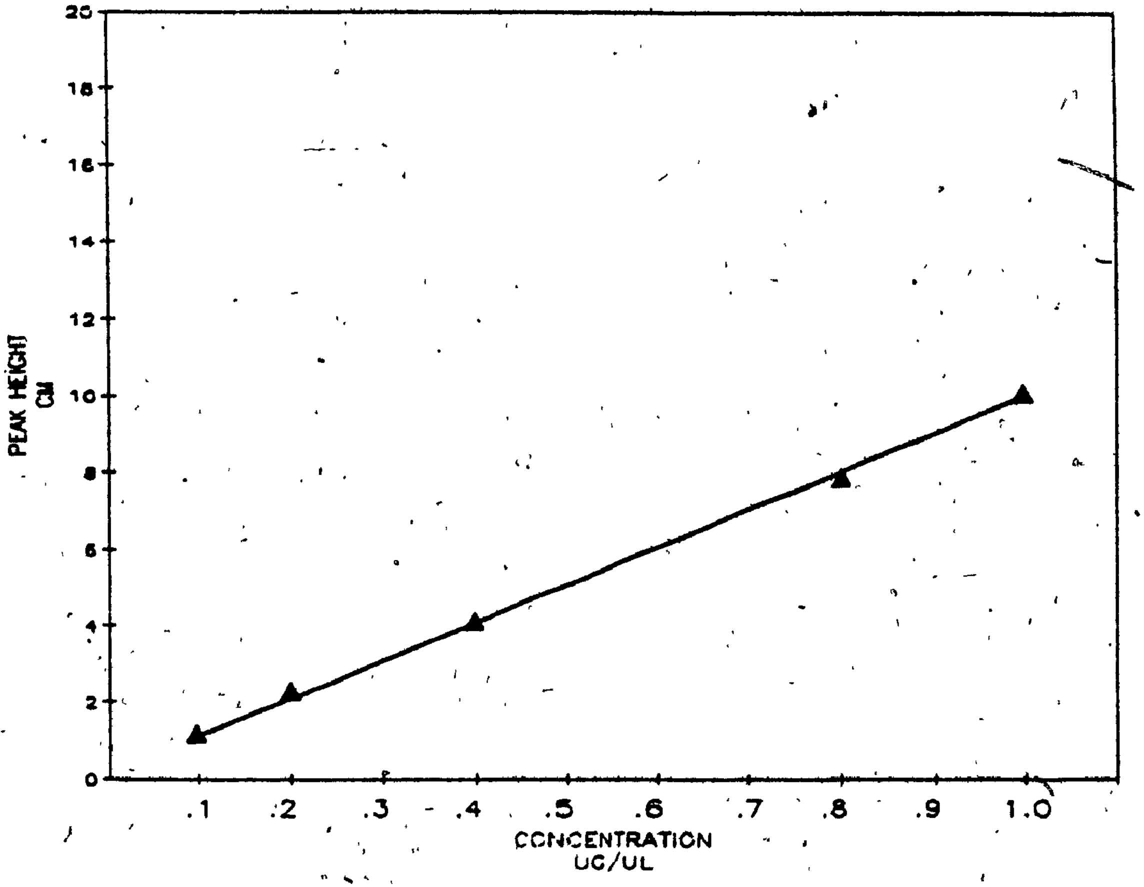


TABLE 48

CALIBRATION CURVES OBTAINED FOR M(DDC)$_n$ ON 1-2-3% OV 210 ON CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing), AT 220°C, CONCENTRATION: 0.1-1.0 ug/ul, N_2 FLOW RATE: 30 ml/min

METAL DDC	RETENTION TIME (min)	SLOPE (cm/ug)	REGRESSION ANALYSIS CORRELATION COEFFICIENT	REGRESSION ANALYSIS Y INTERCEPT (cm)
$Zn(DDC)_2$	1.4	19.2	0.99	- 0.56
$Cd(DDC)_2$	1.9	5.2	0.96	0.65
$Pb(DDC)_2$	1.9	9.9	0.99	- 1.30
$Cu(DDC)_2$	2.6	14.1	1.00	- 0.23
$Ni(DDC)_2$	4.2	14.7	0.99	0.16
$Co(DDC)_3$	10.0	9.2	0.96	2.90

FIG. 64 CALIBRATION CURVES FOR $Zn(DDC)_2$ AND $Cd(DDC)_2$ ON 1-2-3% OV-210 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 220°C, AND 30 ml/min.

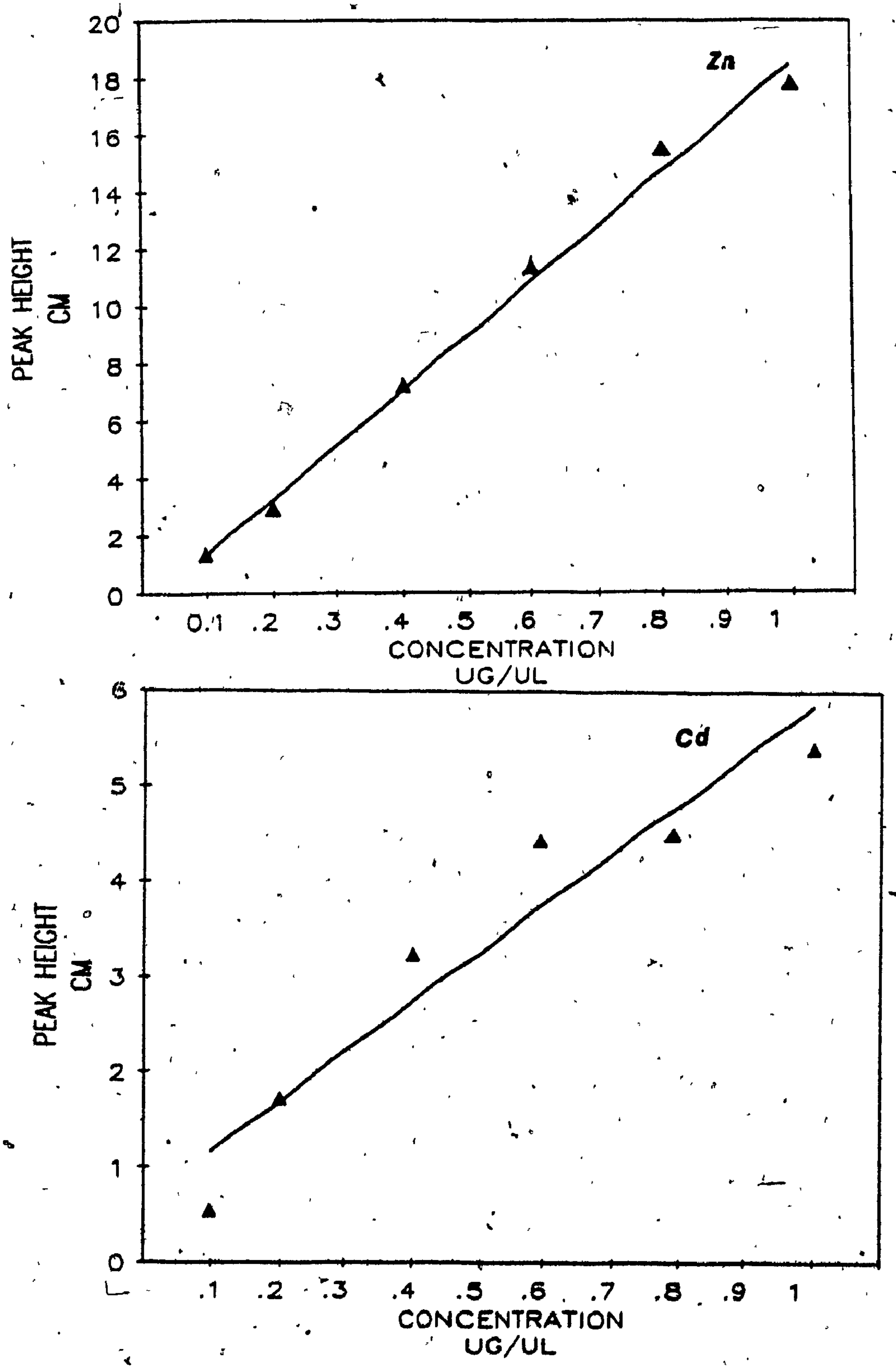


FIG. 65 CALIBRATION CURVES FOR Pb(DDC)$_2$ AND Cu(DDC)$_2$ ON 1-2-3% OV-210 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 220°C, AND 30 ml/min.

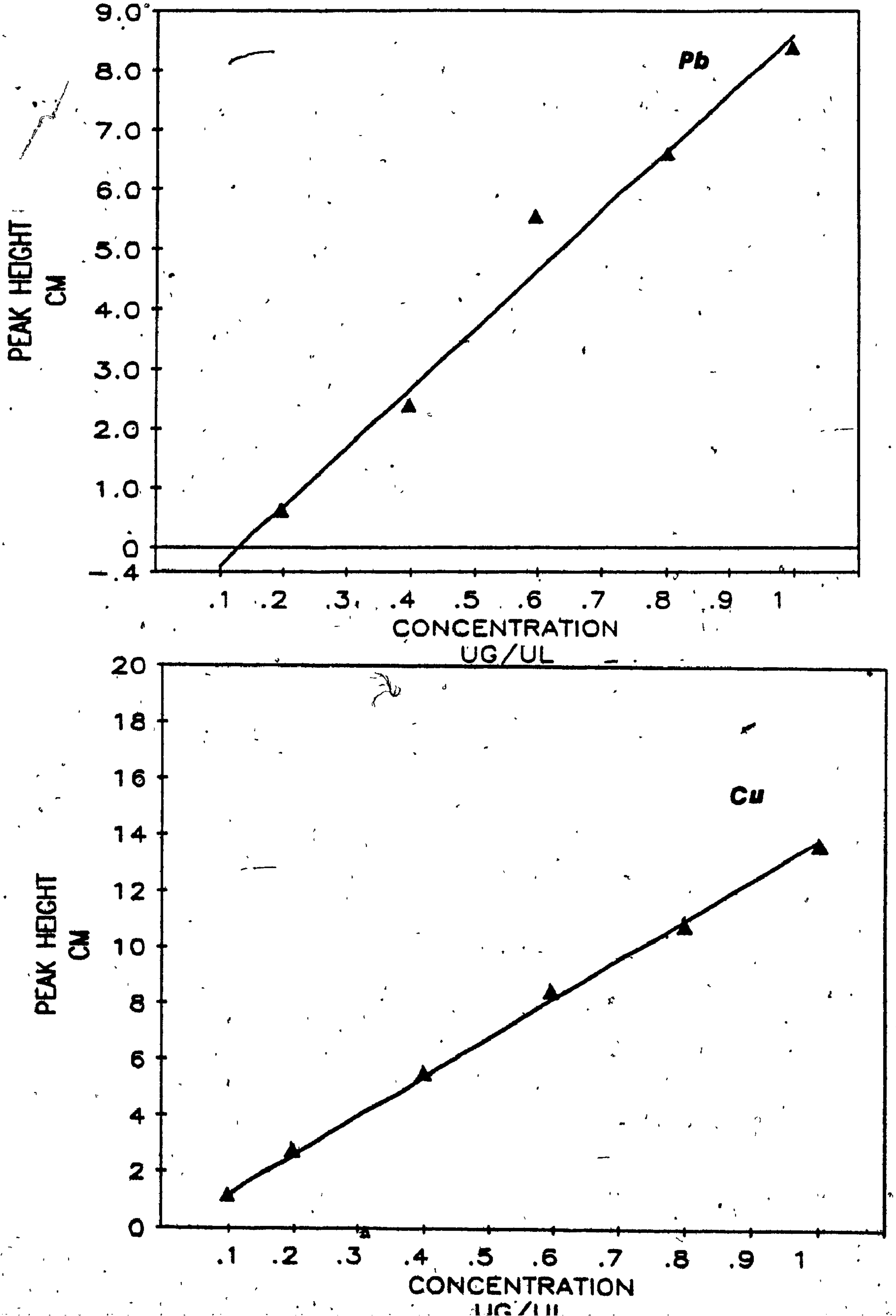


FIG. 66 CALIBRATION CURVES FOR AND Ni(DDC)$_2$ Co(DDC)$_3$ ON 1-2-3% OV-210 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 220°C, AND 30 ml/min.

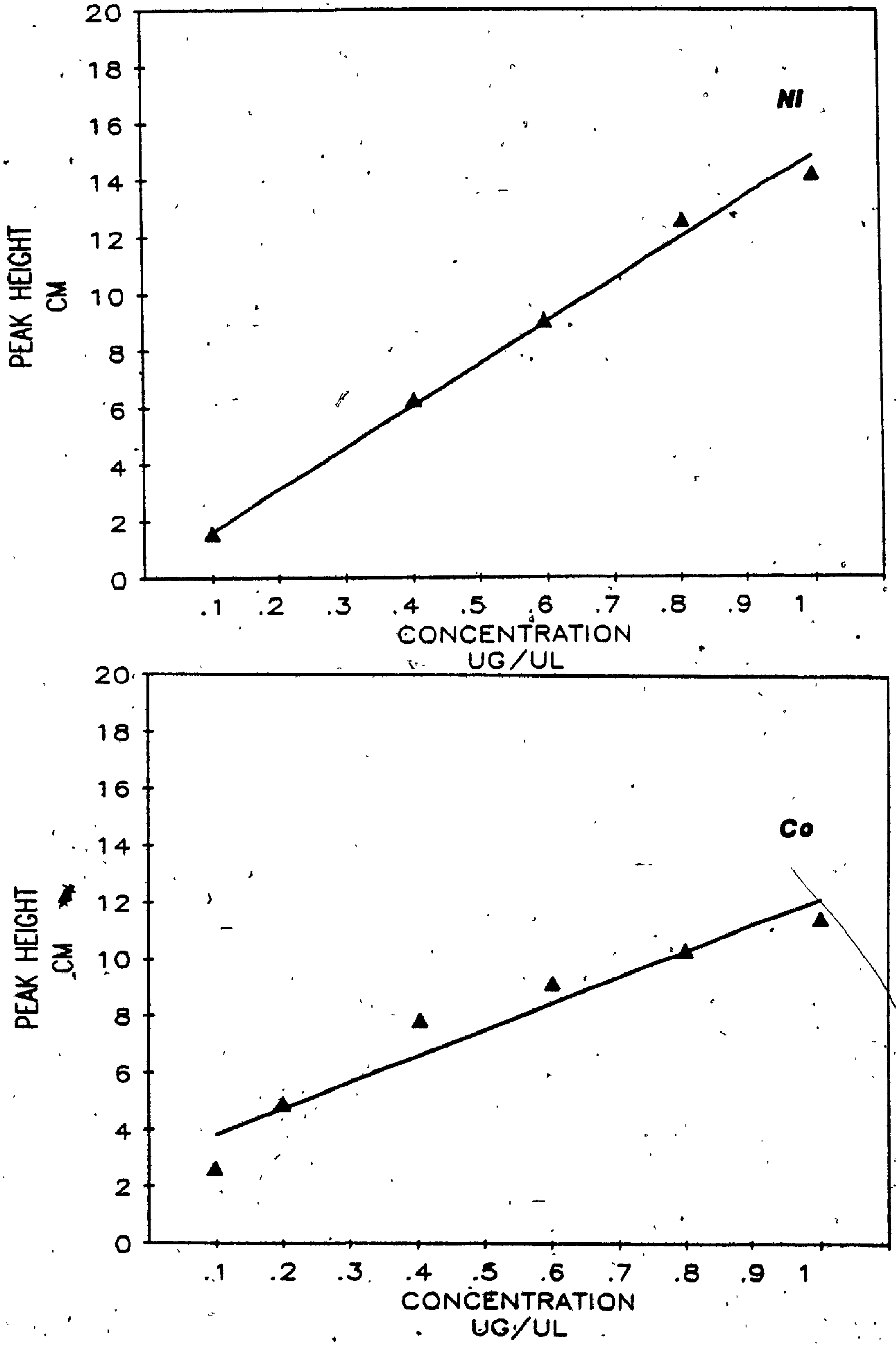


TABLE 49

CALIBRATION CURVES OBTAINED FOR M(DDC)$_n$ ON 3-2-1% OV 225 ON CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing), AT 240°C, CONCENTRATION: 0.1-1.0 ug/ul, N_2 FLOW RATE: 30 ml/min

METAL DDC	RETENTION TME (MIN)	SLOPE (cm/ug)	REGRESSION ANALYSIS	
			CORRELATION COEFFICIENT	Y INTERCEPT (cm)
$Zn(DDC)_2$	3.3	15.4	1.00	- 0.98
$Cd(DDC)_2$	5.1	12.5	0.99	- 0.12
$Pb(DDC)_2$	9.1	4.9	1.00	- 0.29
$Cu(DDC)_2$	6.7	13.1	0.99	- 1.16
$Ni(DDC)_2$	11.7	13.8	0.99	- 0.24

FIG. 67 CALIBRATION CURVES FOR $Zn(DDC)_2$ AND $Cd(DDC)_2$ ON 3-2-1% OV-225 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 240°C, AND 30 ml/min.

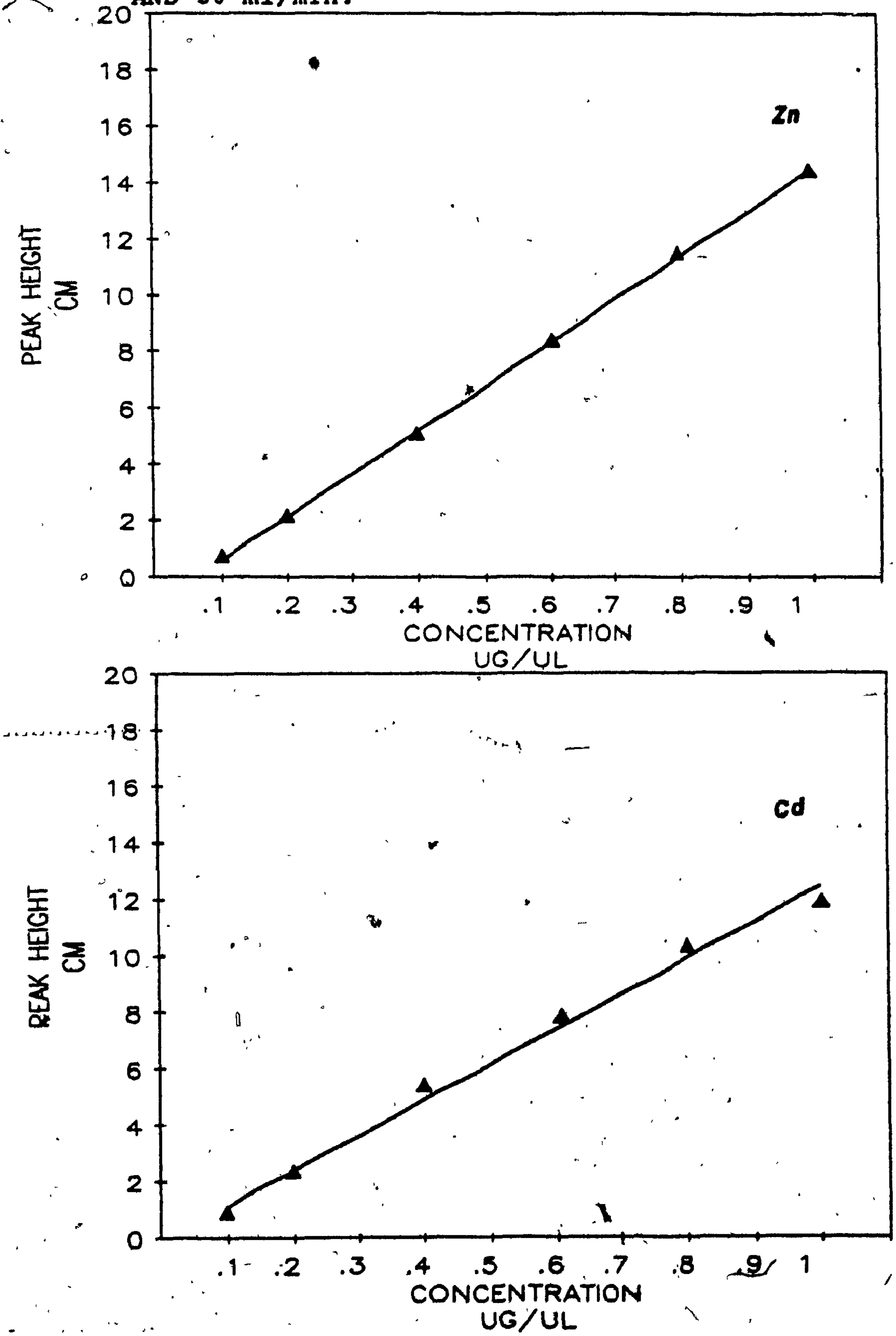


FIG. 68 CALIBRATION CURVES FOR $Pb(DDC)_2$ AND $Cu(DDC)_2$ ON 3-2-1% OV-225 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 240°C, AND 30 ml/min.

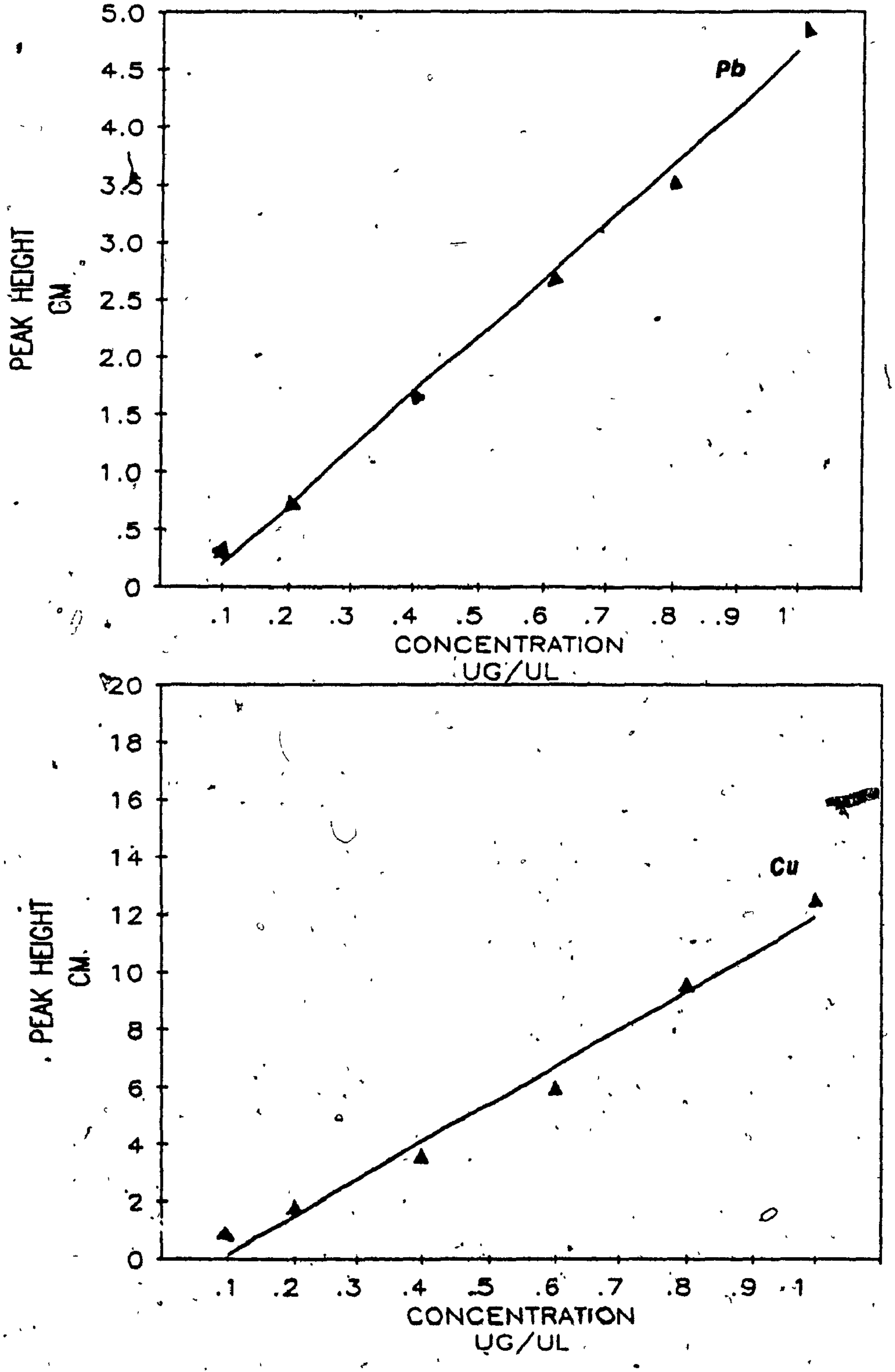


FIG. 69 CALIBRATION CURVE FOR $Ni(DDC)_2$ ON 3-2-1% OV-225 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 240°C, AND 30 ml/min.

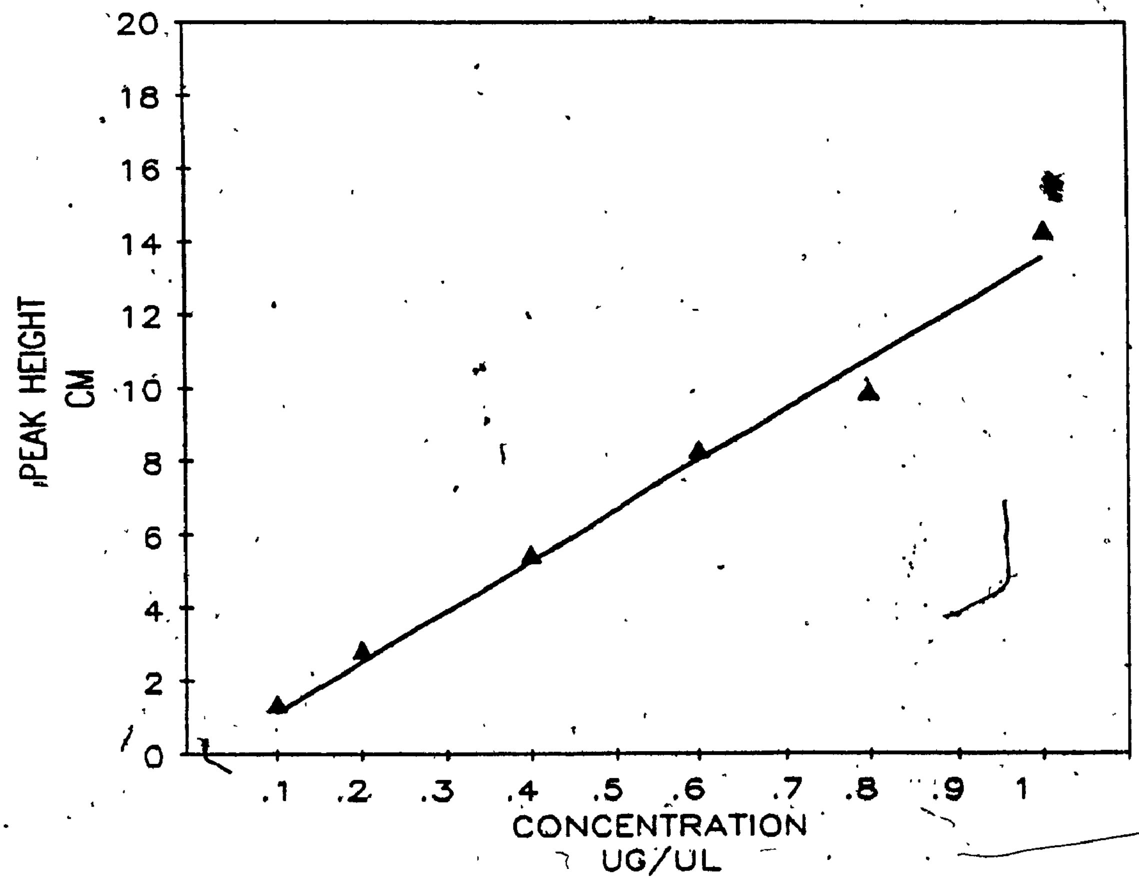


TABLE 50

CALIBRATION CURVES OBTAINED FOR M(DDC)$_n$ ON 1-2-3% EACH OF OV 101 - QF-1 ON CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing), AT 240°C

CONCENTRATION: 0.1-1.0 ug/ul, N_2 FLOW RATE : 30 ml/min

			REGRESSION ANALYSIS	
METAL DDC	RETENTION TIME (min)	SLOPE (cm/ug)	CORRELATION COEFFICIENT	Y INTERCEPT (cm)
$Zn(DDC)_2$	1.2	12.6	1.00	- 0.67
$Cd(DDC)_2$	1.7	12.1	1.00	- 0.32
$Pb(DDC)_2$	2.0	12.1	0.99	- 1.04
$Cu(DDC)_2$	2.0	17.7	1.00	- 0.53
$Ni(DDC)_2$	2.7	28.2	1.00	- 0.56
$Co(DDC)_3$	8.2	13.6	1.00	- 0.02

FIG. 70 CALIBRATION CURVES FOR Zh(DDC)$_2$ AND Cd(DDC)$_2$ ON 1-2-3% EACH OF OV-101-[illegible]F-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 240°C, AND 30 ml/min.

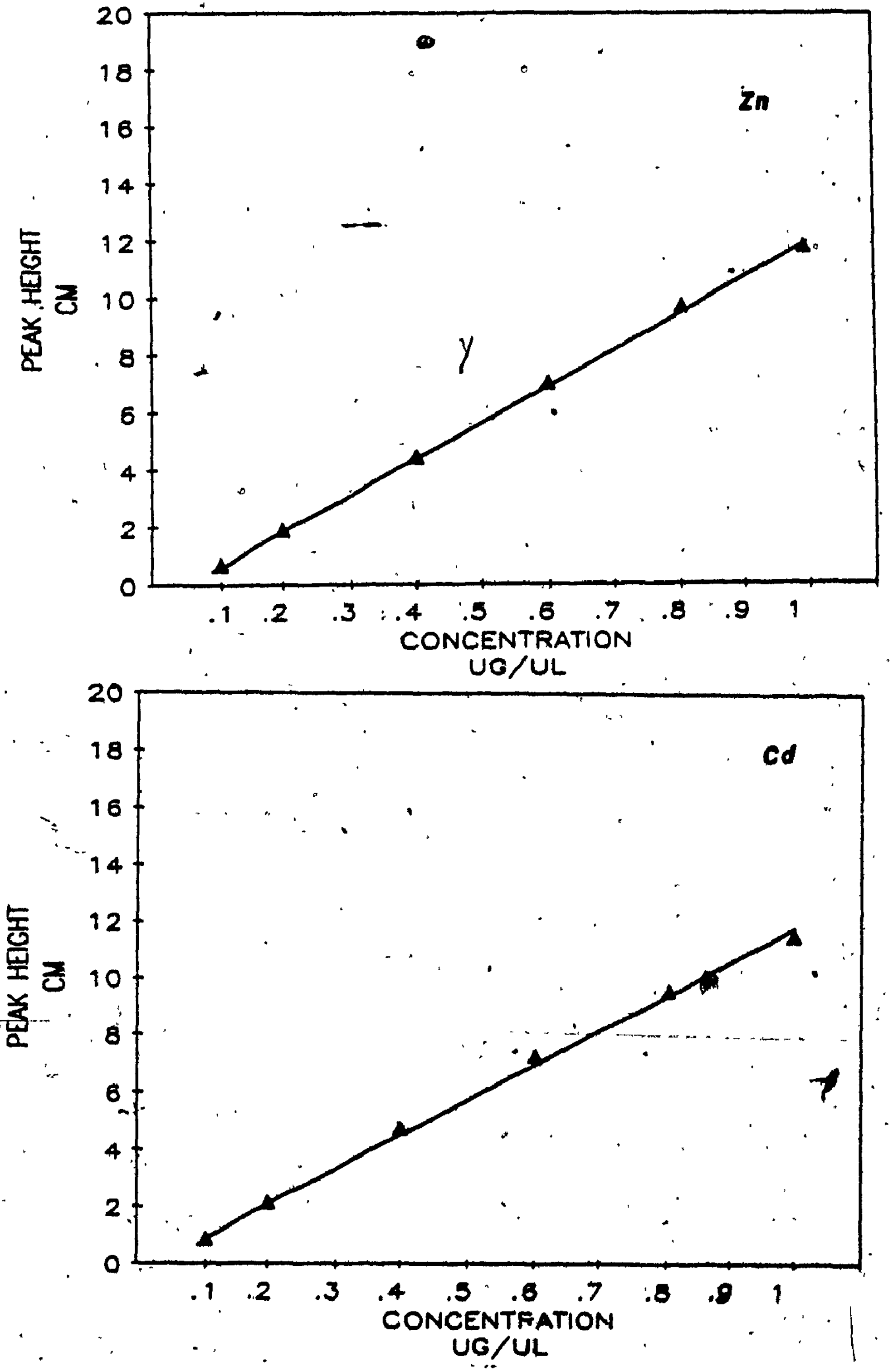


FIG. 71 CALIBRATION CURVES FOR Pb(DDC)$_2$ AND Cu(DDC)$_2$ ON 1-2-3% EACH OF OV-101-QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 240°C, AND 30 ml/min.

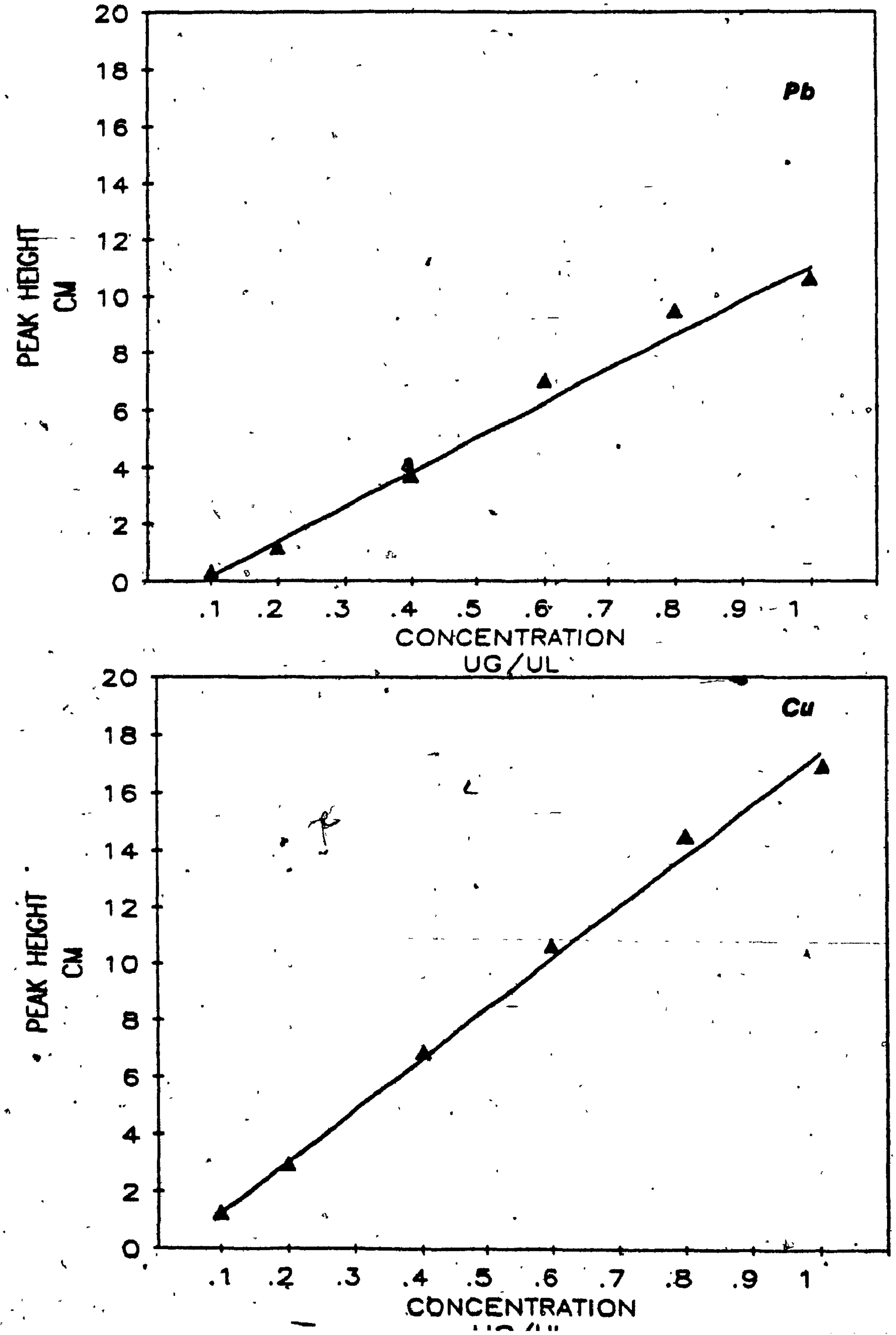


FIG. 72 CALIBRATION CURVES FOR $Ni(DDC)_2$ AND $Co(DDC)_3$ ON 1-2-3% EACH OF OV-101-QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 240°C, AND 30 ml/min.

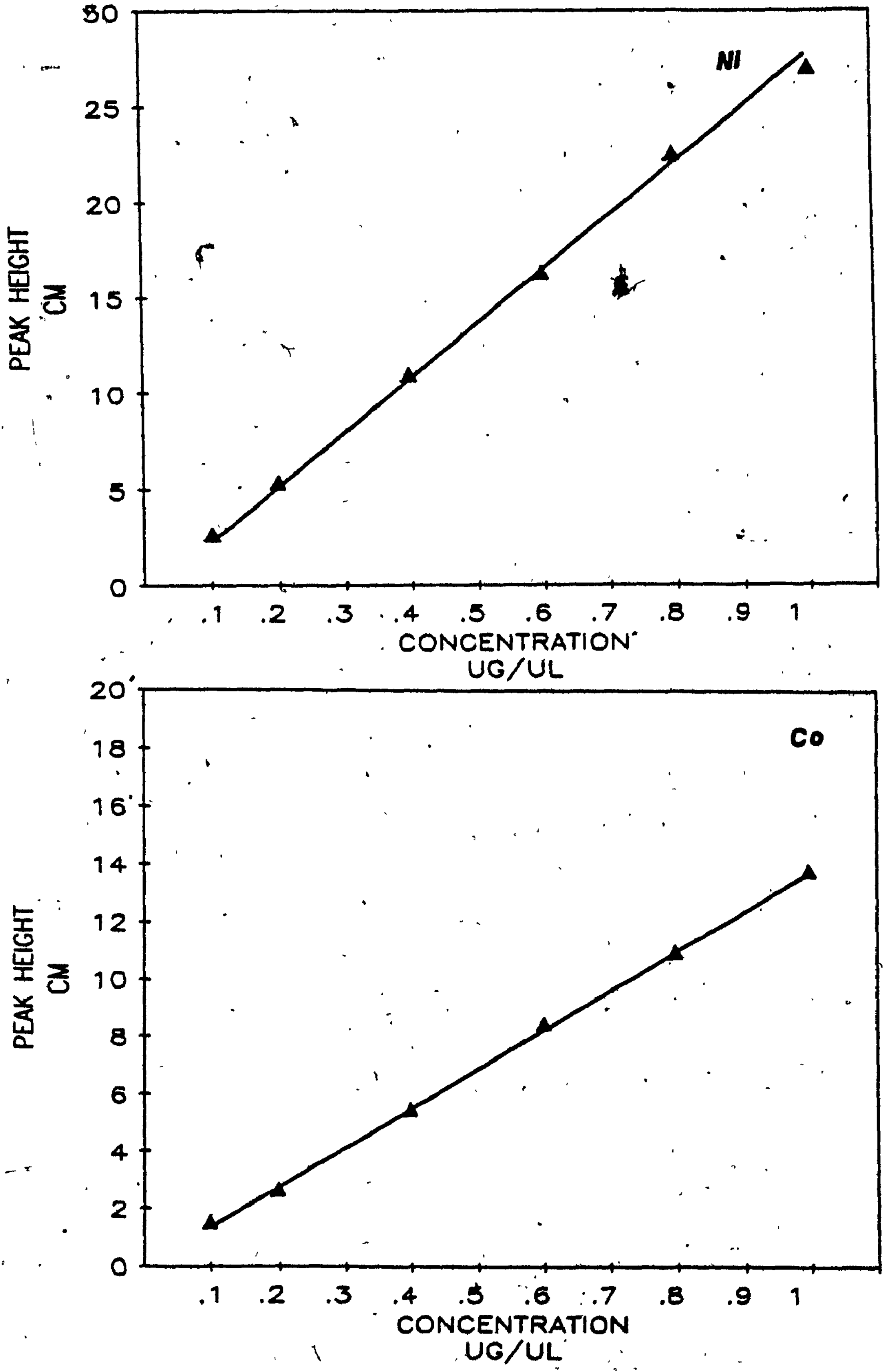


TABLE 51

CALIBRATION CURVES OBTAINED FOR $M(DDC)_n$ ON EACH OF 1-2-3% OV-225-QF-1 ON CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing), AT 240°C, CONCENTRATION: 0.1-1.0 ug/ul, N_2 FLOW RATE: 30 ml/min

METAL DDC	RETENTION TIME (min)	SLOPE (cm/ug)	REGRESSION ANALYSIS CORRELATION COEFFICIENT	REGRESSION ANALYSIS Y INTERCEPT (cm)
$Zn(DDC)_2$	3.0	11.0	1.00	- 0.49
$Cd(DDC)_2$	4.5	12.4	1.00	- 0.31
$Pb(DDC)_2$	7.6	4.8	0.99	- 0.08
$Cu(DDC)_2$	6.1	11.1	0.99	- 0.82

FIG. 73 CALIBRATION CURVES FOR Zn(DDC)$_2$ AND Cd(DDC)$_2$ ON 1-2-3% EACH OF OV-225-QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 240°C, AND 30 ml/min.

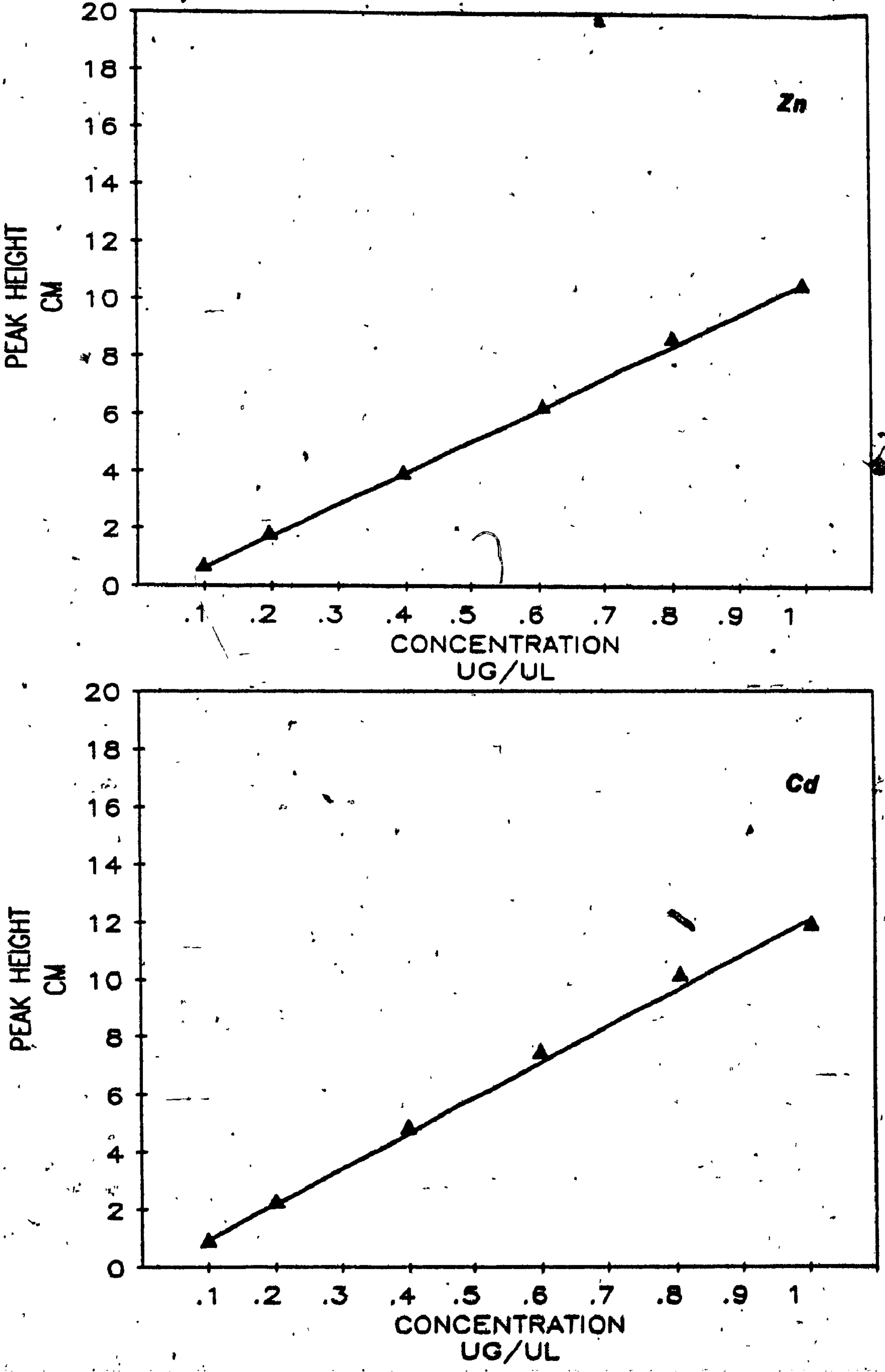
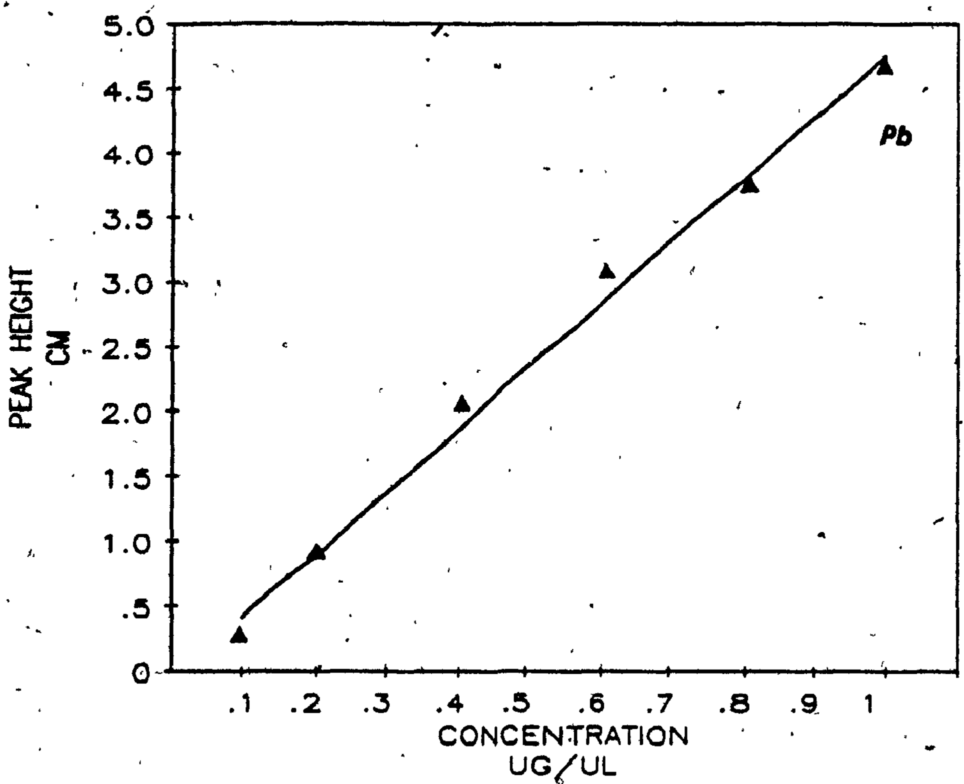


FIG. 74 CALIBRATION CURVES FOR $Pb(DDC)_2$ AND $Cu(DDC)_2$ ON 1-2-3% EACH OF OV-225-QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D. GLASS TUBING), AT 240°C, AND 30 ml/min.



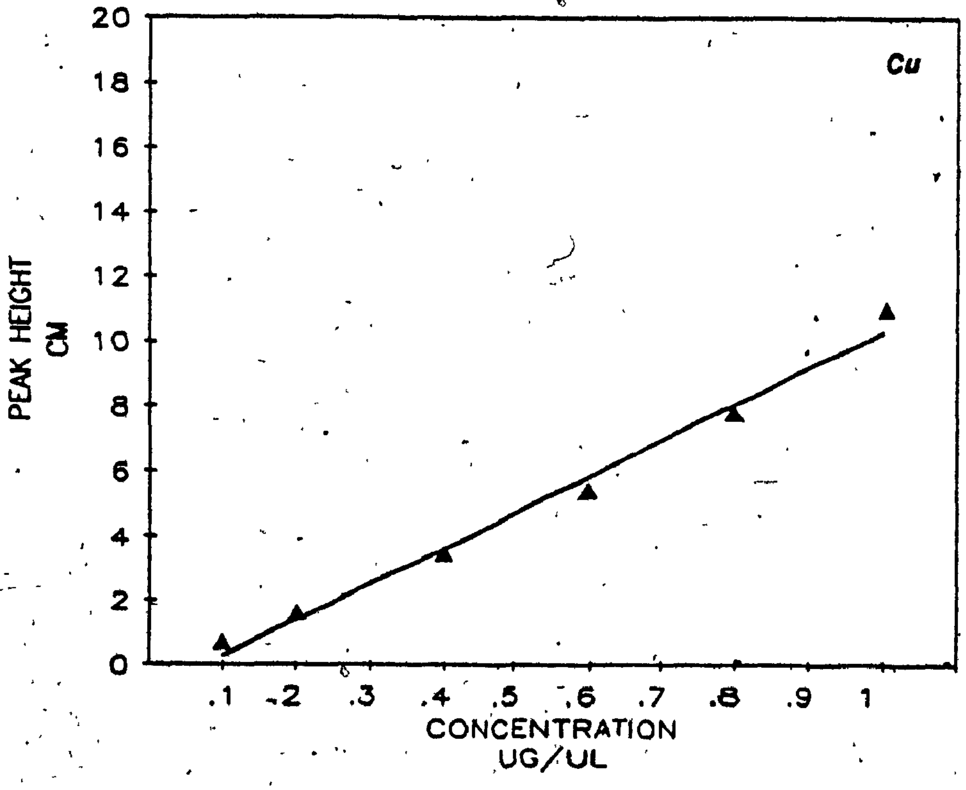


TABLE 52

CALIBRATION CURVES OBTAINED FOR $M(DDC)_n$ ON (1-2-3%) QF-1 + 1% OV-225 ON CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing), AT 240°C, CONCENTRATION RANGE: 0.1-1.0 ug/ul, N_2 FLOW RATE: 30 ml/min

			REGRESSION ANALYSIS	
METAL DDC	RETENTION TIME (MIN)	SLOPE (cm/ug)	CORRELATION COEFFICIENT	Y INTERCEPT (cm)
$Zn(DDC)_2$	1.3	10.3	1.00	- 0.71
$Cd(DDC)_2$	1.9	9.9	1.00	- 0.57
$Pb(DDC)_2$	2.0	16.2	0.98	- 0.65
$Cu(DDC)_2$	2.6	8.3	0.98	0.35
$Ni(DDC)_2$	4.3	8.7	0.99	0.24

FIG. 75 CALIBRATION CURVES FOR $Zn(DDC)_2$ AND $Cd(DDC)_2$ ON (1-2-3%) QF-1 + 1% OV-225 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 240°C, AND 30 ml/min.

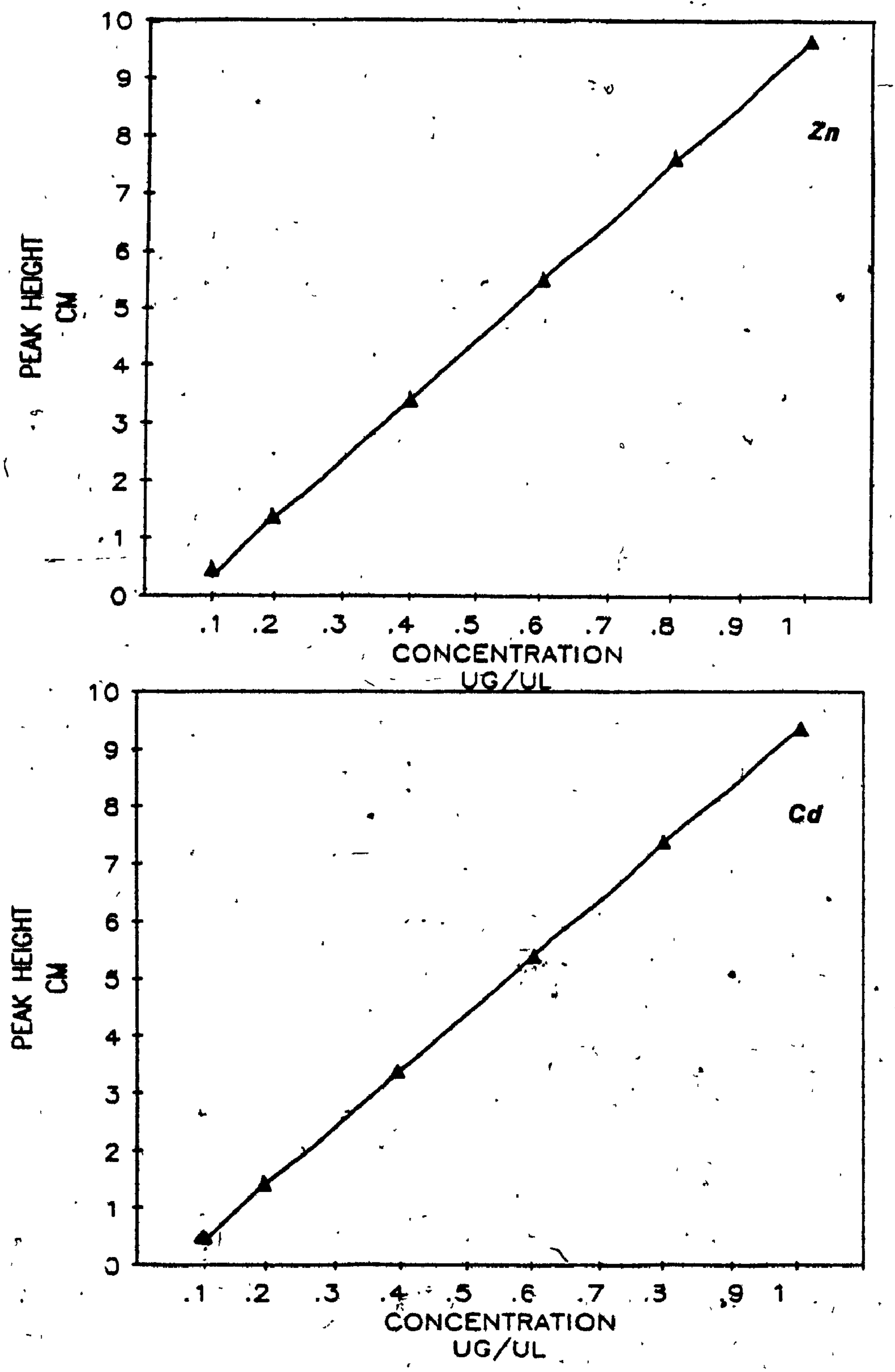


FIG. 76 CALIBRATION CURVES FOR $Pb(DDC)_2$ AND $Cu(DDC)_2$ ON (1-2-3%) QF-1 + 1% OV-225 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 240°C, AND 30 ml/min.

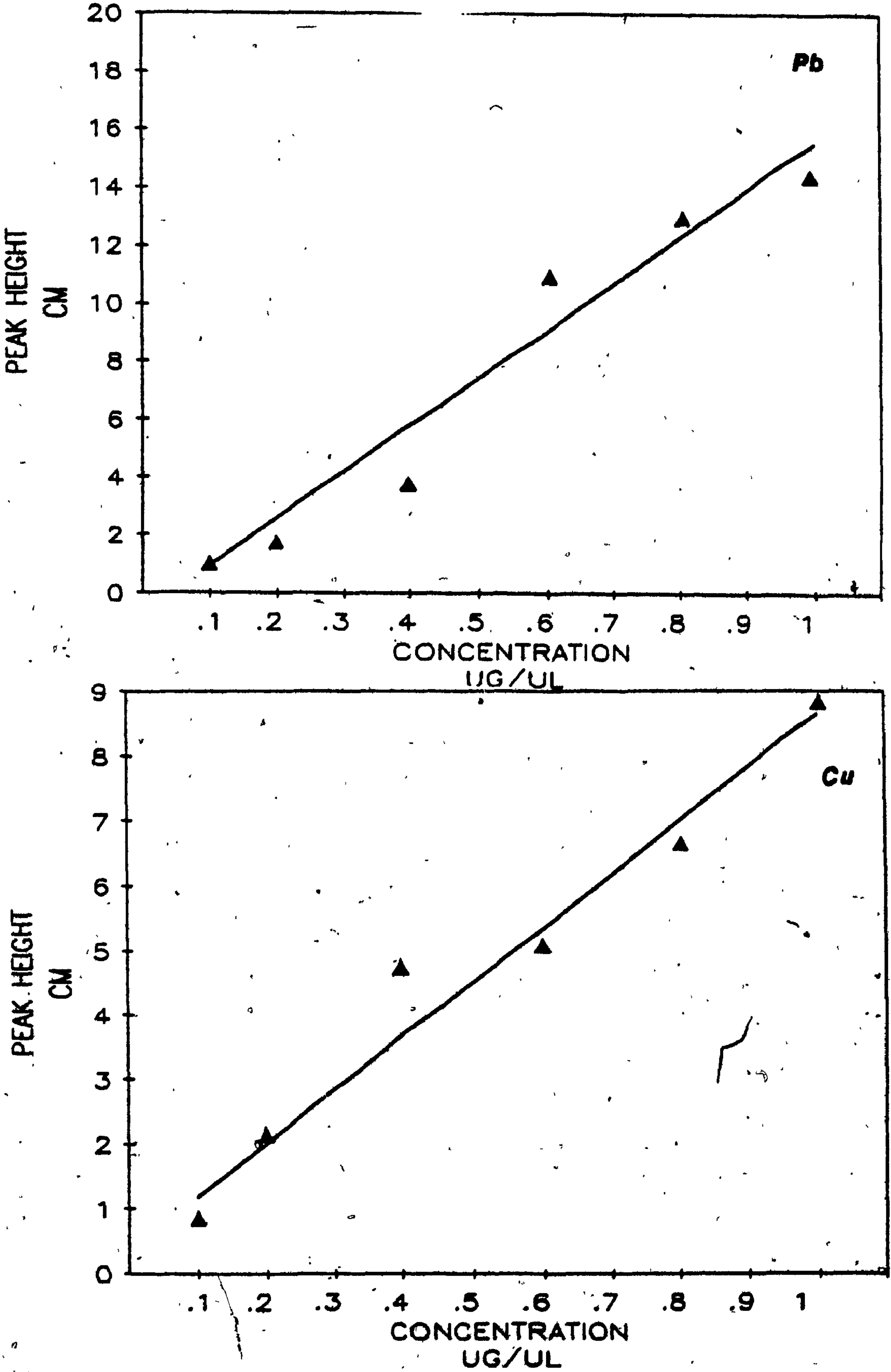
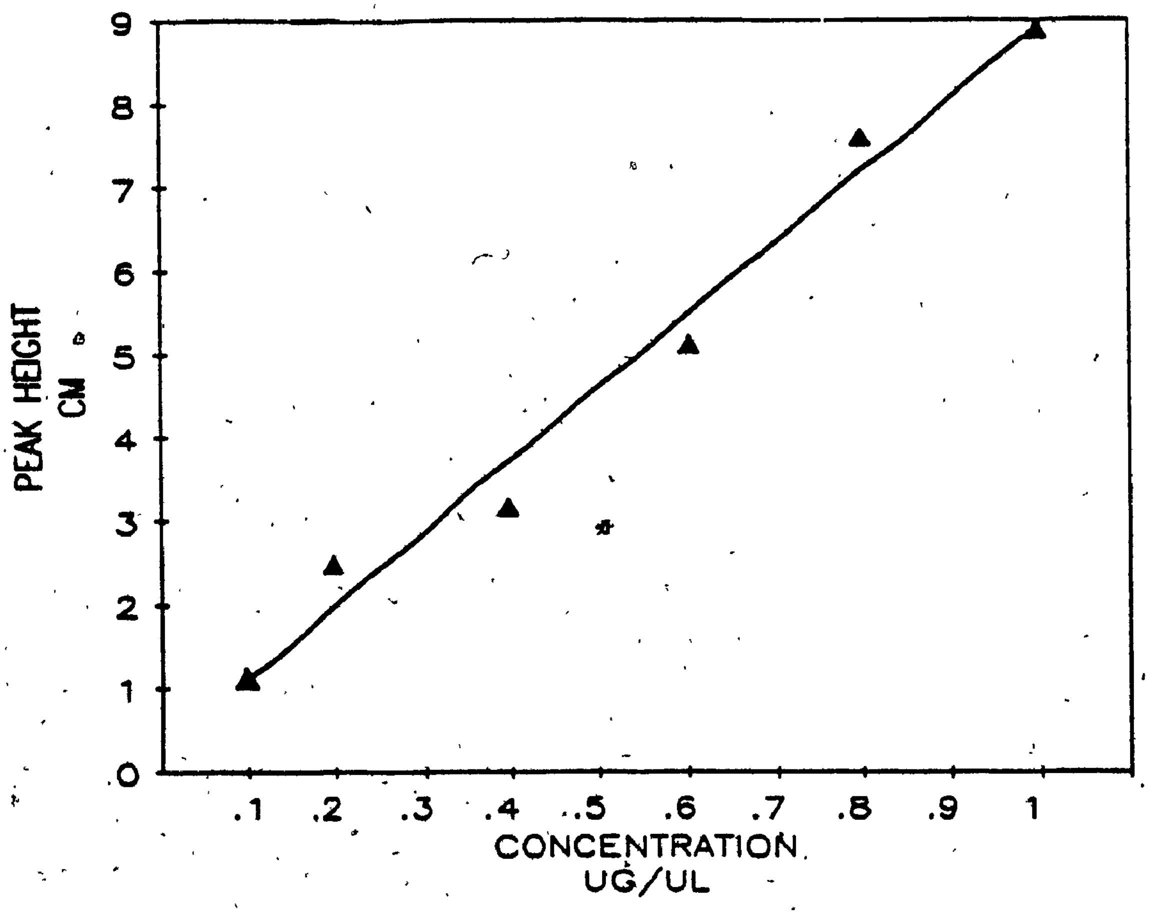


FIG. 77 CALIBRATION CURVE FOR $Ni(DDC)_2$ ON (1-2-3%) QF-1 + 1% OV-225 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 240°C, AND 30 ml/min.



APPENDIX II

SINGLE COLUMN COMPENSATION

When temperature programming is used, the rate of stationary phase bleed from the column increases as the oven temperature rises. This produces a rising baseline which can, in many cases, make quantitation of peaks difficult or impossible.

The standard means for dealing with this effect is to use dual columns with two detectors, an analyzing and a reference column, and measure the difference in signals. the columns must be identical so that the bleed rates will be the same and cancel in the subtraction. In practice this is not possible, so the flow rate in the reference column is adjusted to balance the bleed rates.

Single column compensation provides an electronic substitute for the second reference column. This eliminates the need for a second column and detector.

During a blank run (no sample injection) a mathematical model of the column bleed profile is measured and stored. This is subtracted from subsequent sample runs to produce a flat baseline.

This technique was used in this project with columns such as 5% OV-101 + 5% QF-1 column (Fig. 78) where the bleeding observed was fairly high.

FIG. 78 SINGLE COLUMN COMPENSATION

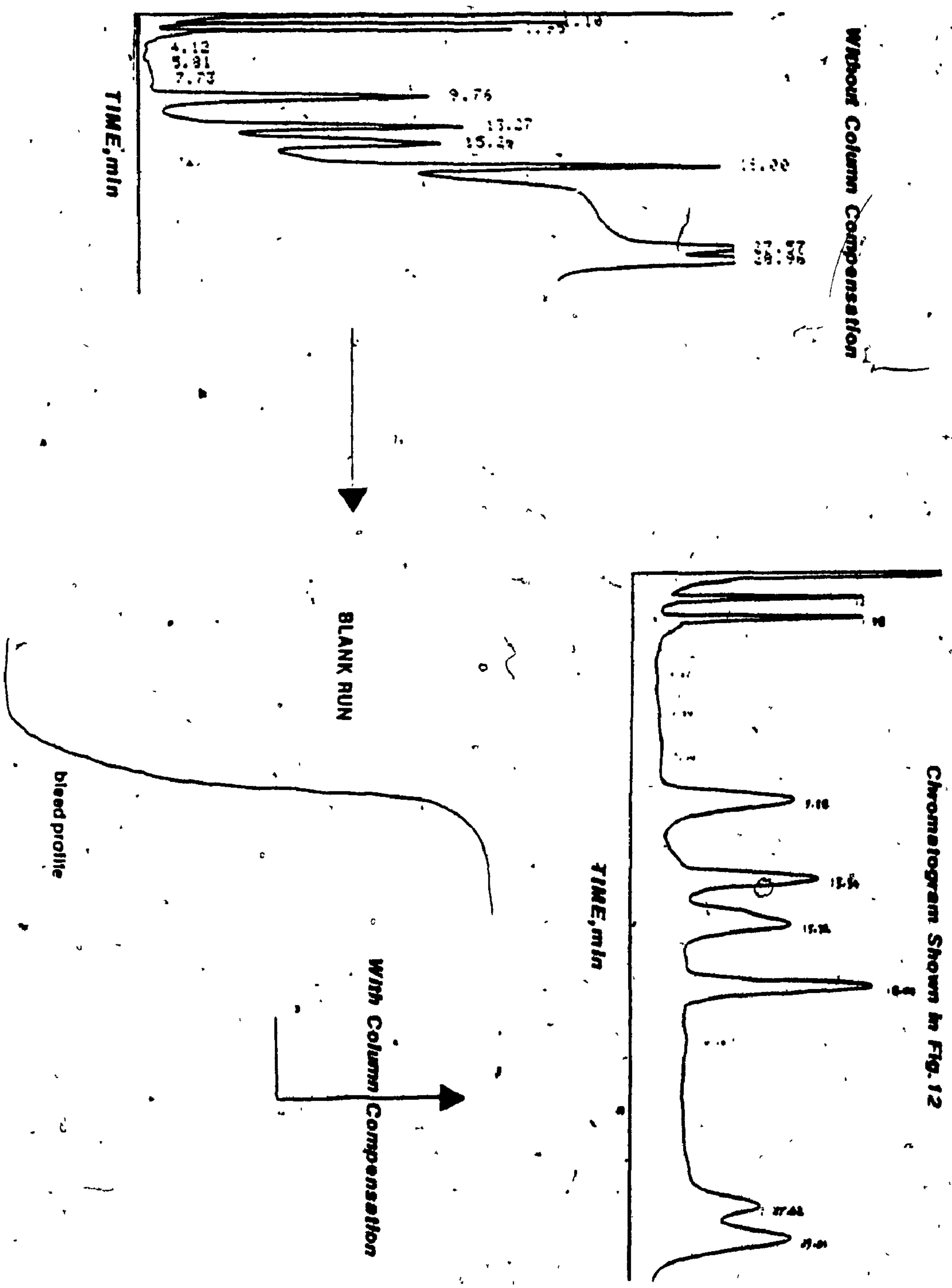


FIG. 15 SEPARATION OF AN EIGHT M(DDC)$_n$ MIXTURE ON 2% OV-101 ON CW-HP, 100/120 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 200°C FOR 10 MINS, THEN PROGRAMMED TO 240°C

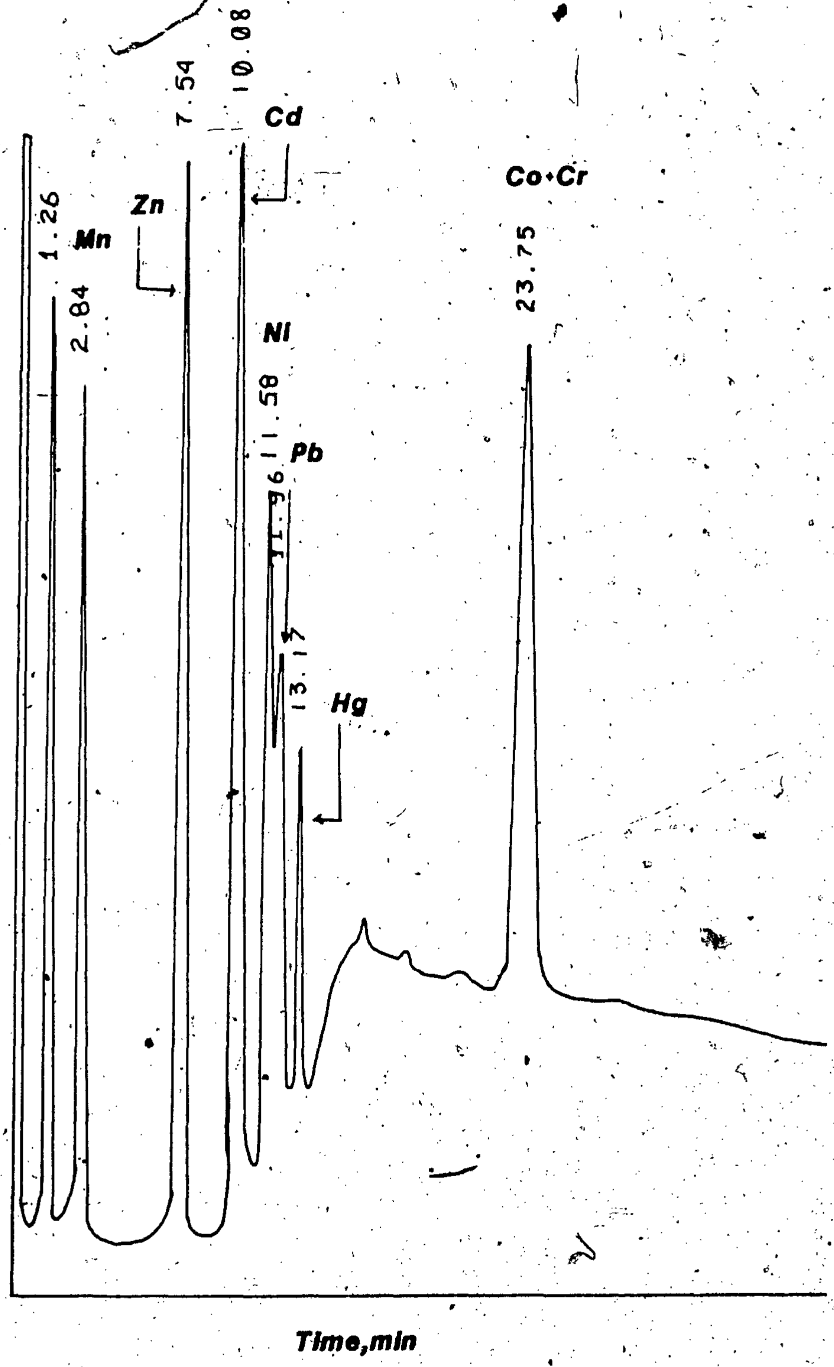


FIG. 16 SEPARATION OF AN EIGHT $M(DDC)_n$ MIXTURE ON 1-2-3% OV-101 ON CW-HP, 100/120 MESH, (60 cm x 2 mm I.D, GLASS TUBING), PROGRAMMED FROM 200°C TO 240°C AT 4°C/min, then at 240°C FOR 10 MINS, AND 25 ml/min.

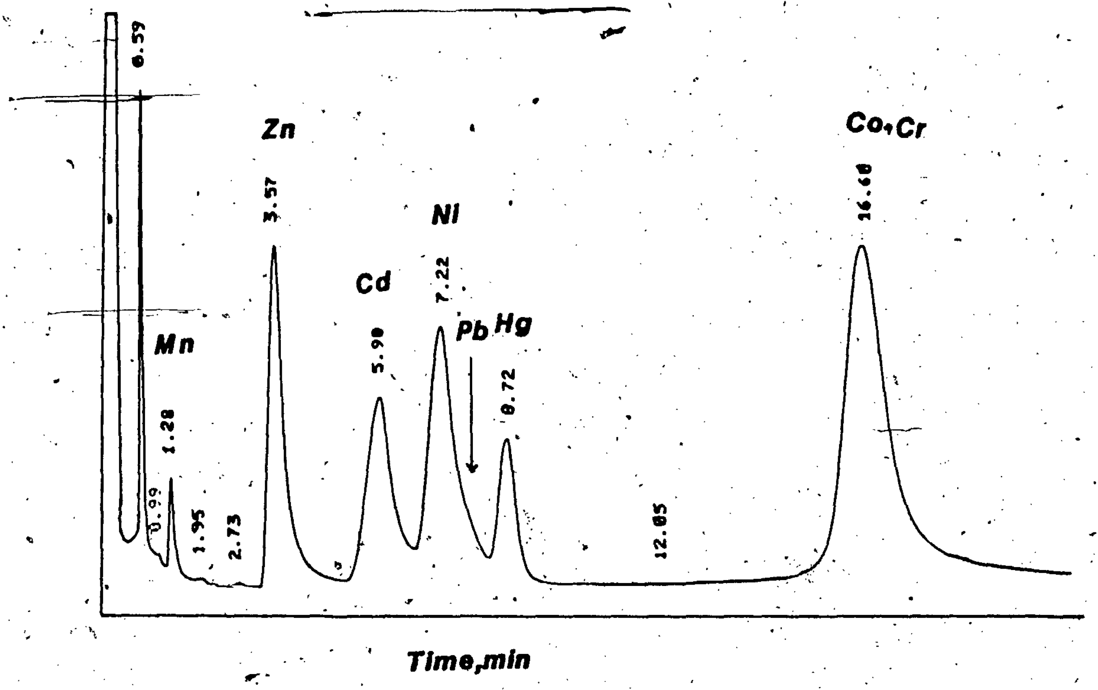
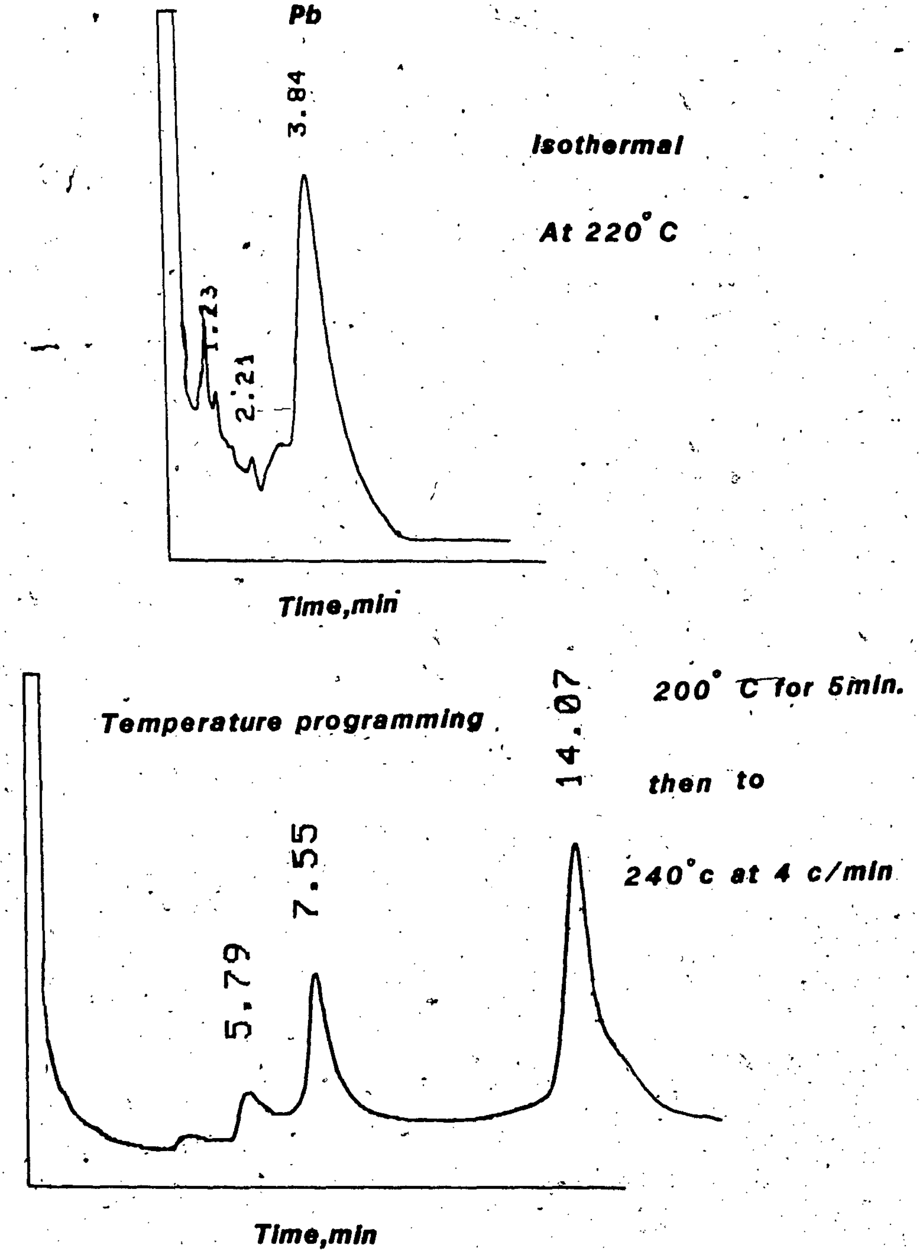


FIG. 17 DECOMPOSITION OF Pb(DDC)$_2$* ON 1-2-3% OV-101 ON CW-HP, 100/120 MESH, (60 cm x 2 mm I.D, GLASS TUBING)



clearly underwent degradation(Fig 17).

7.3.1.2 2-4-6% OV-101 ON CW-HP, 100/120 mesh (60 cm X 2 mm I.D, glass tubing) (Table 10, Figs 18 & 19)

The results of this study are summarized in Table 10. The total analysis time for Zn, Cd, Cu, and Ni DDC on this gradient column (4.8 min), was less than that on the column containing 4% stationary phase which was equivalent to the average loading of the gradient column. The resolution was also superior on the gradient column compared to any of the three conventional columns. The chromatograms on these columns are shown in Fig 18. Although the gradient column provided greater resolution, $Cd(DDC)_2$ and $Cu(DDC)_2$ still did not separate. Therefore, no further experiments were attempted with more complex DDC mixtures. $Pb(DDC)_2$, when injected by itself, gave two peaks on both the 4% column and the gradient column,(Fig 19).

7.3.2 GRADIENT COLUMNS WITH SE-30 STATIONARY PHASE

The use of this stationary phase for the analysis of metal chelates was reported by several workers (1, 2, 8). It is reported that diffusion of low molecular weight n-alkanes

TABLE 10

COMPARATIVE RESULTS OBTAINED ON A 2-4-6% OV-101 GRADIENT COLUMN, AND ON 2%, 4% AND 6% OV-101 CONVENTIONAL COLUMNS AT 220°C

	RETENTION TIME, min			
METAL DDC	2%	4%	6%	2-4-6%
Zn	1.7	3.5	4.0	2.4
Cd	2.6	5.4	6.1	3.7
Cu	2.7	5.4	6.6	3.7
Ni	3.3	7.0	7.9	4.8
RSD% ±	< 0.9	< 1.0	< 0.9	< 0.8
	RESOLUTION			
Zn-Cd	1.2	3.0	2.4	3.3
Zn-Ni	1.9	4.2	3.9	4.4
Cd-Ni	0.7	1.6	1.6	1.6
RSD% ±	< 7.0	< 5.6	< 6.1	< 6.0

FIG. 18 COMPARISON OF CHROMATOGRAMS OBTAINED ON A 2-4-6% OV-101 GRADIENT COLUMN, AND ON 2%, 4%, AND 6% OV-101 CONVENTIONAL COLUMNS, AT 220°C, AND 30ml/min.

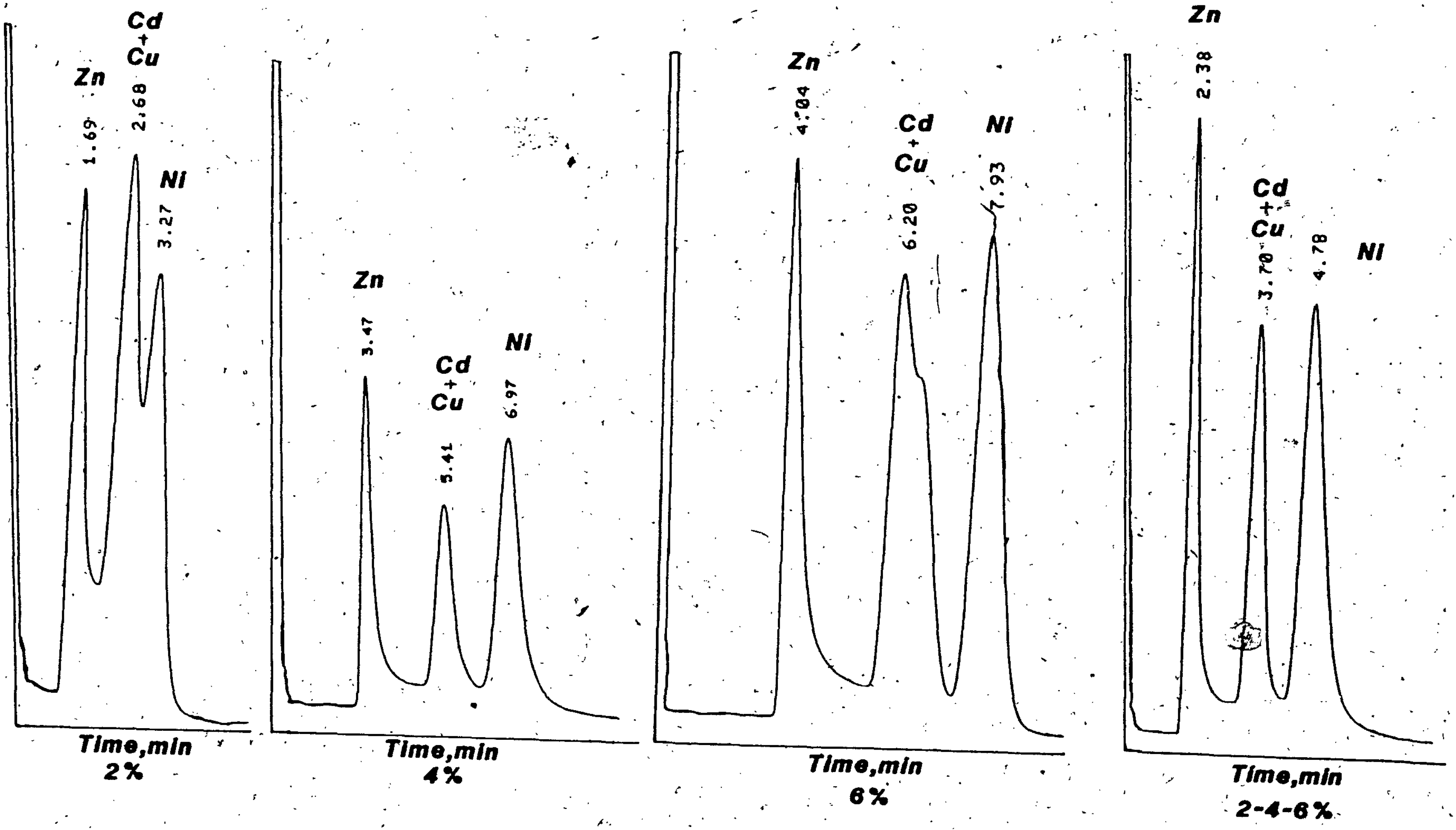
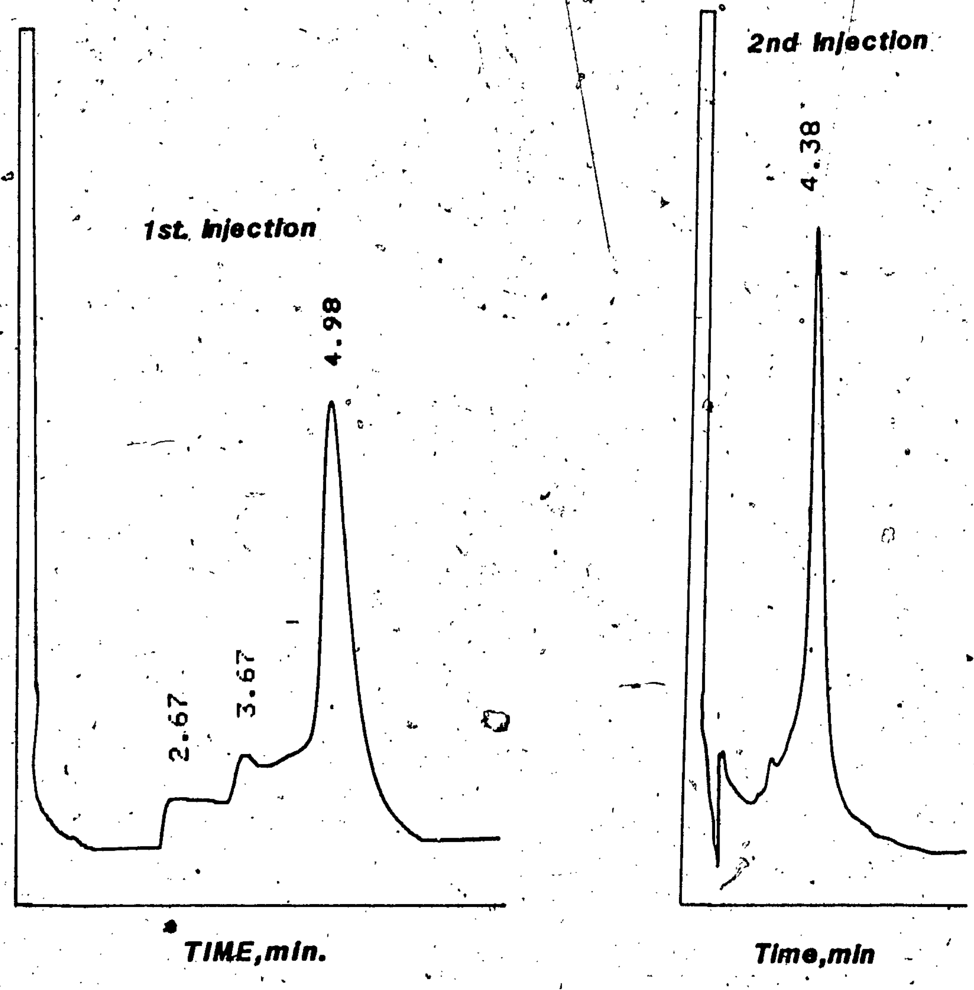


FIG. 19 DECOMPOSITION OF $Pb(DDC)_2$ ON 2-4-6% OV-101 ON CW-HP, 100/120 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 220°C, AND 30 ml/min.



is faster in high molecular weight poly(dimethylsiloxanes) gums such as SE 30, than in methylsilicone oils like OV-101. This being the case more efficient partitioning and better separation efficiency could be predicted using SE-30 as liquid phase (31).

7.3.2.1 1-3-5% SE 30 ON CW-HP, 80/100 mesh, (60 cm X 2 mm I.D, glass tubing) (Table 11, Fig 20)

Results obtained on this column compared to those on 1, 3 and 5% loaded columns are listed in Table 11. The 1-3-5% gradient column was found to give better resolution than any of the conventional columns. Not only was column efficiency higher, but the total analysis time was marginally shorter than on the column containing a loading of 3%, the average of that on the gradient column. The chromatograms are shown in Fig 20. It can be seen that the Cd-Cu pair was still not resolved on these columns. Hence, no further tests were made with this column.

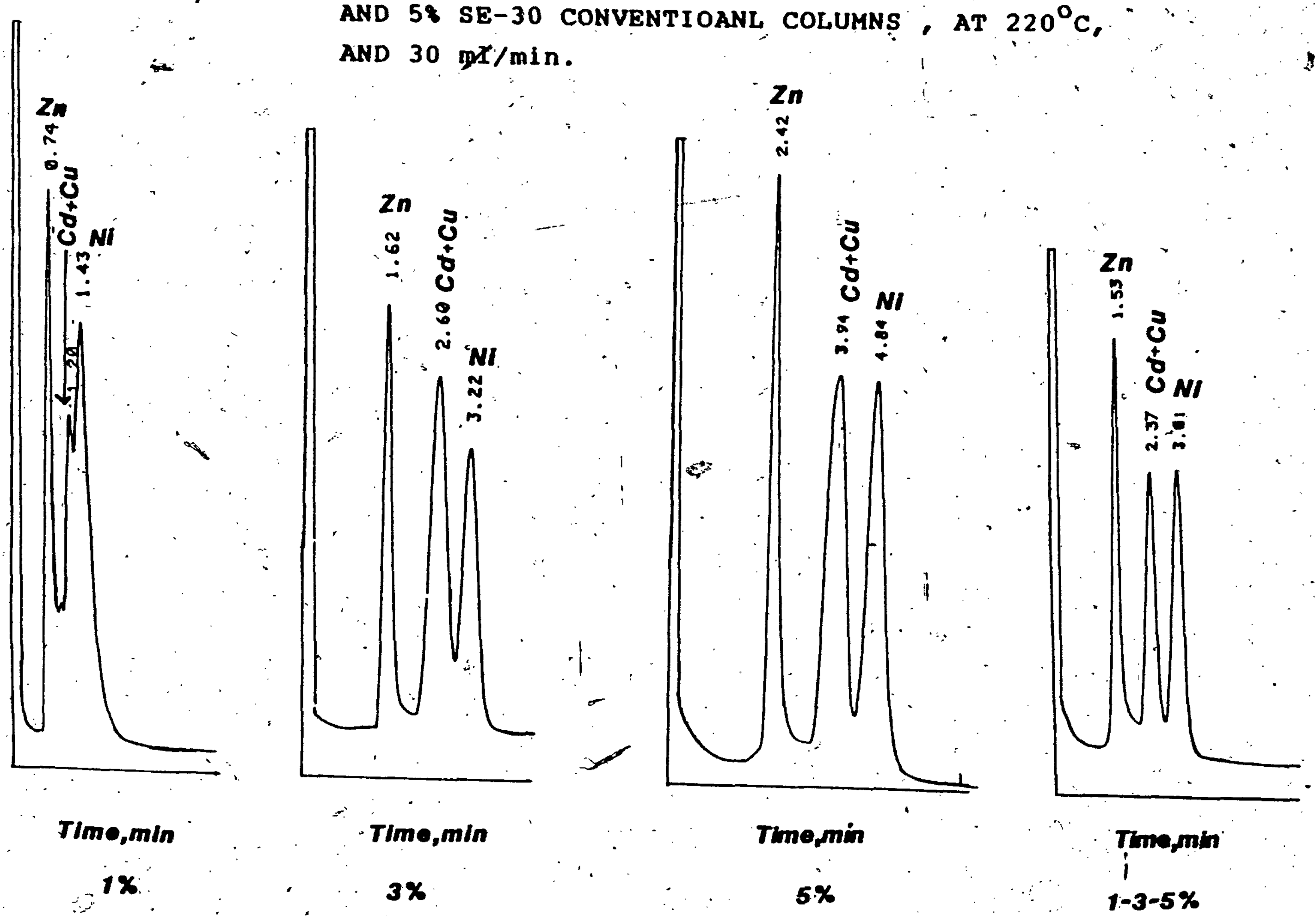
Data is also given for reproducibility (Table 11). Relative standard deviations were all within acceptable levels. This statistical data is typical of what was regularly obtained in the course of experiments with gradient columns.

TABLE 11

COMPARATIVE RESULTS OBTAINED ON A 1-3-5% SE-30 GRADIENT COLUMN, AND ON 1%, 3%, AND 5% SE-30 CONVENTIONAL COLUMNS, AT 220 C

METAL DDC		1%	3%	5%	1-3-5%
RETENTION TIME, min					
Zn	AV±SD	0.7±.01	1.6±.01	2.4±.02	1.5±.01
	RSD%	1.0	1.0	1.0	1.0
Cd		1.2±.02	2.5±.02	3.8±.10	2.4±.01
		1.7	1.0	2.6	0.5
Cu		1.2±.01	2.6±.02	3.9±.01	2.5±.01
		0.5	1.0	0.3	0.5
Ni		1.4±.01	3.2±.03	4.8±.03	3.0±.02
		0.4	1.0	0.5	0.5
RESOLUTION					
Zn-Cd	AV±SD	0.7±.18	1.6±.25	2.1±.02	2.3±.03
	RSD%	21.7	14.8	1.0	1.2
Zn-Ni		1.1±.01	2.5±.40	3.5±.01	4.9±.14
		1.1	15.1	0.4	3.0
Cd-Ni		0.5±0.0	0.9±.10	1.0±.10	1.5±.10
		0.0	10.6	7.3	2.4

FIG. 20 SEPARATION OF Zn, Cd, Cu, AND Ni DDC, OBTAINED ON A 1-3-5% SE-30 GRADIENT COLUMN, AND ON 1%, 3%, AND 5% SE-30 CONVENTIOANL COLUMNS , AT 220°C, AND 30 ml/min.



7.3.2.2 2, 4, 6.....20% SE-30 on CW-HP, 80/100 mesh (60 cm x 2 mm I.D, glass tubing) (Table 12, Fig 21)

Since the use of gradient columns appeared to be beneficial, experiments were performed with a more complex ten stage gradient column. It was compared against a conventional column having 11% loading. Results obtained are shown in Table 12. More theoretical plates were obtained with the gradient column and the Cd/Cu DDC's were separated. Generally speaking, however, resolution was not improved on this complex gradient column compared to simpler, lower loading gradient columns, and analysis times were long. The chromatograms obtained are shown in Fig 21. Broadness of peaks due to the high stationary phase loading was a major disadvantage of this column.

7.3.2.3 CONCLUSIONS BASED ON EXPERIMENTS WITH OV-101 AND SE-30 GRADIENT COLUMNS

1 - It has been demonstrated that gradient columns containing either OV-101 or SE-30 were superior to conventional columns of equivalent loading in terms of resolution and decreased retention times. Also using

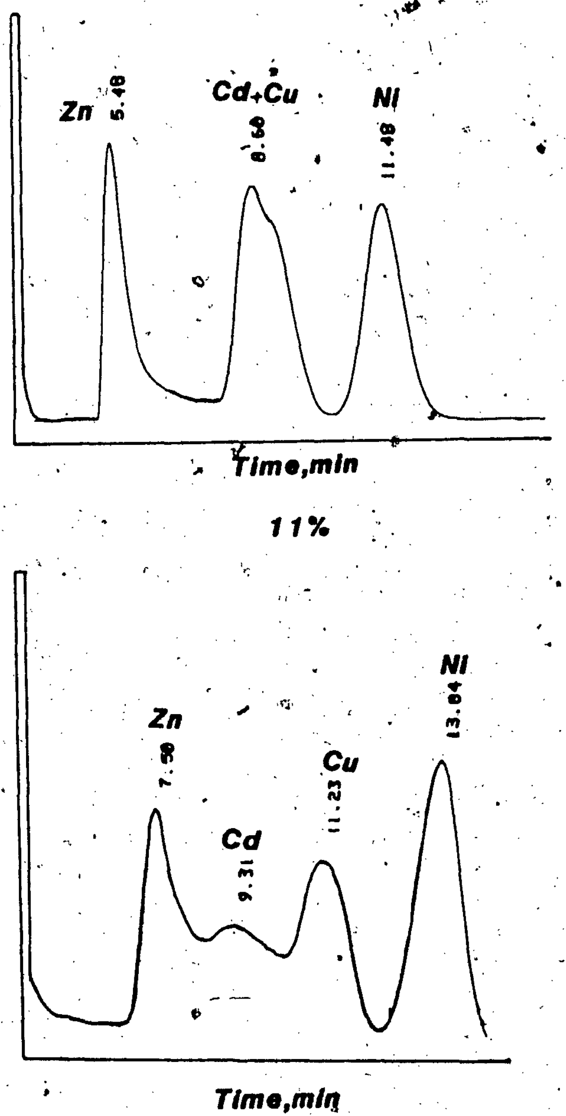
TABLE 12

COMPARATIVE RESULTS OBTAINED ON A 2, 4, 6...20% SE-30 GRADIENT COLUMN AND ON A 11% SE-30 CONVENTIONAL COLUMN AT 220°C

RETENTION DATA				
METAL DDC	11%		(2, 4, 6....20%)	
	N	RT(MIN)	N	RT(MIN)
Zn	595	5.5	744	7.5
Cd	328	8.6	821	9.3
Cu	328	9.0	458	11.2
Ni	652	11.5	949	13.8

RESOLUTION		
Zn-Cd	2.2	1.5
Zn-Cu	2.2	2.4
Zn-Ni	4.4	4.5
Cd-Cu	0.0	1.2
Cd-Ni	1.6	2.9
Cu-Ni	1.6	1.4

FIG. 21 SEPARATION OF Zn, Cd, Cu, AND Ni DDC OBTAINED ON GRADIENT COLUMN 2, 4, ...20% SE-30, AND ON 11% SE-30 CONVENTIONAL CLOLUMN, AT 220°C, AND 30 ml/min.



11%

2-4-6-...20%

temperature programming with the gradient columns led to less bleeding then with conventional columns.

2 - Difficulties in separating DDC of Cd/Cu, Co/Cr, and Pb/Cd pairs were observed with the columns prepared with either OV-101 or SE-30. Only the ten step SE-30 gradient column was successful for the separation of the Cd/Cu pair.

7.3.3 GRADIENT COLUMNS WITH QF-1 STATIONARY PHASE

A series of gradient columns was prepared with QF-1 since this stationary phase had shown promise in conventional columns used for separation of metal DDC.

7.3.3.1 1-2-3% QF-1 ON CW-HP, 80/100 mesh, (60 cm X 2 mm I.D, glass tubing,) (Table 13, Figs 22 & 23)

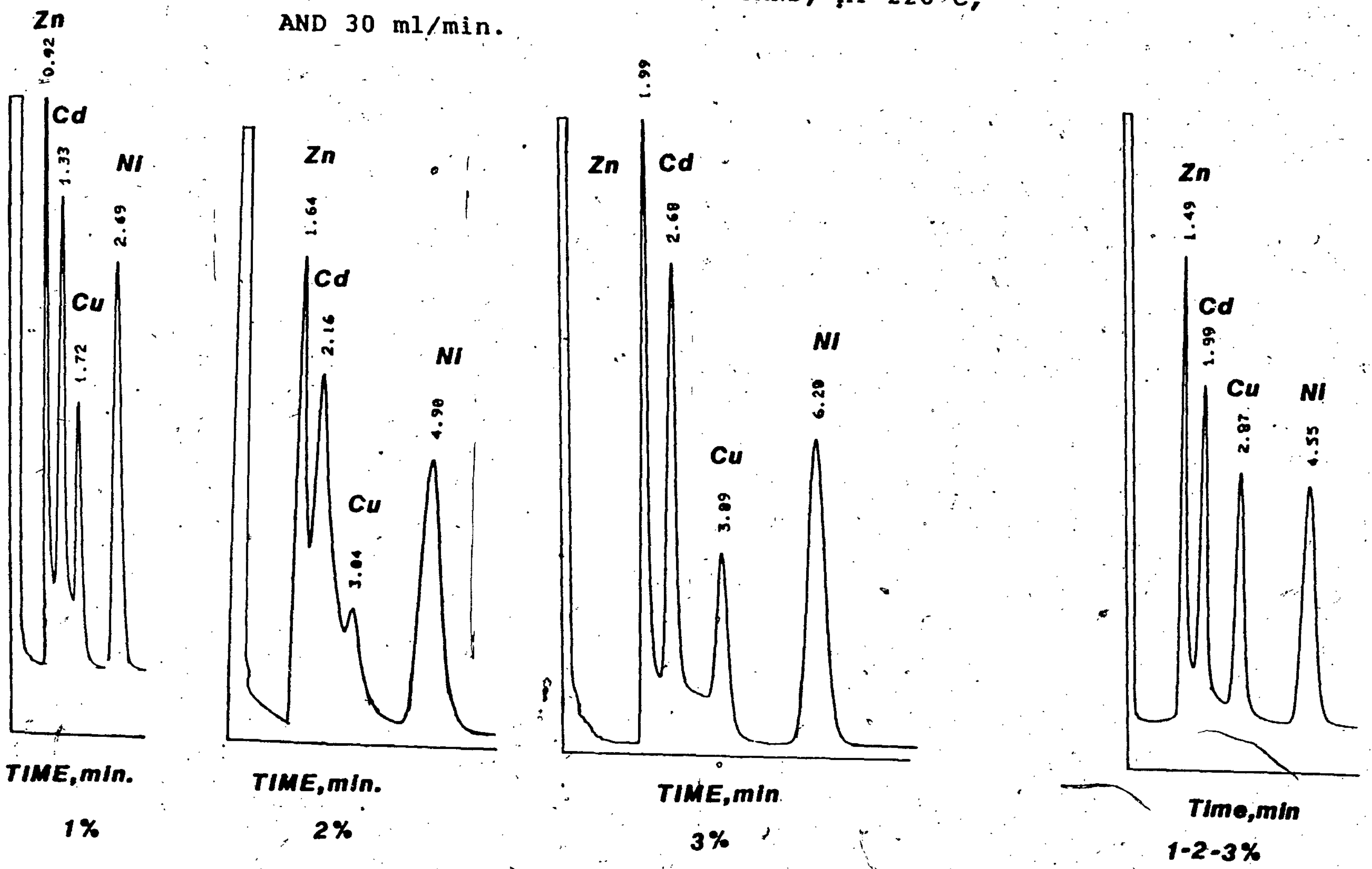
The results obtained with this column and comparisons with 1%, 2% and 3% loading columns are listed in Table 13. Analysis times on the gradient column were reduced compared to the 2% column (equivalent to the mean loading of the gradient column). The resolution obtained for ZN, Cd, Cu, and Ni DDC on the gradient column was superior to that on 1, 2, or 3% columns. The RSD% of the

TABLE 13

COMPARATIVE RESULTS OBTAINED ON A 1-2-3% QF-1 GRADIENT COLUMN, AND ON 1%, 2%, AND 3% QF-1 CONVENTIONAL COLUMNS, AT 220°C

		RETENTION TIME, min			
METAL DDC		1%	2%	3%	1-2-3%
Zn	AV±SD	0.9±.02	1.6±.01	2.0±.01	1.5±.01
	RSD%	2.2	0.7	0.6	0.7
Cd		1.3±.03	2.2±.01	2.9±.04	2.0±.01
		2.4	0.5	1.7	0.3
Cu		1.7±.01	2.2±.01	3.9±.02	2.9±.04
		0.7	0.2	0.5	1.2
Ni		2.7±.02	5.0±.01	6.2±.01	4.6±.01
		1.0	1.4	0.5	0.1
		RESOLUTION			
Zn-Cd	AV±SD	0.9±.04	1.0±.04	1.1±.02	2.0±0.0
	RSD%	3.5	5.4	2.0	0.0
Zn-Cu		2.3±.14	2.4±.11	3.0±.06	5.0±.02
		6.0	5.0	2.0	0.5
Zn-Ni		3.4±.11	4.7±.11	5.3±.25	7.7±.01
		2.5	3.4	4.7	0.2
Cd-Cu		0.9±.06	1.2±.11	1.6±.14	3.9±.02
		6.2	8.7	8.3	1.0
Cd-Ni		2.5±.14	3.6±.03	3.8±.40	7.3±0.0
		5.0	1.0	9.0	0.0
Cu-Ni		2.2±.20	2.1±.20	2.5±.20	4.5±0.0
		8.3	6.7	6.7	0.0

FIG. 22 SEPARATION OF Zn, Cd, Cu, AND Ni DDC OBTAINED ON A 1-2-3% QF-1 GRADIENT COLUMN, AND ON 1%, 2%, AND 3% QF-1 CONVENTIONAL COLUMNS, AT 220°C, AND 30 ml/min.



retention data and resolution obtained for the gradient columns was better than that for any of the conventional columns suggestive of better column stability. In Fig 22, chromatograms are illustrated for the separation of Zn, Cd, Cu, and Ni DDC. It is notable that separation of the Cd/Cu DDC pair was achieved on both gradient and conventional columns. The resolution obtained for Cd/Cu, however, was best on the 1-2-3% gradient column and the analysis time was optimum. In Table 14, the efficiency, number of theoretical plates (N), is compared. The best efficiency was obtained for the gradient column.

No degradation was observed for Pb(II) DDC when it was injected by itself on this gradient column (Fig 23). While this was promising, no separation was obtained for Cd and Pb DDC's when a mixture of Zn, Cd, Pb and Ni DDC was injected.

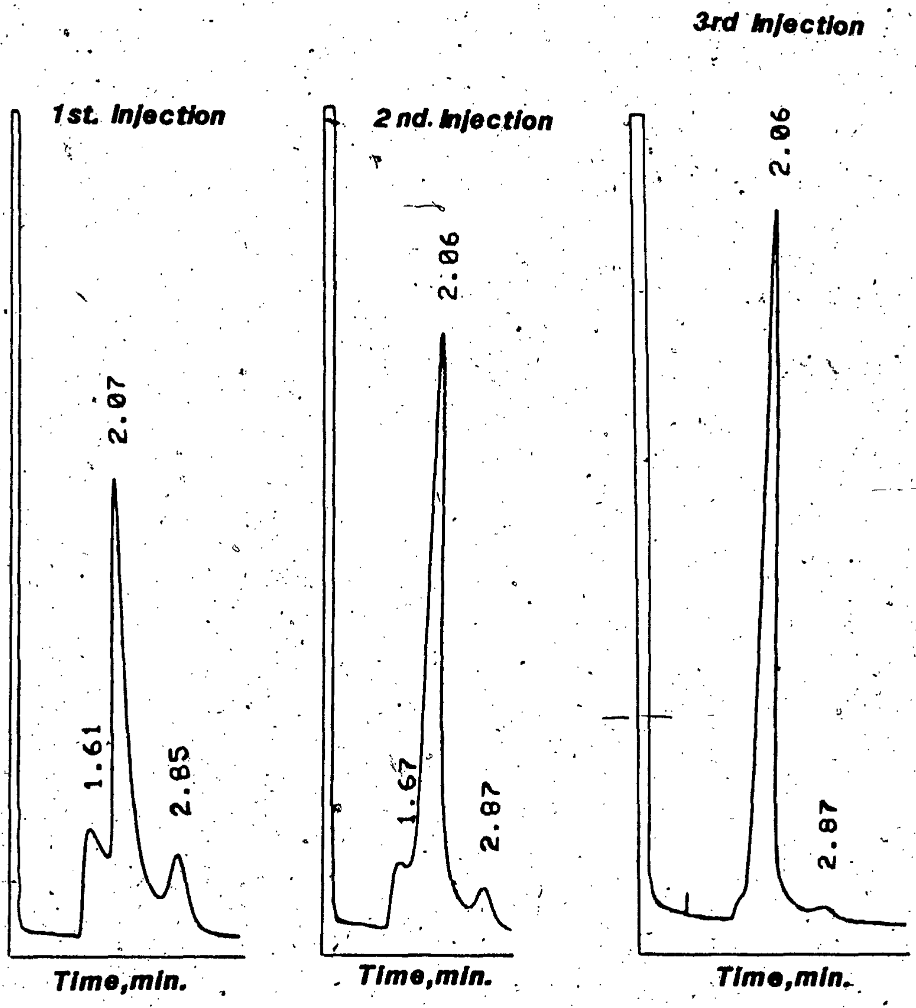
Plots of peak height versus amounts of sample injected on the gradient column were all linear, with correlation coefficients better than 0.99, (Appendix I, Table 46, and Fig 58 to Fig 60).

TABLE 14

COMPARATIVE EFFICIENCY, N, OF CONVENTIONAL 1%, 2%, AND 3% QF-1 COLUMNS AND THE GRADIENT 1-2-3% QF-1 COLUMN AT 220°C

METAL DDC	1%	2%	3%	1-2-3%
Zn	150	103	253	561
Cd	113	117	264	697
Cu	296	595	487	824
Ni	463	476	624	920

FIG. 23 Pb(II) DDC CHROMATOGRAM OBTAINED ON 1-2-3% QF-1 COLUMN, AT 220°C, AND 30 ml/min.



7.3.3.2 1-2-3% QF-1 ON GC-Q, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing) (Table 15, Fig 24)

A column having QF-1, 1-2-3% coated on GC-Q, rather than CW-HP, was tested and results compared with those on the best column reported previously (25) for separating Zn, Cd, Cu, and Ni DDC (Table 15). The resolution achieved was superior to that reported by Carvajal (25) using QF-1/OV-101 on GC- Q (Table 5). It is also important to mention that when the column temperature was increased to 220 C, the performance of the gradient column did not deteriorate, but there was a reduction in the analysis time. Chromatograms obtained in this work, are shown in Fig 24 for analyses at 200°C and 220°C, Note that the analyses times reported by Carvajal (Table 5) were significantly longer.

7.3.3.3 3-2-1%, 2-4-6%, 6-4-2% and 1.0-1.5-2.0-2.5-3.0% QF-1 ON CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing) (Table 16, Figs 25, 26)

Tests with these columns were carried out for a variety of reasons. For example, it was necessary to verify if reversing the gradient, so that the highest

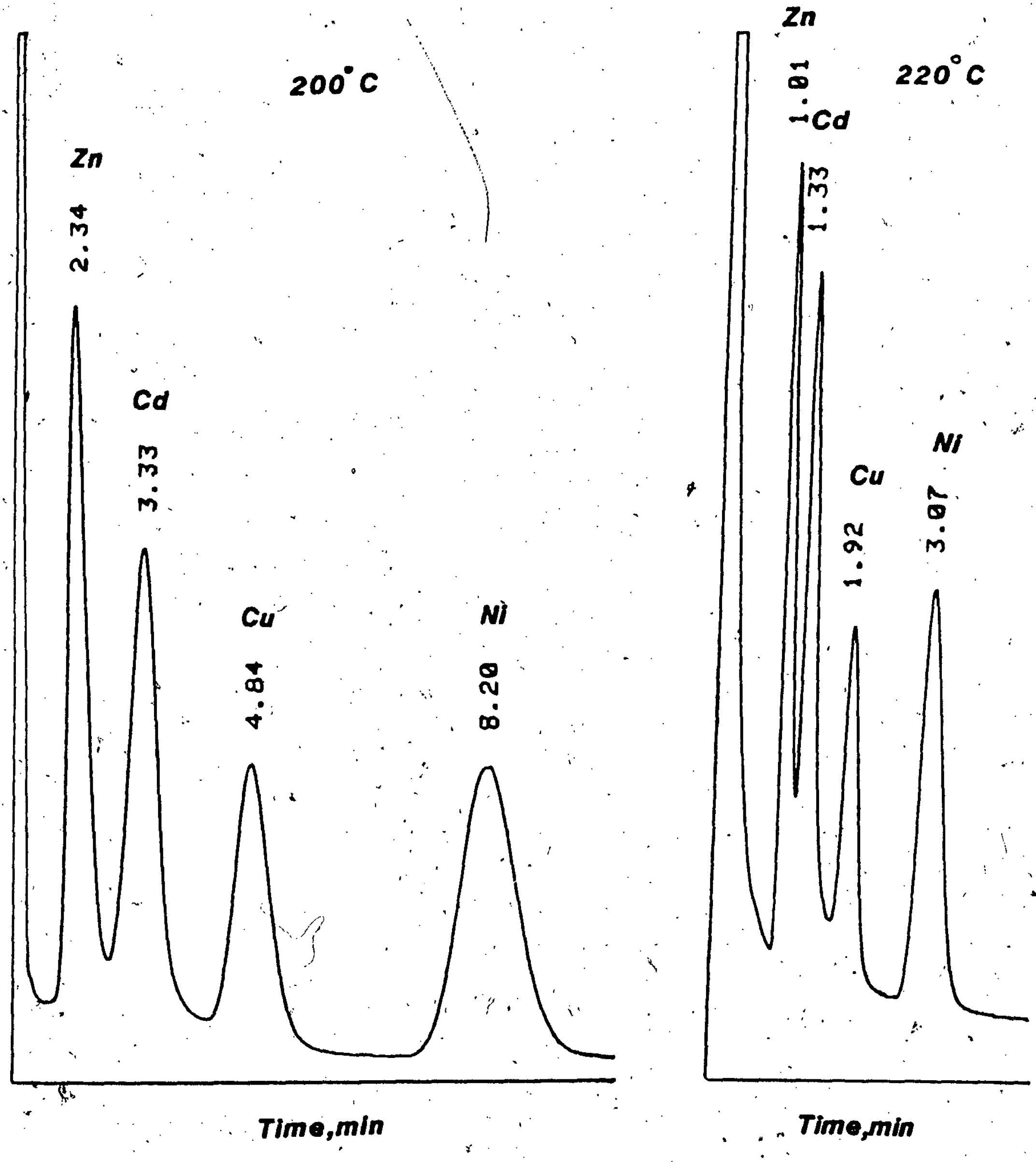
TABLE 15

COMPARATIVE DATA OBTAINED ON 1-2-3% QF-1 ON GC-Q, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING) AT 200^{o}C AND 220^{o}C

RETENTION DATA				
METAL DDC	200^{o}C		220^{o}C	
	N	RT	N	RT
Zn	243	2.3	131	1.0
Cd	279	3.3	300	1.3
Cu	376	4.9	231	1.9
Ni	420	8.2	313	3.1

RESOLUTION		
Zn-Cd	1.4	0.9
Zn-Cu	3.1	2.1
Zn-Ni	5.3	4.0
Cd-Cu	1.7	1.5
Cd-Ni	4.1	3.6
Cu-Ni	2.6	2.0

FIG. 24 SEPARATION OF Zn, Cd, Cu, AND Ni DDC ON 1-2-3% QF-1 ON GC-Q,80/100 MESH,(60 cm x 2mm,GLASS TUBING),AT 200 C AND 220 C ,AND 30ml/min.



concentration segment was at the beginning of the column rather than at the end would improve column capacity as was predicted by Barry (7) and Locke (4). Also, it was necessary to achieve separation of the Cd/Pb DDC pair. Listed in Table 16 are the data obtained by injecting the mixture of Zn, Cd, Pb, and Ni DDC on these columns.

Using the 1.0-1.5-2.0-2.5-3.0 % QF-1 gradient column the separation of Cd and Pb was achieved Fig 25. This column also gave better resolution for the Zn/Cd, Cd/Ni and Zn/Ni DDC pairs than had been obtained with other columns under isothermal conditions Table 16. Plots of peak height versus sample size were linear with correlation coefficients better than 0.99, (Appendix I, Table 47, and Fig 61 to Fig 63).

There was no advantage observed in reversing the gradient direction with QF-1 stationary phase at the 1-2-3% level. The resolution was similar in both directions. Retention times were slightly greater using the 3-2-1% QF-1 gradient. With the higher loading gradient columns, 2-4-6% and 6-4-2%, a difference in sample capacity in favor of the 6-4-2% configuration, using $Zn(DDC)_2$ and $Ni(DDC)_2$ as test samples, was observed similar to that reported by Barry (7). Capacity tests were performed by injecting 1.0 ul samples of both at 220°C. Concentration of samples injected ranged

TABLE 16

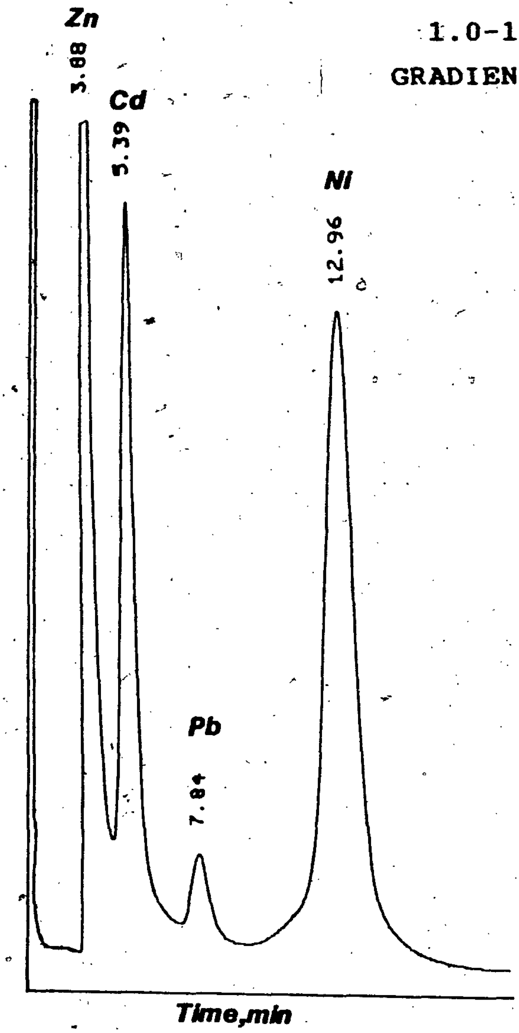
ANALYSIS OF $M(DDC)_n$ ON VARIOUS QF-1 GRADIENT COLUMNS

AT 200°C, N_2 Flow rate: 25 ml/min

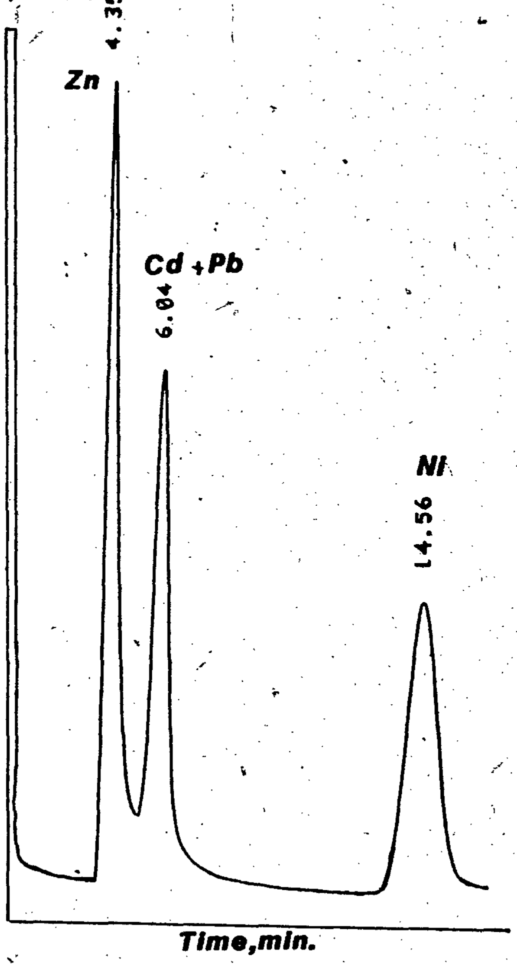
	RETENTION TIME (mins)			
GRADIENT COLUMN	Zn(II)	Cd(II)	Pb(II)	Ni(II)
1-2-3%	4.1	5.8	0.0	13.7
3-2-1%	4.5	6.7	0.0	15.3
2-4-6%	6.7	9.5	0.0	23.0
6-4-2%	7.8	10.8	0.0	26.7
1.0-1.5-2.0-2.5-3.0	3.9	6.5	8.7	13.4

	RESOLUTION				
	Zn-Cd	Cd-Ni	Zn-Ni	Pb-Ni	Pb-Cd
1-2-3%	1.4	4.5	6.1	0.0	0.0
3-2-1%	1.4	4.3	6.1	0.0	0.0
2-4-6%	1.2	3.6	4.9	0.0	0.0
6-4-2%	1.1	3.7	4.7	0.0	0.0
1.0-1.5-2.0-2.5-3.0	1.6	5.2	6.2	2.9	1.9

FIG. 25 SEPARATION OF Zn, Cd, Pb, AND Ni DDC OBTAINED ON 1.0-1.5-2.0-3.0% QF-1, AND ON 1-2-3% QF-1 GRADIENT COLUMNS, AT 220°C, AND 25 ml/min.



1.0-1.5-2.0-2.5-3.0% QF-1



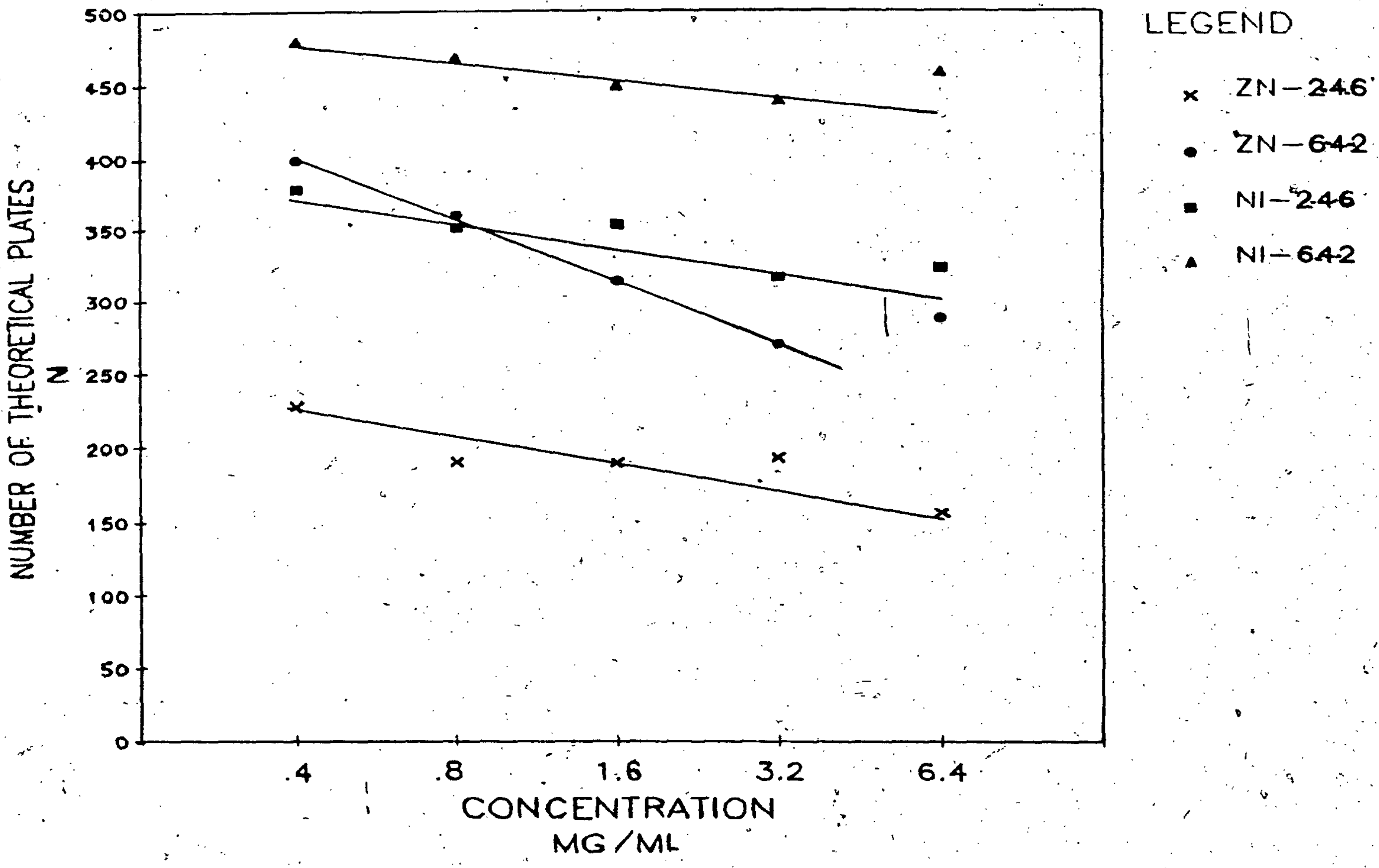
1-2-3% QF-1

between 0.4 to 6.4 mg/ml. Plots of the number of theoretical plates versus the concentration of the sample injected on both 2-4-6% and 6-4-2% columns are shown in Fig 26. From these plots it was concluded that Zn and Ni behaved similarly in that more theoretical plates were obtained using the 6-4-2% QF-1 column configuration. This meant that the 6-4-2% gradient column had a greater capacity. Further tests with larger sample sizes were inconclusive, and therefore, are not reported here.

7.3.3.4 CONCLUSIONS BASED ON TESTS WITH QF-1 STATIONARY PHASE

1. At isothermal column temperature the gradient 1-2-3% QF-1 column showed behavior superior to that of 1, 2 or 3% QF-1 conventional columns.
2. The 1.0-1.5-2.0-2.5-3.0% QF-1 column separated Cd and Pb DDC at isothermal column temperature.
3. The gradient columns containing 1-2-3% and 1.0-1.5-2.0-2.5-3.0% QF-1 were the two best columns for minimizing analysis time and optimizing resolution of metal DDC.
4. In tests with 2-4-6% and 6-4-2% QF-1 gradient columns, increased column capacity was observed for the high to low ,6-4-2% configuration.

Fig. 26 Determination of Column Capacity of 2-4-6% vs.6-4-2% QF1 Columns at 220 deg.C and 30 ml/min.



7.3.4 GRADIENT COLUMNS WITH OV-210 STATIONARY PHASE ON CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing) (Table 17, Figs 27 & 28)

This stationary phase has polarity similar to that of QF-1. It is, however, a higher molecular weight polymer. Its use has not been reported previously for the separation of metal DDC chelates.

Table (17) contains the data obtained for the metal DDC mixture of Zn(II), Cd(II), Cu(II) and Ni(II) on conventional 1, 2, and 3% columns and on the 1-2-3% gradient stationary phase column . Chromatograms are shown in Fig 27. The resolution achieved with the 1-2-3% gradient column was superior by about 30% to that on any of the three conventional columns. Zn and Cd DDC, however, were not completely resolved even by the gradient column (R = 0.9). The total analysis time for separating the four metal DDC on the gradient column was less than on the column containing 2% loading, i.e. the average loading on the gradient column.

Injection of $Pb(DDC)_2$ by itself on the gradient column gave a degradation peak, but when injections were repeated several times, the degradation peak disappeared leaving only one peak for this chelate (Fig 28).

TABLE 17

COMPARATIVE DATA OBTAINED ON A 1-2-3% OV-210 GRADIENT COLUMN, AND ON 1%, 2%, AND 3% OV-210 CONVENTIONAL COLUMNS AT 220°C

RETENTION TIMES (mins)

METAL DDC	1%	2%	3%	1-2-3%
Zn AV±SD	0.7±.01	1.8±.00	2.3±.01	1.4±.01
RSD%	1.0	0.0	0.3	1.0
Cd	1.0±.01	2.4±.01	3.0±.01	1.8±.01
	1.0	0.40	0.2	0.3
Cu	1.3±.01	3.4±.01	4.5±.01	2.6±.02
	1.0	0.2	0.3	1.0
Ni	2.1±.01	5.5±.02	7.2±.02	4.1±.02
	0.5	0.3	0.2	0.5
RESOLUTION				
Zn-Cd AV±SD	0.7±.12	0.7±.01	0.6±.04	0.9±.01
RSD%	15.0	1.0	6.4	1.0
Zn-Cu	1.0±.22	1.8±.17	2.3±.15	2.4±.03
	12.3	8.4	6.3	1.2
Zn-Ni	2.7±.50	3.2±.11	4.5±.08	4.9±.01
	13.1	3.3	1.7	0.3
Cd-Cu	0.0±.11	1.0±.09	1.1±.06	1.8±.04
	12.0	7.7	5.6	2.6
Cd-Ni	2.5±.40	2.1±.07	2.9±.07	3.7±.04
	13.5	2.8	2.4	0.0
Cu-Ni	2.6±.30	1.3±.14	2.2±.06	2.6±.02
	13.2	8.7	2.5	1.0

FIG. 27 SEPARATION OF Zn, Cd, Cu, AND Ni DDC OBTAINED ON A 1-2-3% OV-210 GRADIENT COLUMN, ON 1%, 2%, AND 3% CONVENTIONAL COLUMNS, AT 220°C, AND 30 ml/min.

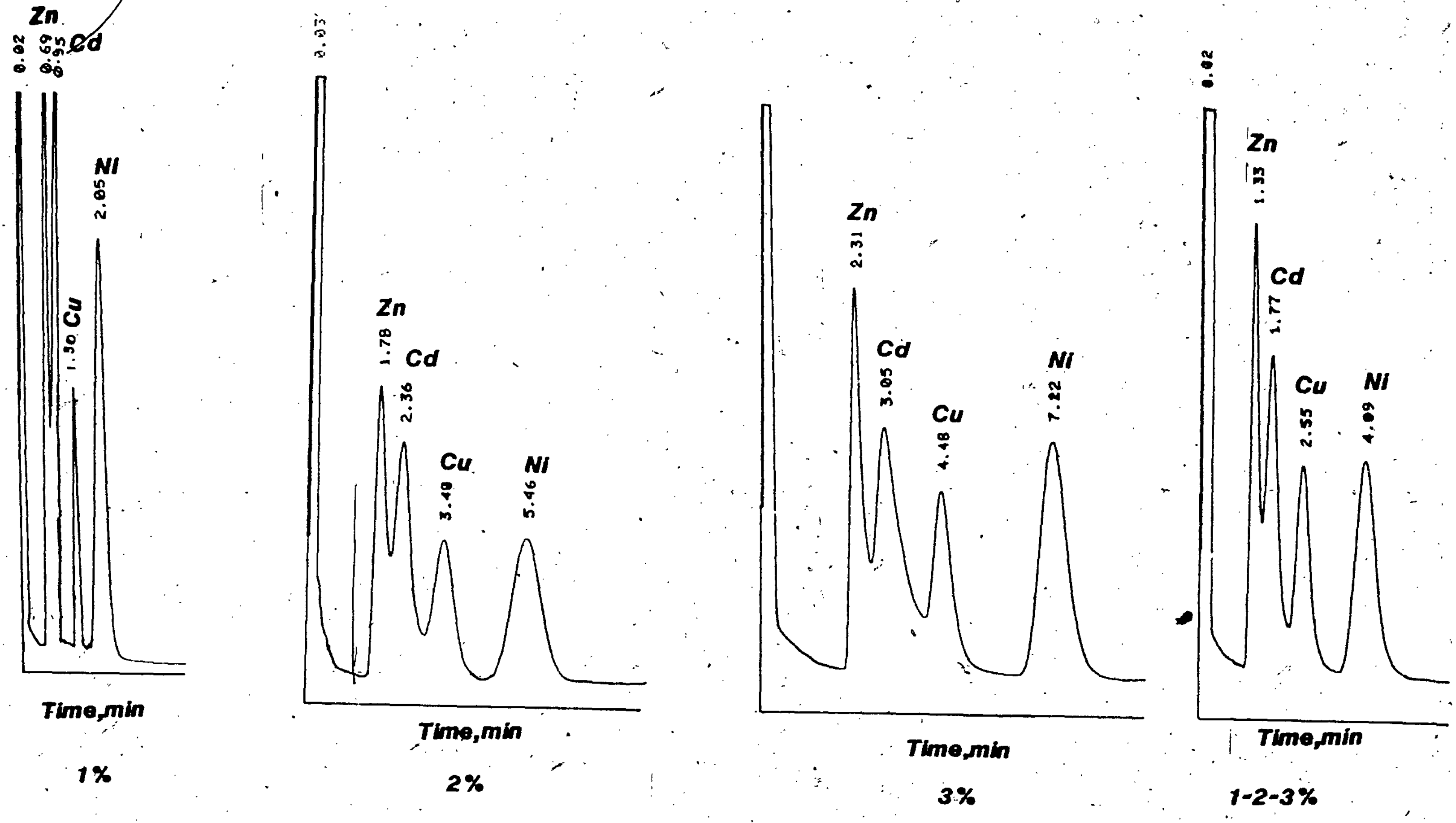
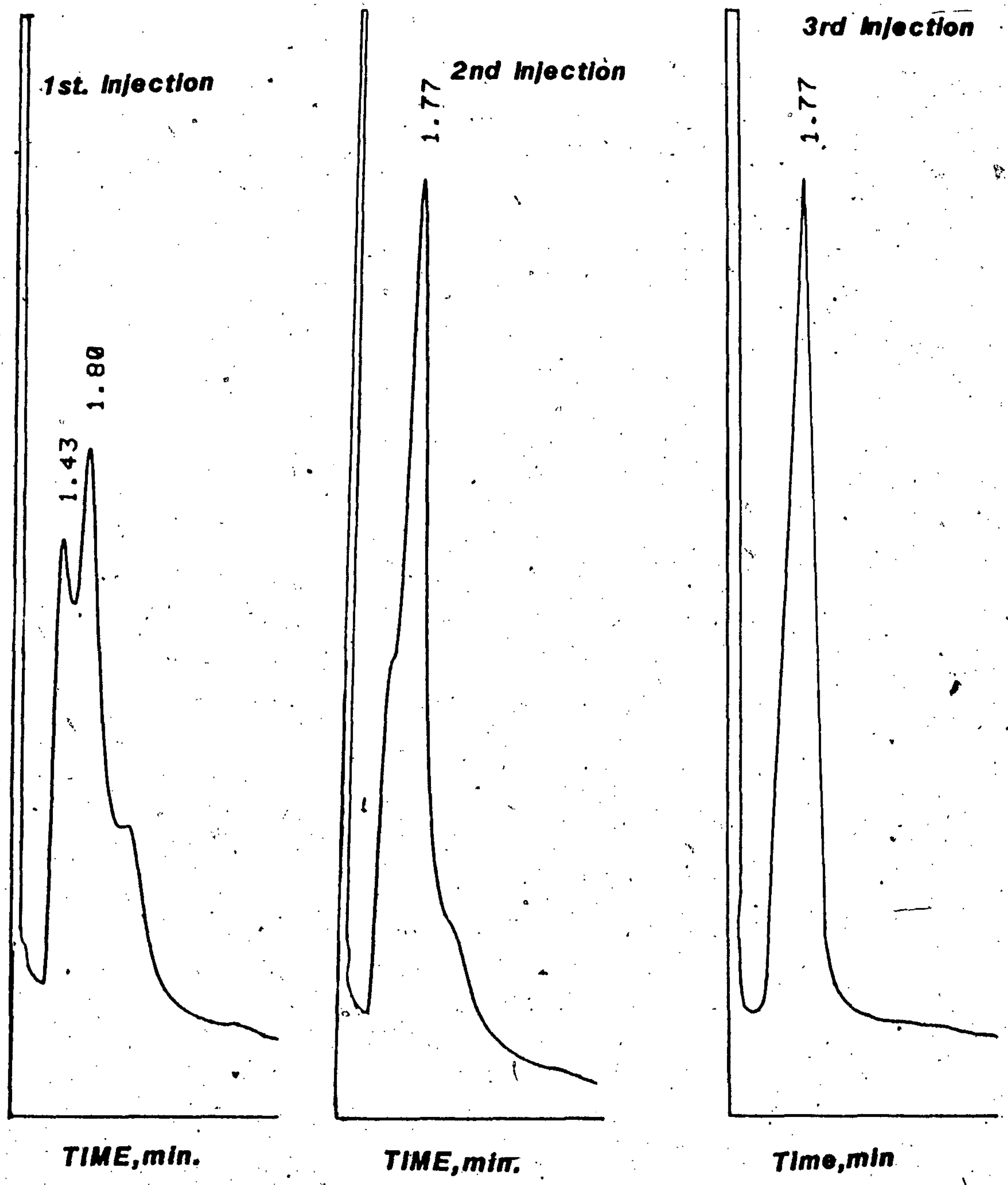


FIG. 28 Pb(DDC)$_2$ DEGRADATION ON 1-2-3% OV-210 GRADIENT COLUMN, ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 220°C AND 30 ml/min.



Plots of peak height versus concentration for Zn(II), Cd(II), Pb(II), Cu(II), Ni(II) and Co(III) DDC were all linear with correlation coefficients better than 0.90 (Appendix I, Table 48 and Fig 64 to Fig 66).

7.3.5 GRADIENT COLUMNS WITH OV-225 ON CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing) (Tables 18 & 19, Figs 29 & 30)

This stationary phase was chosen for evaluation because of its general reported chromatographic selectivity for organic compounds having carbonyl or thiocarbonyl groups (27).

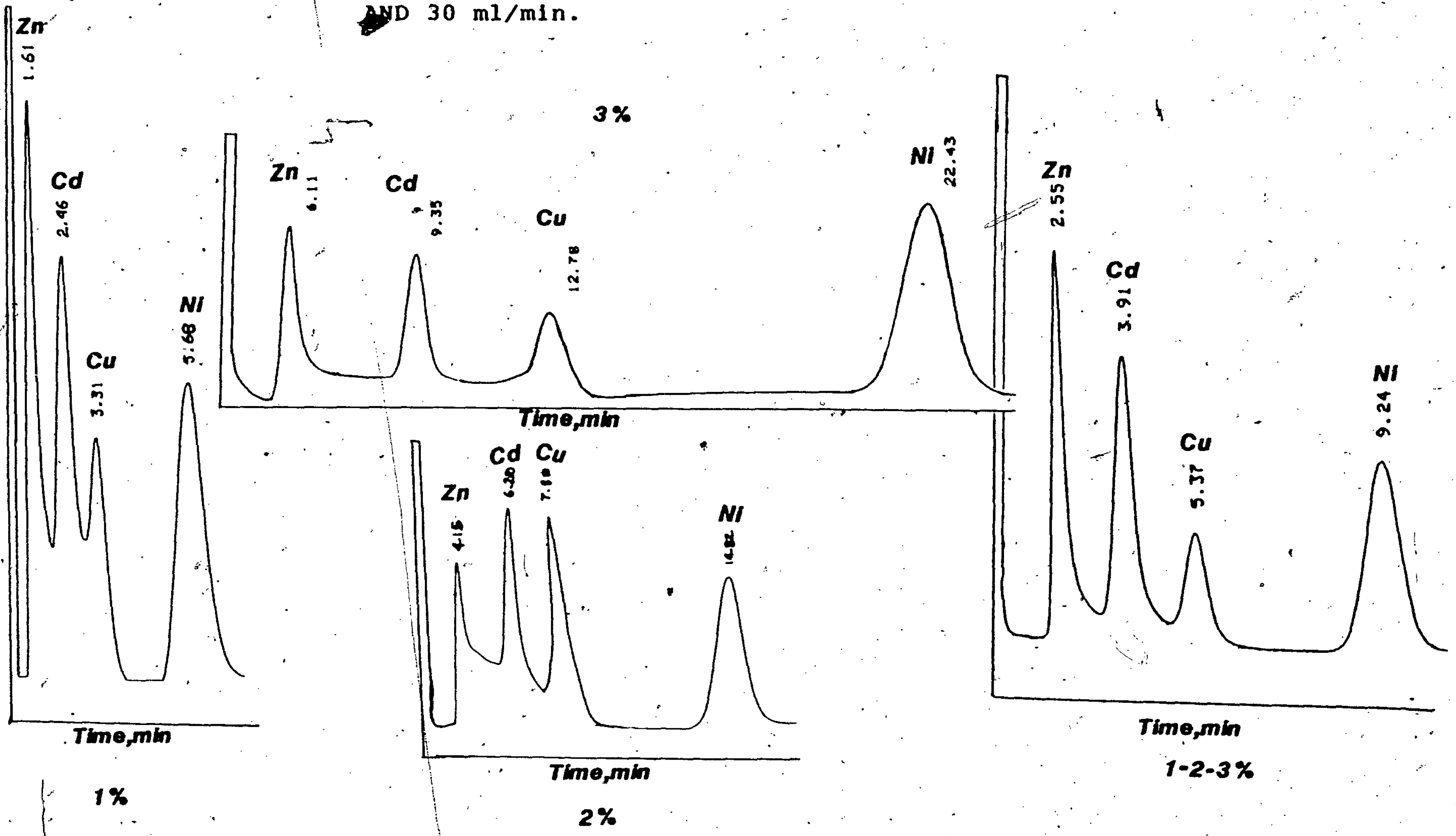
The data obtained upon injection of a mixture of Zn(II), Cd(II), Cu(II) and Ni(II) DDC onto the 1, 2, 3% and 1-2-3% loaded columns are listed in Table 18. Chromatograms appear in Fig 29. The analysis time on the gradient column was less than on the 2% column. Resolution with the gradient column was greater than with the 1% and 2% columns, but less than with 3%. However, the analysis time on the latter was 2.4 times that on the gradient column. Peaks were very broad and spread far apart.

TABLE 18

COMPARATIVE DATA OBTAINED FOR A Zn, Cd, Cu, AND Ni DDC MIXTURE ON A 1-2-3% OV-225 GRADIENT COLUMN, AND ON 1%, 2%, AND 3% CONVENTIONAL COLUMNS AT 240°C

METAL DDC	RETENTION TIMES (min) 1%	2%	3%	1-2-3%
Zn	1.6	4.1	6.1	2.6
Cd	2.5	6.3	9.4	3.9
Cu	3.3	7.8	12.8	5.4
Ni	5.7	14.8	22.4	9.2
		RESOLUTION		
Zn-Cd	1.0	2.0	3.6	2.7
Zn-Cu	2.3	2.7	3.6	4.9
Zn-Ni	3.6	6.5	10.2	8.3
Cd-Cu	1.0	1.3	2.9	2.1
Cd-Ni	2.6	5.8	7.7	5.9
Cu-Ni	2.1	4.1	5.1	4.2

FIG. 29 SEPARATION OF Zn, Cd, Cu, AND Ni DDC OBTAINED ON A 1-2-3% OV-225 GRADIENT COLUMN, AND ON 1%, 2%, AND 3% OV-225 CONVENTIONAL COLUMNS, AT 240°C, AND 30 ml/min.



Reversing the gradient to give a 3-2-1% configuration had a pronounced detrimental effect. The peaks were even further spread out and retention times greatly increased, (Table 19, Fig 30). This effect could be related to the fact that volume flow rate through a column is normally greater at the outlet than at the inlet because of the pressure drop across the column. The lower flow rate at the inlet when combined with a higher loading in the front end, will likely manifest itself as greater sample band broadening leading to poorer resolution, longer retention times, and lower column efficiency. This was also reported by Barry (7). This is a major disadvatange of high front end loaded columns.

It was concluded that OV-225 did indeed have a stronger affinity for metal DDC than did QF-1 or OV-210, but this was not advantageous in terms of $M(DDC)_n$ separations that could be obtained.

Plots of peak height response versus sample concentration for Zn(II), Cd(II), Pb(II), Cu(II), and Ni(II) DDC were all linear with correlation coefficients better than 0.99 (Appendix I, Table 49, Figs 67 to Fig 69).

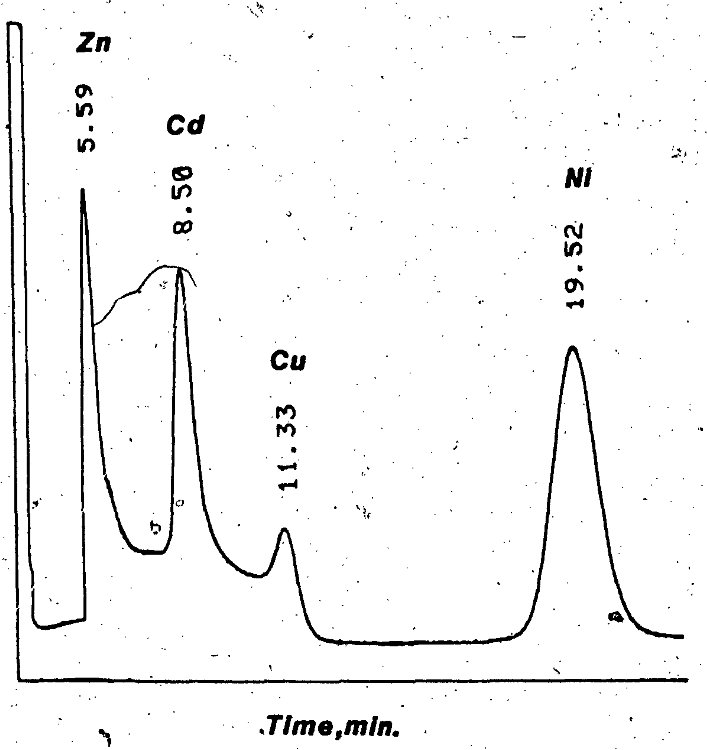
TABLE 19

RESULTS OBTAINED ON 3-2-1% OV-225 FOR THE MIXTURE OF Zn, Cd, Cu, AND Ni DDC ON CW-HP, 80/100 MESH, (60 cm x 2 mm, I.D, GLASS TUBING) AT 240°C

METAL DDC	RETENTION DATA RT (min)	N
Zn	5.6	500
Cd	8.5	803
Cu	11.3	802
Ni	19.5	902

	RESOLUTION
Zn-Cd	2.7
Zn-Cu	4.4
Zn-Ni	7.7
Cd-Cu	2.0
Cd-Ni	5.8
Cu-Ni	3.9

FIG. 30 SEPARATION OF Zn, Cd, Cu, AND Ni DDC OBTAINED ON 3-2-1% OV-225 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 240°C, AND 30 ml/min.



7.3.6 TWO STATIONARY PHASE-GRADIENT COLUMNS

The combination of two phases results in a mixed stationary phase which has intermediate properties between those of the two pure phases. This should enable more precise tailoring of columns to achieve resolution of difficult to separate mixtures. Accordingly, experiments were carried out with mixed phases to attempt improvement in resolution of metal DDC over that which had been obtained up to this point.

7.3.6.1 GRADIENT COLUMNS WITH 1-2-3% WITH EACH OF OV-101-QF-1 STATIONARY PHASES ON CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing) (Tables 20, 23, Figs 31, 34)

The retention time, resolution and efficiency data obtained, at 200 C, on the 1-2-3% gradient column and on the 1, 2, 3% each conventional columns for a mixture of Zn, Cd, Cu, and NI DDC and the number of theoretical plates calculated for each column are shown in Table 20. The gradient column was found to give the best resolution for the four metal DDC mixture (Fig 31). Efficiency in terms of theoretical plates was also best for the gradient column. The analysis time with the gradient column was less than that obtained on the 2% column .

TABLE 20

COMPARATIVE DATA OBTAINED FOR A Zn, Cd, Cu, AND Ni DDC MIXTURE ON A 1-2-3% EACH OF OV-101-QF-1 GRADIENT COLUMN, AND ON 1%, 2%, AND 3% EACH OF OV-101-QF-1 CONVENTIONAL COLUMNS AT 200°C

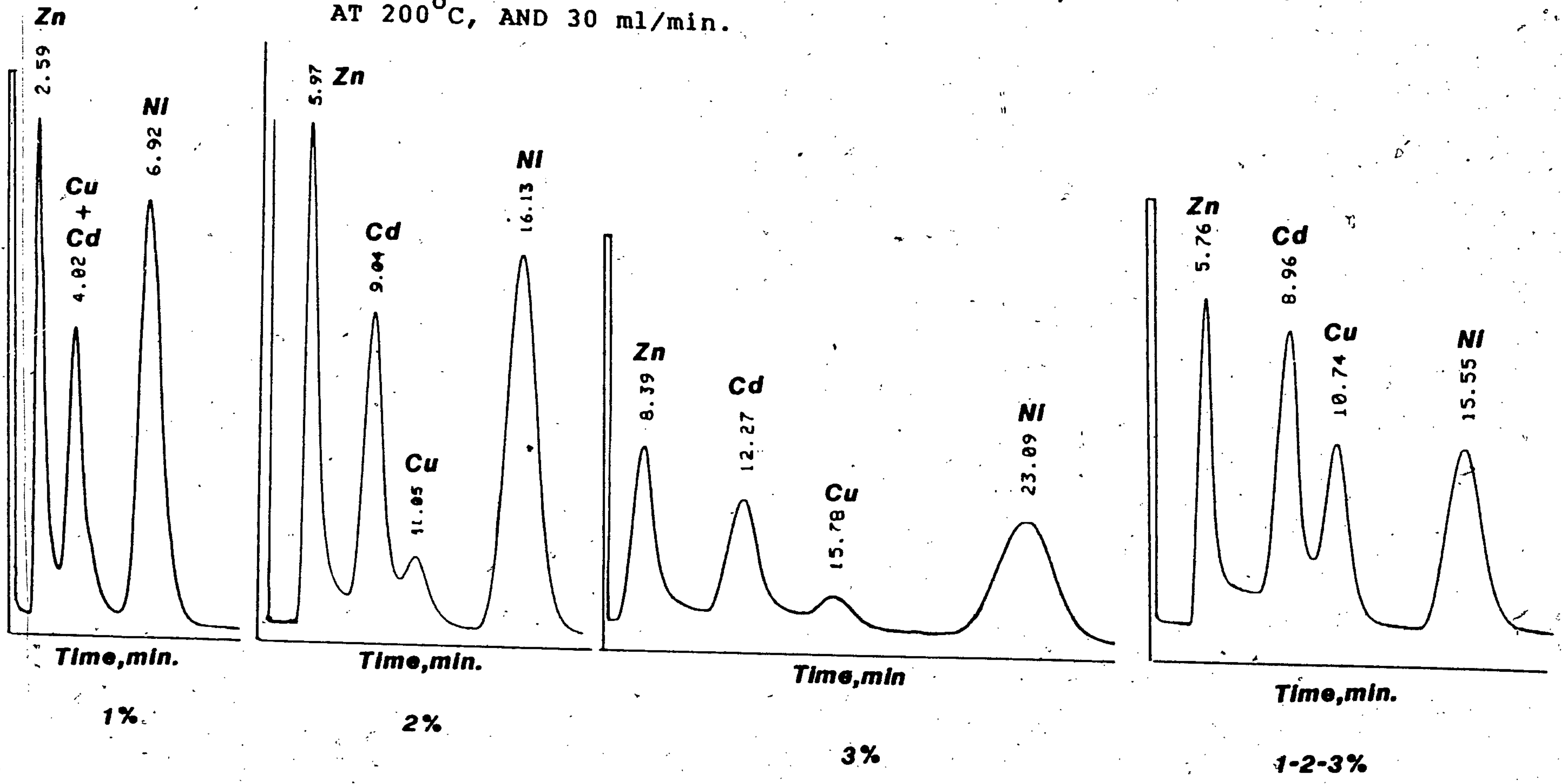
RETENTION DATA

METAL DDC	1%		2%		3%		1-2-3%	
	RT	N	RT	N	RT	N	RT	N
Zn	2.6	107	6.0	291	8.4	348	5.7	829
Cd	4.0	100	9.0	270	12.3	498	8.9	502
Cu	4.7	100	11.0	267	15.8	692	10.7	721
Ni	6.9	158	16.13	406	23.1	507	15.5	799

RESOLUTION

	1%	2%	3%	1-2-3%
Zn-Cd	1.1	1.7	1.9	2.5
Zn-Cu	1.1	2.5	3.1	4.1
Zn-Ni	2.7	4.4	5.0	6.0
Cd-Cu	0.0	0.8	1.4	1.5
Cd-Ni	1.5	2.6	3.4	3.9
Cu-Ni	1.5	1.7	2.2	2.8

FIG.31 SEPARATION OF Zn, Cd, Cu, AND Ni DDC ON A 1-2-3% EACH OF OV-101-QF-1 GRADIENT COLUMN, AND ON 1%, 2%, AND 3% EACH OF OV-101-QF-1 CONVENTIONAL COLUMNS, AT 200°C, AND 30 ml/min.



At higher temperatures, e.g 220°C, the Cd/Cu DDC pair did not separate well (Table 21, Fig 32).

Plots of peak height response versus sample concentration for Zn(II), Cd(II), Pb(II), Cu(II), Ni(II), and Co(III) DDC were all linear and with correlation coefficient better than 0.99 (Appendix I, Table 50, and Fig 70 to Fig 72).

A run was carried out with the stationary phase gradient reversed, i.e. using 3-2-1% each of OV-101-QF-1. The results are shown in Table 22, and Fig 33. There was no improvement in efficiency and hence no evidence for improved capacity. Retention times, however, were increased and all peaks were much broader.

In an attempt to improve further the separation of the Cd/Cu DDC pair, a 1.5-2.5-3.5% each of OV-101-QF-1 and a 3.5-2.5-1.5% each of OV-101-QF-1 columns were prepared and tested. Results are given in Table 23 and Fig 34 for these columns and for comparison for a 2.5% each of OV-101-QF-1 column, which was the equivalent of the average gradient loading. The 2.5% loaded column was also referred to in the literature (25), as one of the best columns for separation of Zn, Cd, Cu, Ni, and Co DDC. Results were no better with the 1.5-2.5-3.5% gradient column than with the 2.5% column. However, improved resolution and efficiency were obtained

TABLE 21

RESULTS OBTAINED FOR A Zn, Cd, Cu, AND Ni DDC MIXTURE ON A 1-2-3% EACH OF OV-101-QF-1 GRADIENT COLUMN AT 220°C

METAL CHELATE	RETENTION DATA RT(min)	N
Zn	2.5	202
Cd	3.7	339
Cu	4.4	313
Ni	6.2	607

	RESOLUTION
Zn-Cd	1.6
Zn-Cu	2.3
Zn-Ni	4.3
Cd-Cu	0.8
Cd-Ni	2.8
Cu-Ni	1.7

FIG. 32 SEPARATION OF Zn, Cd, Cu, AND Ni DDC ON 1-2-3% EACH OF OV-101-QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING) AT 220°C, AND 30ml/min.

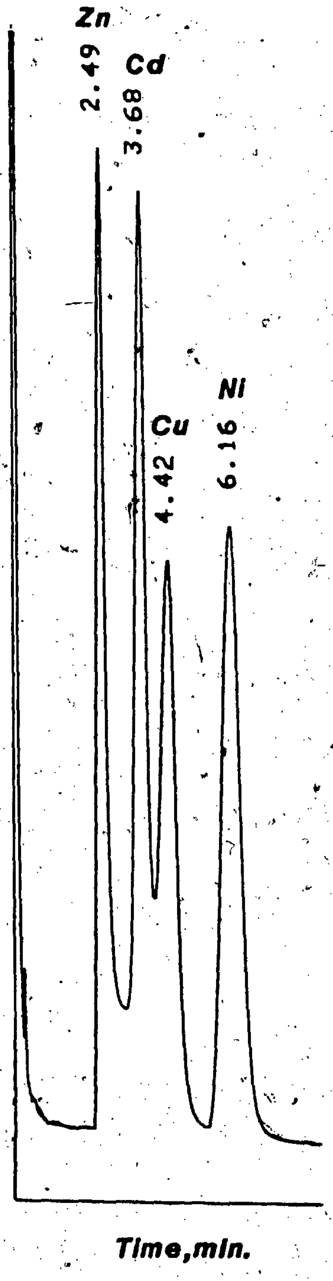


TABLE 22

RESULTS OBTAINED FOR A Zn, Cd, Cu, AND Ni DDC MIXTURE ON 3-2-1% EACH OF OV-101-QF-1 GRADIENT COLUMN AT 200°C

METAL DDC	RETENTION DATA N	RT(min)
Zn	652	10.9
Cd	903	15.8
Cu	941	19.9
Ni	732	28.4

	RESOLUTION
Zn-Cd	2.6
Zn-Cu	4.2
Zn-Ni	6.0
Cd-Cu	1.8
Cd-Ni	4.0
Cu-Ni	2.5

FIG. 33 SEPARATION OF Zn, Cd, Cu, AND Ni DDC ON 3-2-1% EACH OF OV-101-QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING) AT 200°C, AND 30ml/min.

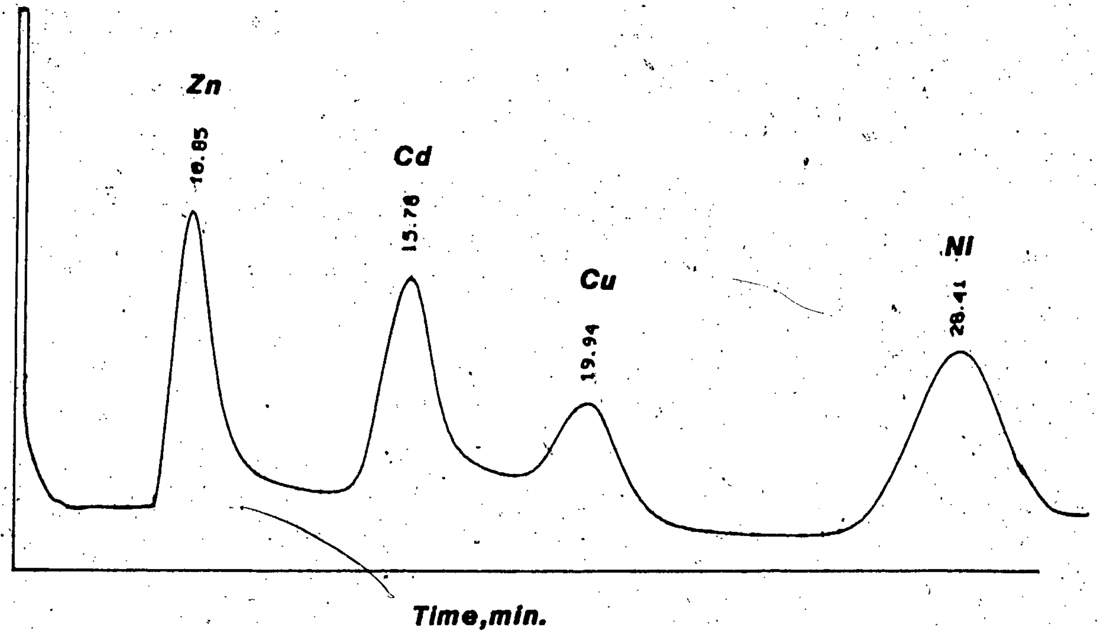


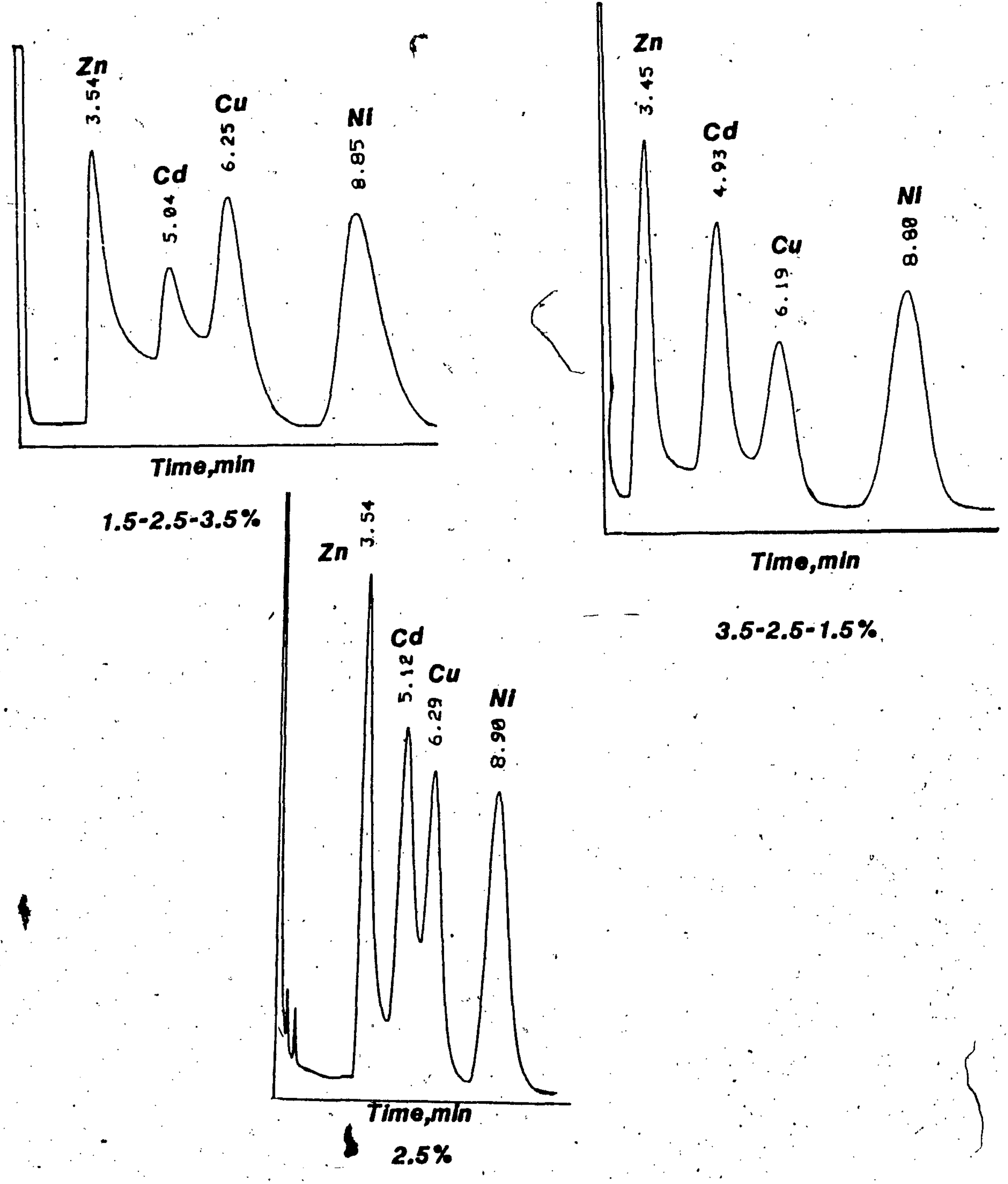
TABLE 23

COMPARATIVE RESULTS OBTAINED FOR THE SEPARATION OF Zn, Cd, Cu, AND Ni DDC ON 1.5-2.5-3.5% AND ON 3.5-2.5-1.5% EACH OF OV-101-QF-1 GRADIENT COLUMNS, AND ON 2.5% EACH OF OV-101-QF-1 CONVENTIONAL COLUMN AT 220°C .

RETENTION DATA						
METAL DDC	1.5-2.5-3.5%		2.5%		3.5-2.5-1.5%	
	N	RT	N	RT	N	RT
Zn	517	3.4	232	3.4	529	3.5
Cd	384	4.9	392	4.9	790	4.9
Cu	164	6.1	341	6.1	563	6.2
Ni	419	8.7	541	8.7	733	8.8

RESOLUTION			
Zn-Cd	1.9	1.6	2.3
Zn-Cu	2.1	2.3	6.4
Zn-Ni	4.6	4.4	3.4
Cd-Cu	0.8	0.9	1.5
Cd-Ni	2.8	3.0	1.9
Cu-Ni	1.5	1.9	2.2

FIG. 34 SEPARATION OF Zn, Cd, Cu, AND Ni DDC ON 1.5-2.5-3.5% AND 3.5-2.5-1.5% EACH OF OV-101-QF-1 ON CW-HP GRADIENT COLUMNS, AND ON A 2.5% EACH OF OV-101-QF-1 CONVENTIONAL COLUMN AT 220°C, AND 30ml/min.



1.5-2.5-3.5%

3.5-2.5-1.5%

2.5%

with the 3.5-2.5-1.5% each of OV-101-QF-1 configuration, a result not expected based on previous tests with high inlet end gradient columns.

7.3.6.2 GRADIENT COLUMNS WITH 1-2-3% EACH OF OV-225-QF-1 STATIONARY PHASE, ON CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing), (Table 24, Fig 35)

Tests with OV-225 stationary phase had demonstrated the very strong affinity of this phase for $M(DDC)_n$ studied (Section 7.3.5). The affinity of QF-1 for these chelates is much lower. Consequently, it was be predicted that a combination of these two liquid phases might give a column which could be tailor made to yield excellent selectivity for particular metal DDC mixtures.

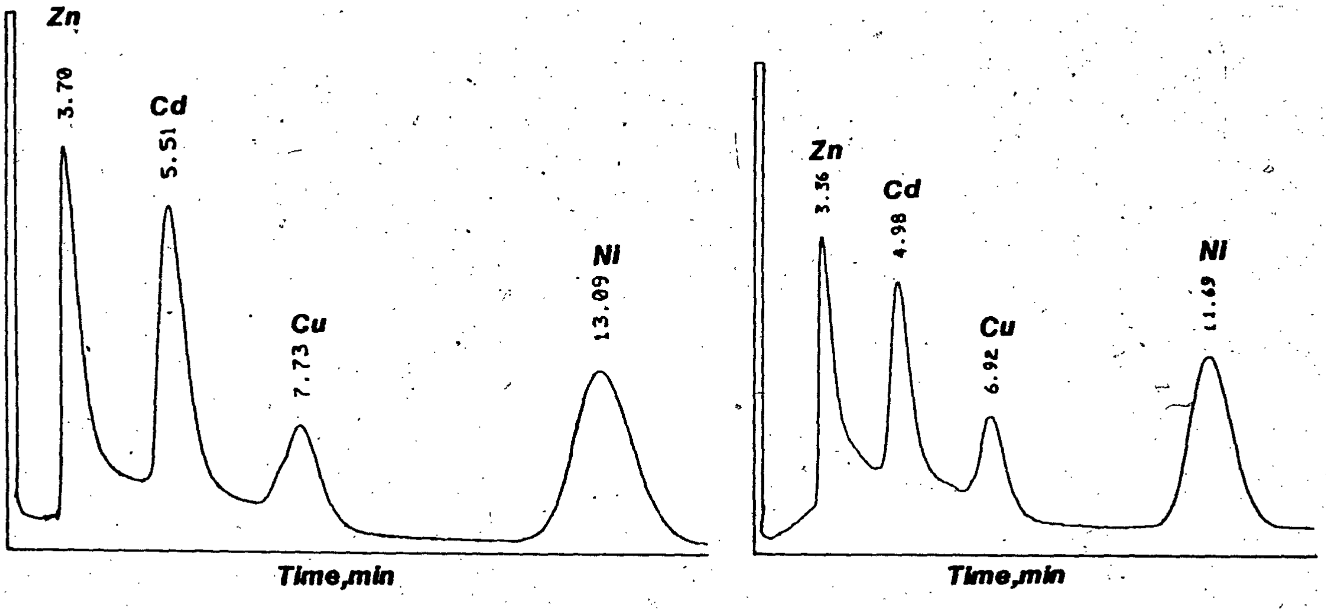
The separation data for a Zn(II), Cd(II), Cu(II), and $Ni(II)(DDC)_2$ mixture on this gradient column and on a 2% column, which is the average loading of the gradient column, are given in Table 24. Both the calculated resolution and efficiency, were somewhat superior on the gradient column even though this is not all that obvious from the chromatograms (Fig 35). Analysis time was reduced on the gradient column. The Cd/Cu DDC pair separation was

TABLE 24

COMPARATIVE DATA OBTAINED FOR Zn, Cd, Cu, AND Ni DDC ON A 1-2-3% EACH OF OV-225-QF-1 GRADIENT COLUMN, AND ON A 2% EACH OF OV-225-QF-1 CONVENTIONAL COLUMN AT 240°C

METAL DDC	2%			1-2-3%		
	N	RT	RSD% ±	N	RT	RSD% ±
Zn	342	3.7 ± .01	0.2	320	3.4 ± .01	0.2
Cd	600	5.5 ± .02	0.5	620	5.0 ± .01	0.2
Cu	664	7.7 ± .04	0.5	694	6.9 ± .01	0.1
Ni	685	13.1 ± .07	0.6	1294	11.7 ± .01	0.1
			RESOLUTION			
Zn-Cu		2.1 ± .07	3.0		2.1 ± .07	2.4
Zn-Cu		4.0 ± .12	3.1		4.0 ± .02	0.5
Zn-Ni		6.7 ± .12	1.8		8.1 ± .06	0.8
Cd-Cu		2.1 ± .07	3.5		2.1 ± .03	1.4
Cd-Ni		5.2 ± .12	2.2		6.4 ± .10	1.9
Cu-Ni		3.4 ± .12	3.6		4.1 ± .12	0.7

FIG. 35 SEPARATION OF Zn, Cd, Cu, AND Ni DDC ON A 1-2-3% EACH OF OV-225-QF-1 GRADIENT COLUMN, AND ON A 2% OV-225-QF-1 CONVENTIONAL COLUMN, AT 240°C, AND 30 ml/min.



2%

1-2-3%

similar on the two columns.

Plots of peak height response versus sample concentration for Zn, Cd, Pb, and Cu DDC were linear, and the correlation coefficients were better than 0.99, see (Appendix I, Table 51, and Figs 73 to Fig 74).

7.3.6.3 GRADIENT COLUMNS CONTAINING (1-2-3%) QF-1 + 1% OV-225) AS STATIONARY PHASE, ON CW-HP, 80/100 mesh, (60 cm x 2 mm I.D, glass tubing) (Tables 25, 26, Figs 36, 37, & 38)

A new gradient column was prepared in which one phase, QF-1, was varied to give a gradient of 1-2-3% while the other, OV-225, was kept constant at 1%. Table 25 summarizes the retention data and the resolution obtained for the Zn, Cd, Cu, and Ni DDC mixture at two different isothermal column temperatures, 220°C and 240°C, and under temperature programming conditions, of 200 - 240°C, at 4°C/min. No column bleeding was observed during temperature programming. A complete separation was achieved at 240°C in five minutes (Fig 36). At 220°C, a higher plate count was obtained, but the analysis time was doubled. Using temperature programming, a very good separation was obtained (Fig 37). This gradient column was compared to one

TABLE 25

RESULTS OBTAINED FOR Zn, Cd, Cu, AND Ni DDC ON A (1-2-3%) QF-1 + 1% OV-225 GRADIENT COLUMN ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING)

RETENTION DATA						
METAL DDC	240C		220C		200-240 TEMP PROG	
	N	RT	N	RT	N	RT
Zn	86	1.6	329	3.6	690	5.6
Cd	350	2.3	582	5.4	1307	7.2
Cu	207	3.2	412	7.6	1431	8.5
Ni	460	5.4	448	13.2	2403	11.0

RESOLUTION			
	240C	220C	200-240 TEMP PROG
Zn-Cd	1.2	2.1	2.0
Zn-Cu	2.0	3.5	3.3
Zn-Ni	4.4	5.8	6.2
Cd-Cu	1.3	1.8	1.5
Cd-Ni	4.0	4.6	4.5
Cu-Ni	2.2	2.8	2.8

FIG. 36 SEPARATION OF Zn, Cd, Cu, AND Ni DDC ON A (1-2-3%) QF-1 + OV-225 GRADIENT COLUMN, AT 220°C, AND 240°C , AND 30ml/min.

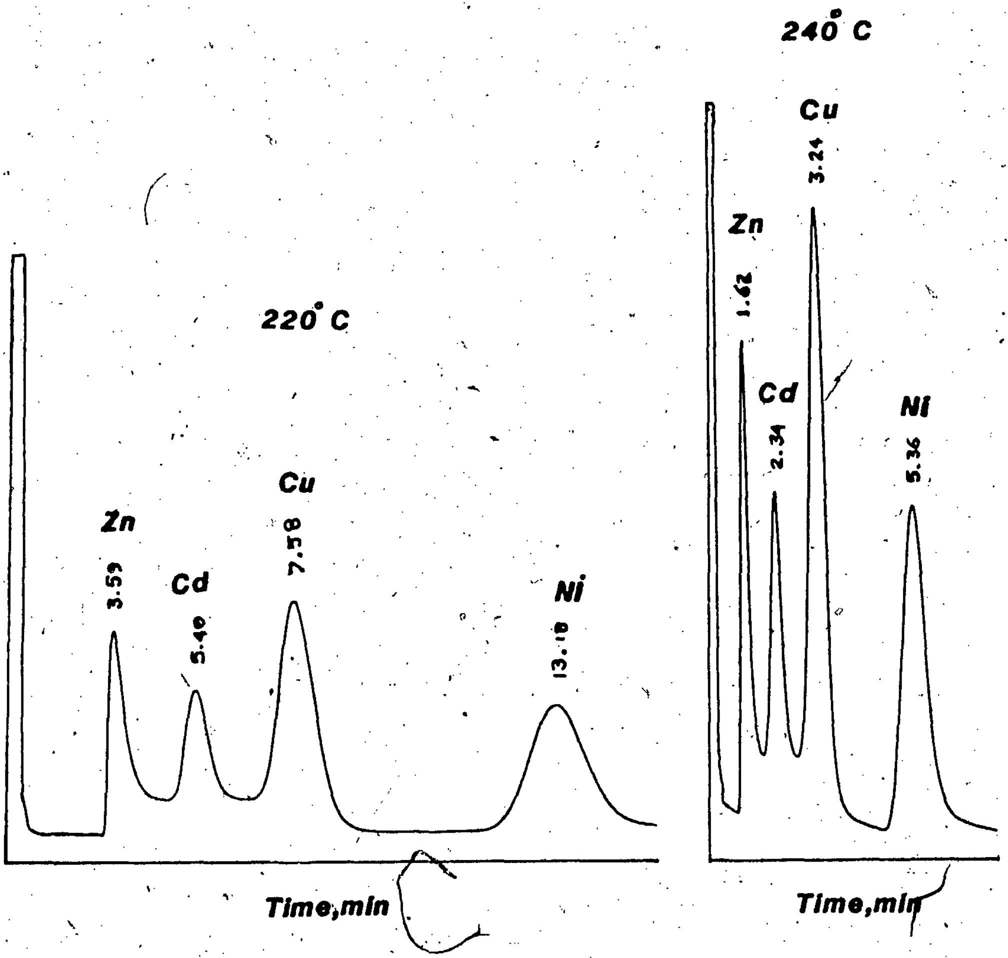
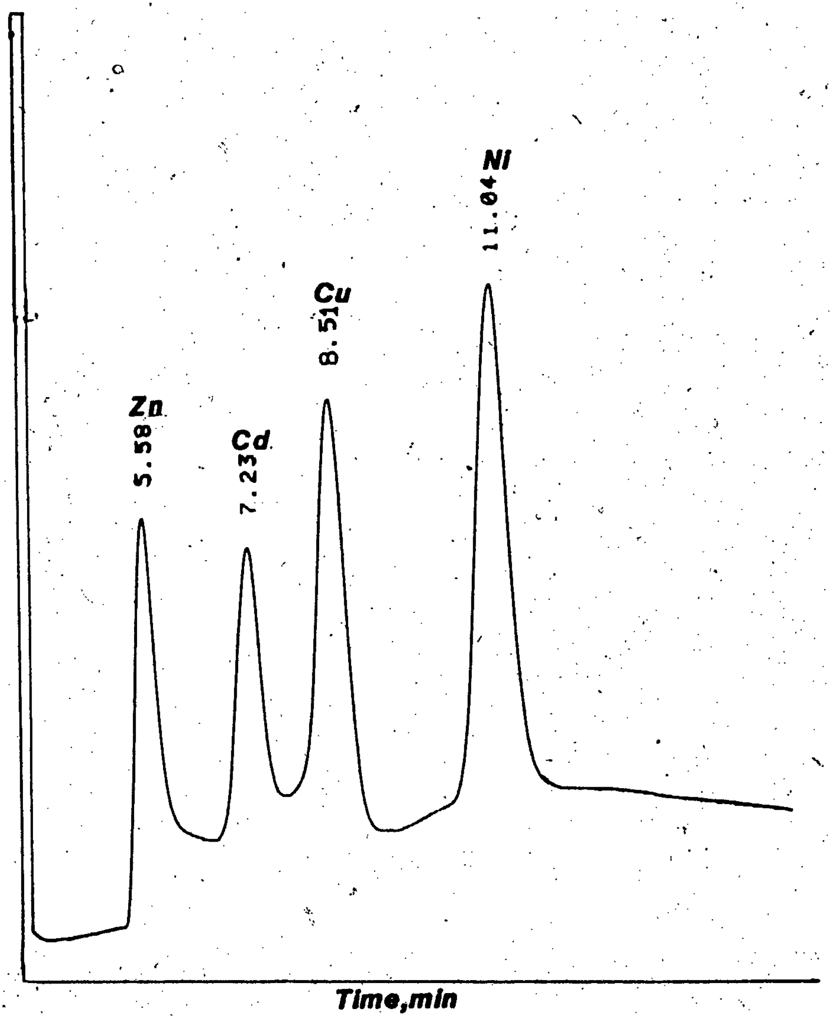


FIG. 37 SEPARATION OF Zn, Cd, Cu, AND Ni DDC ON A (1-2-3%) QF-1 + OV-225 GRADIENT COLUMN, AT TEMPERATURE PROGRAMMING FROM 200°C TO 240°C, AT 4°C/min.



containing a loading of 2% QF-1 and 1% OV-225 (Table 26). The number of theoretical plates and the resolution for the gradient column was significantly higher. The analysis time was similar.

Analysis was next attempted of a mixture of Pb(II), Hg(II), Co(III), Zn(II), Cd(II), Cu(II), and Ni(II) DDC. The mixture was injected at 240°C and the results are shown in Fig 38. Six metals were separated. Cu(II) and Hg(II) DDC, however, eluted together after 3.3 minutes. Pb(II) DDC was separated from Cu(II) DDC, but the resolution was not complete due to interference by Hg(II) DDC.

The plots of peak height response versus sample concentration were all linear, and the correlation coefficients were better than 0.99, (Appendix I, Table 52, and Figs 75 to Fig 77).

TABLE 26

COMPARATIVE DATA OBTAINED FOR Zn, Cd, Cu, AND Ni DDC ON A (1-2-3%) QF-1 + 1% OV-225 GRADIENT COLUMN, AND ON A 2% QF-1 + 1% OV-225 CONVENTIONAL COLUMN, AT 220°C

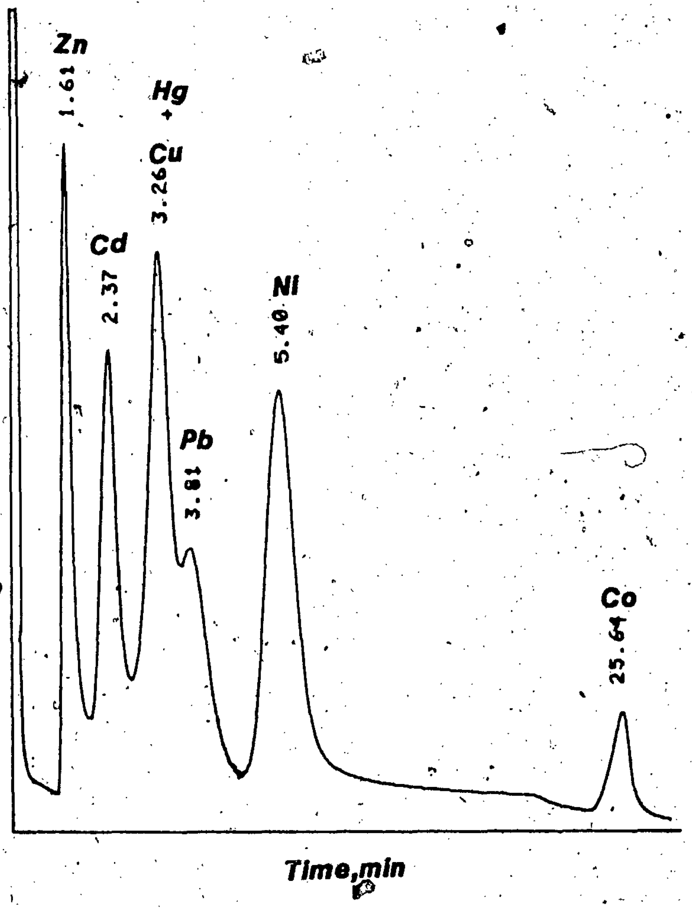
RETENTION DATA

METAL DDC	2% QF-1 + 1% OV-225		1-2-3% QF-1 + 1% OV-225	
	N	RT(min)	N	RT(min)
Zn	270.40	3.70	490.80	3.60
Cd	247.00	5.50	385.60	5.40
Cu	262.80	7.70	471.50	7.60
Ni	431.40	13.50	527.00	12.30

RESOLUTION DATA

Zn-Cd	1.6	2.1
Zn-Cu	2.9	3.9
Zn-Ni	5.6	6.5
Cd-Cu	1.3	1.8
Cd-Ni	4.0	4.6
Cu-Ni	2.6	3.0

FIG. 38 SEPARATION OF Zn, Cd, Cu, Ni, Pb, Hg, AND Co DDC ON A (1-2-3%) QF-1 + OV-225 GRADIENT COLUMN, AT 240°C.



7.4 SEPARATION OF A COMPLEX MIXTURE OF METAL DDC CHELATES

Tests with four component mixtures were useful for purposes of evaluating column efficiency and determining analysis times. To see if any of the gradient columns could be useful for closer to real life separations, it was necessary to determine whether more complex mixtures could be separated. Accordingly, a mixture consisting of: Zn(II), Cd(II), Cu(II), Ni(II), Hg(II), Pb(II), Co(III), and Cr(III), and occasionally MoO_2(II) DDC was then tested on the following columns:

(1) ONE PHASE GRADIENT

- 1-2-3% and 3-2-1% QF-1
- 1.0-1.5-2.0-2.5-3.0% QF-1
- 1-2-3% OV-210

(2) TWO-PHASE GRADIENT

- 1-2-3% (OV-101-QF-1)
- (1-2-3% QF-1) + 1% OV-225
- 1-2-3% (OV-225-QF-1)

OV-225 gradient columns were omitted from further study because of their especially strong affinity towards the metal chelates. This meant that high temperatures would be required in order to elute late peaks such as Co and Cr DDC with enhanced possibilities for thermal degradation.

7.4.1 1-2-3% QF-1 ON GC-Q, 80/100 Mesh, (60 cm X 2 mm I.D, glass tubing) (Table 27, Figs 39 to 42)

On this column no degradation was observed for $Pb(DDC)_2$ and $Hg(QDC)_2$ at 190°C, but at 204°C $Hg(DDC)_2$ gave one major peak and two small peaks amounting to approximately 4% of the injected sample (Fig 39).

Results of analysis of the test mixture at a column temperature of 190°C are illustrated in Fig 40 and Table 27. Temperature programming from 160°C to 200°C, at 2°C/min enhanced the resolution of the Cd/Pb DDC pair and generally improved the chromatogram. However, the Cr/Co DDC pair was still not well resolved (Fig 41).

Temperature programming from 200°C to 240°c, at 4°C/min, led to a good separation of a mixture of Zn, Cd, Pb, Cu, Ni, Hg, Cr and Co DDC in 7.5 minutes, except that Cd and Pb, were not resolved; as well as Co and Cr (Fig 42).

FIG. 39 CHROMATOGRAMS OBTAINED FOR Hg(DDC)$_2$ ON 1-2-3% QF-1 ON GC-Q, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING)

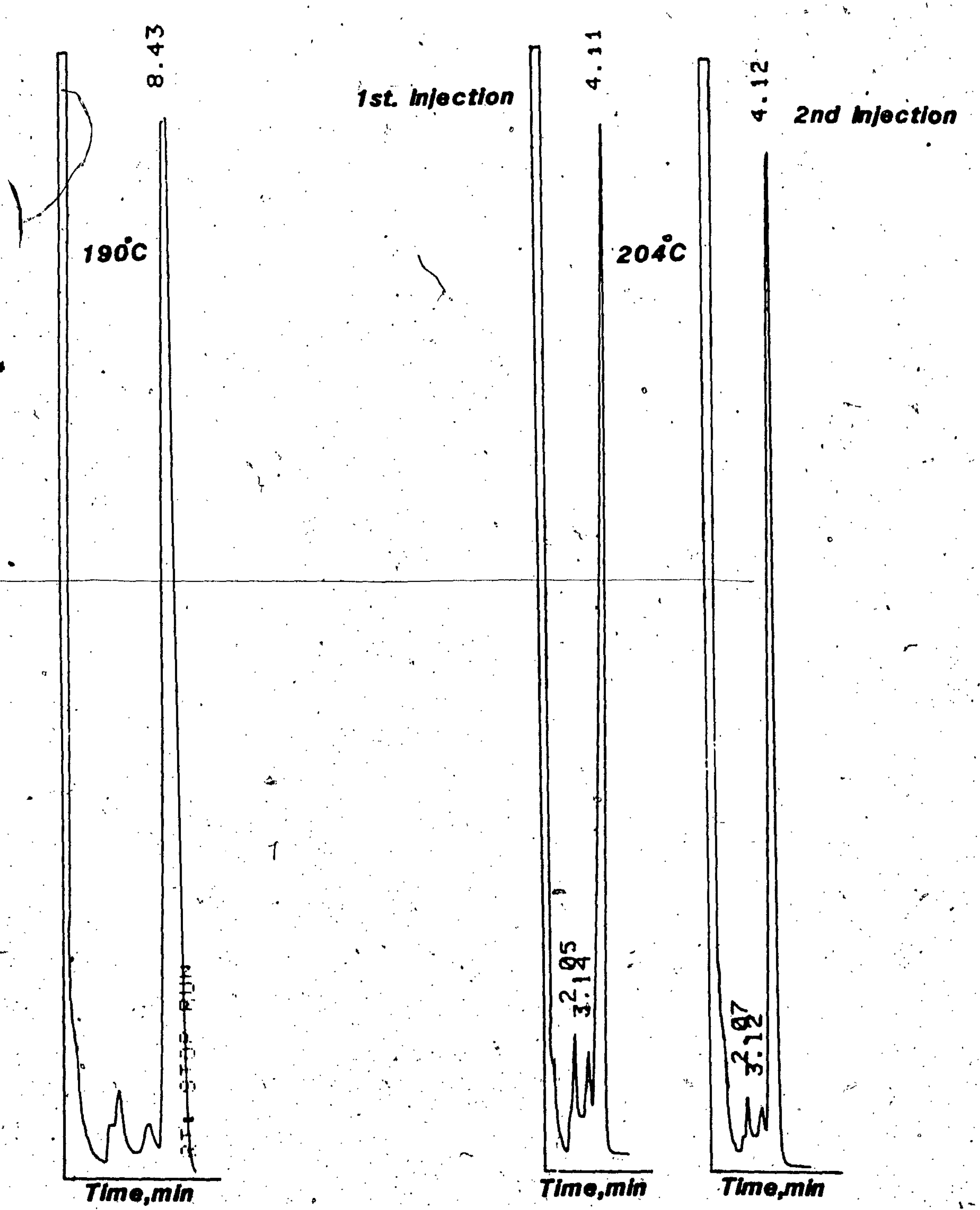


FIG. 40 SEPARATION OF A COMPLEX MIXTURE OF EIGHT $M(DDC)_n$ ON 1-2-3% QF-1 ON GC-Q, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 190°C.

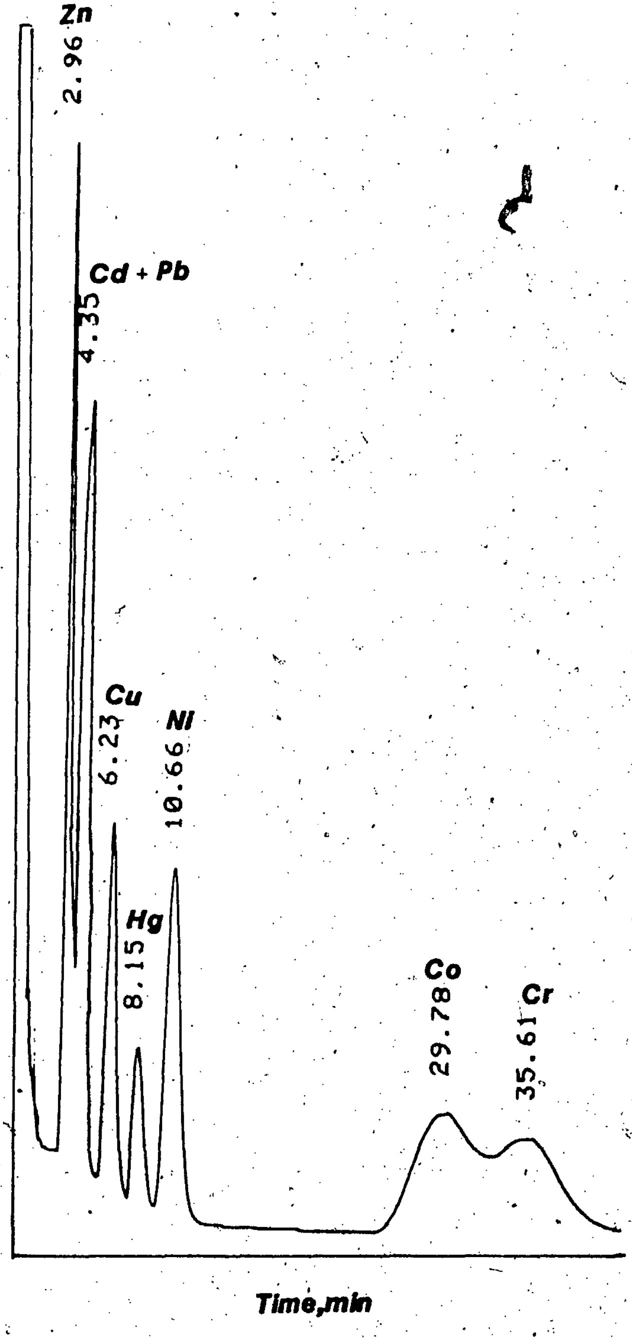


TABLE 27

COMPARATIVE DATA OBTAINED FOR A MIXTURE OF EIGHT $M(DDC)_n$ ON 1-2-3% QF-1 ON GC-Q, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 190°C, AND TEMPERATURE PROGRAMMING FROM 160°C to 200°C, AT 2°C/min

RETENTION DATA

METAL DDC	190°C		TEMP PROGRAM	
	RT	N	RT	N
Zn	3.0	140	9.0	255
Cd	4.4	194	10.8	296
Pb	4.4	194	11.8	731
Cu	6.2	202	14.1	1418
Hg	8.2	472	16.3	2704
Ni	10.7	455	18.4	1759
Co	29.8	142	29.6	206
Cr	35.6	193	32.2	338

RESOLUTION

	190°C	TEMP PROGRAM
Zn-Cd	1.2	1.4
Cd-Pb	0.0	0.5
Cu-Pb	1.3	1.7
Cu-Hg	1.2 ✓	1.5
Hg-Ni	1.4	1.4
Ni-Co	3.2	2.2
Cr-Co	0.6	0.3

FIG. 41 SEPARATION OF A COMPLEX MIXTURE OF EIGHT M(DDC)$_n$
ON 1-2-3% QF-1 ON GC-Q, 80/100 MESH,
(60 cm x 2 mm I.D, GLASS TUBING),
PROGRAMMED FROM 160°C TO 200°C, AT 2°C/min.

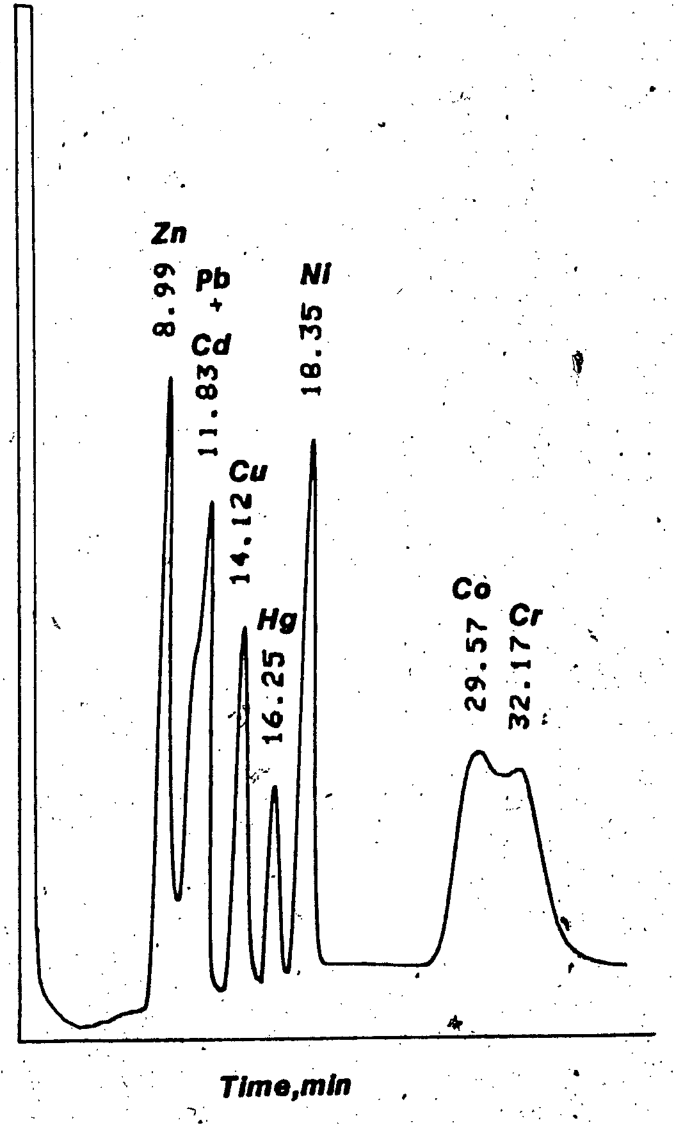
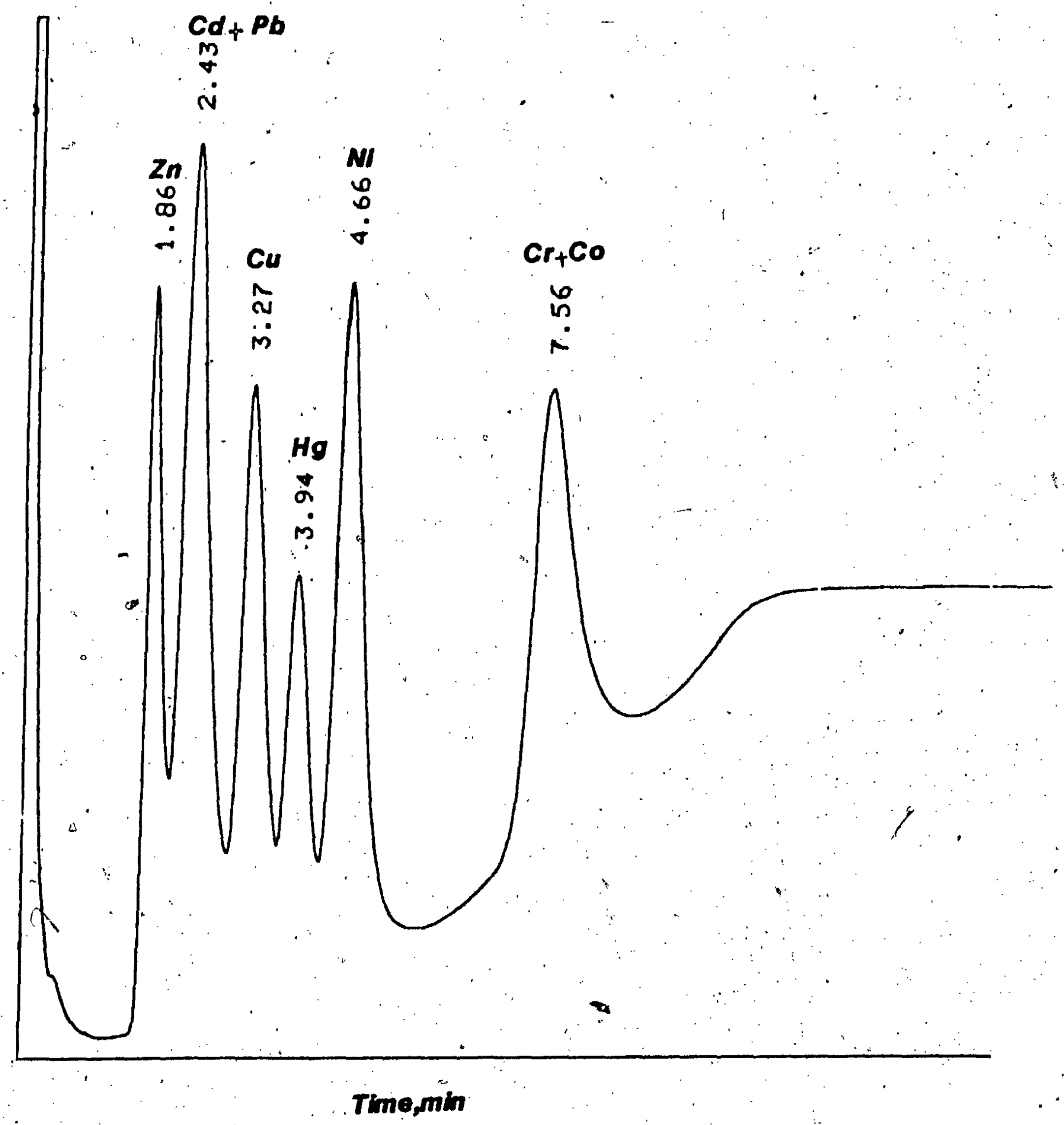


FIG. 42 SEPARATION OF A COMPLEX MIXTURE OF EIGHT $M(DDC)_n$ ON 1-2-3% QF-1 ON GC-Q, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), PROGRAMMED FROM 200°C TO 240°C, AT 4°C/min.



7.4.2 3-2-1% QF-1 ON CW-HP, 80/100 Mesh, (60 cm X 2 mm I.D, glass tubing) (Table 28, Figs 43, 44)

By reversing the QF-1 gradient column configuration, and using temperature programming from 200°c to 230°c, at 1°C/min, a dramatic improvement in the resolution was obtained for a mixture of Zn, Cd, Pb, Cu, Hg, Ni, Co, and Cr DDC. The separation of the Cd/Pb DDC pair, however, was still not achieved. The data are summarized in Table 28 and Fig 43.

With Mo(II) added to the above mixture, and temperature programming from 160°C to 200°C, at 2°C/min, the separation shown in Fig 44 was obtained. $MoO_2(DDC)_2$ was detected but the Cr/Co DDC pair separation deteriorated and the Cd/Pb pair remained unresolved.

Extrapolating from Ettre's work (64), an R = 4.9 obtained for the Ni/Co DDC pair meant that an additional four peaks should be resolvable between peaks for Ni and Co DDC. Theoretically, therefore, it should be possible to add at least four more suitable metal DDC to the above test mixture and still obtain adequate resolution on the above column.

TABLE 28

RESULTS OBTAINED FOR THE SEPARATION OF AN EIGHT $M(DDC)_n$ MIXTURE ON 3-2-1% QF-1 ON GC-Q, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), TEMPERATURE PROGRAMMING FROM 200°C TO 230°C, AT 1°C/min

	RETENTION DATA	
METAL DDC	RT(MIN)	N
Zn	3.3	481
Cd	4.6	334
Pb	4.6	334
Cu	6.3	637
Hg	7.7	1162
Ni	9.6	1028
Co	18.9	847
Cr	20.9	1210

	RESOLUTION
Zn-Cd	1.6
Cd-Pb	1.7
Cu-Pb	0.0
Cu-Hg	1.4
Hg-Ni	1.9
Ni-Co	4.9
Cr-Co	0.8

FIG. 43 SEPARATION OF A COMPLEX MIXTURE OF EIGHT $M(DDC)_n$ ON 3-2-1% QF-1 ON GC-Q, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), PROGRAMMED FROM 200°C TO 230°C, AT 1°C/min.

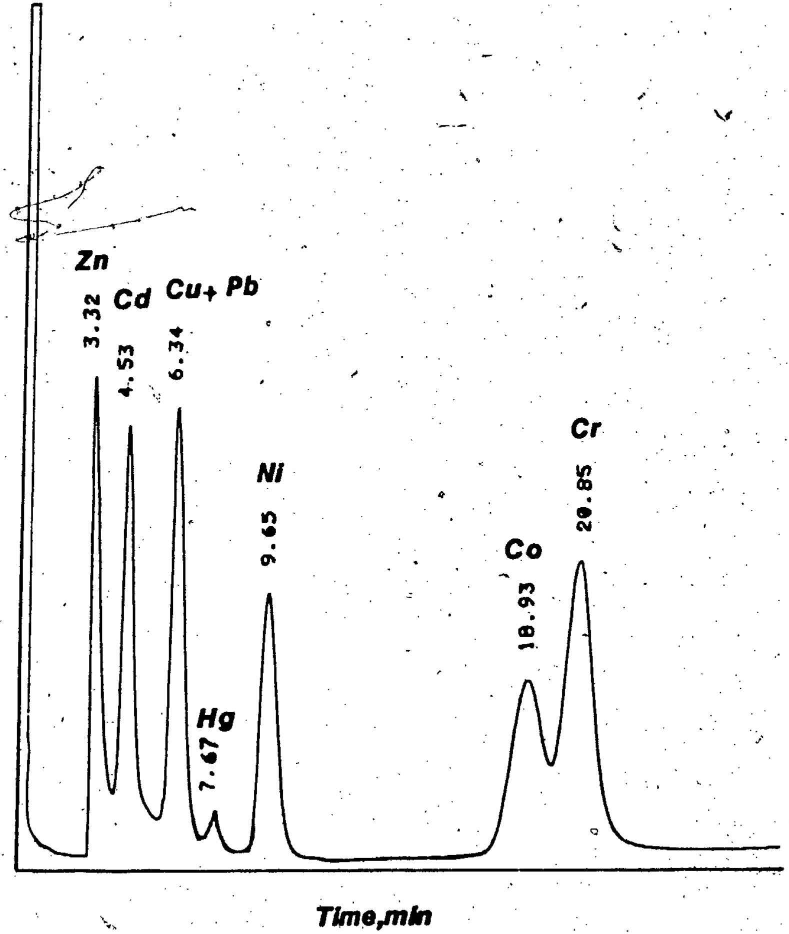
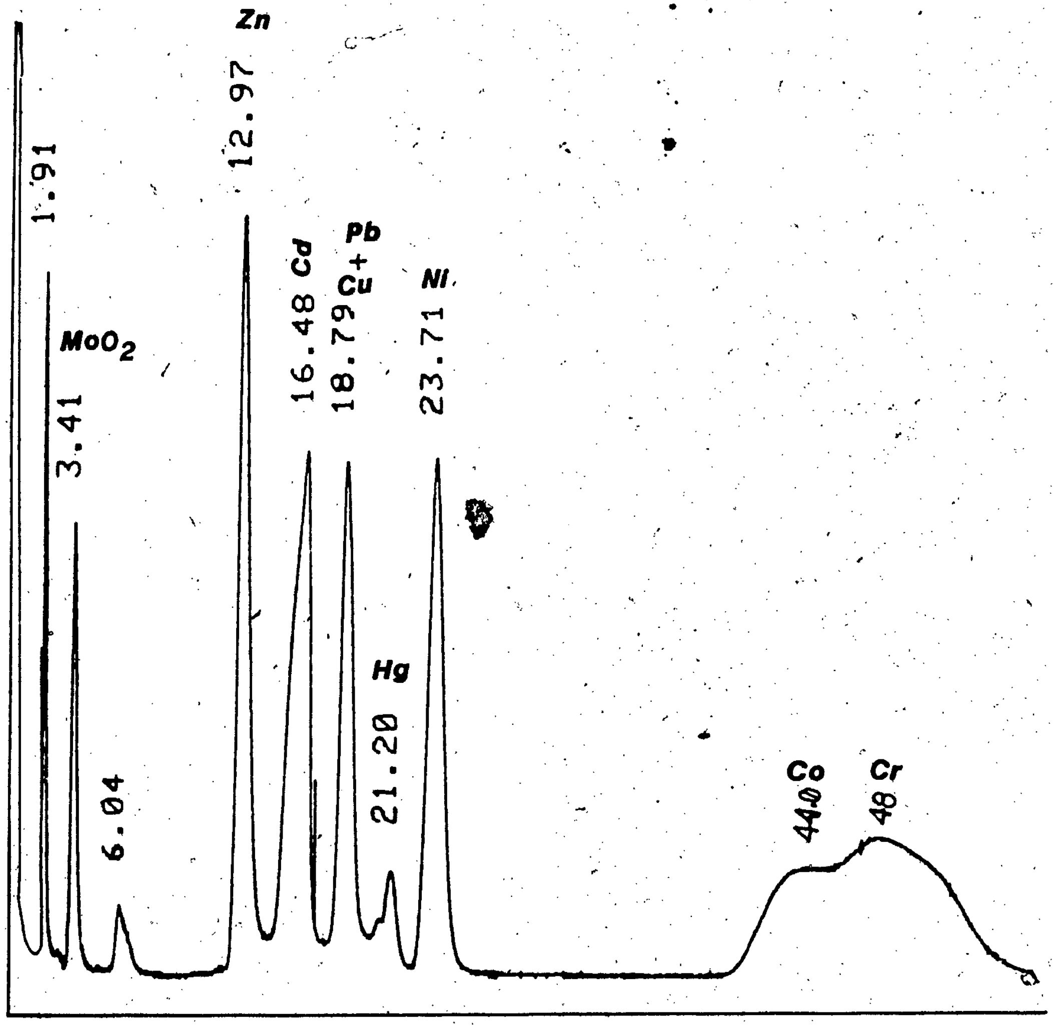


FIG. 44 SEPARATION OF A COMPLEX MIXTURE OF NINE $M(DDC)_n$ ON 3-2-1% QF-1 ON GC-Q, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), PROGRAMMED FROM 160°C TO 200°C, AT 2°C/min.



7.4.3 1.0-1.5-2.0-2.5-3.0% QF-1, ON CW-HP, 80/100 Mesh, (60 cm x 2 mm I.D, glass tubing) (Table 29, Figs 45, 46)

The gradient column 1.0-1.5-2.0-2.5-3.0 % QF-1, was prepared in order to solve the problem of Pb and Cd separation. Good resolution for Pb and Cd had been achieved for a mixture of four $M(DDC)_n$, namely: Zn, Cd, Pb, and Ni, as shown in Fig 25. The data obtained for a more complex DDC mixture are shown in Table 29 and Fig 45. No separation of Cd and Pb was achieved due to the presence of $Cu(DDC)_2$ which apparently caused the Cd and Pb peaks to overlap. The resolution was improved by temperature programming at a lower temperature range, as shown in Fig 46.

7.4.4 1-2-3% OV-210 ON CW-HP, 80/100 Mesh, (60 cm x 2 mm I.D, glass tubing) (Table 30, Fig 47)

The best results with the 1-2-3% OV-210 gradient column for the resolution of the eight metal DDC complex mixture were obtained by temperature programming from 180^{o}C to 240^{o}C at 2^{o}C/min, (Table 30, Fig 47). Two pairs of metal DDC could not be separated on this column; i.e: Cd/Pb and Cu/Pb. In addition resolution of the Co/Cr DDC pair was

TABLE 29

RESULTS OBTAINED FOR THE SEPARATION OF A MIXTURE OF EIGHT $M(DDC)_n$ ON 1.0-1.5-2.0-2.5-3.0% QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), WITH TEMPERATURE PROGRAMMING FROM 200°C TO 230°C, AT 1°C/min

	RETENTION DATA	
METAL DDC	RT(min)	N
Zn	3.5	306
Cd	4.7	292
Pb	4.7	292
Cu	6.5	676
Hg	8.2	1681
Ni	9.9	928
Co	19.2	655
Cr	21.0	784

	RESOLUTION
Zn-Cd	1.3
Cd-Pb	0.0
Cu-Pb	1.7
Cu-Hg	1.9
Hg-Ni	1.6
Ni-Co	4.3
CR-CO	0.6

FIG. 45 SEPARATION OF A COMPLEX MIXTURE OF EIGHT $M(DDC)_n$ ON 1.0-1.5-2.0-2.5-3.0% QF-1 ON GC-Q, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), PROGRAMMED FROM 200°C TO 230°C, AT 1°C/min.

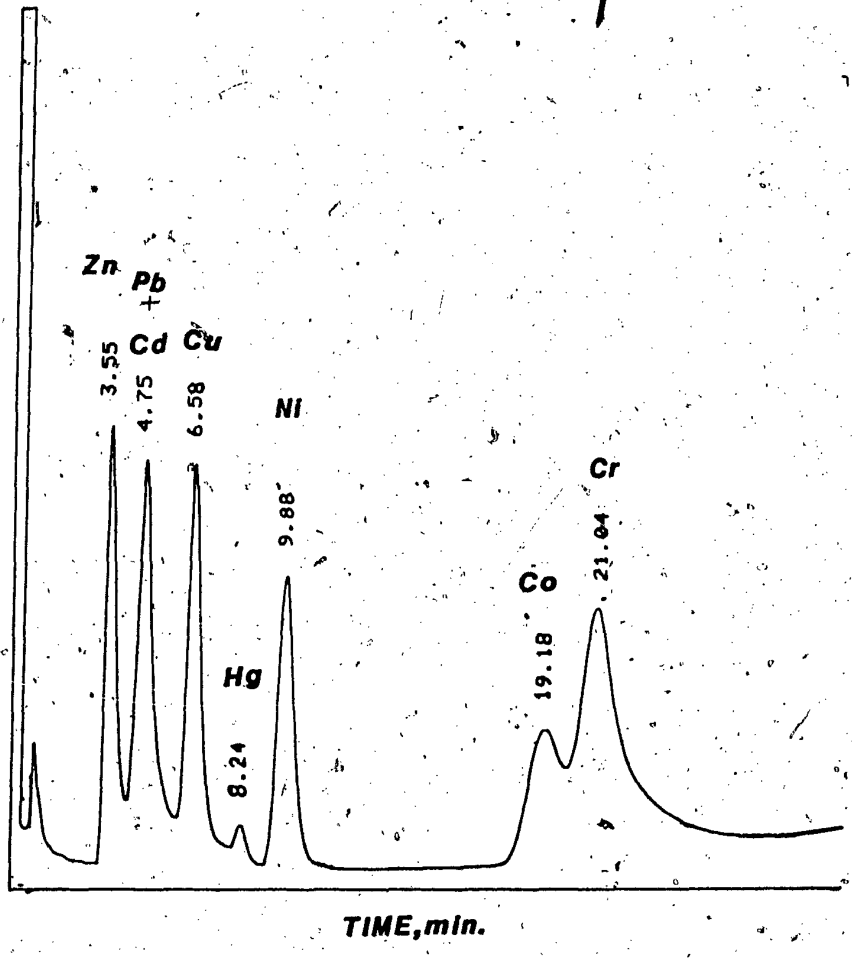


FIG. 46 SEPARATION OF A COMPLEX MIXTURE OF EIGHT $M(DDC)_n$ ON 1.0-1.5-2.0-2.5-3.0% QF-1 ON GC-Q, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), PROGRAMMED FROM 170°C TO 210°C, AT 2°C/min.

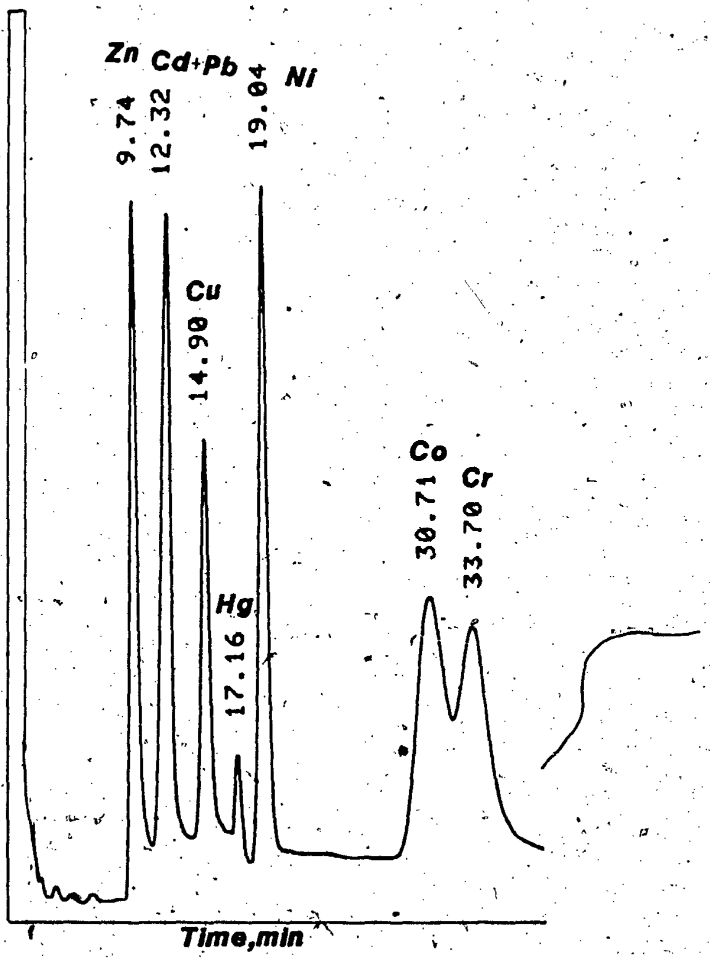


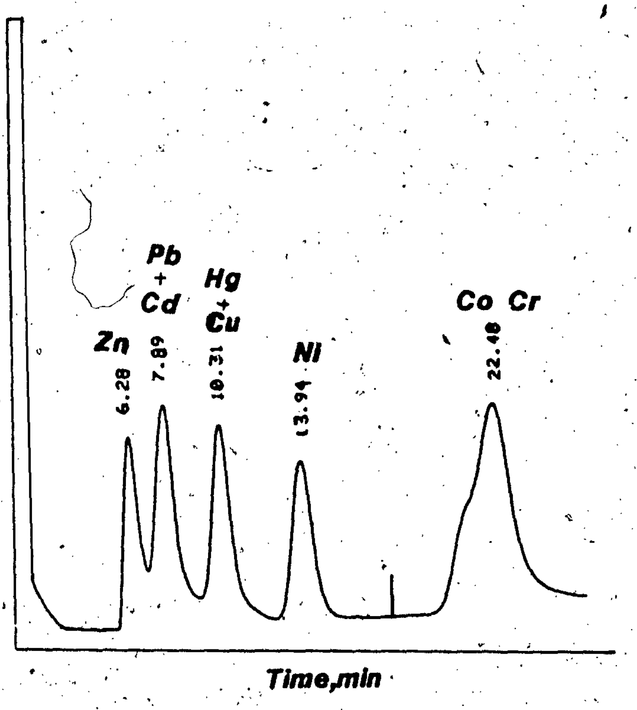
TABLE 30

RESULTS OBTAINED FOR THE SEPARATION OF AN EIGHT $M(DDC)_n$ MIXTURE ON 1-2-3% OV-210 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), WITH TEMPERATURE PROGRAMMING FROM 180°C TO 240°C, AT 2°C/min

METAL DDC	RETENTION DATA RT(min)	N
Zn	6.3	438
Cd	7.9	389
Pb	7.9	389
Cu	10.3	664
Hg	10.3	664
Ni	13.9	966
Co	21.0	489
Cr	22.5	742

	RESOLUTION
Zn-Cd	1.2
Cd-Pb	0.0
Cu-Pb	1.5
Cu-Hg	0.0
Hg-Ni	2.2
Ni-Co	2.5
Cr-Co	0.4

FIG. 47 SEPARATION OF AN EIGHT $M(DDC)_n$ MIXTURE ON 1-2-3% OV-210 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), PROGRAMMED FROM 180°C TO 240°C, AT 2°C/min.



incomplete. Although the other metal DDC's were well separated, this column did not offer much promise overall.

7.4.5 1-2-3% EACH OF OV-101-QF-1 ON CW-HP, 80/100 Mesh, (60 cm X 2 mm I.D, glass tubing) (Table 31, Fig 48)

The 1-2-3% each of OV-101-QF-1 stationary phase column was tested with the eight metal DDC mixture. The results are summarized in Table 31 and in Fig 48. Neither the Cd/Pb nor the Cu/Hg pair could be separated. The maximum separation obtained with this column was of six DDC chelates only.

7.4.6 1-2-3% EACH OF OV-225-QF-1 ON CW-HP, 80/100 Mesh, (60 cm x 2 mm I.D, glass tubing) (Table 32, Figs 49, 50)

On this column good resolution was obtained even for Cd and Pb DDC as shown in Table 32, Figs 49, 50 . The only drawbacks were that the peak for $Pb(DDC)_2$ overlapped that of $Hg(DDC)_2$, and the Co/Cr DDC pair resolution had deteriorated compared to some of the previously tested columns.

TABLE 31

RESULTS OBTAINED FOR THE SEPARATION OF AN EIGHT $M(DDC)_n$ MIXTURE ON 1-2-3% EACH OF OV-101 AND QF-1 ON CW-HP, 80/100 MESH, (60 cm 2 mm I.D, GLASS TUBING) WITH TEMPERATURE PROGRAMMING FROM 200°C TO 250°C, AT 2°C/min

METAL DDC	RETENTION DATA RT(min)	N
Zn	5.8	538
Cd	8.7	374
Pb	8.7	374
Cu	10.7	378
Hg	10.7	378
Ni	14.7	858
Co	28.3	1896
Cr	29.3	2838

RESOLUTION	
Zn-Cd	2.1
Cd-Pb	0.0
Cu-Pb	1.0
Cu-Hg	0.0
Hg-Ni	1.9
Ni-Co	5.9
CR-CO	0.4

FIG. 48 SEPARATION OF AN EIGHT $M(DDC)_n$ MIXTURE ON 1-2-3% EACH OF OV-101-QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), PROGRAMMED FROM 200°C TO 250°C, AT 2°C/min.

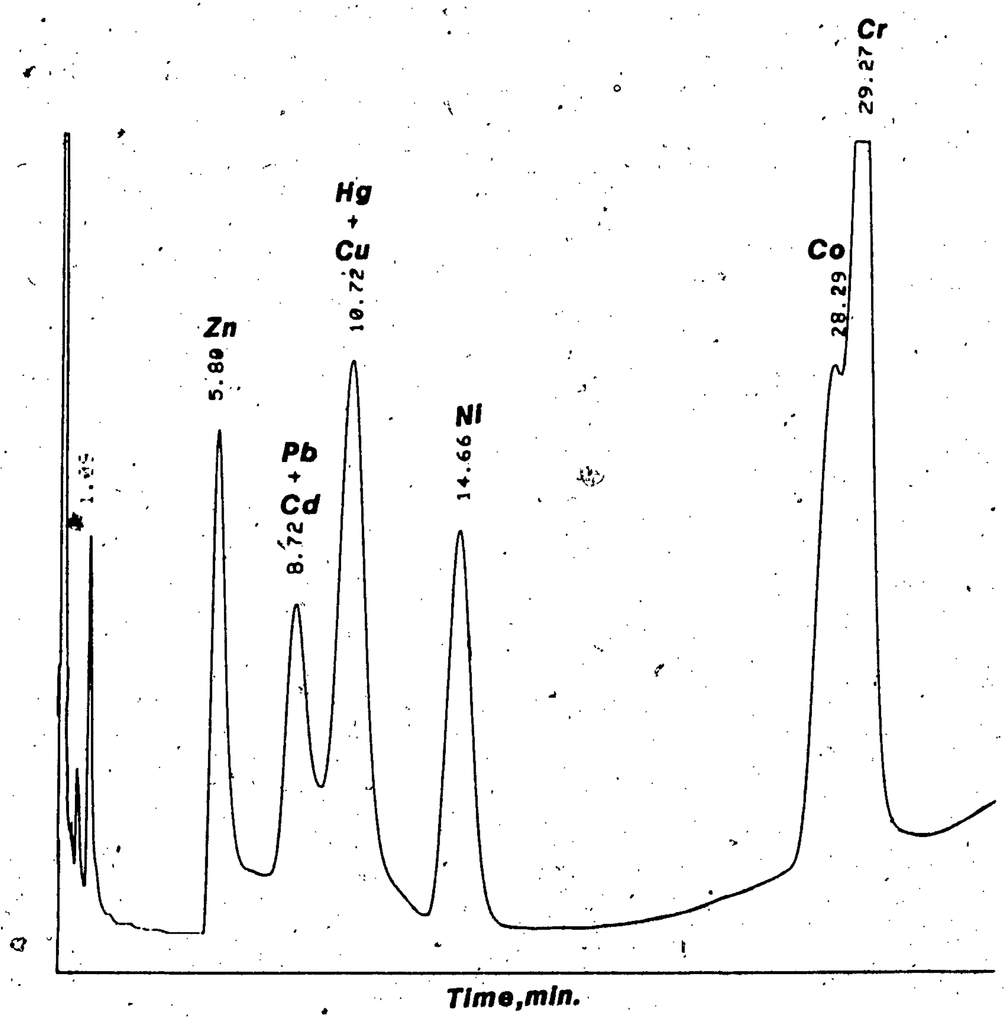


TABLE 32

RESULTS OBTAINED FOR THE SEPARATION OF AN EIGHT $M(DDC)_n$ MIXTURE ON 1-2-3% EACH OF OV-225-QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm, I.D, GLASS TUBING), AT 230°C FOR 15 mins, THEN TEMPERATURE PROGRAMMED FROM 230°C TO 260°C, AT 8°C/min

METAL DDC	RETENTION DATA RT(min)	N
Zn	4.6	529
Cd	6.9	529
Cu	9.5	446
Pb	11.9	468
Hg	11.9	468
Ni	16.2	1050
Co	32.5	1878
Cr	34.5	1469

RESOLUTION	
Zn-Cd	2.3
Cd-Cu	1.7
Cu-Pb	1.2
Pb-Hg	0.0
Hg-Ni	2.1
Ni-Co	6.5
Cr-Co	0.6

FIG. 49 SEPARATION OF AN EIGHT M(DDC)$_n$ MIXTURE ON 1-2-3% EACH OF OV-225-QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 230°C FOR 15 MINS, THEN PROGRAMMED FROM 230°C TO 260°C, AT 8°C/min.

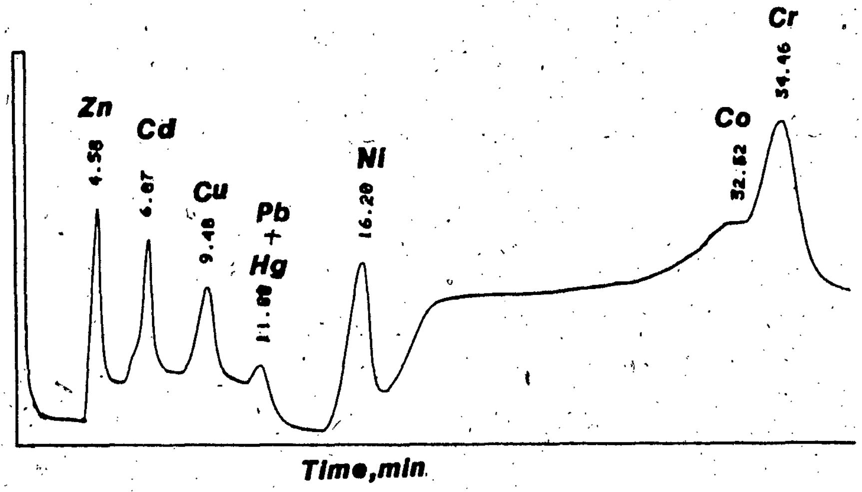
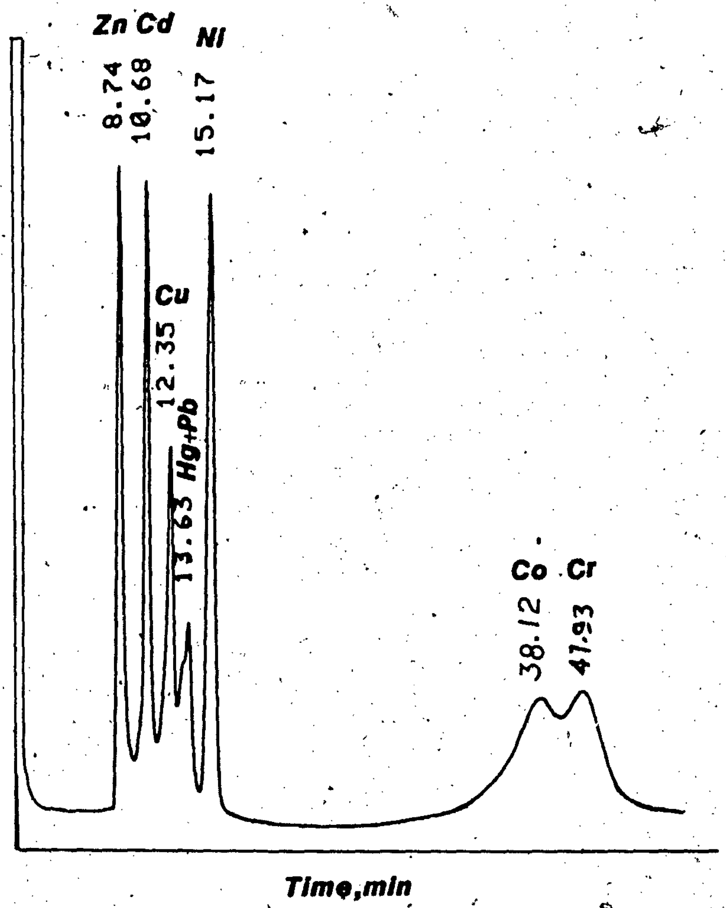


FIG. 50 SEPARATION OF AN EIGHT $M(DDC)_n$ MIXTURE ON 1-2-3% EACH OF OV-225-QF-1 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), PROGRAMMED FROM 200°C TO 250°C, AT 4°C/min.



7.4.7 (1-2-3%) QF-1 + 1% OV-225 ON CW-HP, 80/100 Mesh, (60 cm x 2 mm I.D, glass tubing) (Table 33, Figs 51, 52)

In a further investigation, a mixture of seven metal $(DDC)_n$ was injected onto a (1-2-3%) QF-1 + 1% OV-225 column. The separation achieved was excellent as shown in Fig 51, except that the Hg/Pb pair did not separate.
A mixture of eight metal DDC was injected and the results are shown in Table 33, and Fig 52. The separation obtained was excellent except for the Hg/Pb and Cr/Co pairs.

7.5 Conclusions:

1. Gradient columns with methylsilicone stationary phase, OV-101 or SE-30, were generally superior to conventional columns, but separation of a metal DDC complex mixture was not all that seccessful, only five peaks being obtained as shown for the case of 1-2-3% OV-101.

2. The more polar stationary phase QF-1, proved to be more selective for the separation of metal DDC. The gradient columns prepared with this stationary phase were the best columns for the separation of a complex metal DDC mixture. 3-2-1% QF-1 and 1.0-1.5-2.0-2.5-3.0% QF-1 were

TABLE 33

RESULTS OBTAINED FOR THE SEPARATION OF AN EIGHT $M(DDC)_n$ MIXTURE ON (1-2-3%) QF-1 + 1% OV-225 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), WITH TEMPERATURE PROGRAMMING FROM 200°C TO 250°C, AT 2°C/min

METAL DDC	RETENTION DATA RT(min)	N
Zn	8.4	780
Cd	11.8	866
Cu	14.3	1010
Hg	16.0	1029
Pb	16.0	1029
Ni	19.1	2287
Co	36.7	1012
Cr	36.7	1012

	RESOLUTION
Zn-Cd	2.4
Cd-Cu	1.5
Cu-Hg	0.9
Hg-Pb	0.0
Pb-Ni	1.7
Ni-Co	5.6
Cr-Co	0.0

FIG. 51 SEPARATION OF A SEVEN M(DDC)$_n$ MIXTURE ON (1-2-3%) QF-1 + 1% OV-225 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), PROGRAMMED FROM 200°C TO 240°C, AT 4°C/min.

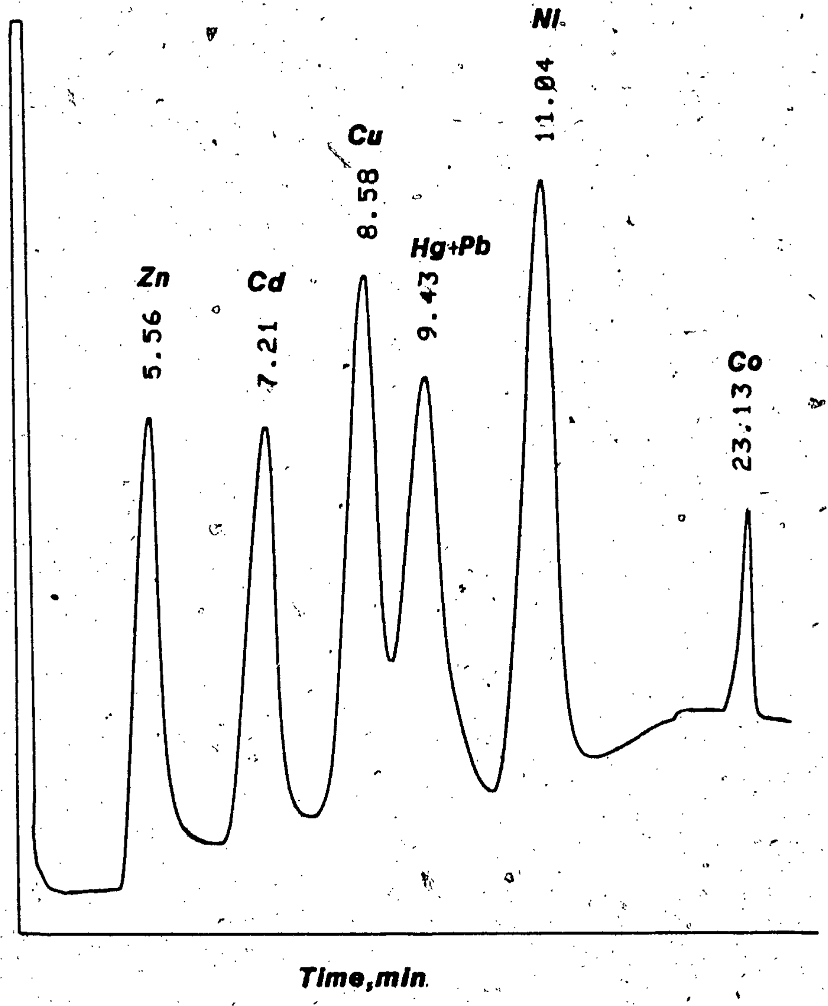
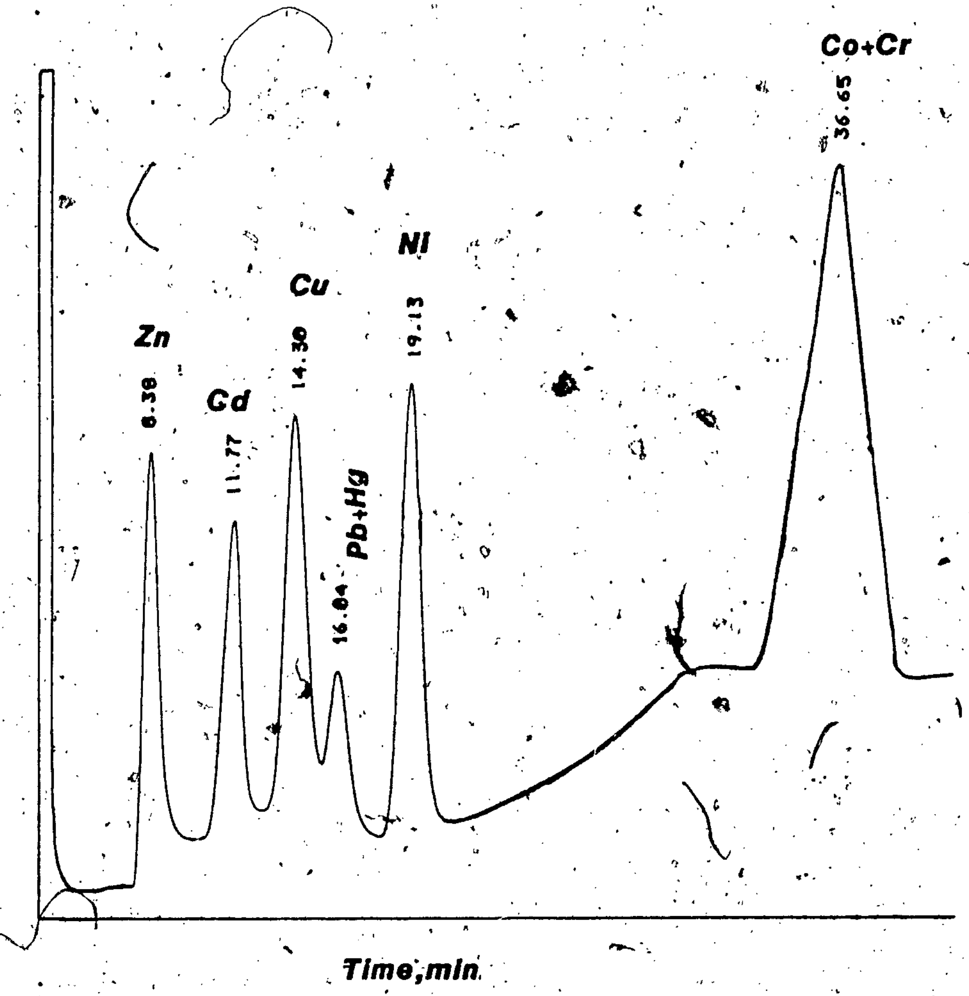


FIG. 52 SEPARATION OF AN EIGHT $M(DDC)_n$ MIXTURE ON (1-2-3%) QF-1 + 1% OV-225 ON CW-HP, 80/100 MESH, (60 cm x 2 mm I.D, GLASS TUBING), AT 200°C FOR 5 MINS, THEN PROGRAMMED FROM 200°C TO 250°C, AT 2°C/min.



the two best columns tested. They can be recommended for the separation of metals in soil, water-pollution and marine bottom sediments.

3. Two-stationary phase gradient columns gave good separation for the complex eight metal DDC mixture. The best combination was that of 1-2-3% (OV-225-QF-1). The separation on this column was better than that obtained on a 1-2-3% (OV-101-QF-1) column. The (OV-225-QF-1) column would be useful in the analysis of marine sedements. Another column which would be useful in this case is (1-2-3%) QF-1 + 1% OV-225 which showed very good resolution for a mixture of eight metal DDC. In both cases retention times are long enough to permit resolution of metal DDC from any large extraneous peaks eluted before the DDC's.

8.0 Characterization of gradient columns using Retention Indices:

The gradient columns studied in this project were primarily intended to achieve the separation of metal chelate complex mixtures. To identify the characteristics of the gradient columns prepared and thereby determine their potential for the separation of other classes of compounds. Retention Indices were determined for selected compounds representing various chemical groups on a number of columns. It was also the intent of the tests carried out to establish the effect of changes in gradient columns on Retention Indices.

The test probes chosen for characterization were those proposed by McReynolds (47) because they adequately characterize the principal molecular interactions responsible for retention in gas chromatography. All columns used for characterization were 180 cm x 2 mm I.D in size.

Two types of gradient columns were characterized:

1. Variable percent Loaded gradient columns:

   Those columns tested were the most efficient gradient columns found for the separation of $M(DDC)_n$.

2. Polarity gradient columns:

These gradient columns had not been used in the separation of $M(DDC)_n$ study. Columns contained three different stationary phases, loaded on the same support, and each at the same percentage loading. The gradient in these columns resulted from differences in polarity of the stationary phases.

8.1 Preliminary work:

Preliminary work was undertaken with the following columns:

1. 20% OV-17 on CW-HP 80/100 mesh
2. 20% Squalane CW-HP 80/100 mesh

These columns were prepared in order to confirm McReynolds constants that have been reported for OV-17 stationary phase, and to optimize GC conditions, and compare them to those reported by McReynolds (48).

The test probes recommended by McReynolds (48) (table 1) were used to characterize the OV-17 column. The Retention Indices were calculated according to EQ. 13. The results obtained with the OV-17 and squalane columns are shown in Table 34. The differences in experimental results and those reported by McReynolds were probably the result of at least three factors:

(1) Tail suppressors were not added during column preparation, as was the case in McReynolds work. This may have affected the retention times of the the test probes.

(2). The column temperature used, 120 °C, was as reported by McReynolds. However, other conditions such as injector, and detector temperature,

TABLE 34

CHARACTERIZATION OF OV17
USING McREYNOLDS' CONSTANTS

SOLUTE	20% OV17		20% SQUALANE		I		
		RSD% ±		RSD% ±	OBTAINED	REP*	DIFF
BENZENE	750	.70	596	1.3	155	119	+35.5
N-BUTANOL	735	.30	581	2.1	158	154	- 3.7
1,4DIOXANE	831	.15	605	2.0	184	226	+42.0
1-NITROPROPANE	901	.13	644	0.6	243	257	+14.3
PYRIDINE	896	.14	694	0.3	202	202	+ 0.2
2-METHYL- 2-PENTANOL	805	.16	690	0.5	112	115	+ 3.4
1-IODOBUTANE	938	.10	824	0.1	119	114	- 4.7
PENTANONE	785	.22	634	0.5	162	151	-10.7
CIS-HYDRINDANE	1160	.10	1026	0.01	69	58	-10.8

* REPORTED BY McREYNOLDS (47).

column length, and carrier gas flow rate may not have been the same.

(3) Lack of reproducibility in reproducing retention indices on Squalane columns has been reported in the literature (57). It has been shown to be the result of either oxidation of,or, impurities in the Squalane.

8.2 Polarity gradient columns

8.2.1 Phenylsilicon stationary phase:

By combining equal length of packing containing OV-1 (0% phenyl), OV-3 (10% phenyl) and OV-7 (20% phenyl), at 3% loading each, within a 180 cm x 2 mm I.D column, an ascending polarity gradient column is obtained. A second column is created simply by reversing the order of the configuration to give a descending polarity gradient. The Retention Indices obtained for the McReynolds chemical probes for these two columns are shown in Table 35. They were compared with an OV-3 column which has a polarity corresponding to the average polarity of the gradient columns, i.e. 10% phenyl. Differences in Retention Index values on both OV-1-3-7 and OV-7-3-1 from those for OV-3 were observed for polar compounds such as 1,4 dioxane,

TABLE 35

COMPARATIVE RETENTION INDICES OBTAINED FOR OV7-OV3-OV1 AND OV1-OV3-OV7 GRADIENT COLUMNS, AND FOR AN OV3 CONVENTIONAL COLUMN

SOLUTE	OV7-OV3-OV1		OV1-OV3-OV7		OV3	
		RSD% ±		RSD% ±		RSD% ±
BENZENE	572	1.0	571	1.7	557	3.0
N-BUTANOL	639	0.5	639	1.5	685	6.0
1,4 DIOXANE	625	0.6	639	1.0	819	2.5
1-NITROPROPANE	671	1.0	677	0.5	819	1.5
PYRIDINE	691	0.2	685	0.2	801	0.5
2-METHYL-2-PENTANOL	691	0.4	685	3.0	841	0.7
1-IODOBUTANE	748	1.6	746	0.5	921	1.0
OCTYNE	792	0.3	792	1.0	950	0.8
PENTANONE	886	1.0	526	2.0	551	4.0
CIS-HYDRINDANE	924	1.0	929	0.5	1216	2.0

pyridine , pentanone , octyne , and 1 -iodobutane. Where differnces existed, the Retention Indices values for the gradient columns were generally lower than the values obtained for the OV-3 column. This was due mainly to the shorter retention times for the test probes as well as for reference paraffins on the gradient columns.

By replacing OV-7 with SE-52 (5% phenyl) the average polarity for the column was reduced. In Table 36, the Retention Index data obtained for the OV-1-SE-52-OV-3 and OV-3-SE-52-OV-1 columns are illustrated.

8.2.2 Conclusions based on tests with polarity gradient columns:

1. Polarity gradient columns are useful for analysis of polar compounds such as dioxane, pyridine and 2-methyl-2-pentanol since they provide selectivity with reduced analysis time.
2. RI values for polar substance varied depending on the polarity gradient order. The RI were greater for columns construcuted to have increasing polarity from inlet to outlet ends. This increase in RI values was more pronounced in the case of the lower polarity gradient column OV-1-SE-52-OV-3.

TABLE 36

COMPARATIVE RETENTION INDICES OBTAINED FOR OV1-SE52-OV3 AND OV3-SE52-OV1 GRADIENT COLUMNS, AND AN OV-3 CONVENTIONAL COLUMN

SOLUTE	OV1-SE52-OV3		OV3-SE52-OV1		OV-3	
		RSD% ±		RSD% ±		RSD% ±
BENZENE	528	3.0	533	3.9	557	3.0
N-BUTANOL	663	1.0	613	3.0	685	6.0
1,4 DIOXANE	662	3.0	670	1.5	819	2.5
1-NITROPROPANE	754	1.0	670	2.0	819	1.5
PYRIDINE	754	2.0	680	3.0	801	0.5
2-METHYL-2-PENTANOL	781	1.0	680	1.0	841	1.0
1-IODBUTANE	885	0.2	891	1.0	921	1.0
OCTYNE	886	0.7	843	1.0	950	1.0
PENTANONE	662	1.0	542	1.0	551	4.0
CIS-HYDRINDAN	1006.7	2.0	1018.0	2.0	1215.6	2.0

8.3 Gradient loaded columns:

8.3.1 OV-17 Gradient columns, (Table 37)

Two columns were prepared in the 2-5-10% OV-17 and the 10-5-2% OV-17 configurations. In Table 37 the Retention Indices values on these two columns are compared to those on 2%, 5%, and 10% OV-17 conventional columns. Under the same chromatographic conditions the behavior of the 2% column indicated some solid support activity which caused the RI values to be higher. In general, the two gradient columns showed behavior different from that on the 2, 5 or 10% columns.

8.3.2 Conclusions based on the OV-17 gradient loaded columns:

1. The percent loading variation had a significant effect on the retention index values for various organic substance groups used as testing probes.
2. Reversing the gradient gave different Retention Index values for aromatics, aromatic bases and branched alcohols.
3. At low loadings, less than 5%, the Retention Index values obtained were different from what McReynolds (47) reported at 20% loading. This deviation was also,

TABLE 37

COMPARATIVE RETENTION INDICES OBTAINED FOR 2-5-10% OV-17 AND 10-5-2% OV-17 GRADIENT COLUMNS, AND 2%, 5%, AND 10% OV-17 CONVENTIONAL COLUMNS

SOLUTE	2%		2-5-10%		10-5-2%		5%		10%	
		RSD% ±		RSD% ±		RSD% ±		RSD% ±		RSD% ±
BENZENE	746	1.0	623	0.6	700	0.5	603	1.4	603	1.0
N-BUTANOL	746	1.1	745	0.5	715	0.5	654	1.3	657	0.4
1,4DIOXANE	802	0.5	824	0.5	812	0.6	661	1.0	682	0.4
1-NITROPROPANE	942	0.6	635	0.1	630	0.2	707	0.6	705	0.6
PYRIDINE	877	0.1	800	0.8	894	0.2	815	2.1	811	1.0
2-METHYL-2-PENTANOL	802	0.5	824	0.5	816	0.5	759	0.5	753	0.4
1-IODOBUTANE	937	0.5	947	0.5	947	0.2	897	1.0	844	0.1
PENTANONE	774	1.4	773	0.6	759	0.7	772	2.0	761	1.0
CIS-HYDRINDANE	1090	0.3	1091	0.9	1092	0.8	1058	0.2	1050	0.2

reported by Yancey (58) and other workers (49, 55, 59) for low loading columns.

8.3.3 1-2-3% QF-1 ON GC-Q, 80/100 mesh, (180 cm x 2 mm, I.D, glass tubing) (Table 38)

In Table 38 the RI values obtained for this gradient column were compared to those for the column containing the average gradient loading, 2%. The gradient column showed more selectivity for butanol, dioxane, iodobutane and pentanone test probes. Therefore, the gradient 1-2-3% QF-1 column will be more useful than a conventional column for separating chemical compounds similar to these test probes.

8.3.4 1-2-3% OV-225 ON CW-HP, 80/100 mesh, (180 cm x 2 mm I.D, glass tubing) (Table 39)

The RI values obtained for this column are shown in Table 39. RI values were slightly greater for benzene, butanol, dioxane, pyridine, 2-methyl-2-pentanol and pentanone on the gradient column. Again, the gradient

TABLE 38

CHARACTERIZATION OF A 1-2-3% QF-1 GRADIENT COLUMN, AND A 2% QF-1 CONVENTIONAL COLUMN ON GC-Q, 80/100 MESH, (180 cm x 2 mm I.D, GLASS TUBING) USING KOVATS RETENTION INDICES

SOLUTE	2%		1-2-3%	
		RSD% ±		RSD% ±
BENZENE	676	1.7	676	1.5
N-BUTANOL	865	1.7	873	1.0
1,4DIOXANE	840	0.4	889	1.2
PYRIDINE	970	0.4	922	0.6
2-METHYL-				
2-PENTANOL	908	0.5	911	0.7
1-IODOBUTANE	943	0.5	953	1.0
2-PENTANONE	943	0.5	953	1.0
CIS-HYDRINDANE	1067	1.0	1046	1.5