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DEVELOPMENT OF HIGH-PRESSURE LIQUID CHROMATOGRAPHIC TECHNIQUES --ETC(U)
APR 78 J R MATTHEWS, G D PIERCE F29601-76-C-0015

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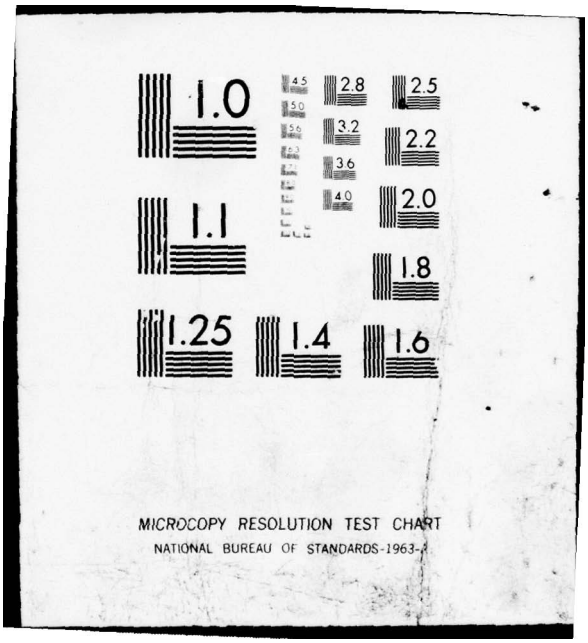
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DEVELOPMENT OF HIGH-PRESSURE LIQUID CHROMATOGRAPHIC TECHNIQUES (PHASE II)

LEVEL III

JAMES R. MATTHEWS
GLENN D. PIERCE

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ERIC H. WANG CIVIL ENGINEERING RESEARCH FACILITY
UNIVERSITY OF NEW MEXICO
P.O. BOX 25, UNIVERSITY STATION
ALBUQUERQUE, NEW MEXICO 87131

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CIVIL AND ENVIRONMENTAL ENGINEERING DEVELOPMENT OFFICE
(AIR FORCE SYSTEMS COMMAND)
TYNDALL AIR FORCE BASE
FLORIDA 32403

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The Organics-Carbon Adsorbable minifilter procedure was used to recover refractory organics from treated wastewater. Ten carbon chloroform extracts were recovered with this procedure. A parallel run using pulsed versus nonpulsed carbon beds was made to compare the recovery efficiencies of the beds. Five parallel runs were made to compare the ability of activated carbon and XAD-2 macroreticular resin to concentrate trace organics. An improved recovery system was developed and evaluated; this system recovered a greater organic mass per gram of carbon and per unit volume of sample than the Organics-Carbon Adsorbable

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minifilter and gave measurable recoveries in 24 hours. The carbon and resin extracts were separated and characterized by reverse-phase and gel permeation high-pressure liquid chromatography. The ultraviolet spectra thus obtained appeared independent of plant operating conditions except for one well resolved ultraviolet peak observed only in extracts recovered from nonchlorinated effluents. This material was collected in microgram amounts; however, this was insufficient for infrared and mass spectroscopy identification. Nonvolatile total organic carbon was used as the parameter to evaluate organic breakthrough of the adsorption columns as well as to perform mass balances through the adsorption systems. Correlations of carbon-chloroform extract, nonvolatile total organic carbon, and chemical oxygen demand were developed. An experimental beta-induced luminescence detector developed in previous research was evaluated. Elemental analyses of extracts and a mass balance through the high-pressure liquid chromatography system were accomplished.

PREFACE

This report documents work performed during the period July 1976 September 1977 by the University of New Mexico under contract F29601-76-C-0015 with the Air Force Civil Engineering Center, Air Force Systems Command, Tyndall Air Force Base, Florida 32403. This study was performed under program element 63723F JON 21032C44. The project officer was Major Michael G. MacNaughton of the Environmental Sciences Division, Civil and Environmental Engineering Development Office, ADTC, Tyndall Air Force Base, Florida 32403.

This report has been reviewed by the Information Officer and is releasable to the National Technical Information Service (NTIS). At NTIS it will be available to the general public, including foreign nations.

This report has been reviewed and is approved for publication.

MICHAEL G. MACNAUGHTON
Major, USAF, BSC
Chief, Environmental Sciences

PETER A. CROWLEY
Major, USAF, BSC
Director of Environics

JOSEPH S. PIZZUTO
Colonel, USAF, BSC
Commander

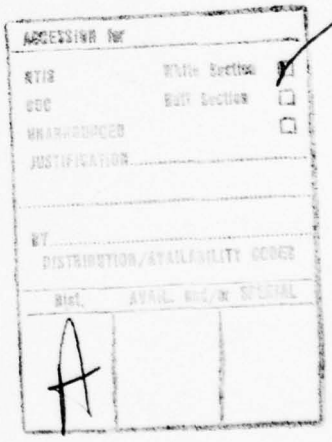

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TABLE OF CONTENTS

Section	Title	Page
I	INTRODUCTION	1
II	LITERATURE REVIEW	5
	Characterization of Treated Effluents	5
	Analysis and Identification of Low Molecular Weight Compounds	7
	Analysis and Identification of High Molecular Weight Compounds	8
III	RECOVERY OF ORGANICS AND CHARACTERIZATION OF TREATED WASTEWATER	11
IV	DEVELOPMENT OF AN IMPROVED RECOVERY SYSTEM	25
V	EXTRACT CHARACTERIZATION	33
	Preparation of Extracts for Chromatography	33
	Operating Procedures for the Waters ALC200 Series High-Pressure Liquid Chromatography System	33
	Reverse-Phase Separation With Single Eluent	36
	Microstyragel Separation	38
	Beta-Induced Luminescence Detector	39
	Gradient Elution With Reverse-Phase Column	41
	Characterization by Serial Reverse-Phase HPLC-GPC	41
	Elemental Analysis	44
	Spectrographic Analysis	45
VI	CONCLUSIONS AND RECOMMENDATIONS	49
REFERENCES		51
BIBLIOGRAPHY		53
APPENDIX A	REVERSE-PHASE SPECTRA	55
APPENDIX B	GEL PERMEATION SPECTRA	71
ABBREVIATIONS, ACRONYMS, AND SYMBOLS		81

LIST OF FIGURES

Figure	Title	Page
1	Extended-Aeration, Activated-Sludge Treatment Plant	11
2	NVTOC Versus Time (Parallel Carbon Resin)	16
3	Carbon Chloroform Extract Correlations	23
4	NVTOC Versus COD Correlations	23
5	Activated Carbon Kinetics	26
6	Improved Recovery System (Activated Carbon)	28
7	Improved Recovery System (Column Effluent NVTOC)	29
8	Gel Permeation Spectra - CCE - Parallel Run 3	30
9	Gel Permeation Spectra - CCE - Improved Recovery System 2	31
10	Gel Permeation Spectra - Resin Extracts - Improved Recovery System 2	32
11	Beta-Induced Luminescence Detection Spectrum	40
12	Gradient Elution and Isocratic Chromatograms for Carbon Chloroform Extract (Parallel Run 3)	42
13	Gradient Elution for Resin Extracts (Parallel Run 3)	43
14	Infrared Spectrum of Isolated Reverse-Phase Peak	46

LIST OF TABLES

Table	Title	Page
1	Sample Collection Data	13
2	Effluent CCE, COD, and NVTOC for Activated Carbon Runs	14
3	Effluent RErE, RAcE, and NVTOC for Nonparalleled Resin	14
4	Effluent CCE, RErE, RAcE, and Sample Characterization for Parallel Runs	15
5	Treated Wastewater Characterization	21
6	Relationships Between Effluent Parameters	22
7	Elemental Analyses	44

SECTION I
INTRODUCTION

BACKGROUND

The broad scope of Air Force activities demands that both industrial and domestic wastewaters be treated and discharged in accordance with Federal or more stringent regional standards and local industrial waste ordinances. The analysis of most metals, independent of oxidation state and bonding, may be accomplished routinely; however, the environmental significance of a substance often requires a determination of the chemical nature of the wastestream components with which the substance is associated in solution or to which it is bonded.

In traditional sanitary engineering practice, the organic content of domestic secondary treatment plant effluents is described in terms of Biochemical Oxygen Demand (BOD), Chemical Oxygen Demand (COD), or Total Organic Carbon (TOC). These are useful parameters for plant design and control, but they disclose little information concerning the organics or inorganics present in a flow. In selecting a wastewater treatment process, it must be determined whether it will be more appropriate to lower carbonaceous oxygen demand or to reduce phosphorous, nitrogen, or heavy metal content; therefore, a knowledge of the compounds present in the organic matrix, or of their general character, is desirable.

Various Environmental Protection Agency, state, and university laboratories are attempting to further identify the specific nature of the organic carbon by gas chromatography/mass spectrography and other sophisticated analytical procedures. These investigations are directed toward establishing the prevailing levels of pesticides, hydrocarbons from industrial wastes, and naturally occurring trace organics. In recent years, consideration has been given to halo organics, which form a very large class of organic compounds that may be persistent, bioaccumulable, and/or toxic on a short- or long-term basis.

Concern for the nature and amount of trace organic substances in finished drinking water and water sources is now evident. A high degree of sophistication is required to analyze the spectra of organics present in these waters.

Reference 1 presents the Organics-Carbon Adsorbable (O-CA) procedure as a tentative standard for evaluating the gross organic content of raw and finished waters. Although it is recognized that the activated carbon medium recovers only a portion of the naturally occurring trace organics, the method does provide enough information to signal a degradation in the quality of raw water, water purification, or water distribution, or a potential hazard from unsuspected chemical content.

An efficient recovery of trace organics from aqueous systems has also been accomplished by weakly bonding the solute onto polymer resins, which are later rinsed with selected organic solvents to obtain a gross extract. Despite rather intensive effort in this area of investigation, ion exchange is generally not recognized as the method of choice for concentrating a broad range of trace organic materials. In general, however, the resin procedure is simpler, less time consuming, and less dependent upon the judgment of the analyst than the activated carbon recovery procedure.

Studies performed on natural and finished waters show that both resin and activated carbon recovery procedures are specific in their ability to concentrate or remove organics. However, limited data are available for estimating the relative fractions of the organics present in the effluent from an activated sludge facility that are recovered as resin or activated carbon extracts. Also, the development of correlations between extract concentration, COD, and TOC has not been reported for a functioning activated sludge plant that treats primarily domestic sewage.

Another approach to analyzing the organics refractory to secondary biological treatment is to introduce the aqueous effluent sample directly into a high-pressure liquid chromatograh (HPLC) fitted with appropriate columns and detectors to separate and identify the components of the complex mixture. Most HPLC systems are very versatile in terms of column capabilities, solvents,

Reference

1. *Standard Methods for the Examination of Water and Wastewater*, 14th edition, American Public Health Association, American Water Works Association, and Water Pollution Control Federation, Washington, D.C., 1975.

temperature, and mode of operation. Unlike gas/liquid chromatography, HPLC can often be employed to separate high molecular weight polymer substances, which may then be identified or characterized by other techniques.

OBJECTIVES

The following were the objectives of this project: 1) development of HPLC techniques for the separation and measurement of refractory organics in biologically treated effluents, 2) evaluation of the capability of activated carbon and macroreticular resin media for recovering residuals from the effluents, 3) characterization of the effluents by conventional wastewater parameters, 4) correlation of conventional parameters with carbon and resin recoveries, and 5) characterization and identification of selected fractions.

SCOPE

Ten carbon chloroform extracts (CCE) were recovered from the effluent of a secondary treatment plant. A carbon mass balance employing total organic carbon as the major parameter was used. The capability of the minisampler procedure for providing a meaningful quantitative estimate of low turbidity secondary effluents was evaluated. The work also included fractionation of the organics by solvent extraction, gel-permeation chromatography, and functional group analysis.

Five of the extracts were recovered by carbon adsorption and five by parallel ion exchange recovery on XAD-2. The quantity of carbonaceous organics passing through the systems was measured by effluent TOC values. The work also included development of an optimized activated carbon recovery system for refractory organics, which uses a significantly smaller volume of sample and less active carbon. Continued effort was made on the fractionation of organics by HPLC, gel-permeation chromatography, and functional group analysis.

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SECTION II
LITERATURE REVIEW

This literature survey presents studies pertaining to the characterization of treated effluents by traditional and more sophisticated procedures and to the analysis of wastewater samples and extracts by the HPLC and the Gas Chromatography/Mass Spectroscopy (GC/MS) techniques. Section IV of Reference 2 contains a summary of procedures for the recovery and characterization of trace organics.

CHARACTERIZATION OF TREATED EFFLUENTS

The process behavior of two paper mill treatment facilities with different systems is reported by Keith (Reference 3). The author states: "To our knowledge this study represents the first attempt to characterize a wastewater chemically, trace the dissolved volatile organics through the treatment system, and correlate this information with the traditional collective pollution parameter measurements (BOD, TOC)." Total fatty acids in the effluent increased 17 percent with biological treatment, and the total number of fatty acids increased relative to the raw wastewater. Direct solvent extraction with chloroform was employed to concentrate the organics from the sample stream.

Dunlap, et al. (Reference 4) concentrated organic pollutants from a landfill well by means of XAD-2, Polyamide Woelm propanol-water (PAW-PW), PAW-2-propanol (PAW-P), carbon chloroform extracts (CCE), and carbon

References

2. Caton, Jr., Roy D., Matthews, James R., and Walters, Edward A., *Development of High Pressure Liquid Chromatographic Techniques*, AFCEC-TR-76-26, Air Force Civil Engineering Center, Tyndall Air Force Base, Florida, 1976.
3. Keith, L. H., "GC/MS Analyses of Organic Compounds in Treated Kraft Paper Mill Wastewaters." In L. H. Keith (Ed.), *Identification and Analysis of Organic Pollutants in Water*, Ann Arbor Science Publishers, Inc., Ann Arbor, Michigan, 1976.
4. Dunlap, W. J., et al., "Isolation and Identification of Organic Contaminants in Ground Water." In L. H. Keith (Ed.), *Identification and Analysis of Organic Pollutants in Water*, Ann Arbor Science Publishers, Inc., Ann Arbor, Michigan, 1976.

alcohol extracts (CAE). Each of the adsorbents was found to have both advantages and disadvantages. PAW was particularly effective for recovery of intermediate to high molecular weight compounds containing phenolic hydroxyl groups, and it was as effective as XAD-2 for recovery for all compounds tested which had a molecular weight greater than 200. XAD-2 was far superior to PAW for the lower molecular weight compounds. Activated carbon appears to be much more effective than either PAW or XAD-2 for removal of carboxylic acids from solutions of pH greater than 6. Also, it was found that although the first column in the series of two XAD-2 or PAW columns appeared to recover most of the organic matter that was present, significant quantities of material were present on the second column in the series of two carbon columns.

The standard O-CA minifilter and a step-flow reactor were used by Hewitt (Reference 5) to characterize four shallow Rio Grande Valley wells for trace organics. Two wells exceeded the recommended value for CCE in drinking water (0.7 mg/ℓ) with concentrations of 1.27 and 0.87 mg/ℓ. The CCE recovered by the O-CA minisampler showed significant correlation with well depth, nonvolatile total organic carbon (NVTOC), and total and fixed residue.

Chian and DeWalle (Reference 6) characterized the soluble organic matter in leachate collected below solid waste fills by ultracentrifugation, ultrafiltration, gel permeation, gas chromatography, and specific chemical analyses. After the leachate sample had been subjected to 30,000 rpm for 30 minutes, the largest organic fraction consisted of free fatty acids capable of permeating a 500-molecular weight ultrafiltration membrane. The authors state that their results agree with those of less comprehensive studies of humic substances in soil or water. They state that samples from widely different environments indicate a similar distribution of specific organics in the different molecular-weight fractions. This uniformity implies that similar processes control the composition of organics in natural environments.

References

5. Hewitt, Jr., J. L., *The Development and Characterization of a Step-Flow Reactor for the Recovery of Trace Organics*, MS Special Problem, Department of Civil Engineering, University of New Mexico, Albuquerque, New Mexico, 1976.

6. Chian, E. S. K., and DeWalle, F. B., "Characterization of Soluble Organic Matter in Leachate," *Environmental Science and Technology, Vol. 11*, p. 158, 1977.

The apparent molecular weights of organics present in the effluent of a trickling filter and contact stabilization activated sludge plant were investigated by Sachdev, et al. (Reference 7). The effluent was filtered through 0.45-micrometer membrane filters, mechanically freeze dried, and resolved on G-10, G-15, and G-25 Sephadex gels. The organic carbon concentrations in the samples and fractions were determined with a carbonaceous analyzer. It was concluded that no organics of apparent molecular weights between 1,500 and 5,000 existed in the two wastewater effluents investigated.

ANALYSIS AND IDENTIFICATION OF LOW MOLECULAR WEIGHT COMPOUNDS

Although it is generally agreed that the bulk of organic matter—over 75 percent in most waters—is nonextractable, nonvolatile, and mostly nongas-chromatographable, the most successful work in identifying and quantifying specific soluble organics in municipal wastewaters has involved the application of GC/MS and GC systems equipped with capillary columns.

Garrison, et al. (Reference 8) studied the nature of the residues from parallel activated sludge and physical-chemical pilot-plant treatment systems to identify extractable volatile organics in representative raw and treated domestic wastewaters. Samples were collected in glass containers, adjusted to pH 4 to 5, and stored for 1 to 3 days at 4°C before extraction with methylene chloride. The appropriate fractions were then partitioned and analyzed by GC/MS as concentrated organic neutrals, methyl esters, and organic bases. Eighty volatile organic substances were identified in the raw and treated wastewaters, and the authors concluded that these methylene chloride-extractable compounds constitute less than 25 percent of the total organic components of raw and treated domestic waste.

References

7. Sachdev, D. R., et al., "Apparent Molecular Weights of Organics in Secondary Effluents," *Journal of the Water Pollution Control Federation, Vol. 48*, p. 570, 1976.

8. Garrison, A. W., et al., "GC/MS Analysis of Organic Compounds in Domestic Wastewaters." In L. H. Keith (Ed.), *Identification and Analysis of Organic Pollutants in Water*, Ann Arbor Science Publishers, Inc., Ann Arbor, Michigan, 1976.

Giger, et al. (Reference 9) employed similar techniques for enriching and identifying substances in effluents that were filtered through fritted glass, solvent-extracted with ethylene chloride, and split into three fractions by adsorption-column chromatography on silica. Gas chromatography and GC/MS systems with glass capillary columns were used for analyses. These authors state that studies of chemical ecology necessitate analyses for single constituents; however, since environmental samples are of extraordinarily high compositional complexity and single components occur in trace quantities, very efficient enrichment, separation, and detection techniques are needed. Further, comparisons with TOC values of primary and secondary effluents showed that only 5 percent of the organics are amenable to gas chromatography.

ANALYSIS AND IDENTIFICATION OF HIGH MOLECULAR WEIGHT COMPOUNDS

The analytical technique that provides the most promise for determining the specific organic compounds present in various waste effluents and natural waters is high-resolution liquid chromatography (HRLC). Pitt, et al. (Reference 10) concentrated wastewater effluent 5- to 100-fold in a vacuum still or rotary evaporator and then lyophilized the 100 mℓ to an unreported fraction. Before the concentration steps were performed, the sample was filtered through a 0.45-micrometer membrane filter and was then passed through weak cation exchange beads in the H^+ form, which acidified it and reduced the quantity of precipitated inorganic salts. The HRLC system employed high-resolution anion exchange chromatography to analyze individual aquatic pollutants. The ion exchange columns were 150-cm lengths of No. 316 stainless steel tubing (0.22 to 0.62-cm i.d.) packed with strongly basic ion-exchange resin. The ultraviolet (UV) analyzer chromatograms were developed by elution with an ammonium acetate-acetic acid buffer solution (pH 4.4) whose acetate concentration gradually

References

9. Giger, W., et al., "Separation and Analysis of Refractory Pollutants in Water by High-Resolution Liquid Chromatography." In L. H. Keith (Ed.), *Identification and Analysis of Organic Pollutants in Water*, Ann Arbor Science Publishers, Inc., Ann Arbor, Michigan, 1976.

10. Pitt, Jr., W. W., et al., "Separation and Analysis of Refractory Pollutants in Water by High-Resolution Liquid Chromatography." In L. H. Keith (Ed.), *Identification and Analysis of Organic Pollutants in Water*, Ann Arbor Science Publishers, Inc., Ann Arbor, Michigan, 1976.

increased from 0.015 to 6.0 molar. The carbohydrate analyzer employed a sodium borate-boric acid buffer (pH 8.9) as the eluent. The boron concentration varied from 0.0845 to 0.845 molar during an analysis. Positive identification of compounds was accomplished by collection of the separate fractions from the chromatographic run; removal of the eluent by freeze drying; taking up the organics in methanol; and examination by UV spectrometry, gas chromatography, mass spectroscopy or other techniques. By use of the UV analyzer, over 100 compounds have been separated and detected in samples of primary effluents, and as many as 50 compounds have been detected in samples of concentrated secondary effluents. The authors emphasize that this procedure significantly reduces the likelihood that the nature of the chemical compounds will be altered since the samples are analyzed in the same media in which they are present in the environment.

A recent publication edited by Simpson (Reference 11) is a lucid, informative book on HPLC. The authors point out that a universal detection method is not available at present, so bulk or solute property detectors must be employed.

A major concern in the study of aqueous environments is the generation of carcinogenic or life-shortening halo organics during the chlorination of raw sewage, treated effluents, or drinking water. In an excellent review of halo organics in water supplies, McConnell (Reference 12) rationally defines the concern over these compounds in the environment. The etiology of chloroform in water has not been determined. However, Symons, et al. (Reference 13) state in an interim treatment guide for the control of chloroform and other trihalomethanes that chloroform concentrations are lower when chlorine disinfectant

References

11. Simpson, C. F., Editor, *Practical High Performance Liquid Chromatography*, Heyden and Son, Ltd., London, 1976.

12. McConnell, George, "Halo-Organics in Water Supplies," *Journal of the Institute of Water Engineers and Scientists* (G.B.), Vol. 30, No. 8, p. 431, 1976.

13. Symons, J. W., et al., *Interim Treatment Guide for the Control of Chloroform and Other Trihalomethanes*, Water Supply Research Division, MERL, Office of Research and Development, U.S. Environmental Protection Agency, Cincinnati, Ohio 45268, June 1976.

is applied to water having the lowest possible organic content. These findings should place increased significance on the ability to recover, separate, and identify those organic compounds discharged through secondary effluents into raw water supplies.

SECTION III

RECOVERY OF ORGANIC EXTRACTS AND CHARACTERIZATION OF TREATED WASTEWATER

The extended-aeration, activated-sludge treatment plant selected as the source for treated wastewater is illustrated in Figure 1. The following recovery runs were made using the O-CA minifilter (O-CA-mf) method and the XAD-2 macroreticular resin procedure: ten O-CA runs, one nonparalleled resin run, and five parallel O-CA and resin runs.

The first four O-CA-mf runs were made with the minifilter installed at the activated-sludge treatment plant. Several problems, including plant filter shutdown, sample solenoid valve failure, and varying minifilter flow rates, occurred during these initial runs. So the recovery procedure could be more closely monitored, all subsequent runs were carried out at the University of New Mexico Civil Engineering Department laboratories. Samples were collected from the Rio Rancho effluent wet well in 24-liter borosilicate glass carboys and

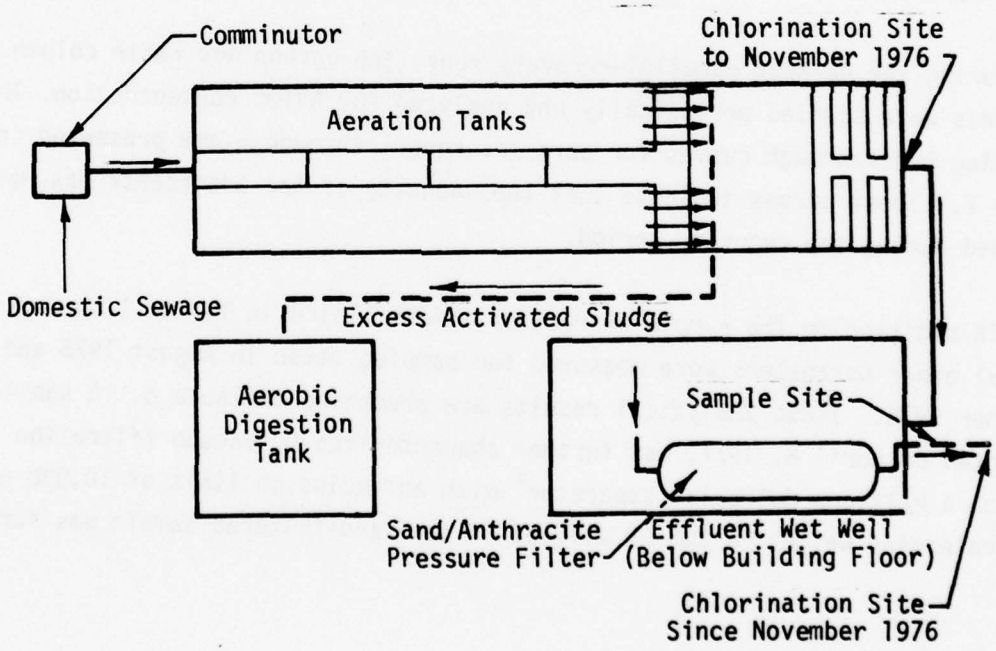

Figure 1. Extended-Aeration, Activated-Sludge Treatment Plant

were transported to the laboratory. Upon arrival, the samples were rapid sand filtered. The filter was constructed from a metal 5-gallon reagent drum with the bottom removed and a fine stainless steel screen soldered over the pour spout. The filter was filled with a 12-inch layer of sand that passed a 600-micrometer sieve and was retained on a 425-micrometer sieve. Before each sample was filtered, the filter bed was washed with cold tap water, stirred, and rinsed until the filter overflow was clear. The elapsed time from sample collection to the completion of filtration was approximately 1 hour. All samples except the first four O-CA samples were filtered in this manner.

The sample dates, flow rates, and multimedia filter and chlorination information are given in Table 1. The organic recoveries, as measured by CCE, resin ether extract (REtE), and resin acetone extract (RAcE), with corresponding NVTOC, COD, and residue data, are shown in Tables 2, 3, and 4.

Extraction blanks were run on the carbon and resin; the appropriate elution procedure was used for each. The value for the carbon chloroform blank was 2.6 mg per 70 g carbon. The values for the resin ether and resin acetone blanks were each less than 0.1 mg per 15 g resin.

During the 48-hour parallel-recovery runs, the carbon and resin column effluents were sampled periodically and analyzed for NVTOC concentration. The resulting breakthrough curves for parallel runs 1 through 5 are presented in Figure 2. These curves indicate that the capacity of the adsorbants was not exceeded during the recovery period.

In addition to the sample characterizations listed in Tables 1 through 4, several other parameters were measured for samples taken in August 1975 and December 1976. These analytical results are presented in Table 5. A sample collected on April 8, 1977, was further characterized by vacuum filtration through a Pellicon molecular separator[1] with an exclusion limit of 10,000 nominal molecular weight. A 200-μℓ aliquot of the sand-filtered sample was further

Footnote

[1]Millipore Corporation, Bedford, Massachusetts 01730.

TABLE 1. SAMPLE COLLECTION DATA

Run Number	Collection Date	Flow, Millions of Gallons Per Day	Plant Sand/Anthracite Filters Operational	Sample Chlorinated
Carbon 1	11-26-75	0.280	Yes	Yes
2	1-13-76	0.134	Yes	Yes
3	1-14-76	0.136	No	Yes
4	1-27-76	0.142	No	Yes
5	5-4-76	0.365	Yes	Yes
6	5-24-76	0.370	No	Yes
7	6-9-76	0.340	No	Yes
8	6-19-76	0.340	No	Yes
9	8-10-76	0.260	No	Yes
10	8-10-76	0.260	No	Yes
Resin 1	10-4-76	0.290	No	Yes
Parallel 1	12-12-76	0.330	No	No
2	1-10-77	0.340	No	No
3	1-18-77	0.350	No	No
4	2-1-77	0.340	No	No
5	3-26-77	0.350	Yes	No
Improved 1	5-2-77	0.380	Yes	No
2	6-1-77	0.370	Yes	No

TABLE 2. EFFLUENT CCE, COD, AND NVTOC FOR ACTIVATED CARBON RUNS

Run Number	CCE, mg/ℓ	COD, mg/ℓ	NVTOC, mg/ℓ
1	1.62	82	25.0
2	1.87	111	33.5
3	17.05	276	120.0
4	9.31	225	-
5	1.94	98	-
6	2.80	115	36.0
7	1.17	82	24.5
8	0.90	82	25.5

TABLE 3. EFFLUENT RErE, RAcE, AND NVTOC FOR NONPARALLELED RESIN

RErE, mg/ℓ	RAcE, mg/ℓ	COD, mg/ℓ	NVTOC, mg/ℓ
0.33	0.42	90.0	29.5

TABLE 4. EFFLUENT CCE, RErE, RAcE, AND SAMPLE CHARACTERIZATION FOR PARALLEL RUNS

Parameter	Parallel Recovery Runs									
	1		2		3		4		5	
	mg	mg/ℓ	mg	mg/ℓ	mg	mg/ℓ	mg	mg/ℓ	mg	mg/ℓ
CCE-mf	114.3	1.91	125.4	2.24	183.4	3.01	197.8	3.41	148.5	2.70
RErE	17.9	0.32	35.3	0.68	27.3	0.50	26.5	0.51	18.6	0.38
RAcE	14.3	0.26	17.7	0.34	29.6	0.54	25.5	0.49	14.7	0.30
COD		54		81		112		50		45
NVTOC		29		30		36		35		15
Residue,										
Total		534		-		534		488		816
Fixed		442		-		364		493		517
Volatile		92		-		170		95		299

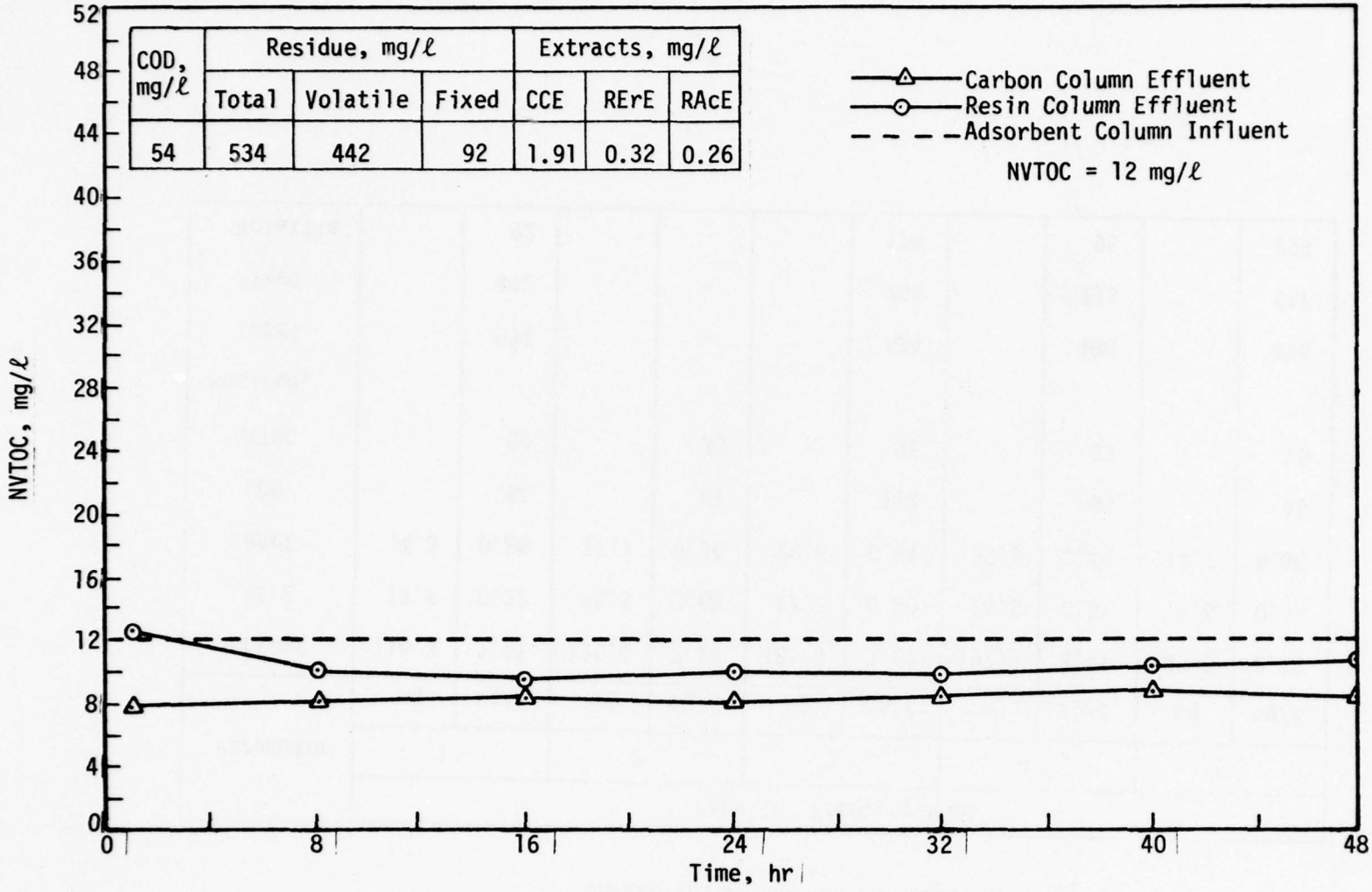

Figure 2. NVTOC Versus Time
(a) Parallel Run 1

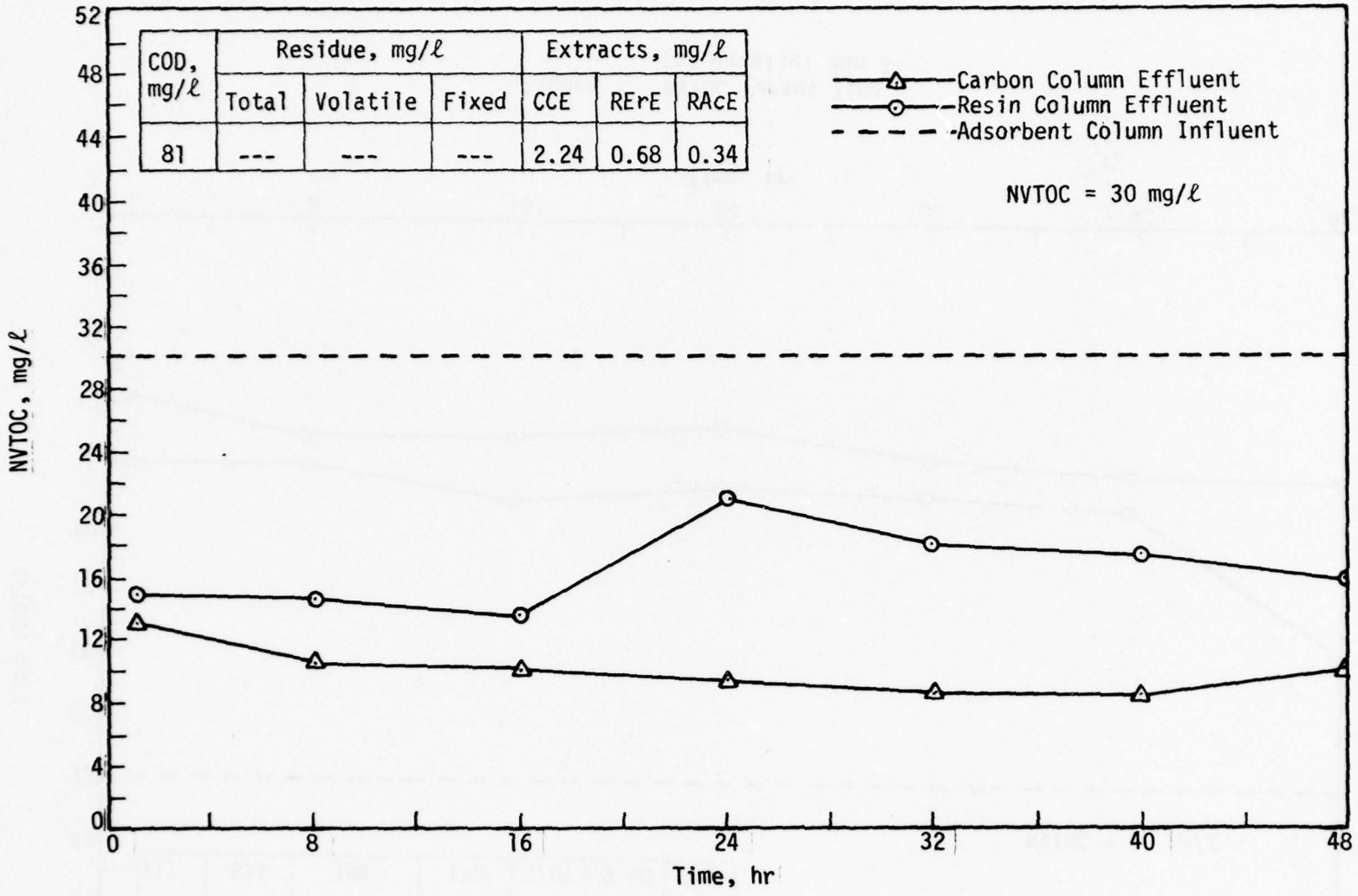

Figure 2. NVTOC Versus Time
(b) Parallel Run 2

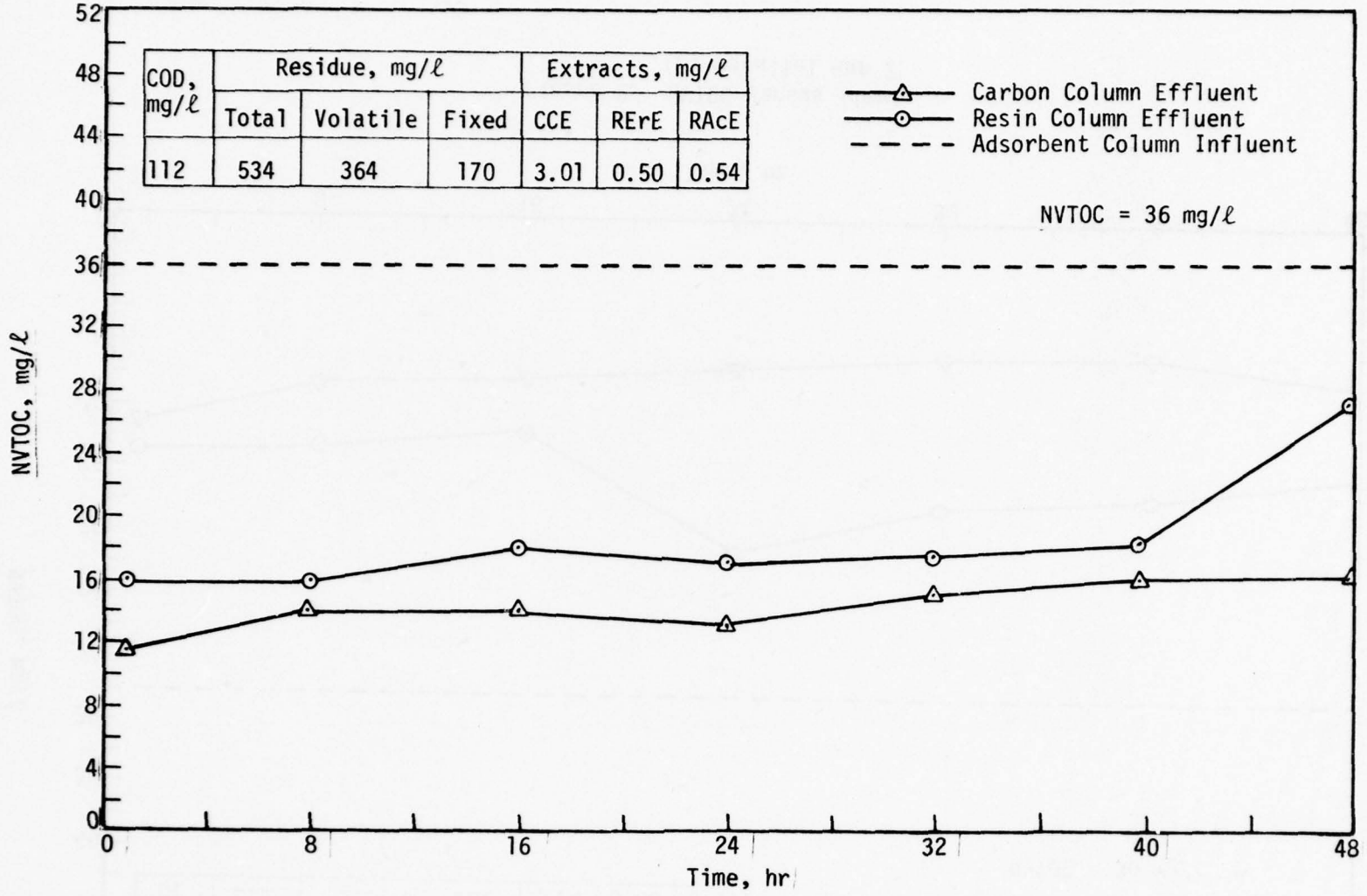

Figure 2. NVTOC Versus Time
(c) Parallel Run 3

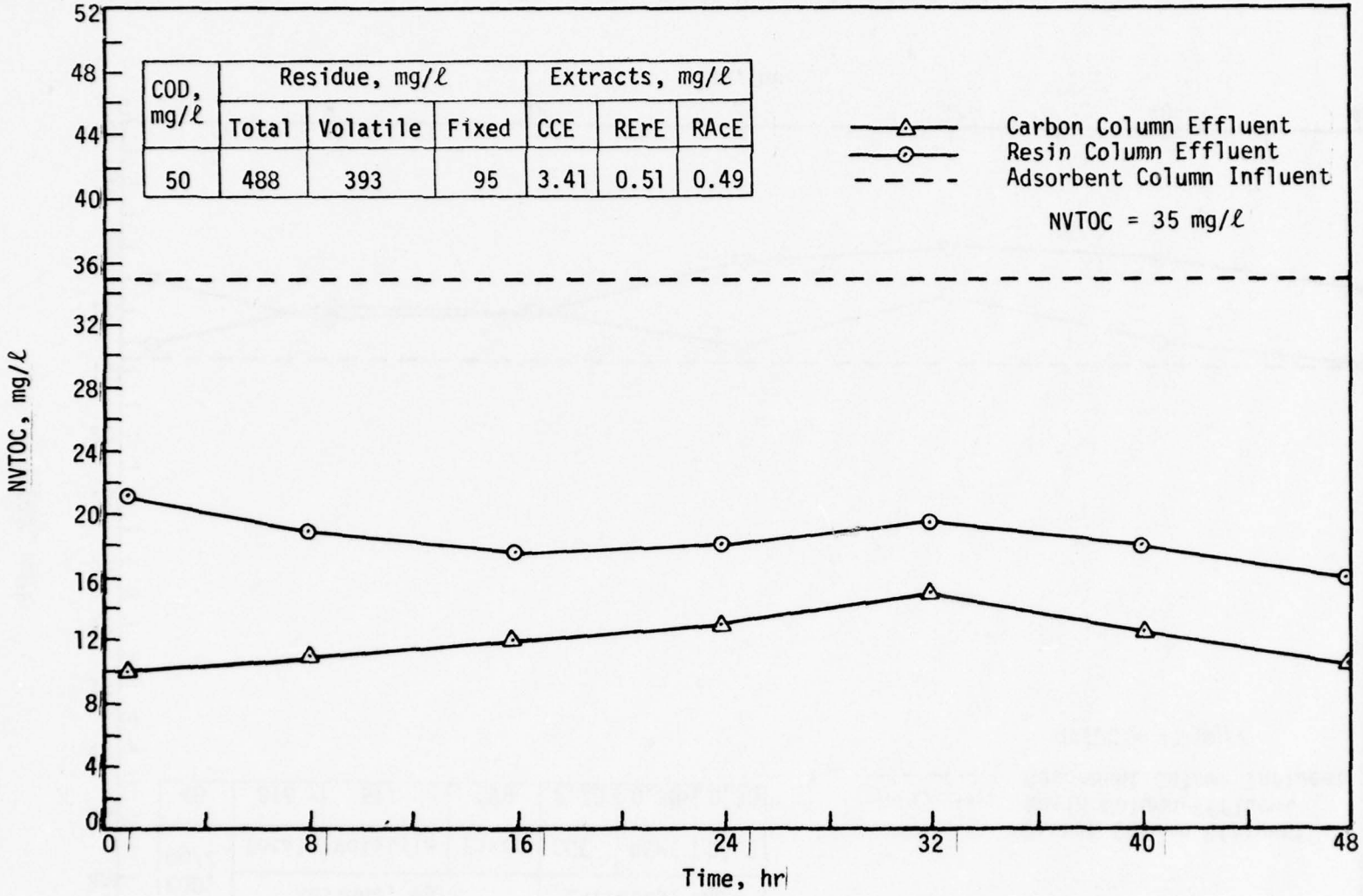

Figure 2. NVTOC Versus Time
(d) Parallel Run 4

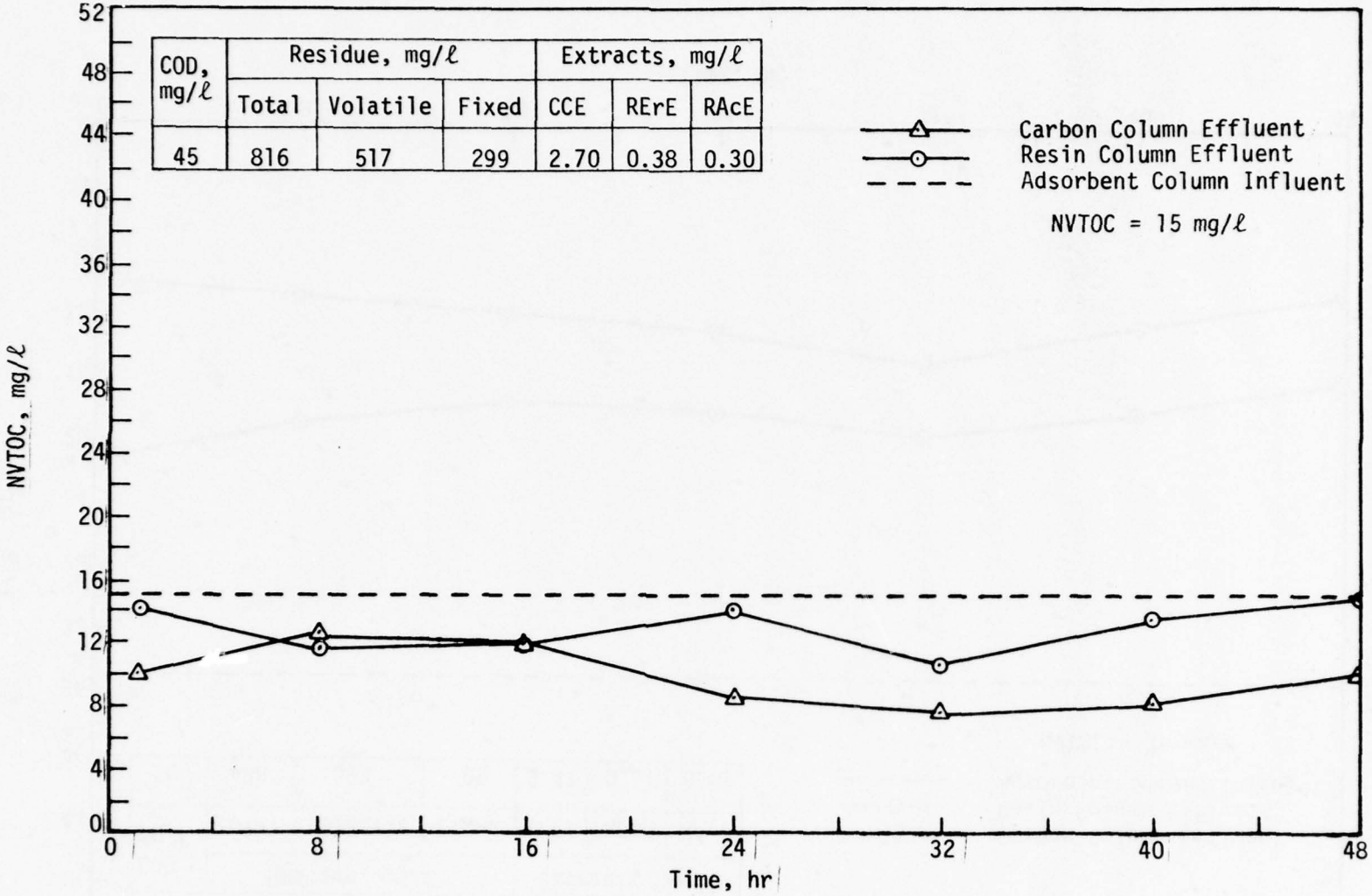

Figure 2. NVTOC Versus Time (Concluded)
(e) Parallel Run 5

TABLE 5. TREATED WASTEWATER CHARACTERIZATION

Parameter	August 1975	December 1976
pH	9.9	7.3
Suspended Solids, mg/ℓ	4	--
Turbidity, FTU	7	1.7
Specific Conductivity, μmhos/cm	820	610
Dissolved Solids, mg/ℓ	521	--
Sodium, mg/ℓ	114	--
Potassium, mg/ℓ	16.2	--
Calcium, mg/ℓ	15.5	--
Magnesium, mg/ℓ	3.4	--
Hardness, mg/ℓ as $CaCO_3$	95	--
Total Alkalinity, mg/ℓ as $CaCO_3$	130	70
Carbonate Alkalinity, mg/ℓ as $CaCO_3$	130	78
BOD_5, mg/ℓ	--	9
COD, mg/ℓ	--	54
Total Phosphorous, mg P/ℓ	11.5	4.3
Ortho Phosphate, mg P/ℓ	8.5	--
Chloride, mg/ℓ	81	--
Sulfate, mg/ℓ	96	--
Fecal Coliform, CFU/100 mℓ	0	0
Total Residue, mg/ℓ	--	534
Fixed Residue, mg/ℓ	--	442
Volatile Residue, mg/ℓ	--	92
Total Nitrogen, mg N/ℓ	9.6	--
Ammonia Nitrogen, mg N/ℓ	1.5	1.2
Nitrate Nitrogen, mg NO_3/ℓ	14.6	--

filtered through a 1-micron glass fiber filter. The Pellicon filter was immersed in the glass fiber filtered sample and vacuum was applied until a volume of 100 mℓ of filtrate was collected. The Pellicon filtrate was analyzed and was found to contain 80 percent of the COD and 85 percent of the NVTOC of the glass fiber filtrate. This result indicates that most of the organic material present is of nominal molecular weight (less than 10,000).

Linear regression analysis yielded relationships with regression coefficients of 0.9 or greater for the correlations tabulated in Table 6. The NVTOC-versus-COD correlation given in the last row of Table 6 was reported in a previous study (Reference 14) of activated sludge effluent from a different treatment plant. The similarity of the regression curves for the two sources of activated sludge effluent, the small plant used for this study treating 0.35 MGD and the other plant (Reference 14) treating more than 25 MGD, would indicate that these relationships may be valid for a broad range of activated sludge effluents. Efforts to correlate CCE with total, fixed, or volatile residue gave regression coefficients of less than 0.4 (not presented). The tabulated relationships are presented graphically in Figures 3 and 4.

TABLE 6. *RELATIONSHIPS BETWEEN EFFLUENT PARAMETERS*

Related Parameters	Relationships	Regression Coefficient	Sample Size
CCE Versus COD	CCE = 0.058 COD - 2.65	0.90	15
CCE Versus NVTOC	CCE = 0.141 NVTOC - 2.19	0.90	14
NVTOC Versus COD	NVTOC = 0.41 COD - 3.24	0.96	14
[a]NVTOC Versus COD	NVTOC = 0.45 COD - 2.01	0.98	16

[a]From Reference 14.

Reference

14. Pierce, G. D., *Pilot Plant Activated Sludge Study Using High Rate Trickling Filter Effluent*, MS Special Problem, Department of Civil Engineering, University of New Mexico, Albuquerque, New Mexico, 1975.

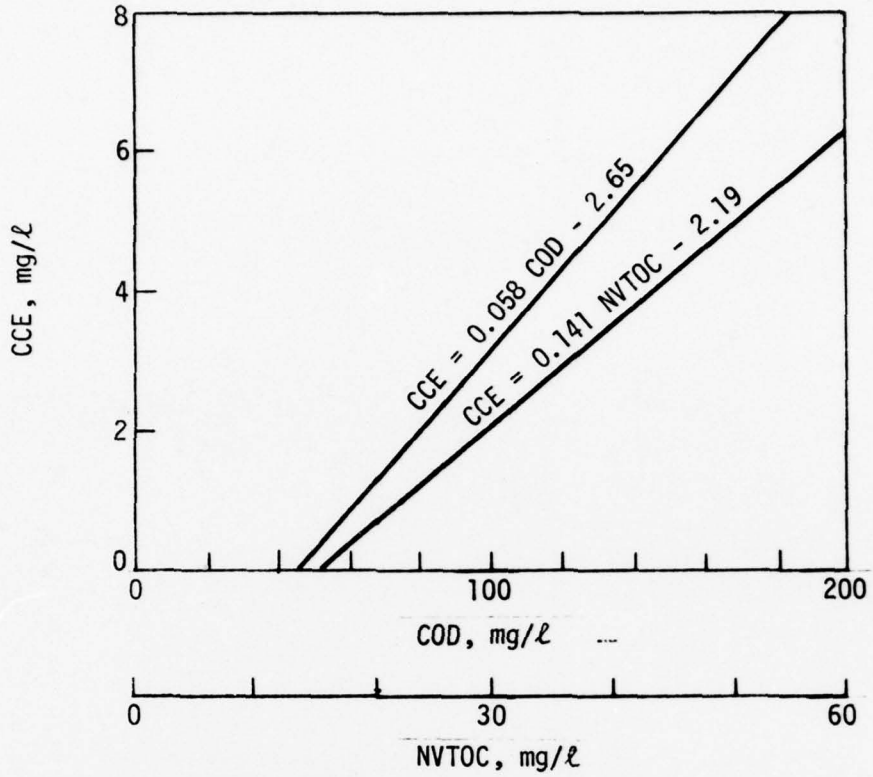

Figure 3. Carbon Chloroform Extract Correlations

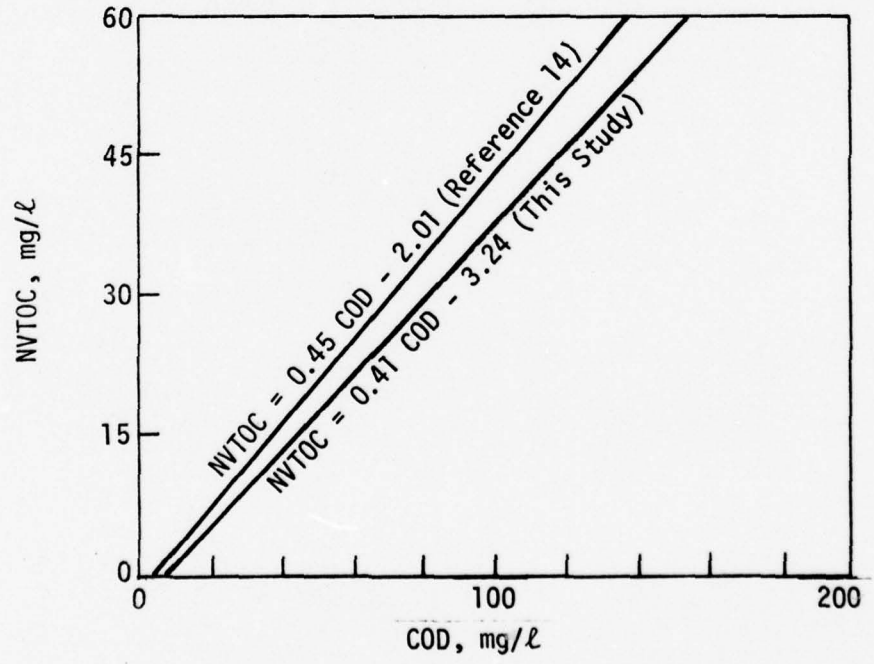

Figure 4. NVTOC Versus COD Correlations

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SECTION IV
DEVELOPMENT OF AN IMPROVED RECOVERY SYSTEM

The limitations of the O-CA-mf apparatus and procedure were apparent after the first recovery run. Since the minifilter is designed to recover trace organics from potable and natural waters, which should contain less than 0.7 mg/ℓ organics, the volume of sample must be large to recover an accurately weighable quantity. Also, the minifilter is not easily cleaned unless rinsing at a high rate of flow is satisfactory. The timer and solenoid assemblies are not standard items except in major metropolitan areas; this factor complicates the fabrication of the standard unit. These limitations and the low recovery efficiency of the unit (approximately 40 percent) were major reasons for seeking an improved system.

Preliminary efforts were directed toward determining whether a hydraulically pulsed carbon bed is significantly more effective than a nonpulsed carbon bed for recovering organics from biological effluents. The parallel recovery run in which two standard minifilters were used yielded CCE concentrations of 1.22 and 1.01 mg/ℓ for the pulsed and nonpulsed carbon beds, respectively. Because the minifilter column does not satisfy the 8-to-1 column-length-to-diameter ratio considered to be essential for many extraction and adsorption processes, the pulsing was eliminated and was replaced by a more slender column geometry.

To establish the ratio of activated carbon to total effluent volume through the unit, batch kinetic studies were performed with NVTOC as the control parameter (Figure 5). The O-CA-mf procedure specifies that 70 g of Filtrasorb 200[1] must be exposed to 60 liters of sample (a ratio of 1167 mg carbon per liter sample). The percent removal of NVTOC attained after 24 hours, compared to the NVTOC removals shown in Figure 2, indicates that the batch kinetic tests at 1000 mg carbon per liter reasonably simulated the standard minifilter characteristics. It is apparent that higher carbon-to-effluent-volume ratios yielded lower residue-NVTOC values, indicating greater recovery.

Footnote

[1]Calgon Corporation, Pittsburg, Pennsylvania.

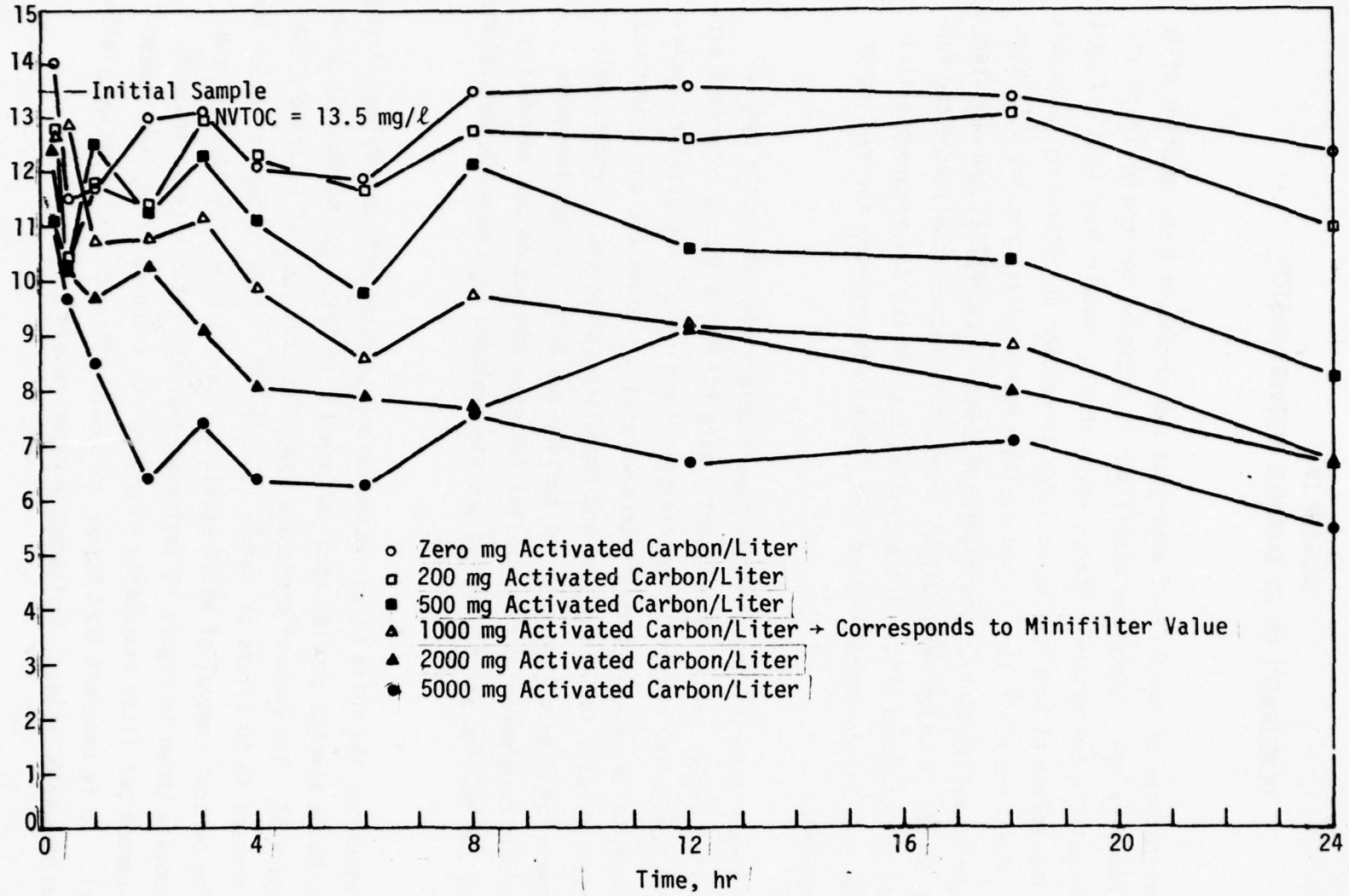


Figure 5. Activated Carbon Kinetics

The improved recovery system (IRS) (Figure 6) was fabricated of components carried by major chemical equipment suppliers. The basic system characteristics include the following features:

Carbon weight	- 35 grams
Sample volume	- 16 liters
Sampling time	- 24 hours
Column length:diameter	- 13.7:1
Tubing	- Teflon® except for silicon at pump
Flow rate	- 11.1 mℓ/min

The 0.25-inch o.d. rigid Teflon® tubing is connected at splices by a 3-inch section of 0.25-inch i.d. flexible Teflon® tubing, which provides an excellent low-pressure coupling. The unit is simple to maintain and operate, and the recovery of CCE and the residual NVTOC values (Figure 7) was significantly more desirable than that obtained with the standard minifilter. On the basis of the relative UV response of the HPLC spectra (Figures 8 and 9), one would expect a significant fraction of the additional components recovered by the IRS to be of low molecular weight and nonpolar in nature.

Efforts were also made to reduce the scale of the resin recovery system in order to lower the investment in solvents and test apparatus. The smaller unit, however, showed no obvious advantage in recovery performance (Figure 10) and proved operationally unreliable due to the low sample-flow rate and the small amount of extract recovered. The low recovery efficiency for NVTOC by the XAD-2 is apparent from the high NVTOC values in the sample effluent (see Figure 7).

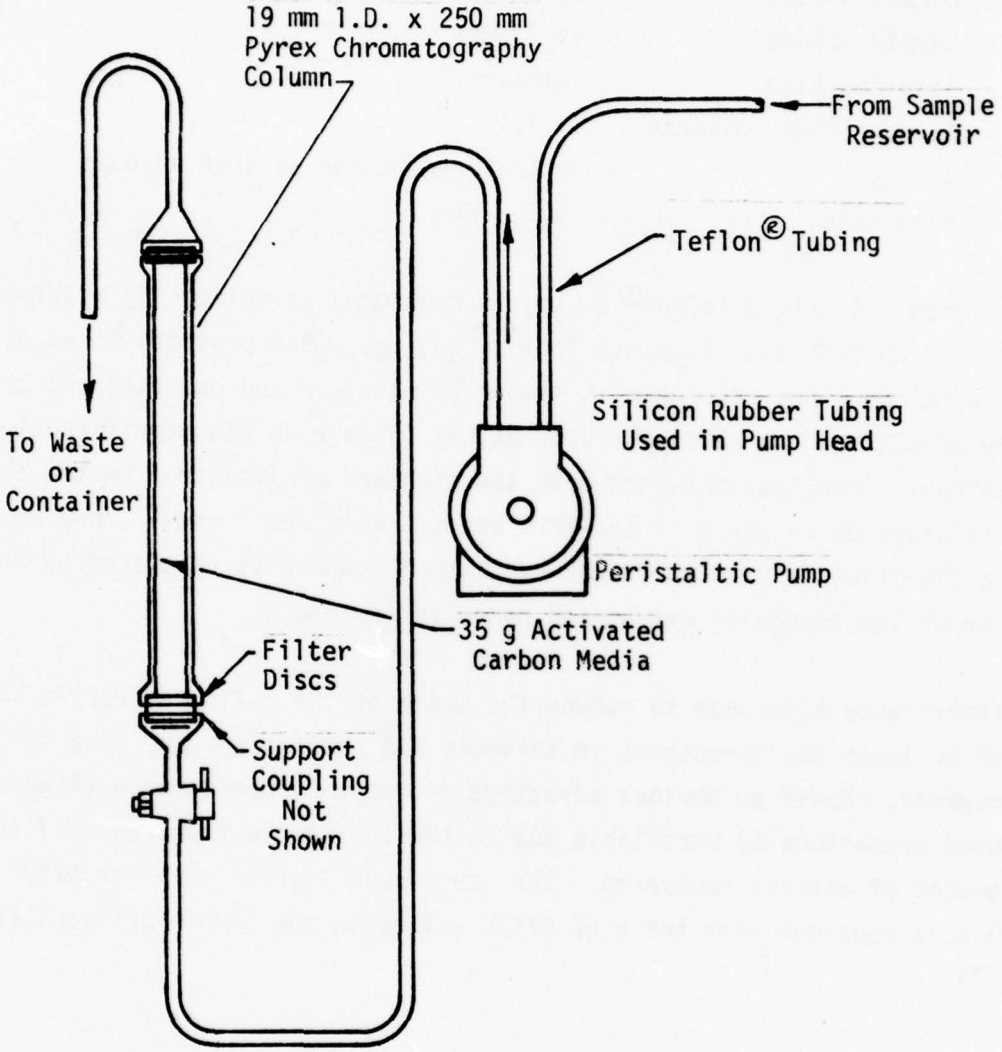


Figure 6. Improved Recovery System (Activated Carbon)

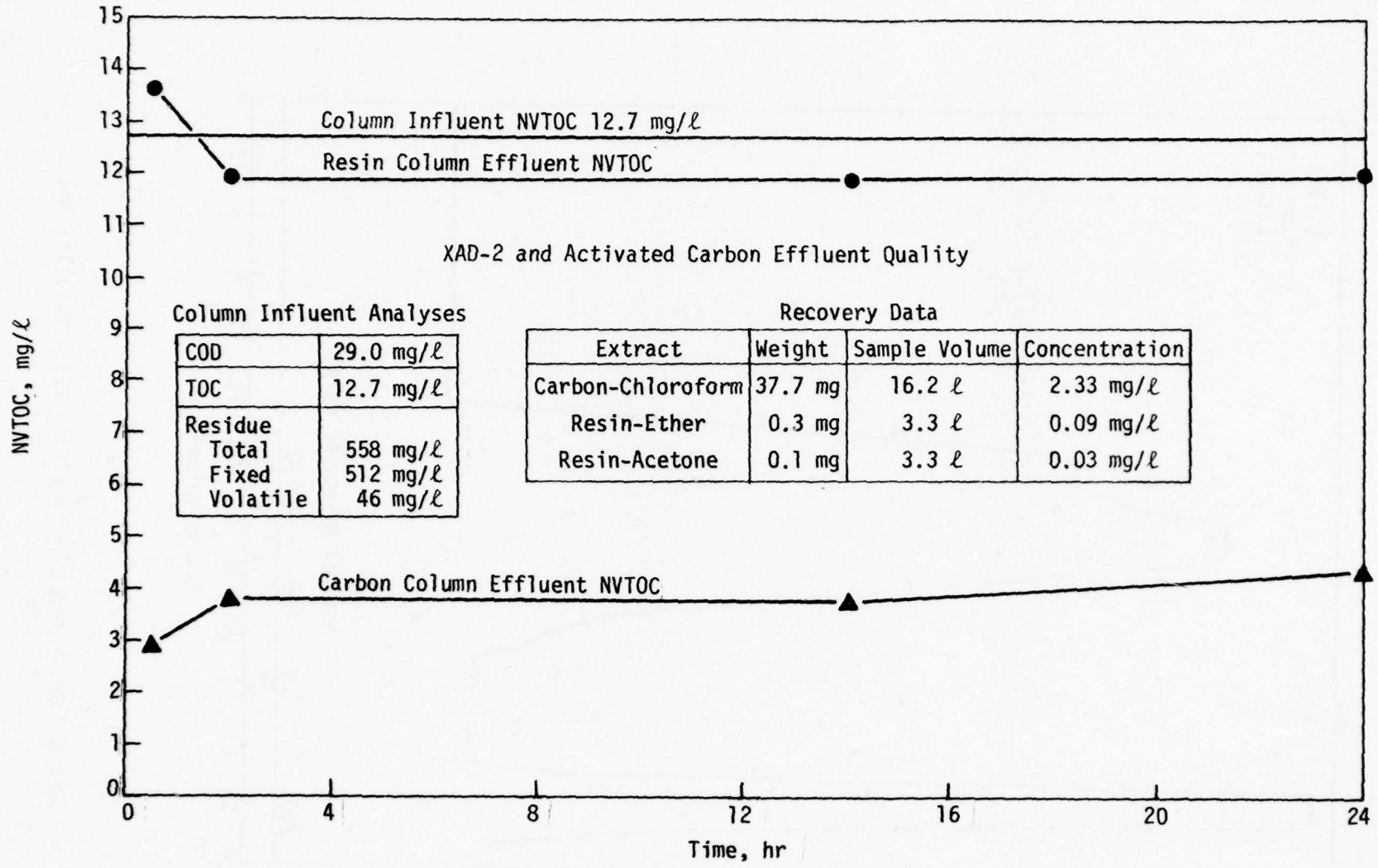


Figure 7. Improved Recovery System (Column Effluent NVTOC)

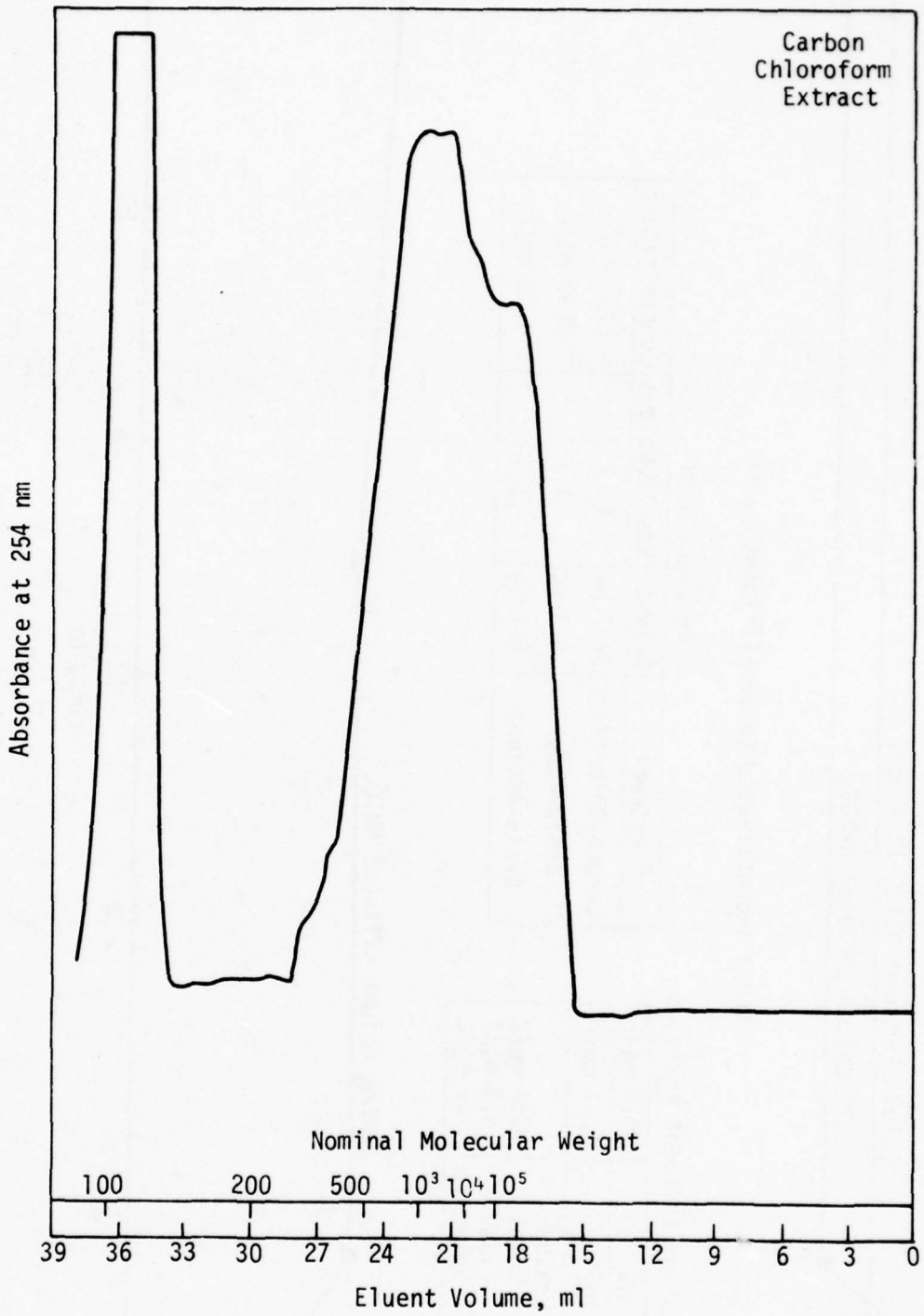


Figure 8. Gel Permeation Spectra - CCE - Parallel Run 3

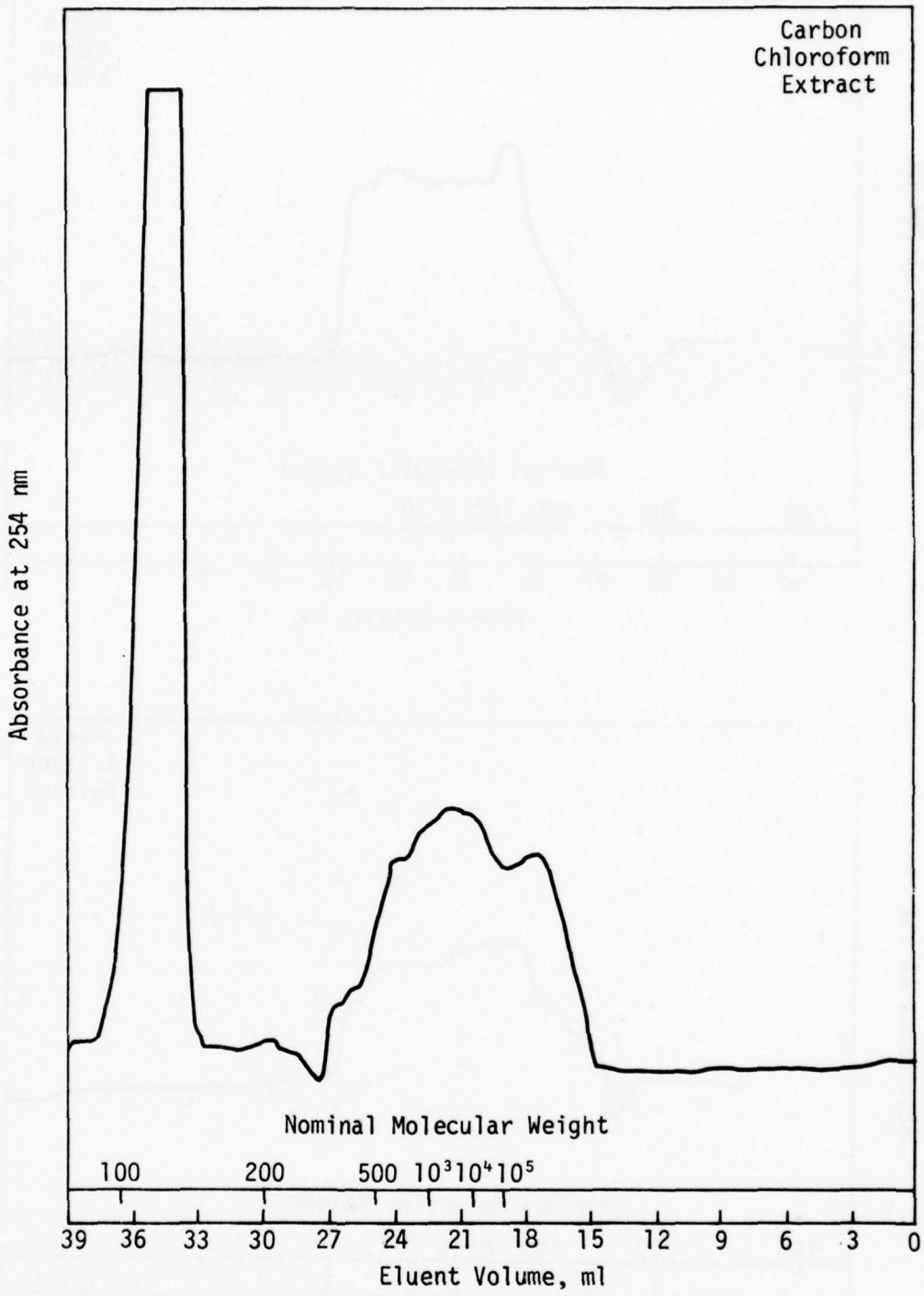
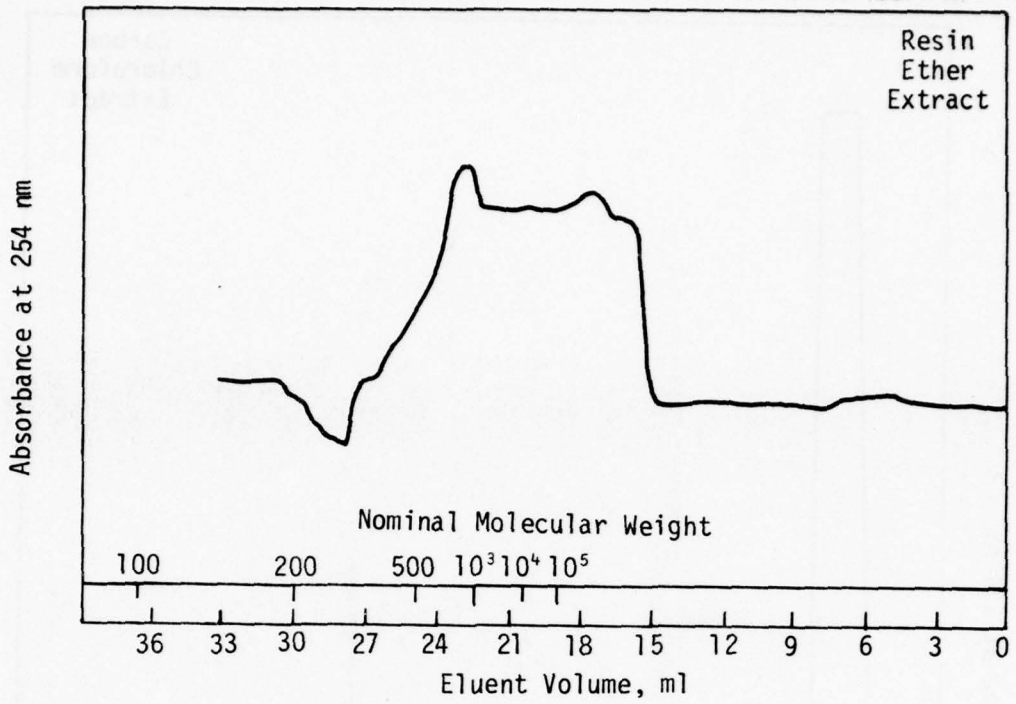


Figure 9. Gel Permeation Spectra - CCE - Improved Recovery System 2



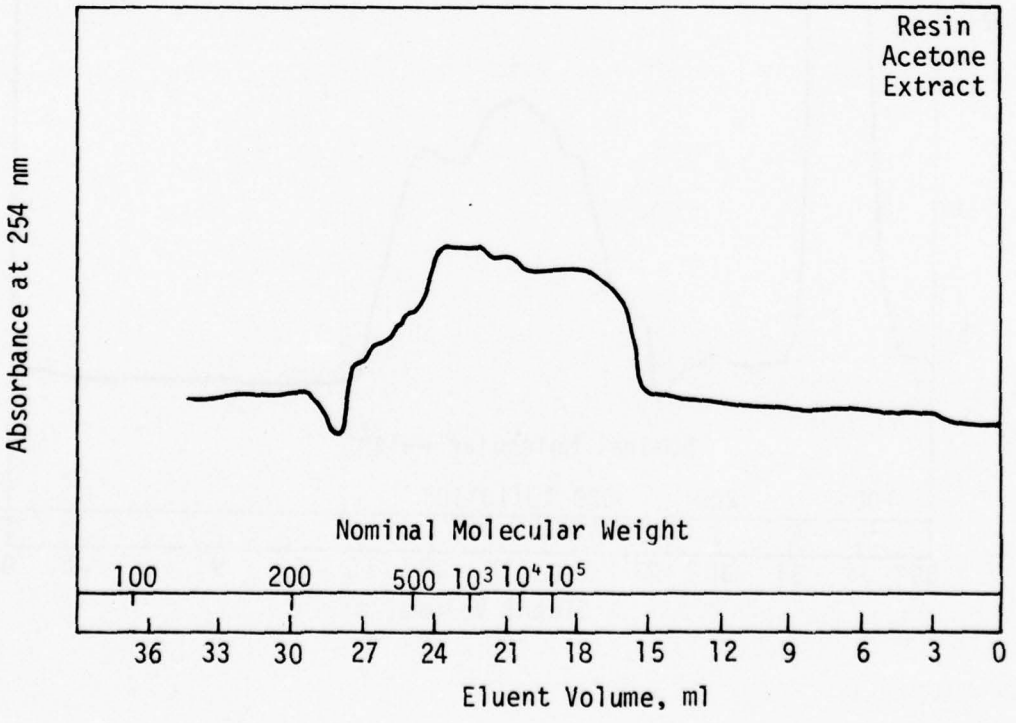


Figure 10. Gel Permeation Spectra - Resin Extracts - Improved Recovery System 2

SECTION V
EXTRACT CHARACTERIZATION

PREPARATION OF EXTRACTS FOR CHROMATOGRAPHY

All the dried carbon and resin extracts were dissolved in 10 mℓ of tetrahydrofuran (THF), except that for the CCE of run 3, an aliquot of approximately 52 mg was used. The sample solutions were filtered through a 1-micrometer Teflon® filter. A glass syringe was used to apply pressure. The Teflon® filters were mounted in a stainless steel holder. (Filters, syringe, and filter holder are manufactured by Millipore Corporation and supplied as a kit by Waters and Associates.) The samples were filtered into cleaned and fired (550°C, 1 hour) borosilicate glass vials fitted with 1/32-inch-thick Teflon® gaskets in the caps.

An attempt was made to dissolve minute aliquots of several extracts in methanol, but this resulted in the formation of a disperse, cloudy material which quickly plugged the Teflon® filters. This material presumably would also have clogged the injection syringe or the column heads, so only THF solutions of the extracts were used for chromatography.

OPERATING PROCEDURES FOR THE WATERS ALC200 SERIES HIGH-PRESSURE LIQUID CHROMATOGRAPHY SYSTEM

In general, the procedures recommended in the manufacturer's manuals were found to be suitable. However, intensive use of the chromatographic system led to the development of several procedural changes that improved the resolution, reproducibility, and baseline stability of the system.

Cleaning the System

Residues of previous solvents or samples may bleed into the system and give spurious peaks on the chromatogram. This may usually be avoided by a thorough cleaning with a highly efficient solvent such as THF. A short piece of tubing is installed in place of the column(s), and the solvent is pumped through the system at a high rate ($\geq$ 5 mℓ/min). Pump at least 25 mℓ to ensure that no traces

of previous solvents remain in the pump. Then pass eight to ten dead volumes of solvent through the sample injector the loading coils, and the refractometer reference cell. The sample loading port on the injector should be opened with the injector selector valve in the inject position. This allows solvent to flow from the port to clean the Teflon® septumless injector and the wire injector plug. Remove and reinsert the plug 6 to 8 times during this procedure.

If stable detector baselines are not obtained after two repetitions of this procedure, a 6N nitric acid wash may be used. The acid solution should not be pumped through the detectors, but only through the pump and injector. After 5 minutes, the acid should be thoroughly washed out with at least 200 mℓ distilled water.

Changing Solvents

In changing solvents, particular attention must be given to removing the old solvent from the HPLC system. If the old and new solvents are immiscible, one or more intermediate solvents must be used. If this step is ignored, minute droplets of emulsified solvent may be deposited in tiny spaces at fittings and may bleed slowly into the system. If the columns must remain installed and there is a question as to the miscibility of the two solvents, the solvent should be changed slowly; otherwise, excessive column pressure or heat of mixing may occur and reduce column efficiency and life. At least 5 volumes of new solvent must be passed through the injector and loading loops. The injection port should be opened under pressure as described under "cleaning the system."

Changing Columns

Column changing is no problem as long as the new column is compatible with the solvent in the system. If not, the old column is removed, and the change to a compatible solvent is made before the new column is installed. It may be necessary to pump 8 to 50 column volumes of solvent through the new column to achieve stable detector baselines.

Detection of Leaks

Even the most minute leak in the system will greatly decrease resolution and reproducibility. Significant leaks may not be detectable, especially if volatile solvents, such as THF, are in use. The pressure transducer built into the Model 6000A Solvent Delivery System is invaluable for leak detection. The manufacturer's manual describes how the transducer is to be used and how the recorded pressure data are to be interpreted. If a leak is indicated, it may be most easily located by operating the system at high pressure (> 3000 psi), without the column, and using a non-volatile solvent such as distilled water. A leak that was particularly difficult to find was finally located in the reference valve, where the impurities passed into and through the refractometer reference cell and then to the waste container several feet away from the valve. Care must always be taken that the reference valve is tightly shut.

Gradient Elution

Gradient elution demands extraordinary attention to system cleanliness and solvent purity. When the operator is using only a single pump and solvent, impurities present may bleed into the system indetectably and at a nearly constant rate. In gradient elution, each solvent is delivered by a separate pump at a constantly changing rate, and impurities present in either the pumps or the solvents appear as a sharply drifting baseline or as spurious peaks. A rapidly fluctuating baseline in gradient elution is due to refractive index changes in the mixed solvent. Such changes were found to occur where an elution scheme using a gradient from water to methanol was tried, but they were not observed in an elution gradient going from 20-percent water in methanol to pure methanol. The magnitude of such refractive index effects can be reduced by additional mixing of the solvents before they are injected. The mixing may be conveniently accomplished by routing the mixed solvent through the high-pressure flow-through filters on both pumps in series (ordinarily, flow is through only one of these filters). The second filter should be in line before the injector.

Gradient elution is much more sensitive than is a single solvent system to solvent degassing, either in the pumps or in the detector. Therefore, the solvents should be thoroughly degassed before they are used; stirring the solvents under the vacuum of an aspirator for 30 minutes was found to be adequate.

General Precautions

The highest standards of cleanliness must be applied to the handling of the sample injection syringe. Three rinses with THF, followed by three rinses with the solvent in use, between samples and at the beginning and end of a day, will give a stable baseline following an injection blank.

Directional changes in all tubing should be smooth and gradual. Sharp or irregular bends in the tubing result in localized turbulent flow conditions, mixing within the tubing, and subsequent peak broadening and loss of resolution on the chromatogram. Sharp bends in the Teflon® tubing of the UV detector were also observed to be sites of solvent degassing and attendent bubble problems. All tubing from the injector, to and from the column, through the detectors, and to the fraction collection point should be of the same internal diameter (0.09 inch). This system is normally assembled with larger tubing from the detector outlet to the collection port on the pump. Mixing that occurs in this larger line yields a complete loss of resolution of collected fractions. It also severely reduces the effectiveness of recycle. The best point for fraction collection was found to be at the discharge of the UV detector; therefore, the differential refractometer was not used.

REVERSE-PHASE SEPARATION WITH SINGLE ELUENT

At the beginning of the HPLC operations, both normal and reverse-phase columns were investigated. A normal-phase Porasil column gave marginal resolution (3 or 4 peaks) with THF as the mobile phase, but this column retained some highly polar components for over 50 column volumes. As a result, chromatogram times were extremely long, and random peaks occurred on subsequent

injections. For these reasons, and the low number of theoretical plates (< 1000) on the available Porasil column, normal-phase HPLC was not continued. A reverse-phase μC18[1] column with over 4000 theoretical plates gave chromatograms of 10 or more peaks under various solvent and flow conditions. The following solvents were used: pure methanol, 95 percent ethanol, pure acetonitrile, distilled water, and mixtures of methanol/water and acetonitrile/water in 10-percent water increments from 10 percent to 90 percent water (all mixtures on a volume/volume basis). Pure methanol gave superior resolution and had the additional advantages of being more readily available and less toxic than pure ethanol. Various flow rates were investigated, with resolution increasing as the flow rates decreased to 0.5 mℓ/min. Further reduction in the flow rate caused a slight loss of apparent resolution and increased elution times. Experiments with different injection volumes showed peak broadening as injections exceeded 10 μℓ (roughly 30 μg). A mass balance performed through the μC18 column with a microbalance gave a recovery of 102.1 percent of a 500-μg injected sample of the CCE of parallel run 5.

As the result of these trials, the following operating conditions were selected for obtaining all of the μC18 HPLC chromatograms in this report: solvent, methanol; flow rate, 0.5 mℓ/min; chart speed, 30 cm/hr; UV-detector, 0.1 absorbance unit full scale deflection at 254 nm; injection size, 2 to 6 μℓ. The resulting spectra are presented in Appendix A.

The differential refractometer was not used because it was less sensitive than the UV detector and demonstrated excessive baseline drift with subtle ambient temperature changes.

Several observations can be made from examination of the reverse-phase spectra. The chromatograms of the first eight carbon runs are remarkably similar in spite of considerable variation in plant effluent quality and CCE concentration. Chromatograms of extracts eluted with the same solvent, i.e., chloroform, ether, or acetone, for the five parallel runs are also remarkably similar. However, neither the REtE nor the RAcE displays components covering the full range of CCE polarities. The ether extract contained less of the more polar (earlier eluting) material than did the CCE, and the acetone extract contained less of the more apolar (later eluting) material than did the CCE.

Footnote

[1]Waters Corporation, Walpole, Massachusetts.

Each CCE of the parallel runs had a large, well resolved apolar UV peak. The peak is less polar than toluene but more polar than hexane. Three recycles through the μC18 column failed to further resolve the peak. This same peak, when collected and reinjected onto the microstyragel columns, elutes last and is well resolved. On the microstyragel columns it is retained for over two void volumes, indicating solute-styragel interaction and making molecular weight determination questionable. This apolar peak is missing on initial carbon runs 1, 2, 3, 7, and 8 and is present to only a slight degree on runs 4, 5, and 6. One possible explanation is that the appearance of this peak is related to periods of effluent chlorination. After runs 1 through 8, but before parallel runs 1 through 5, the plant personnel changed the effluent chlorination site (see Figure 1). These investigators were not advised of the change; consequently, the effluent samples for runs 1 through 8 were chlorinated, and the samples for parallel runs 1 through 5 were unchlorinated. A possible cause for this peak's appearance primarily on chromatograms of extracts of unchlorinated samples is that the peak material reacts with the chlorine to form non-UV-transparent and/or more polar compounds.

MICROSTYRAGEL SEPARATION

Molecular weight determinations were made by gel permeation chromatography (GPC) using a set of four columns in series: one 500Å microstyragel column followed by three 100Å microstyragel columns. Tetrahydrofuran was used throughout as the mobile phase. The theoretical plate count of the column set was determined to be 12,000. Previous trials with a column set of one 500Å and two 100Å columns showed no resolution difference between flow rates of 1.0 and 2.0 mℓ/min. Increasing the flow to the maximum allowable of 3.0 mℓ/min caused loss of resolution, and decreasing the flow below 1.0 mℓ/min increased the time required to obtain a single chromatogram to more than an hour. Since the same conditions were found with the four-column set, flow rates of 1.5 or 2.0 mℓ/min were adopted for all GPC spectra in this report.

A series of defined molecular weight polystyrenes and polyethylene glycols were used to calibrate the column set. Four low molecular weight polypropylene glycol standards borrowed from Los Alamos Scientific Laboratories (LASL) were used to complete the calibration curve. To ensure precise calibration, the

actual pump delivery was checked several times during the calibration period. Delivery drift was found to be ±5 percent. In addition, a stopwatch was used to measure the time from the standard injection to the detection of the peak maximum.

Under the flow condition selected (2.0 mℓ/min), a complete GPC can be obtained in less than 20 minutes if no sample-column interaction other than size sorting occurs. It can be seen in the GPC spectra of the parallel run CCEs that the prominent, well resolved final peak elutes in more than 20 minutes, indicating that some additional interaction mechanism with the cross-linked divinyl-benzene column packing has occurred.

The GPC spectra (Appendix B) show that, although the relative amounts of material at any specific molecular weight differ, the maximum and minimum molecular weights of the extracts are independent of the extraction method. The exception to this observation is the final discrete peak seen only on the CCE chromatograms. This peak requires such a large elution volume that one must assume it interacts with the microstyragel by some mechanism other than size sorting. It has been demonstrated that this peak is the same material which elutes last from the μC18 column (discussed more fully above). Chloroform, diethyl-ether, acetone, toluene, xylene, benzene, and methanol were injected into the microstyragel column set in an unsuccessful effort to duplicate the elution time of this peak.

BETA-INDUCED LUMINESCENCE DETECTOR

As reported in Reference 2, the beta-induced luminescence detector (BILD) is best used in the quenching mode. Xylene is a highly luminescent, relatively apolar solvent suitable for use in the quenching mode of this detector in a normal-phase HPLC system. When the BILD was used with a possibly defective Porasil column, results were not reproducible. The performance of the BILD was then evaluated in the fluorescent mode. A μC18 reverse-phase column with methanol as the eluent was used. The BILD was placed in series following the UV detector. The BILD gave no response in this mode. Xylene was again used as the solvent, with the μC18 column installed and the BILD in the quenching mode. Figure 11 shows that the BILD responded to two of the four peaks detected

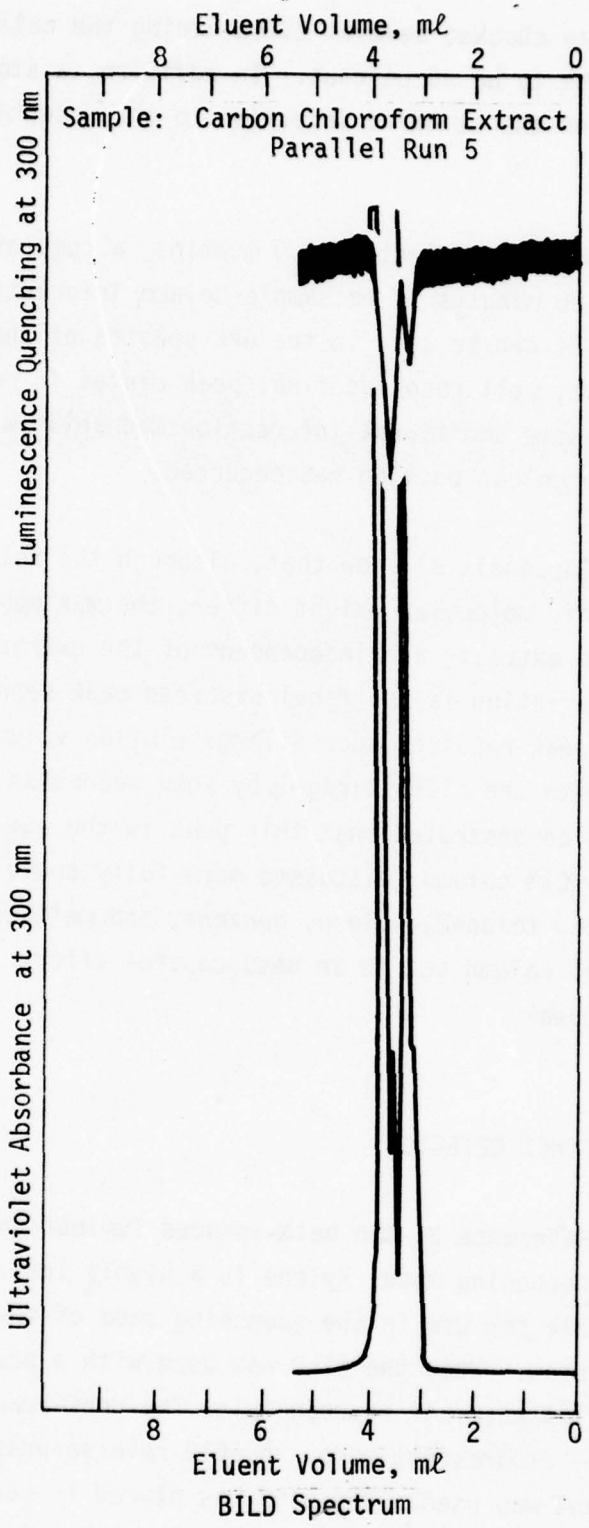


Figure 11. Beta-Induced Luminescence Detection Spectrum

by the UV instrument at 300 nm. The BILD must be developed further before it can be of significant use in detecting components of complex mixtures such as these wastewater extracts.

GRADIENT ELUTION WITH REVERSE-PHASE COLUMN

The spectra obtained when the μC18 column is used in a single eluent system reveal groupings of poorly resolved components of similar polarities. Principles of HPLC indicate that resolution in a reverse-phase system should be enhanced by an increase in the polarity of the mobile phase and that peak broadening should occur, especially for the later eluting components, when elution times are increased (Reference 15). Work with the single eluent system has indicated that the peak spreading effect decreases resolution more markedly than increased solvent polarity improves it.

With gradient elution, it is possible to increase the solvent polarity for only part of the elution volume. With water as a suitable, more polar second solvent, over 30 variations of operating conditions were made to improve the resolution obtained with the single eluent system. Spectra of three extracts obtained under optimum conditions are shown in Figures 12 and 13. It is apparent from these spectra that very small amounts of water, relative to the total elution volume, improve the resolution of the resulting chromatogram. It was also found that one may improve resolution in one portion of the chromatogram without improving resolution in another polarity region.

CHARACTERIZATION BY SERIAL REVERSE-PHASE HPLC-GPC

Samples of approximately 100 μg of the extracts of parallel run 5 were injected onto the μC18 column, and column effluent fractions were collected. The

Reference

15. Simpson, C. F., "Practical Experiments in HPLC with Typical Results." In C. F. Simpson (Ed.), *Practical High Performance Liquid Chromatography*, Heyden and Son, Ltd., London, 1976.

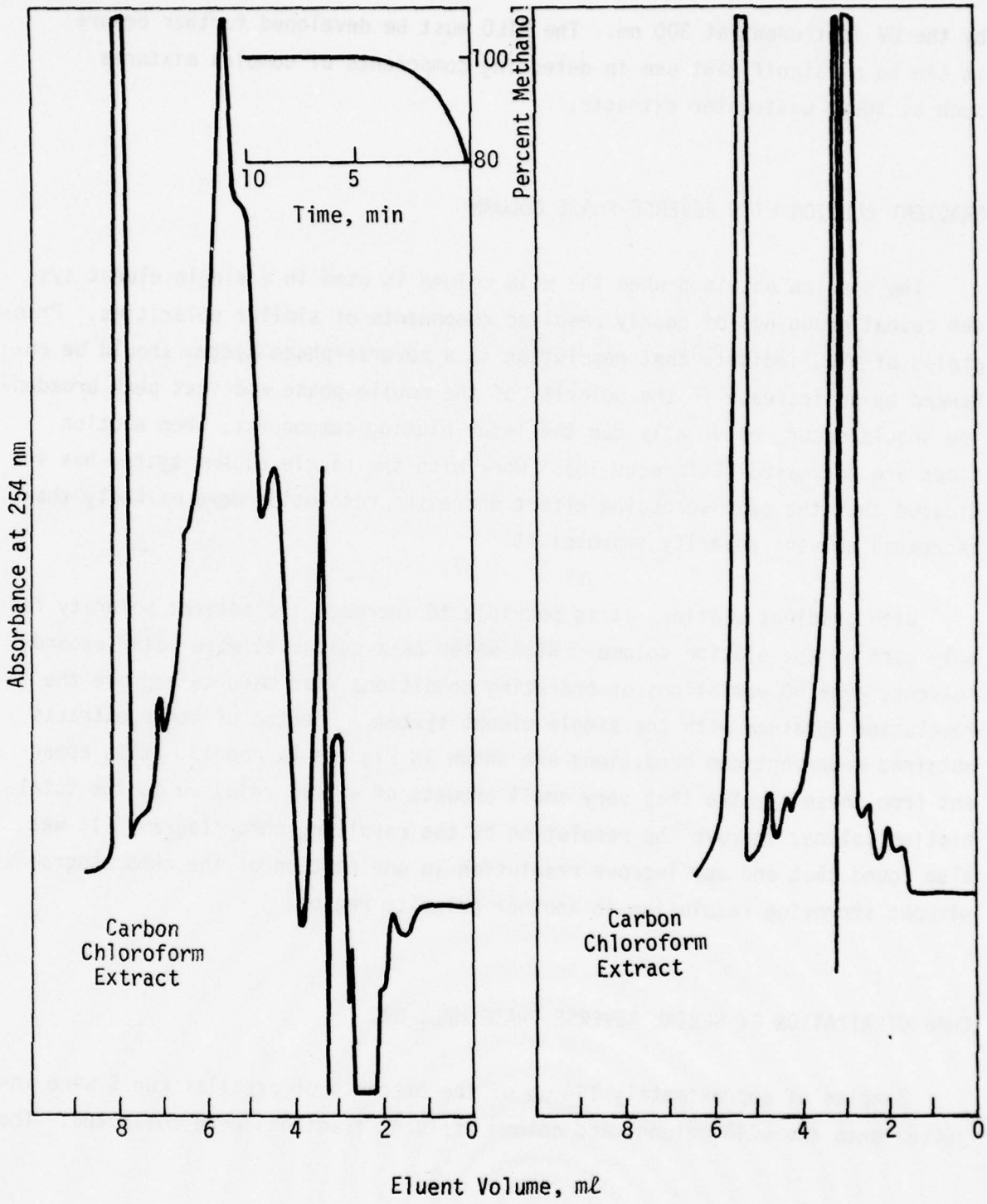


Figure 12. Gradient Elution and Isocratic Chromatograms for Carbon Chloroform Extract (Parallel Run 3)

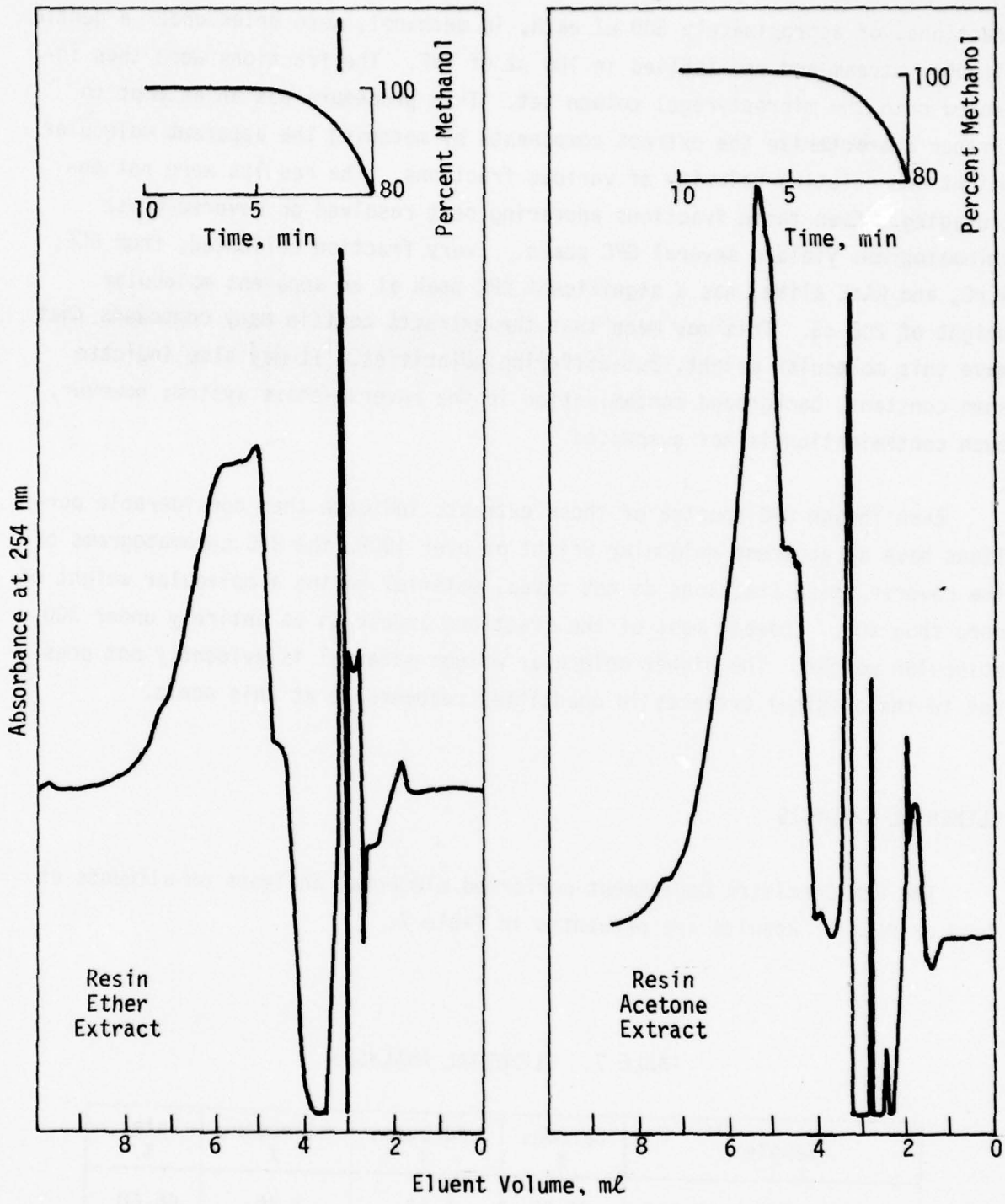


Figure 13. Gradient Elution for Resin Extracts (Parallel Run 3)

fractions, of approximately 500 μℓ each, in methanol, were dried under a gentle nitrogen stream and redissolved in 100 μℓ of THF. The fractions were then injected onto the microstyragel column set. This procedure was an attempt to further characterize the extract components by matching the apparent molecular weight and relative polarity of various fractions. The results were not encouraging. Even those fractions appearing best resolved on reverse-phase chromatograms yielded several GPC peaks. Every fraction collected, from CCE, RErE, and RAcE alike, has a significant GPC peak at an apparent molecular weight of 200 ±5. This may mean that the extracts contain many compounds that have this molecular weight, but differing polarities. It may also indicate some constant, background contamination in the reverse-phase system; however, such contamination is not suspected.

Even though GPC spectra of these extracts indicate that considerable portions have an apparent molecular weight of over 1000, the GPC chromatograms of the reverse-phase fractions do not reveal material having a molecular weight of more than 500. Indeed, most of the fractions appear to be entirely under 300 molecular weight. The higher molecular weight material is evidently not present in the original extracts in quantities recoverable at this scale.

ELEMENTAL ANALYSIS

The UNM Chemistry Department performed elemental analyses on aliquots of four extracts. Results are presented in Table 7.

TABLE 7. ELEMENTAL ANALYSES

Sample	Carbon, %	Hydrogen, %	Nitrogen, %	Total, %
Improved System 2 CCE	39.59	5.63	1.46	46.68
Parallel 5 CCE	53.09	8.00	1.35	62.44
Parallel 5 RErE	60.92	7.80	1.05	69.77
Parallel 5 RAcE	55.19	7.85	1.46	64.50

SPECTROGRAPHIC ANALYSIS

Several attempts were made to perform infrared spectroscopy of isolated μC18 peaks collected from injections of extract samples. However, the infrared (IR) instrumentation available for use in the UNM Chemistry Department did not provide meaningful spectra from the microgram amounts of material in each fraction. A successful IR spectrum was obtained by combining a particular fraction from four successive large injections. The spectrum is shown in Figure 14. The peak collected for IR analysis was the last eluting peak on the μC18, indicating that it is even less polar than toluene. Its late elution (greater than 2 void volumes) from the microstyragel column set means that it undergoes some solute-column interaction with the polydivinylbenzene styragel. Although these behaviors are consistent with aromaticity, the IR spectrum does not show aromatic character. The carboxylate IR peak may indicate that the solute-styragel interaction is of an ion-exchange nature. Preliminary mass spectroscopy and recycle HPLC results from LASL indicate that this fraction is a single compound.

The normal size injection for the μC18 column is approximately 25 μg, and the maximum injection that will not cause undesirable column effects is about 250 μg. If a 250-μg injection is collected in 10 equal fractions of 25 μg, the fractions are an order of magnitude too small for IR spectroscopy and at least two orders of magnitude too small for nuclear magnetic resonance spectroscopy or elemental analysis with the equipment available.

Mass spectroscopy can be performed on these microgram fractions, and some preliminary mass spectroscopy has been done on two fractions by LASL. Interpretation of these data has proved difficult since it appears that at least one of the fractions is still a complex mixture rather than a single compound. Further mass spectroscopy would be an appropriate means of identifying these compounds.

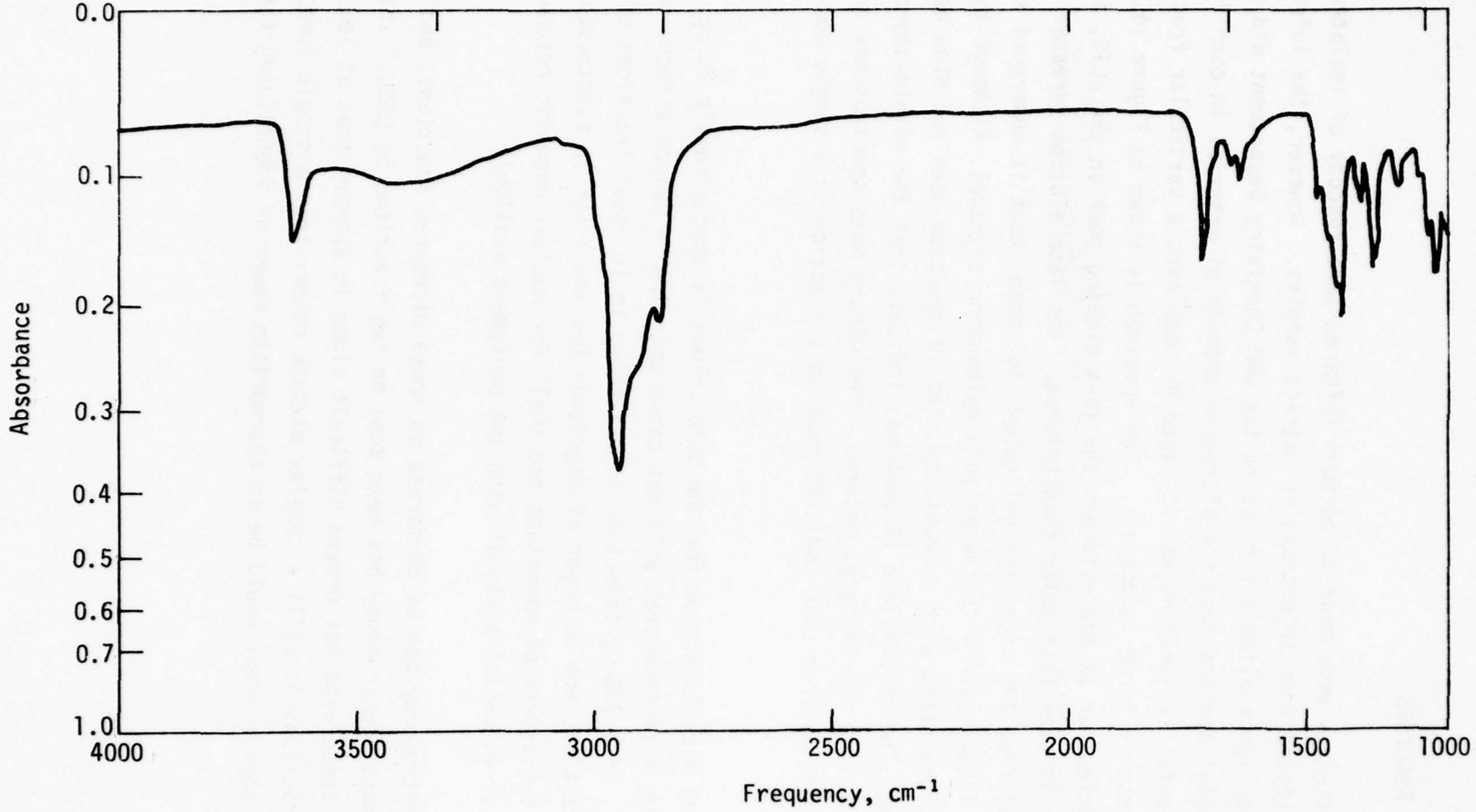


Figure 14. Infrared Spectrum of Isolated Reverse-Phase Peak

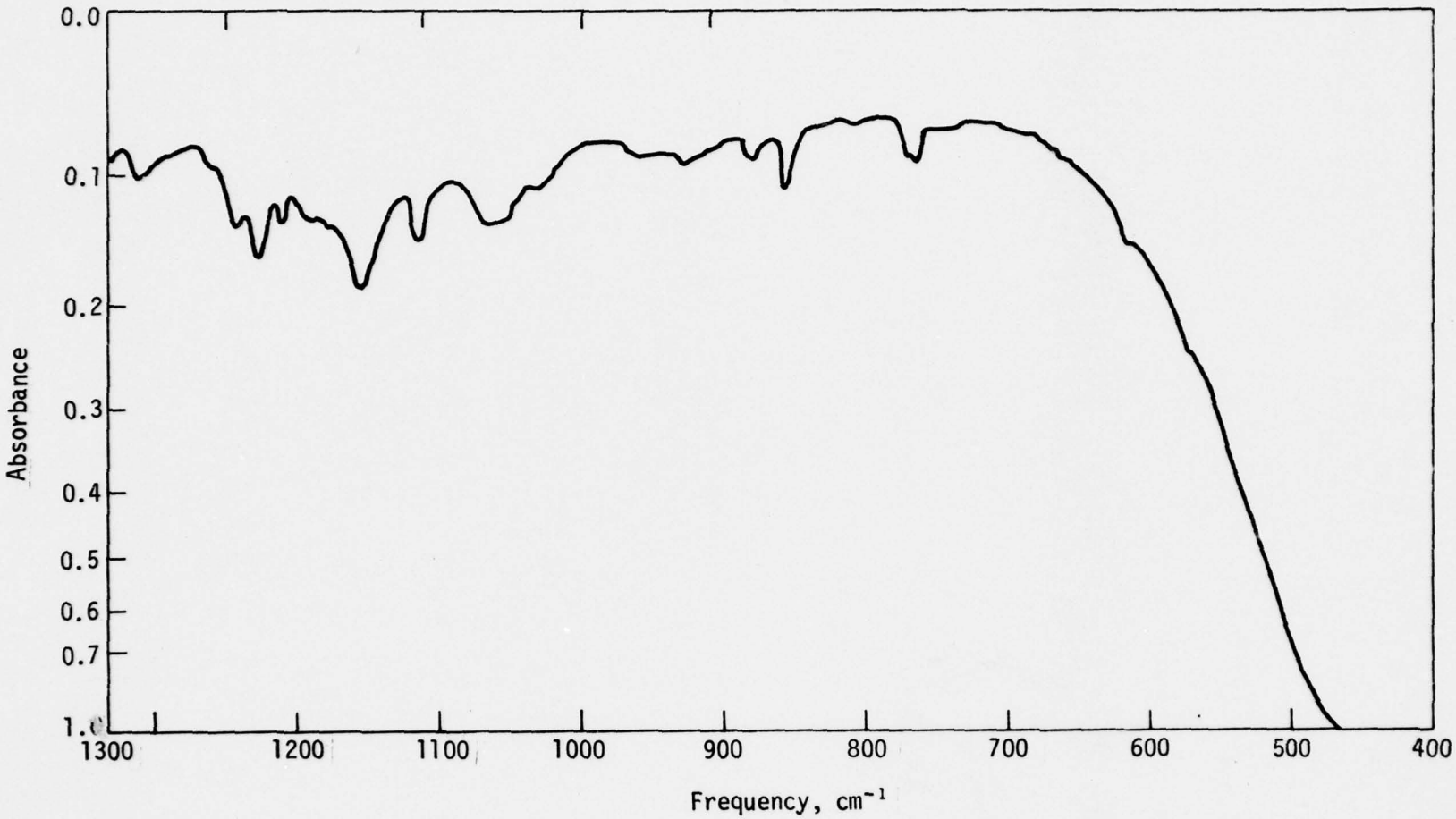


Figure 14. Infrared Spectrum of Isolated Reverse-Phase Peak (Concluded)

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SECTION VI
CONCLUSIONS AND RECOMMENDATIONS

CONCLUSIONS

The O-CA minifilter procedure is unsatisfactory for the routine recovery of residual organics from activated sludge effluents because of low recovery efficiency, difficult operational sequence, and time-consuming maintenance duties.

Discrete separation of the extract components was achieved by employing HPLC with reverse-phase μC18 and/or microstyragel columns; however, the mass spectrometric data are highly complex, and a well resolved and separated substance has not yet been identified.

Correlations were found among the CCE, COD, and NVTOC values. These correlations are given in Table 6.

Concentration of residual organics on XAD-2 macroreticular resin proved to be very inefficient (less than 7 percent) in removing trace organics from the sample stream.

The XAD-2 HPLC UV spectra of the RAcE and RErE blanks yielded several prominent responses indicating that organic release from the resin is a vital portion of the extracts.

The general forms of the CCE, RAcE, and RErE UV spectra are remarkably similar in each of the five parallel runs.

The improved system developed for recovering residual organics from activated-sludge effluents demonstrated a significantly higher recovery efficiency than that of the minifilter system. It is also easier to operate and maintain.

The amount of material that can be resolved and separated by HPLC from a single injection is insufficient for any other analytical identification procedure except mass spectroscopy.

Gradient elution employing a methanol/water eluent system significantly improves the resolution of the carbon and resin extracts. This method, however, is more demanding in operator skill, solvent preparation, and operator time.

A single, prominent, low molecular weight peak occurring on both the reverse-phase and microstyragel UV spectra contained major fractions of compounds with molecular weights of 32 and 64; the peak is much less evident in, or is absent from, extracts recovered after the effluent has been chlorinated.

RECOMMENDATIONS

The improved recovery system should be employed to concentrate trace organics of nonvolatile substances and to evaluate the effluent quality of secondary treatment plants.

The status of the beta-induced luminescence detector for HPLC should be monitored by the Air Force Environics Group in view of the detector's sensitivity in the quenching mode for halo organics.

A procedure employing centrifugation at 20,000 rpm, concentration by strongly anionic resin eluted with inorganic buffers, and final concentration by lyphilization prior to injection onto a microBondagel E-linear column or equivalent should be evaluated for its efficiency in recovering nonvolatile organics from effluents.

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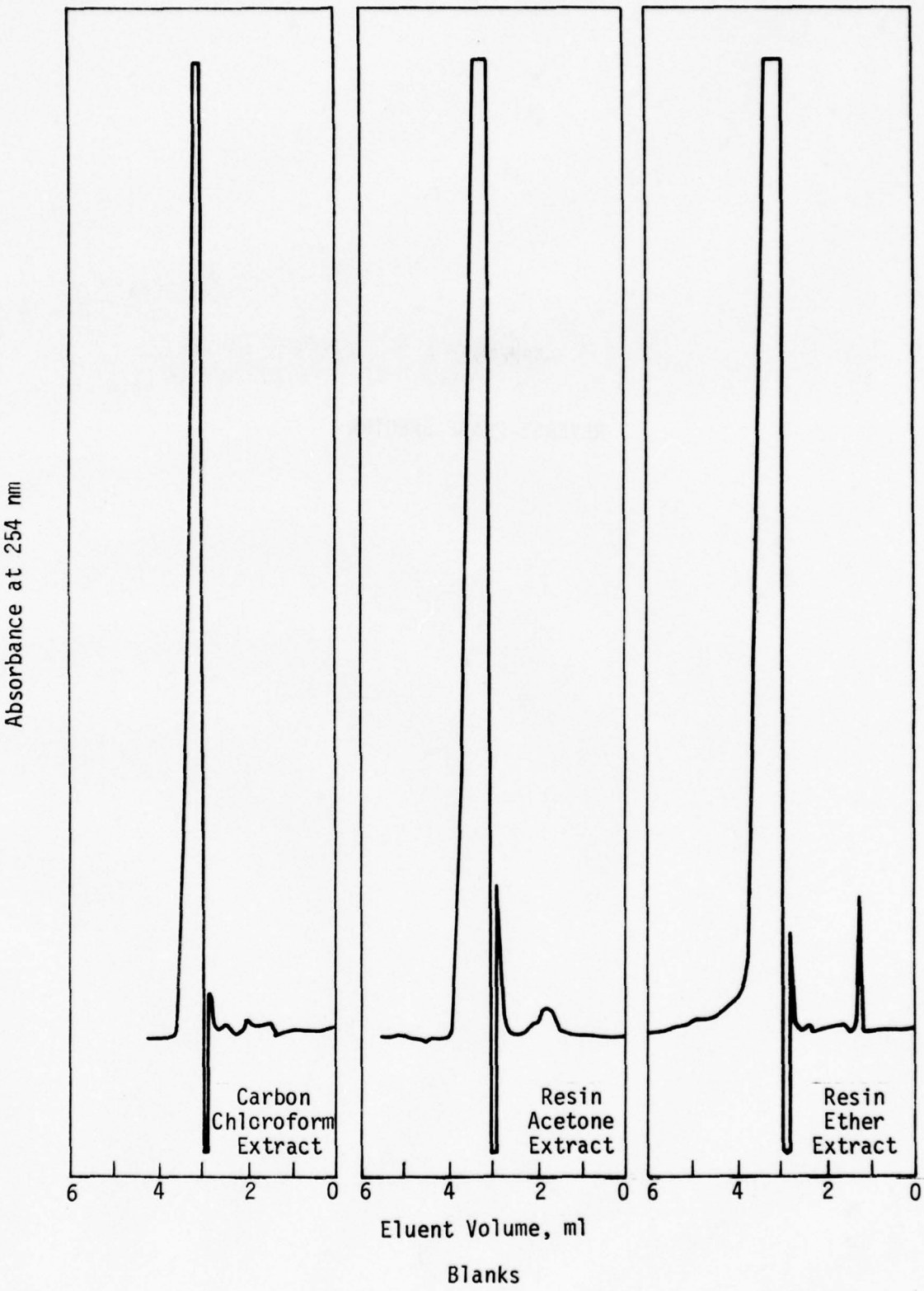
APPENDIX A

REVERSE-PHASE SPECTRA


Absorbance at 254 nm
Carbon
Chloroform
Extract
Resin
Acetone
Extract
Resin
Ether
Extract
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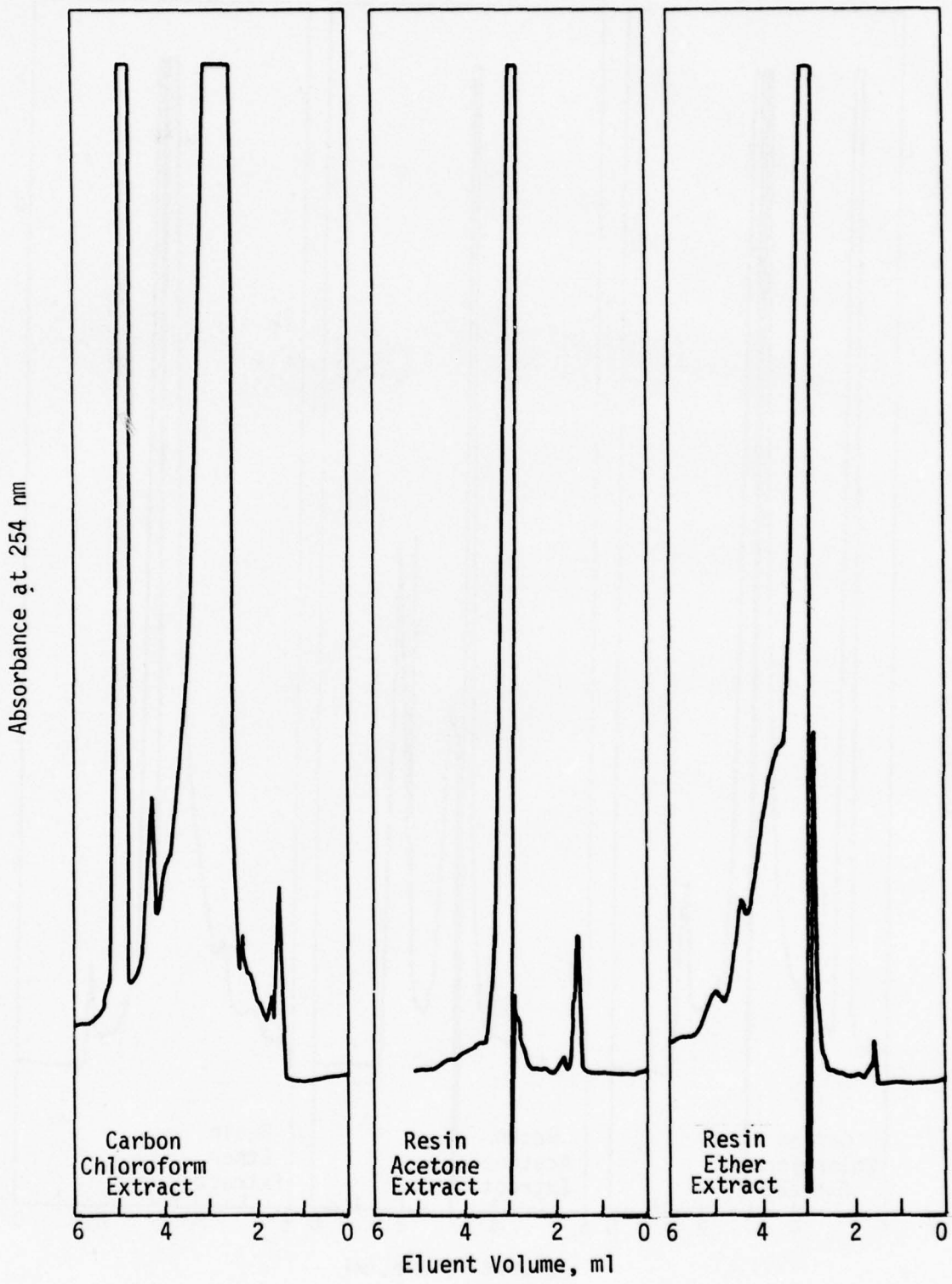
4
2
0
Eluent Volume, ml
Blanks



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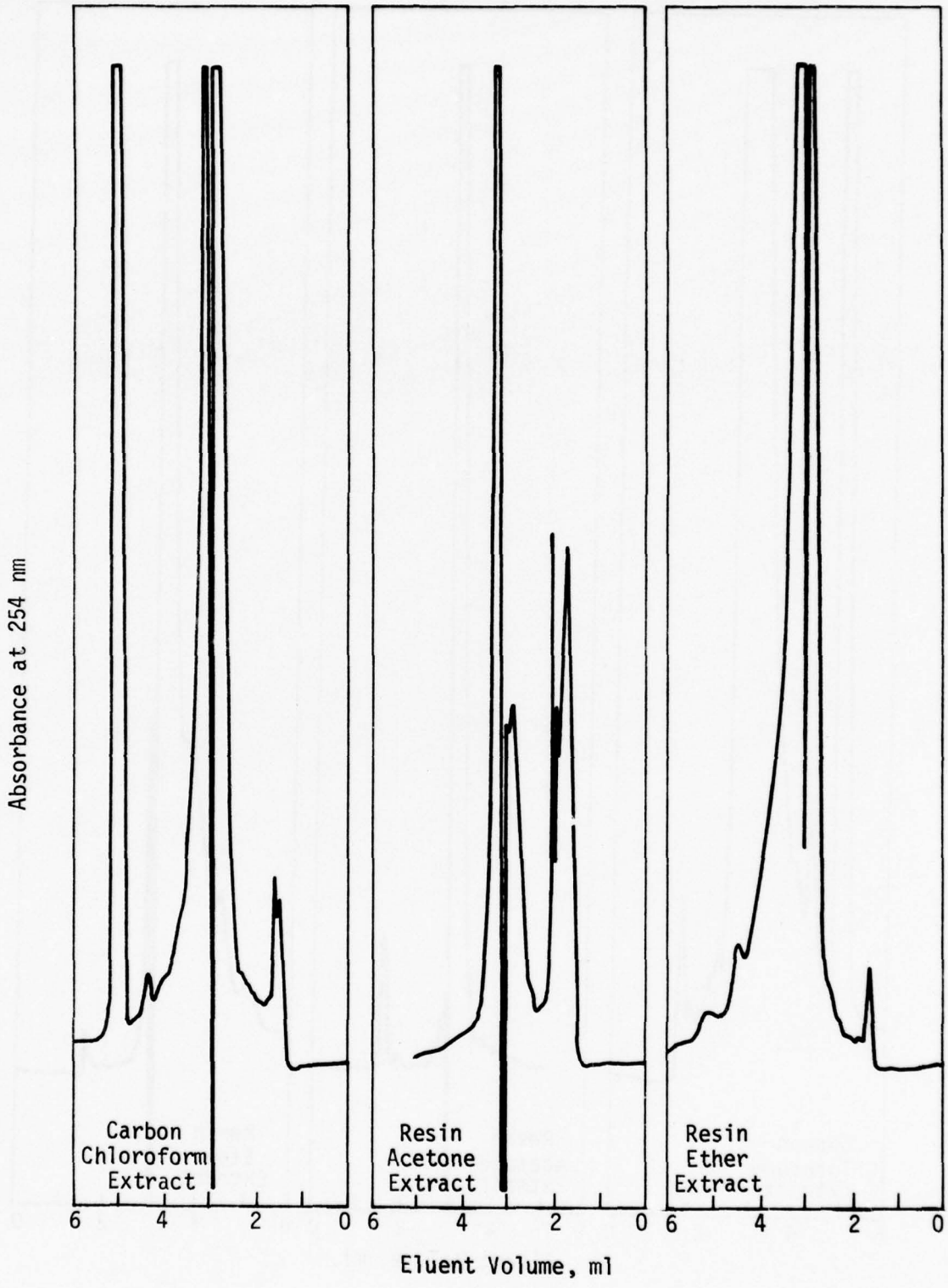

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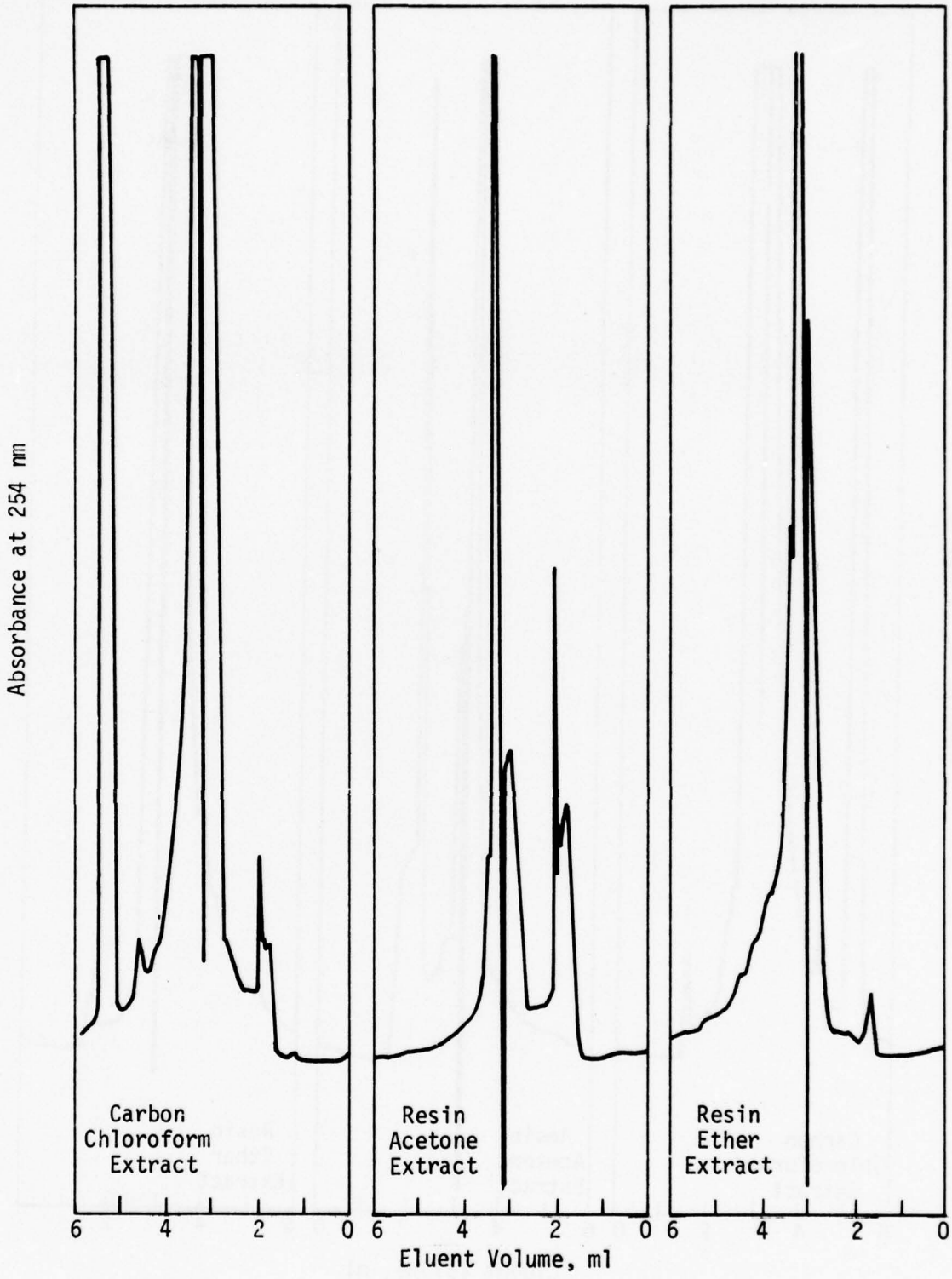

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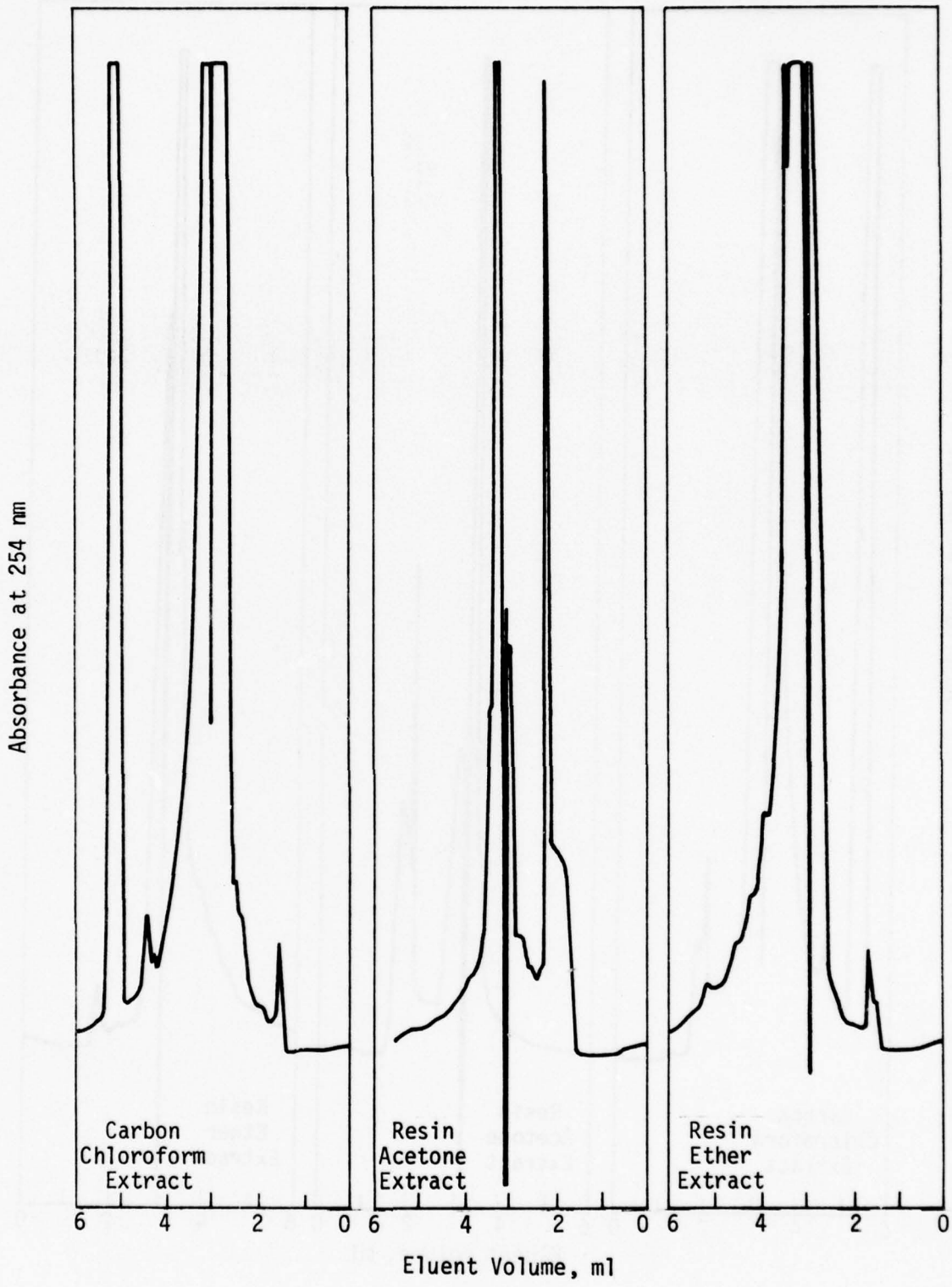

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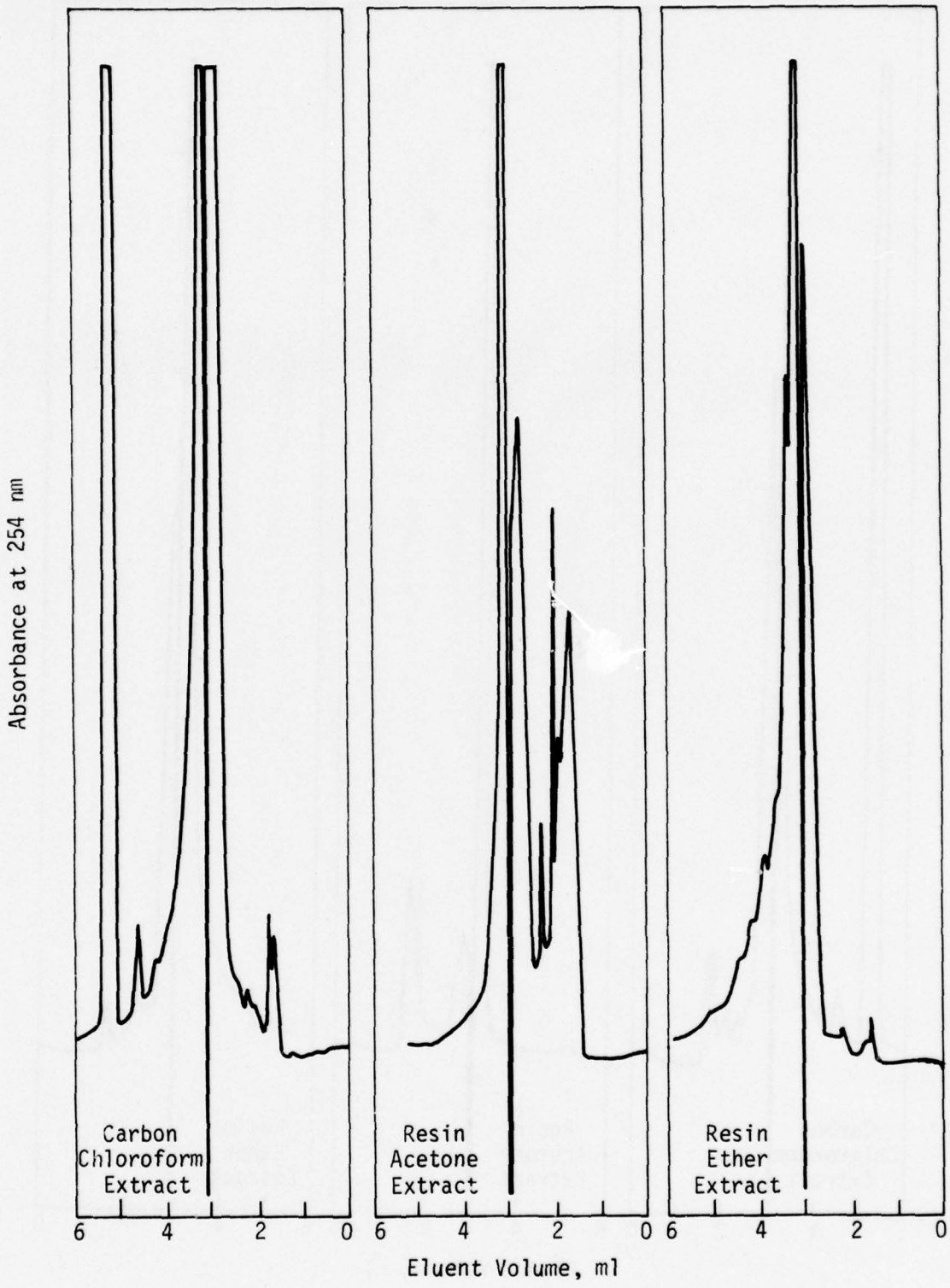

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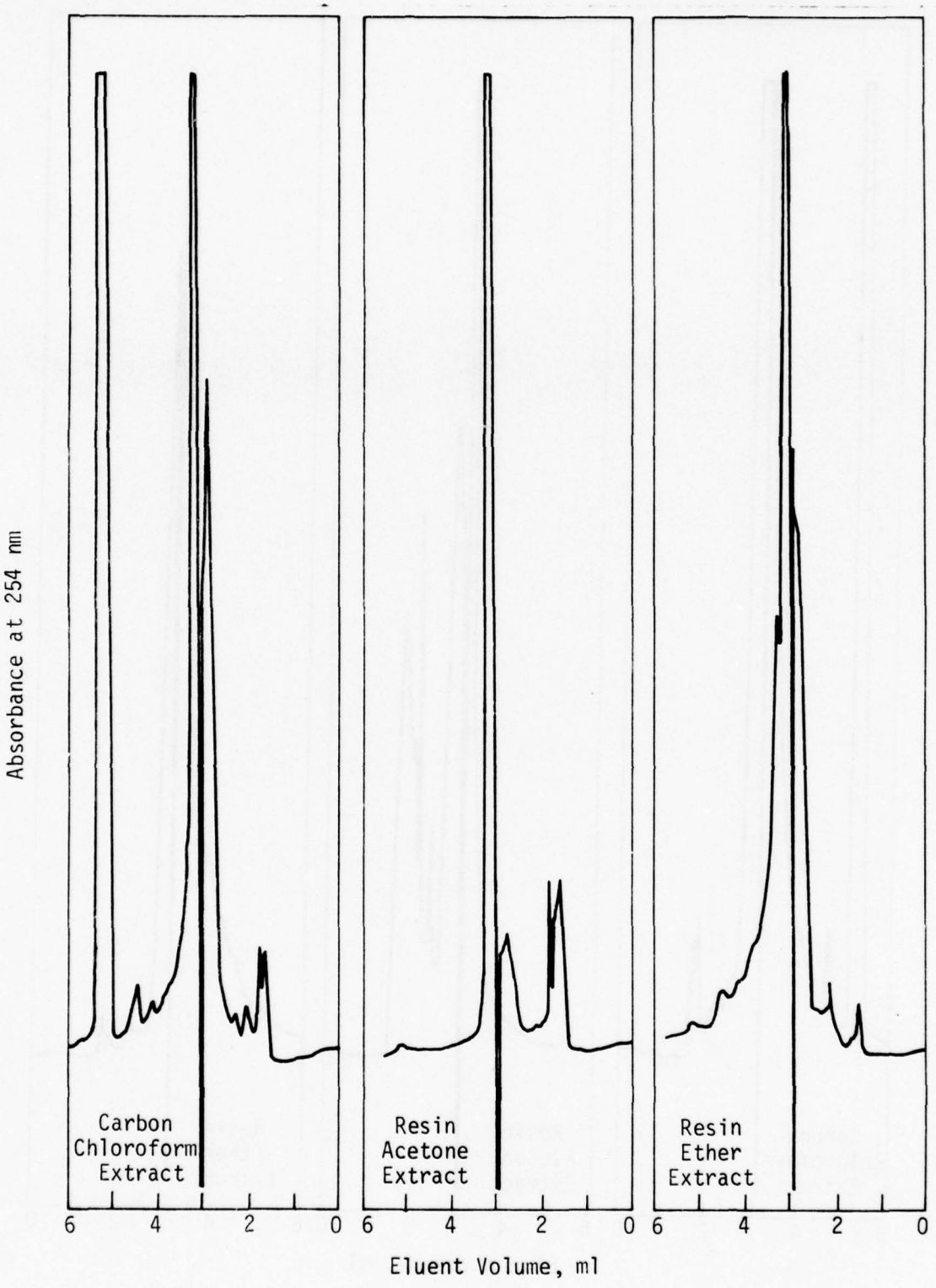

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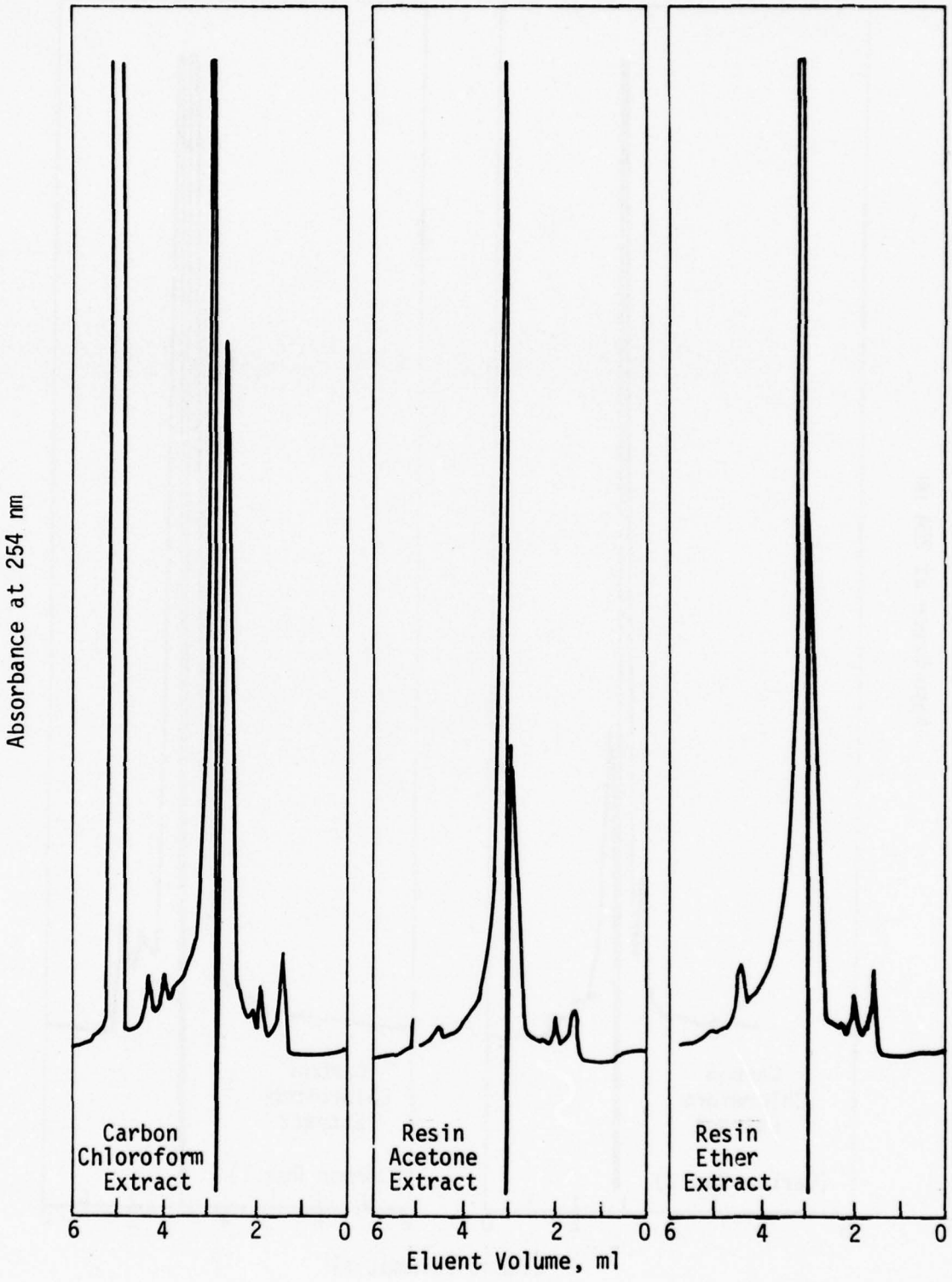

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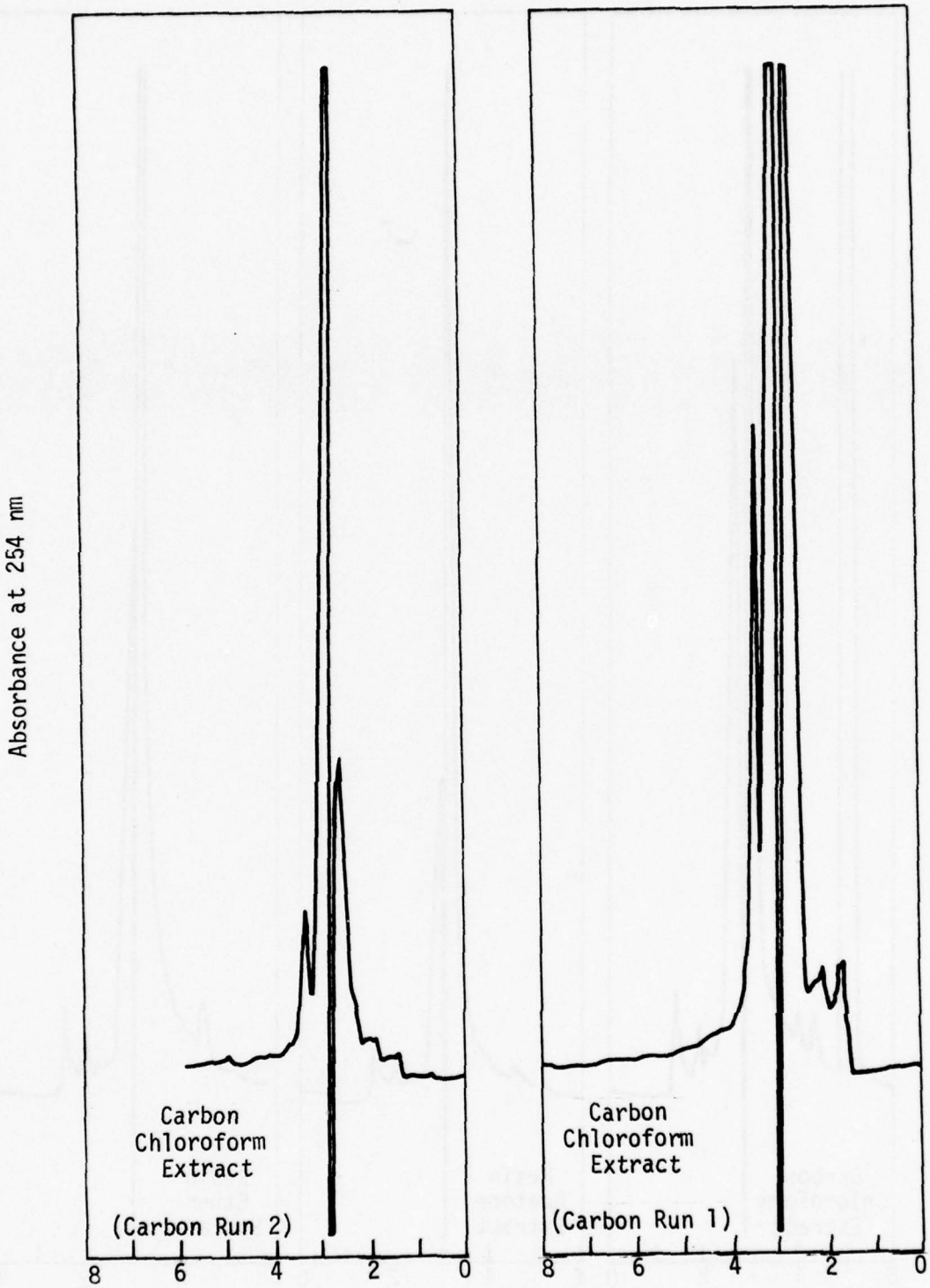

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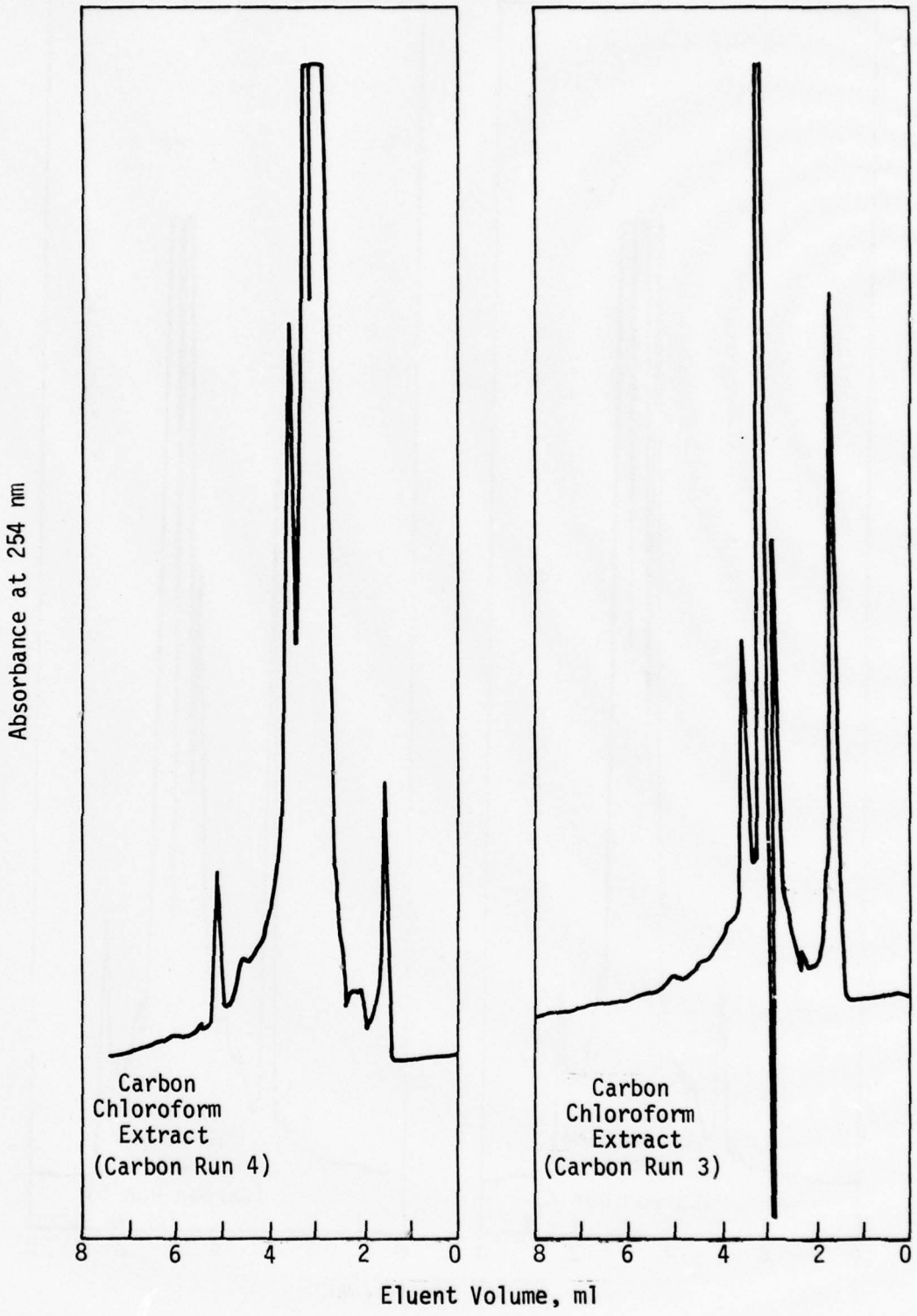

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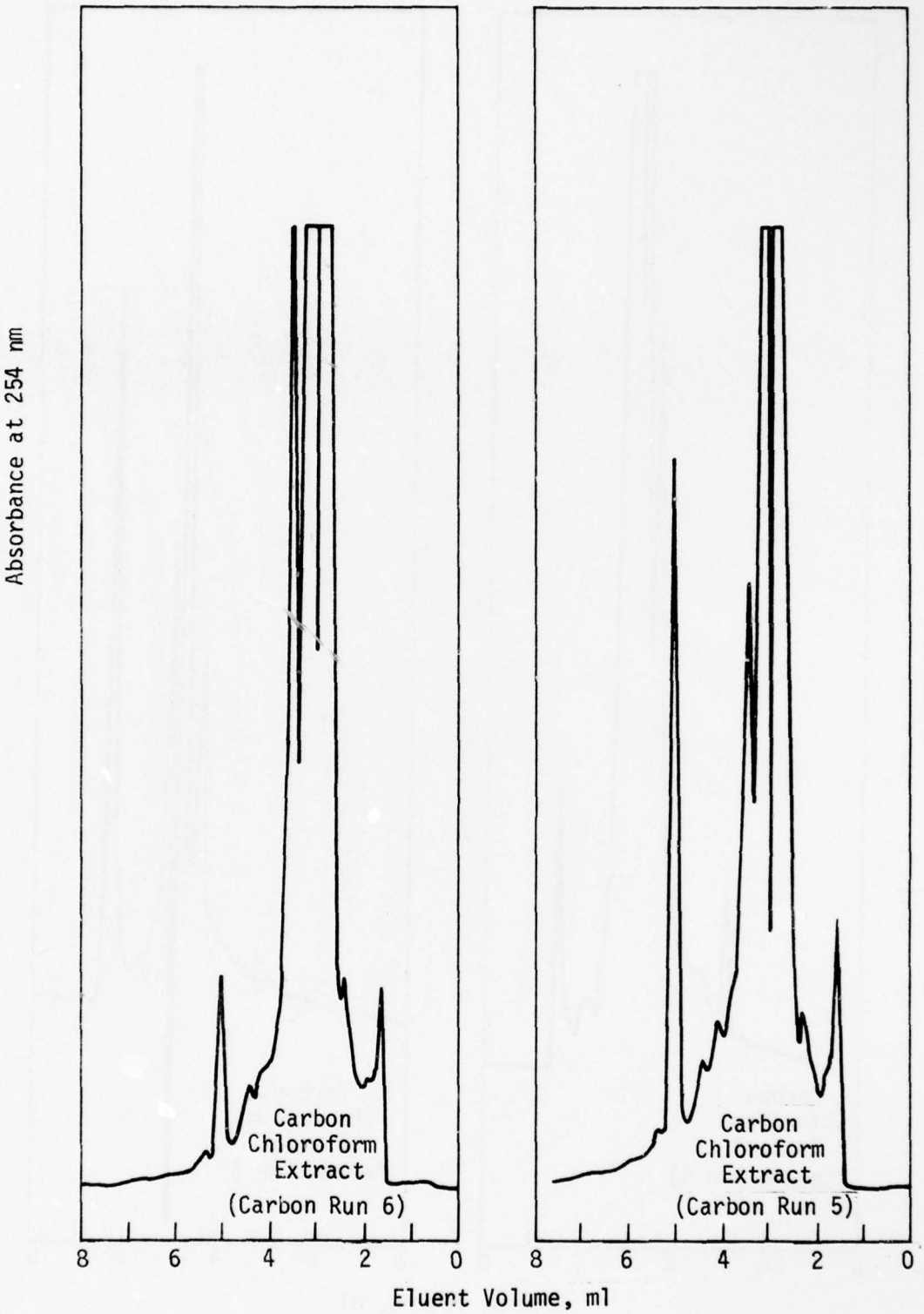
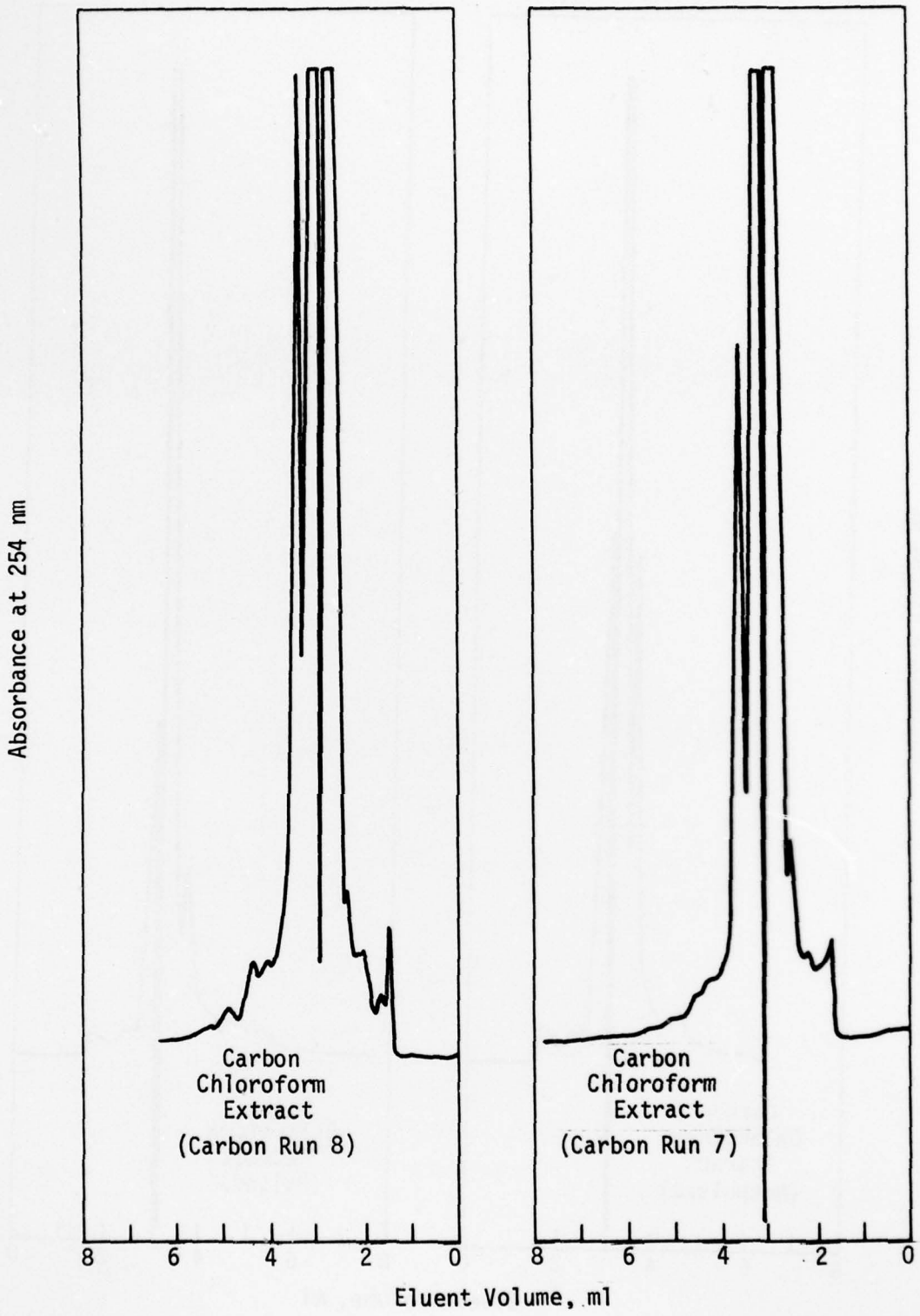
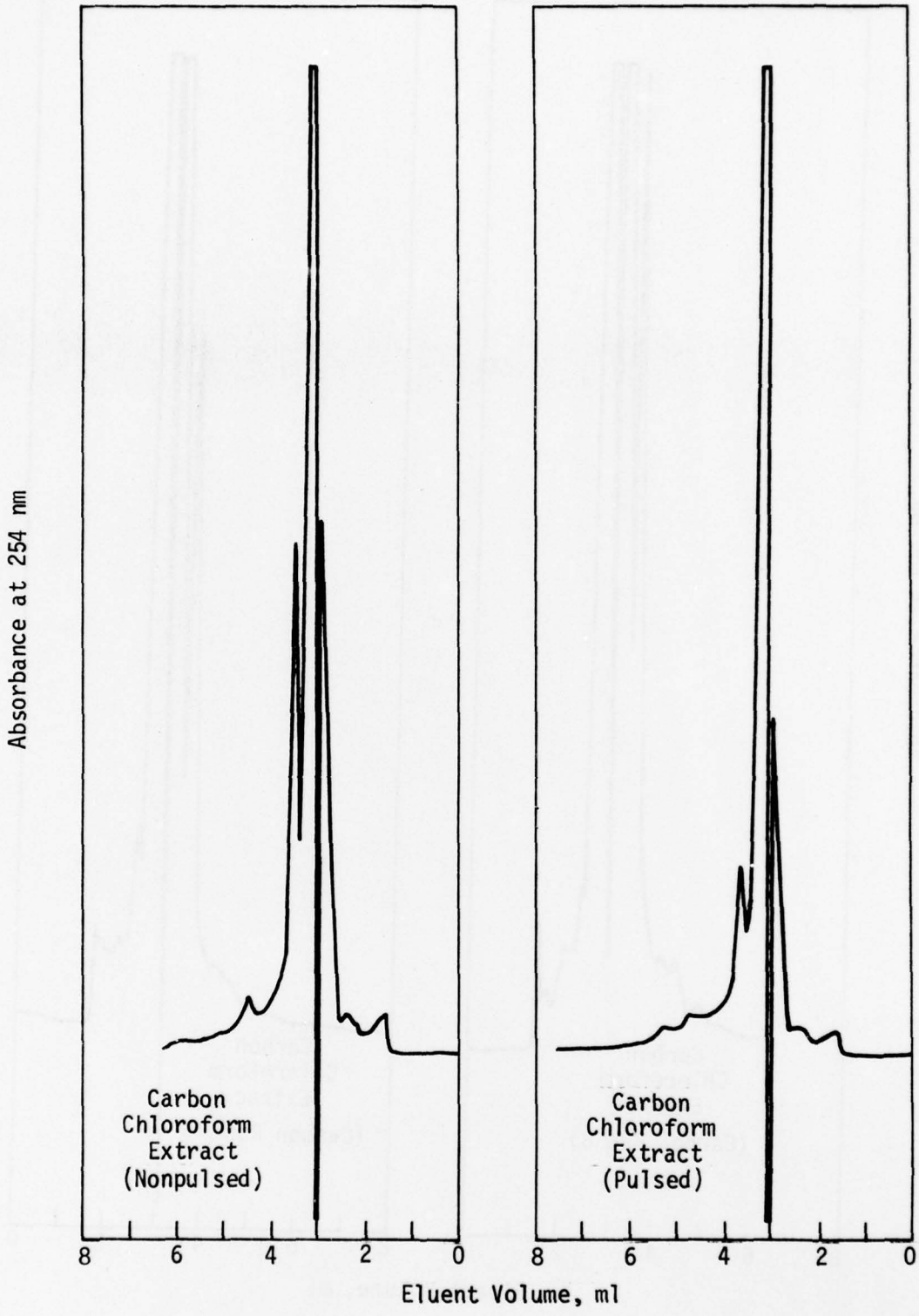


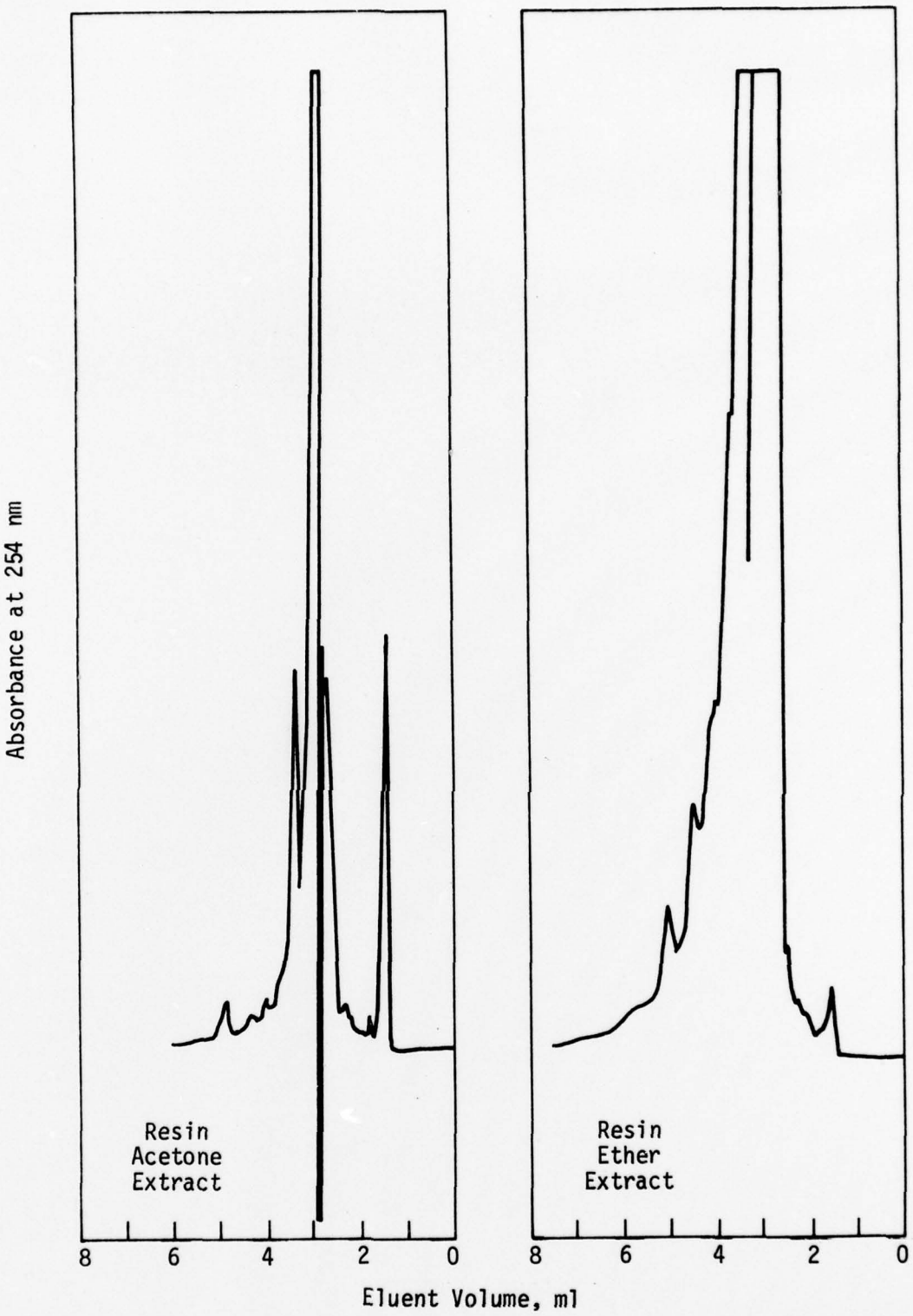
Carbon Runs 5 and 6



Carbon Runs 7 and 8



Pulsed and Nonpulsed
Bed Carbon Runs



Resin Run 1

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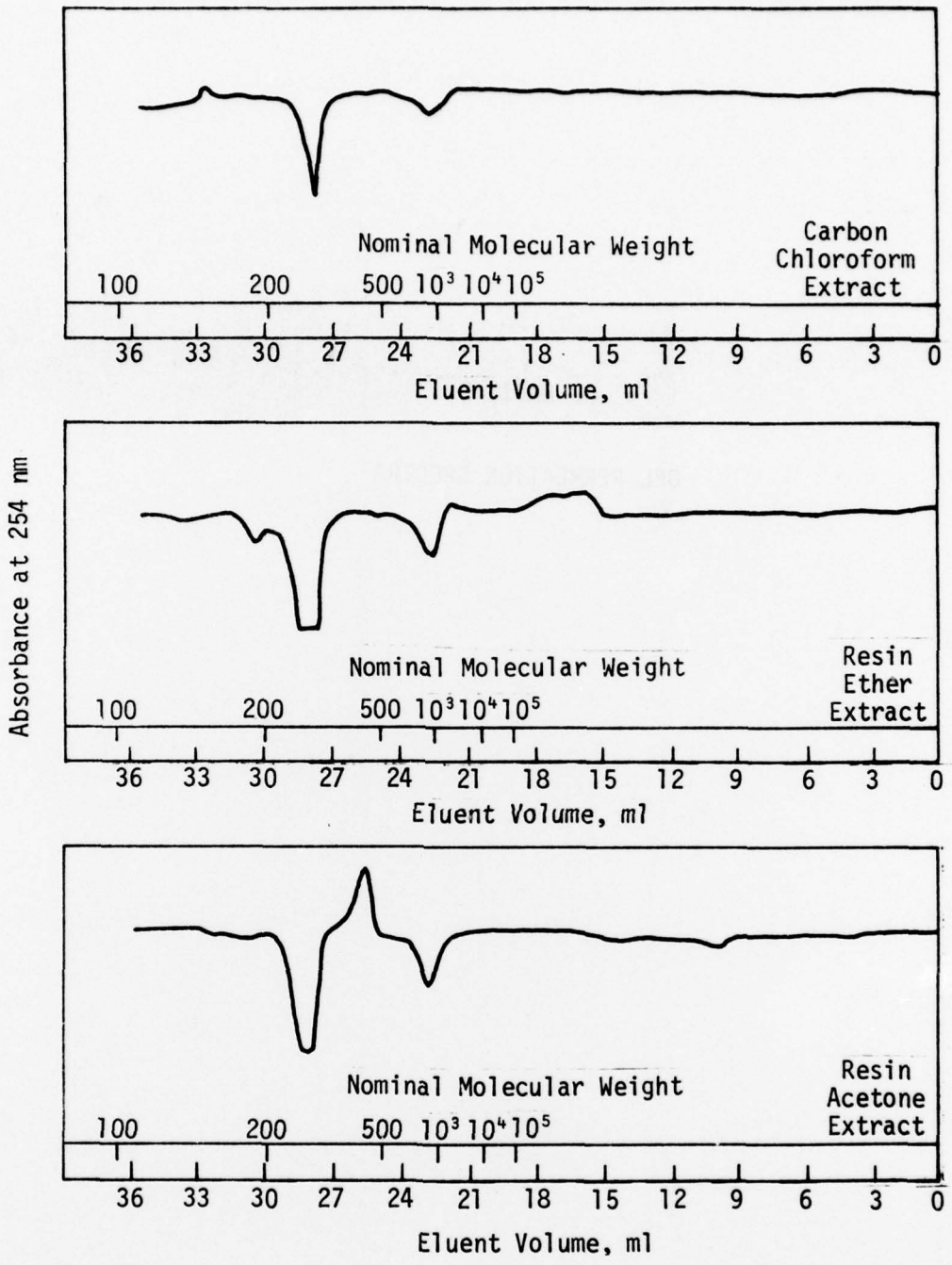
APPENDIX B

GEL PERMEATION SPECTRA


Absorbance at 254 nm
Nominal Molecular Weight
100
200
500
10^3
10^4
10^5
Carbon Chloroform Extract
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Eluent Volume, ml
Resin Ether Extract
Resin Acetone Extract

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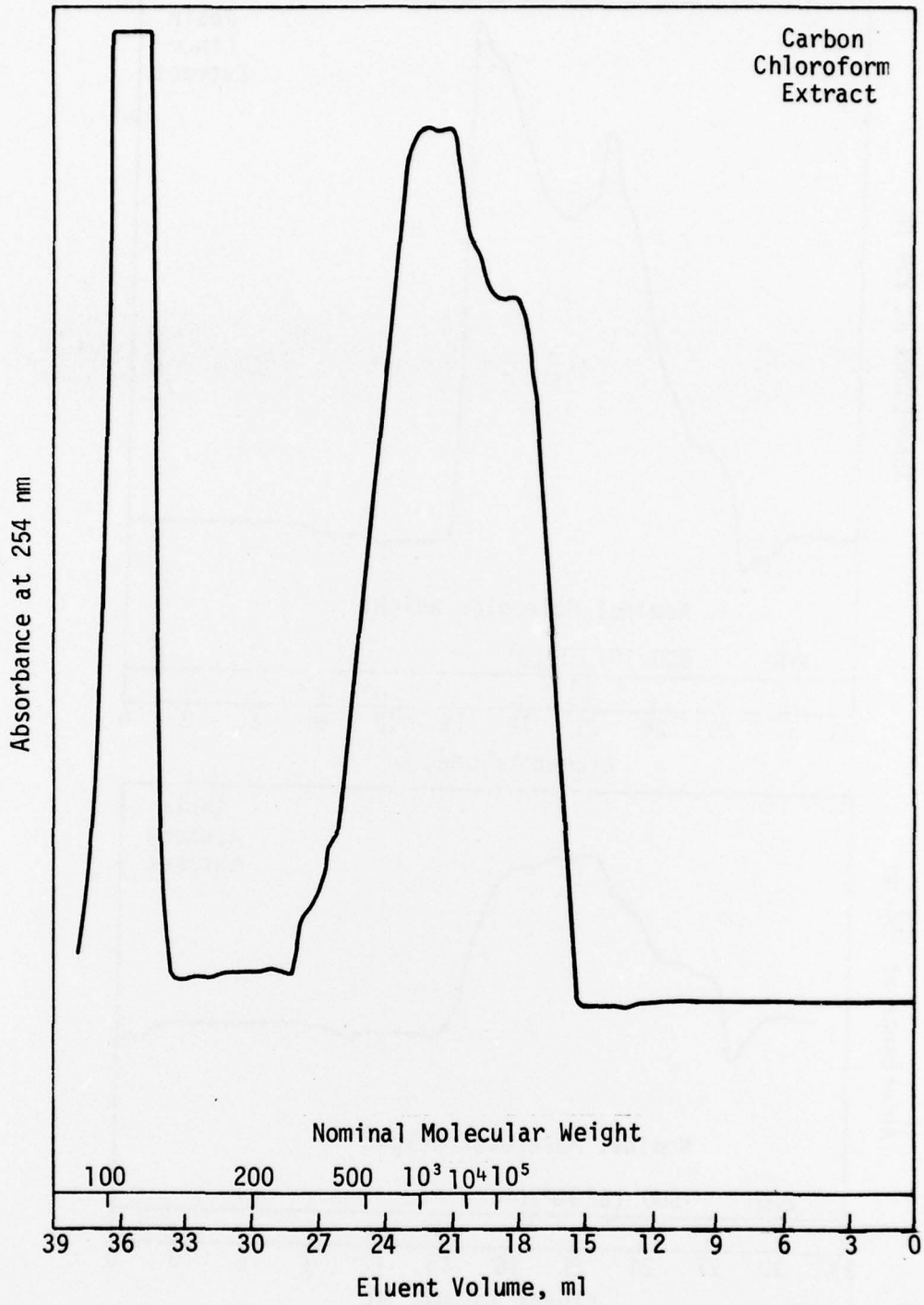

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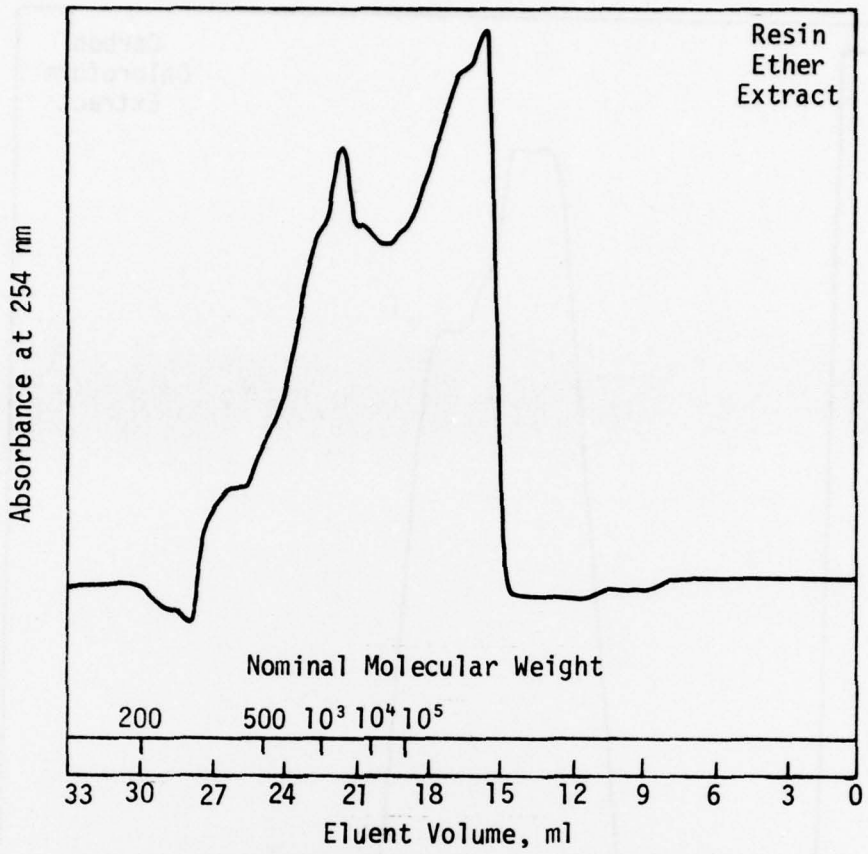

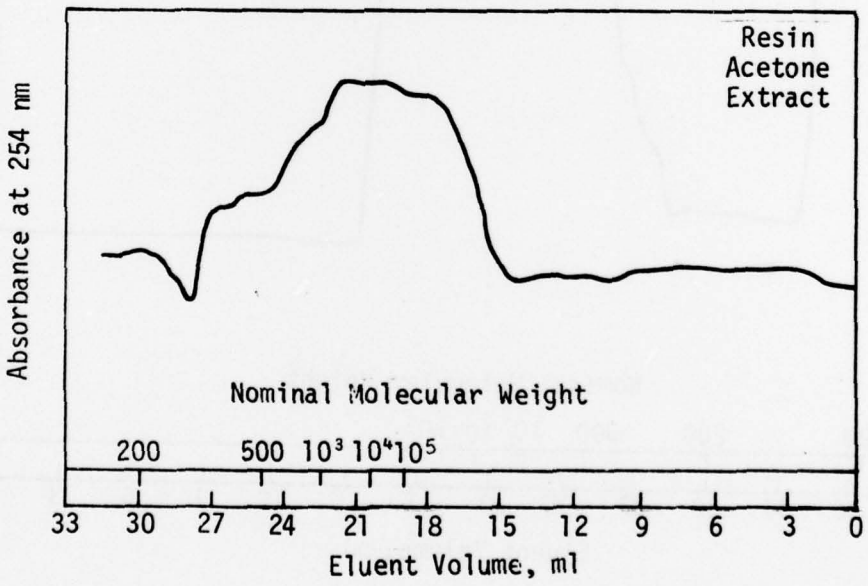

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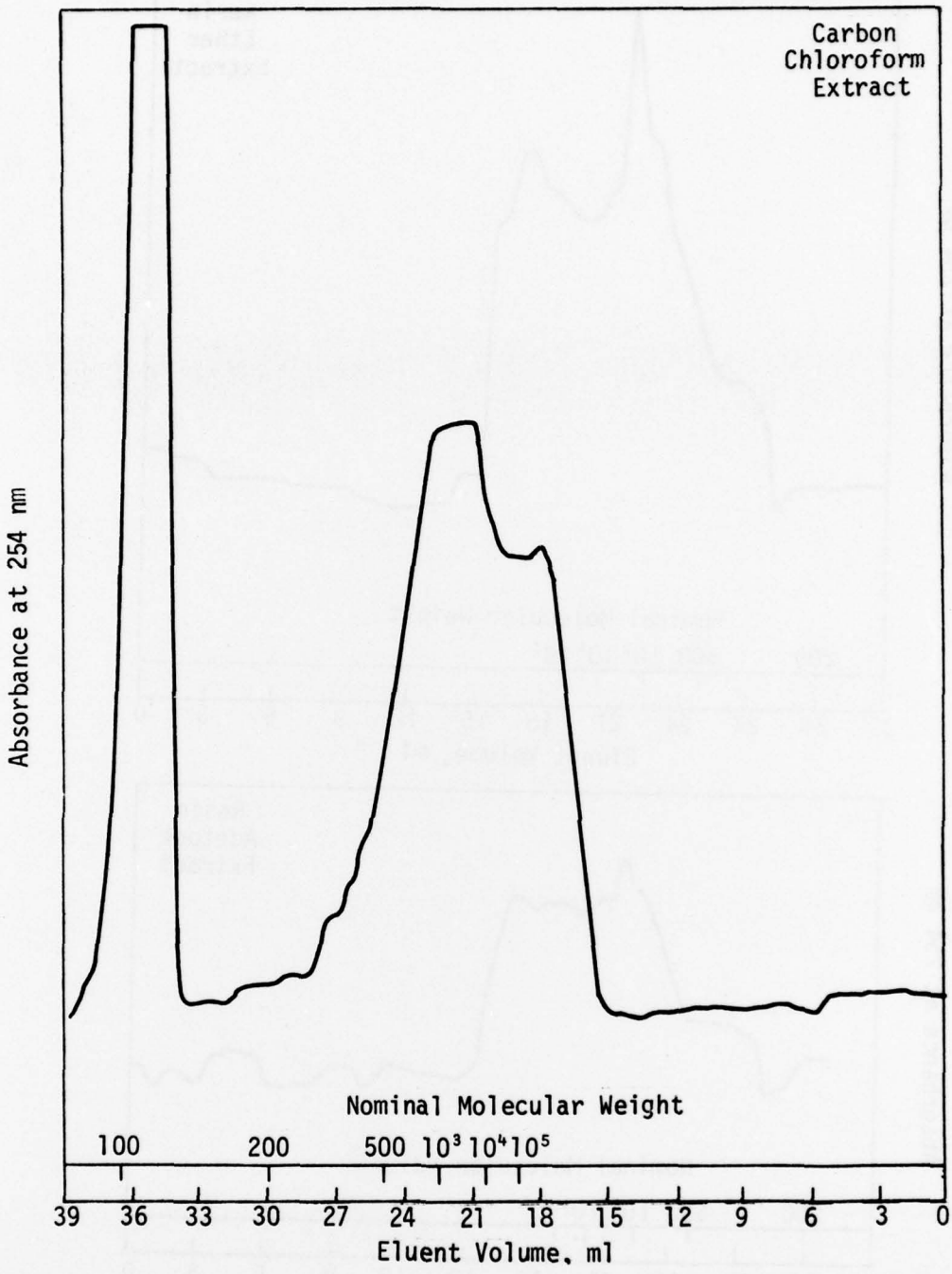

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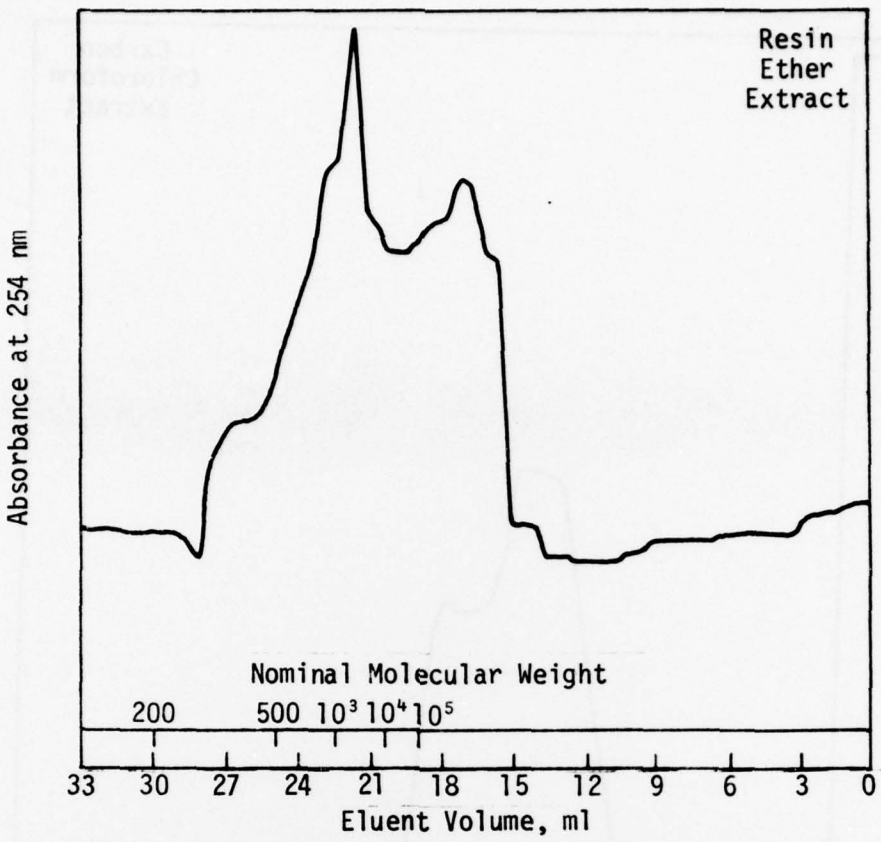

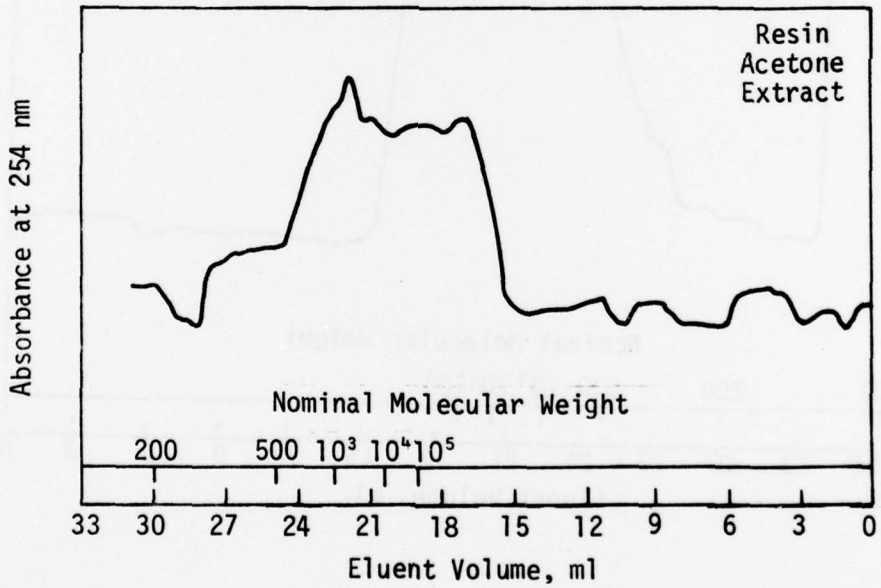

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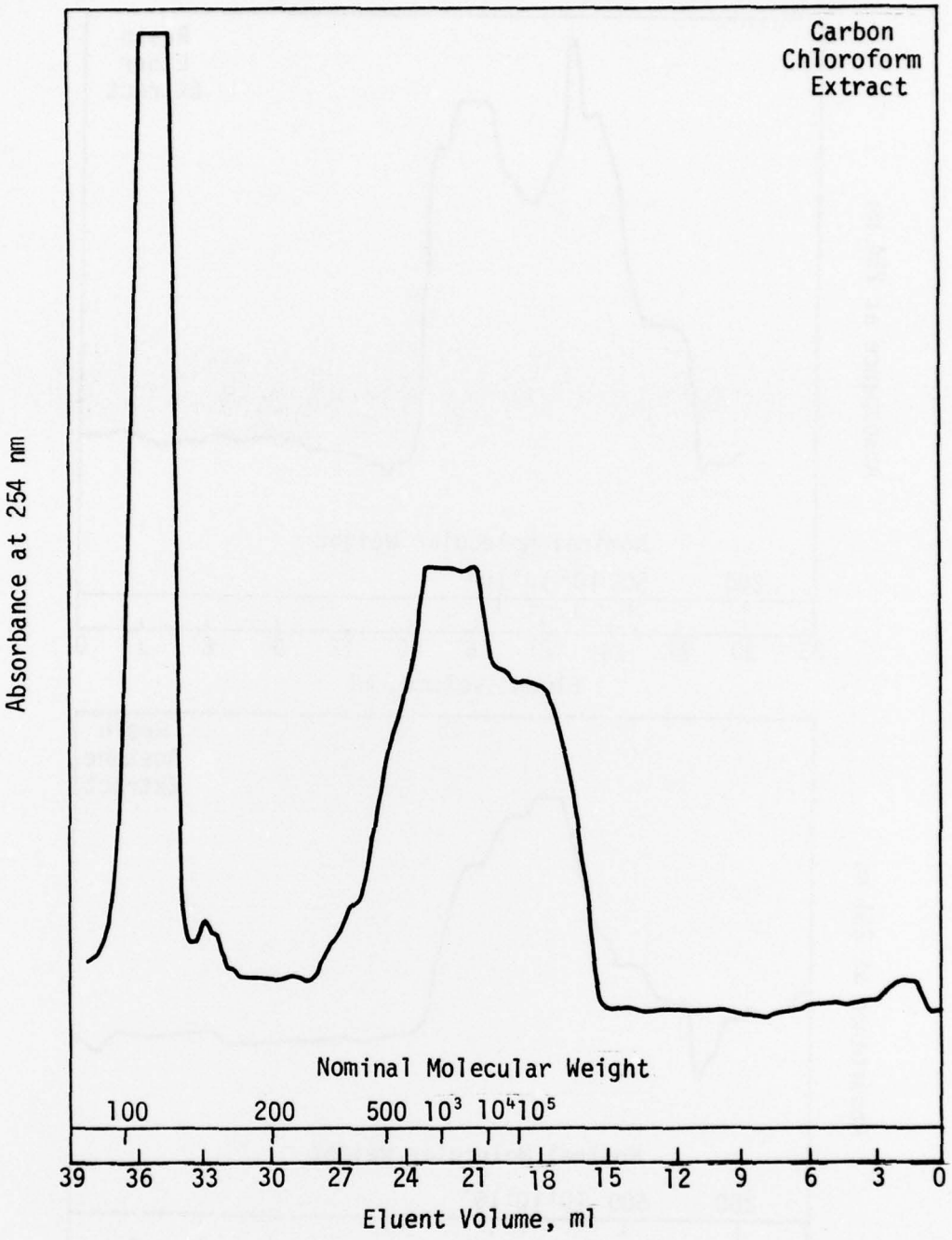

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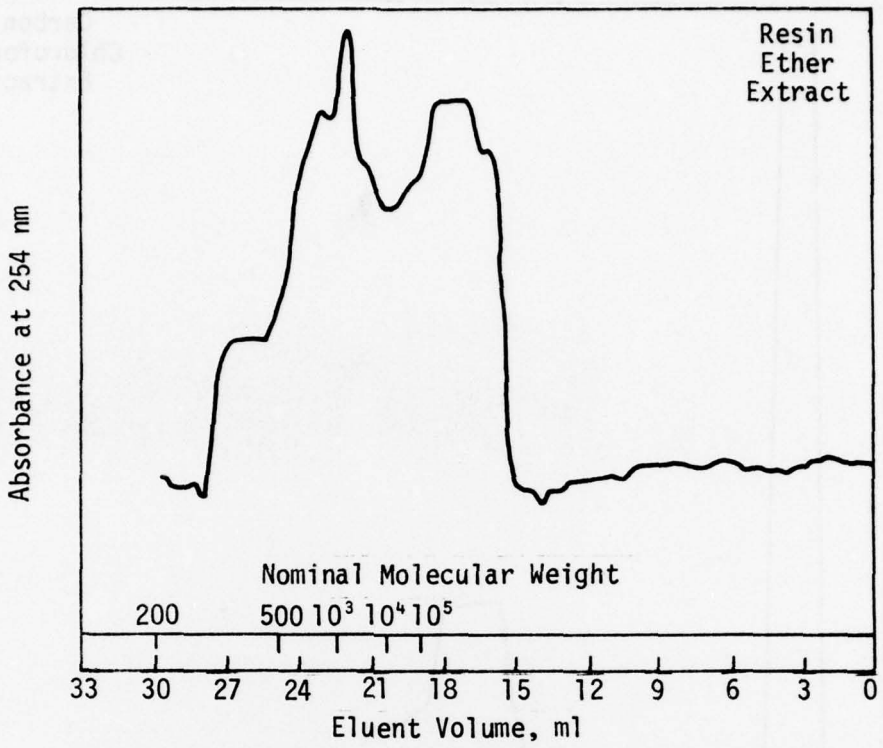

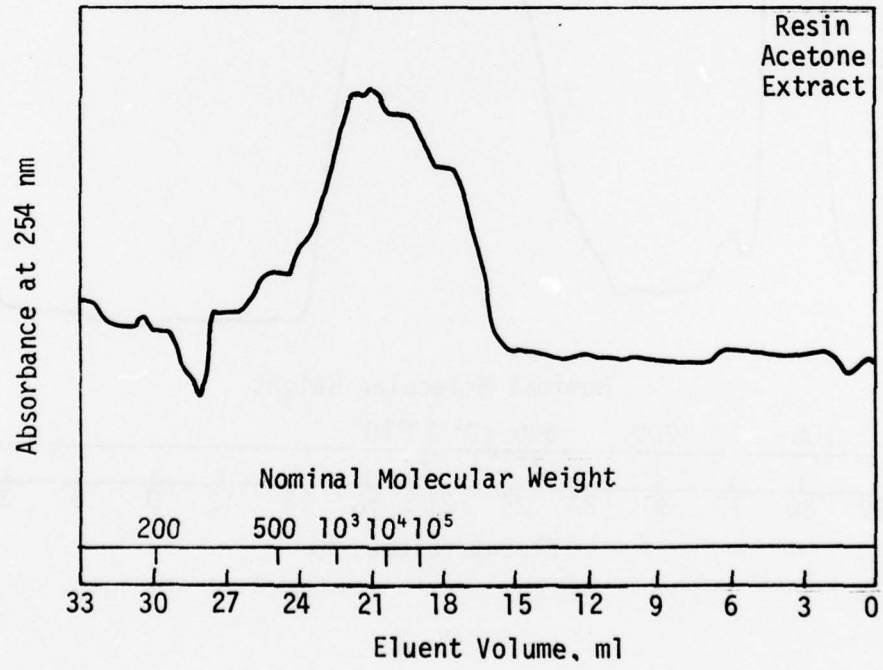

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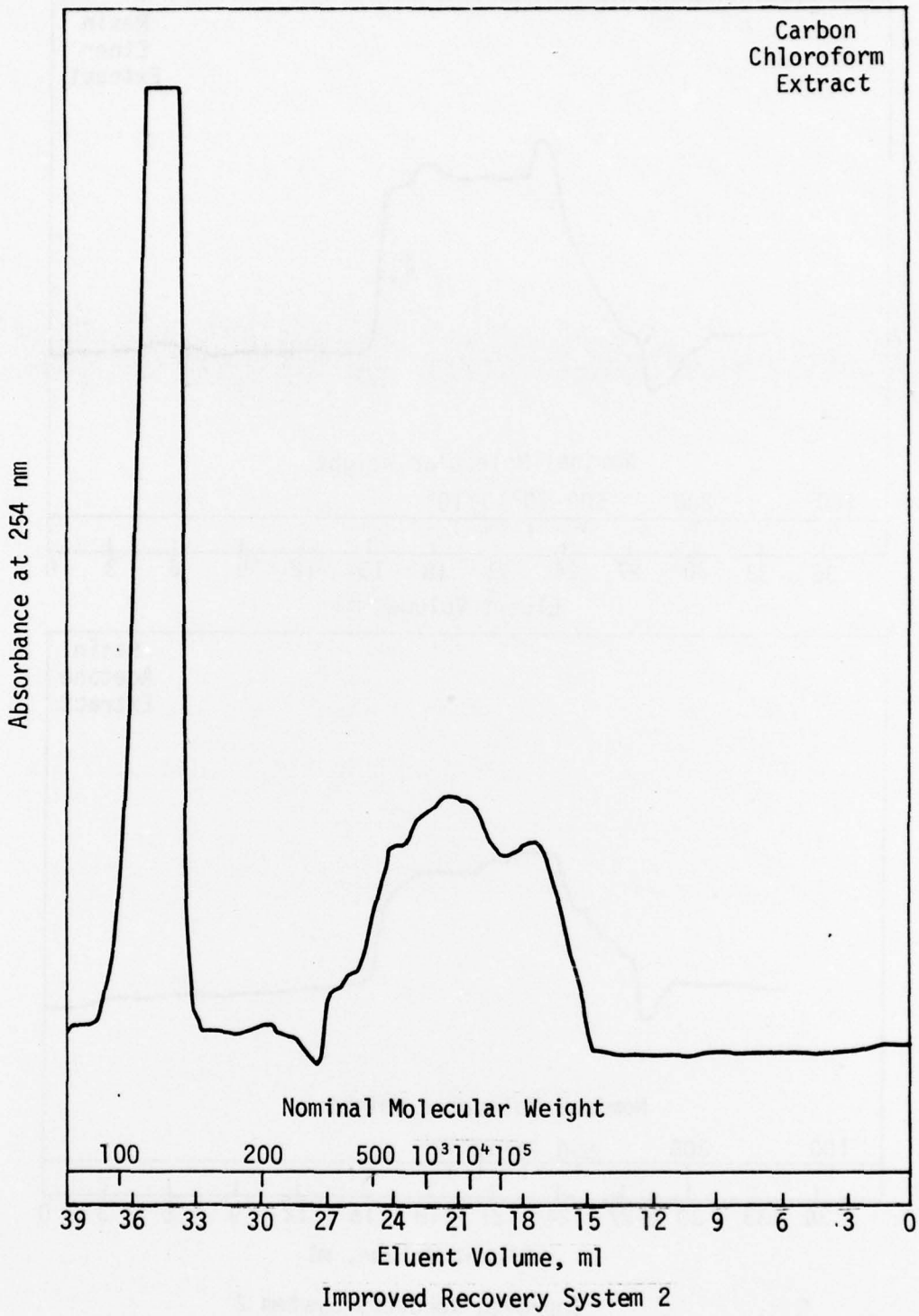
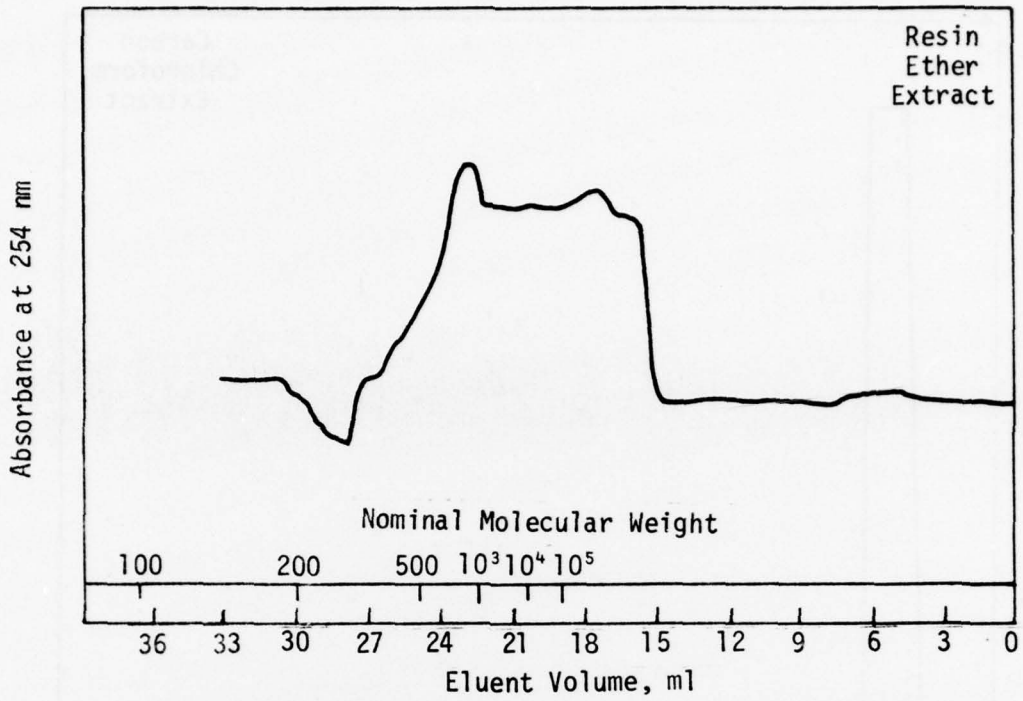
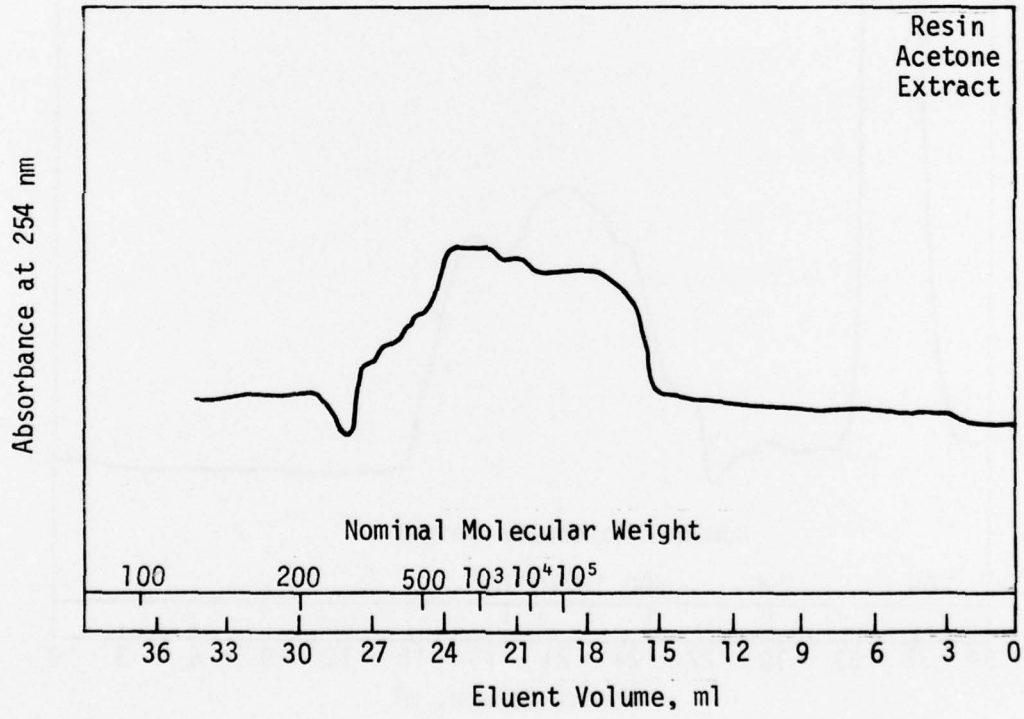


Improved Recovery System 2





Improved Recovery System 2

ABBREVIATIONS, ACRONYMS, AND SYMBOLS

BILD	beta-induced luminescence detector
BOD	biochemical oxygen demand
CAE	carbon alcohol extract
CCE	carbon chloroform extract
COD	chemical oxygen demand
GC	gas chromatography
GC/MS	gas chromatography/mass spectroscopy
GPC	gel permeation chromatograph(y)
HPLC	high-pressure liquid chromatograph(y)
HRLC	high-resolution liquid chromatograph(y)
IR	infrared
IRS	improved recovery system
LASL	Los Alamos Scientific Laboratories
MGD	millions of gallons per day
NVTOC	nonvolatile total organic carbon
O-CA	organics-carbon adsorbable
O-CA-mf	O-CA minifilter
PAW	Polyamide Woelm
PAW-P	PAW-2-propanol
PAW-PW	PAW propanol-water
RAcE	resin acetone extract
RErE	resin ether extract
THF	tetrahydrofuran
TOC	total organic carbon
UV	ultraviolet
UNM	University of New Mexico
XAD-2	XAD-2-ether

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HQ AFISC/SGMS	1
HQ USAFA/Library	1
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AMD/RDU	1
AMD/RDB	1
ADTC/CS	1
AMRL/THE	1
OEHL/CC	3
1 MSEW/SGB	1
Dept of Environmental Sciences California Inst of Technology	1
Dept of Civil Engineering Mass Institute of Technology	1
Toxic Matls Information Center	1
Southeast Environmental Rsch Lab	1
Pacific Northwest Envmtl Rsch Lab	1
Det 1 ADTC/PRT	1
Det 1 ADTC/EC	6
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USAFSAM/EDE	2
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National Science Foundation	1
US Army Med Bioengrg R&D Lab	2
Dept of Civil Engineering Standford University	1
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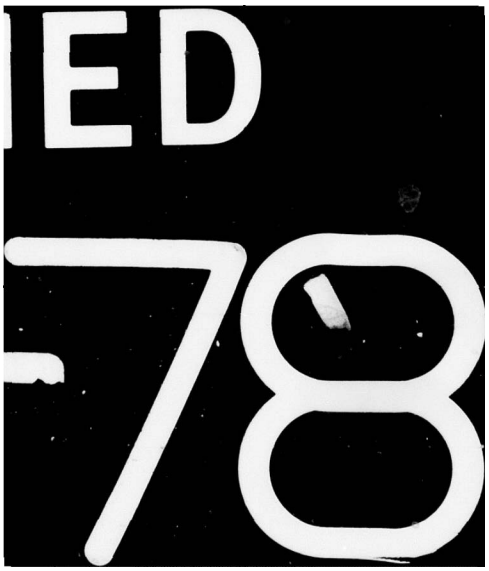
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