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Analysis of Esters and Acids from Petroleum By Combined Techniques

By

Elsa Lacey

A THESIS SUBMITTED
IN PARTIAL FULFILLMENT OF THE
REQUIREMENTS FOR THE DEGREE OF

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Thesis Director's Signature:

Houston, Texas

May, 1972

Dedication

To my husband, Jim.

Acknowledgments

I would like to express my sincere gratitude to Professor J. L. Franklin for his invaluable guidance and support throughout the period of this research.

I am grateful to Dr. B. G. Harnsberger who contributed in innumerable ways to the developments reported herein and to Mrs. Rae Royle who was of great assistance in obtaining the data. I thank Dr. P. A. Haug and Dr. J. A. McCloskey for the mass spectrometric type analysis.

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Introduction

The estimated fifty billion barrels of heavy oil in place in the U. S. have acquired tremendous importance in the last few years. This importance will increase greatly as other types of petroleum reserves become exhausted.

Heavy oils are oils of high density, 0.9037 g/cc or higher,* whose low mobility make their recovery by conventional means extremely difficult. Practical means for recovering significant amounts of the oils that cannot be produced by conventional water displacement have not yet been found.[1]

It has long been agreed that unrecovered heavy oil is retained by capillary forces that are large relative to the viscous forces imposed by water. Differences in chemical composition of the heavy oils, especially those affecting surface activity, may be a factor in heavy oil production by thermal recovery methods. It is conceivable that if certain polar functional groups such as exist in petroleum acids interact with the porous medium, then they would affect displacement processes. It is desirable to investigate some of the polar, surface-active constituents of

---

*Heavy oils are defined as oils having a gravity of 25° API or less.[2]

$$^{\circ}\text{API Gravity} = \frac{141.5}{\text{Sp. Gr. } 60^{\circ}\text{ F}/60^{\circ}\text{ F}} - 131.5$$

where, sp. gr. $60^{\circ}/60^{\circ}$ F = specific gravity of the oil in question at 60° F compared with that of water at 60° F.

heavy oils and their adsorption characteristics. The data obtained may help engineers to design models for future experiments on a more scientific basis and will assist in the interpretation of results already obtained.

The purpose of this research is to study the polar components of heavy oils, particularly the petroleum acids. According to Seifert[3] and others[4], carboxylic acids are the main surface-active components of petroleum. It is possible that carboxylic acids form tenacious films at the interface between heavy oils and sand grains. Adsorption of the acids on the formation may play an important role in heavy oil production. It may be possible to alter "in situ" some of the acids with a consequent improvement in thermal recovery response. Very little research has been conducted in this area.

The study of the polar components of heavy oils was conducted according to the following program:

1. Separation of fractions.

A heavy oil was separated into fractions by several different methods. Various adsorbents and eluents were used.

2. Adsorption studies of the isolated fractions.

Studies of relative heats of adsorption were carried out on selected polar fractions of the heavy oil. The fractions were diluted to standard concentrations and comparative heats of adsorption were determined. A micro-adsorption

detector was used for these measurements. The number average molecular weight of the polar fractions was determined by vapor-phase osmometry.

3. Characterization of the isolated fractions.

One of the fractions, Fraction D, was isolated from a heavy oil by caustic extraction, followed by a series of ion exchange separations. Compared with fractions isolated by other methods, Fraction D exhibited the highest relative heat of adsorption and the value for its number average molecular weight indicated that practically no high molecular weight materials or "asphaltenes" were present. On this basis, Fraction D was selected for further studies. Fraction D was methylated and its methyl esters were further separated by urea, thiourea and gas chromatography; mass spectrometry was used in an attempt to identify and characterize individual components.

Historical

Although carboxylic acids are considered to be the logical precursors of petroleum hydrocarbons,[5,6] very little has been known of the structure of naphthenic acids with ten or more carbons.[7] Chemically, the naphthenic acids have been classified as carboxylic acids of the formulas $C_nH_{2n-2}COOH$ and $C_nH_{2n-4}COOH$, with the largest group falling into the former class.[8]

The naphthenic acid mixture is one of the most complex yet examined. Separation of the components of higher molecular weight present in the mixture is quite difficult.[9,10]

Carboxylic acids are of great geochemical significance because of the role they are thought to have played in the origin of life on earth[11,12] and on the origin of petroleum.[13-16] Many investigators have pursued the study of these materials but most of them have not dealt with whole crude oils. The work before 1940 on the occurrence and composition of naphthenic acids in petroleum is summarized by Ellis[17] and Littmann[8] and the work up to 1955 by Lochte and Littmann.[18] Lochte and Littmann concluded that "naphthenic acids" range from 6 to more than 20 carbon atoms and consist predominantly of cyclopentyl acids.

Bock and Behrend[19] studied commercial naphthenic acid mixtures by gas chromatography and found that about 33% of the mixture consists of branched-chain fatty acids with a

methyl group in position 3 and a considerable amount of branched chain fatty acids with an ethyl group in the same position. They also found C_{11} and C_{12} branched carboxylic acids; normal fatty acids were not detected. Cason and Liauw[20] identified a C_{11} monocyclic acid. Cason and Graham[21] identified C_{14}-C_{15}, C_{19}-C_{20} isoprenoid acyclic acids as well as normal fatty acids from C_6-C_{18}. Cason and Khodair[22] found 3-ethyl-4-methylcyclopentyl acetic acid which was the first nonisoprenoid branched C_{10} or larger acid that had been identified. They also found C_{10}-C_{12} normal acids and a C_{11} acyclic isoprenoid acid, i.e. 4,8-dimethyl nonanoic acid. The material on which the investigation by J. Cason and co-workers was carried out was a refinery-treated sample of commercial naphthenic acids.[23]

The most recent work has been that of Seifert[24], who extracted whole crude oils with sodium hydroxide-alcohol-water and separated the resulting acids by ion-exchange and silica gel chromatography. He studied a low viscosity oil (95 centistokes at 100° F) and found acids amounting to 2.5 wt. % of the total crude oil. The number average molecular weight for his acid fraction is 300-400. His was the first semiquantitative attempt to analyze a fraction of the carboxylic acids from a total virgin crude oil. A carboxylic acid fraction isolated from crude oil by Seifert consisted of about 1500 compounds.[25] The most abundant species identified in this fraction contained 2, 3, 4, and 5 saturated

rings and fused polynuclear structures. Seifert's findings illustrate the complexity of the acid fractions isolated from petroleum.

It should be noted that the following acids have been isolated from petroleum: 2-methyl pentanoic, 2-methyl hexanoic, cyclopentanoic carboxylic, 2-methyl cyclopentanoic, 3-methyl cyclopentanoic, cyclopentyl acetic, 3-methyl-cyclopentyl acetic, 2,3-dimethyl cyclopentyl acetic, cyclohexyl acetic,[20-22] trans 2,2,6-trimethyl cyclohexyl acetic,[20] and benzoic acids.[20] Several series of aromatic carboxylic acids have been isolated from the extract of Colorado Green River shale.[26]

Part I: Crude Oil Samples

Discussion

A - Description of Heavy Oil Samples

The starting material for our studies consisted of samples of produced heavy crude oils from Midway Sunset Field*, California; they exhibit a range of API gravities from 12.1 to 20.4. Available crude oil data are given in Tables I and II. (Note: All the results listed in Table II were determined experimentally.) Data for the water associated with the oils are found in Table III. New, unused containers were selected for sampling the heavy oils to insure against contamination. The samples were taken from the wellhead.

It has commonly been noted that crude oils produced from relatively young geological formations tend to be rather heavy (viscous) and naphthenic in composition while crude oils from older rocks generally are light and paraffinic in nature.[28,29] Our data are in agreement with the above; the younger oils (Pleistocene) show a higher viscosity at 100° F than the older oils (Pliocene and Upper Miocene**), with one exception, Heavy Oil #8 (Tables I and II). It appears that

---

*Midway Sunset Field is in the southwest end of the San Joaquin Valley, Kern County, California.[27]

**The Miocene epoch occurred from 10 million to 25 million years ago; the Pliocene epoch occurred from one million to 10 million years ago; the Pleistocene epoch occurred from ten thousand to one million years ago.

TABLE I

CRUDE OIL RESERVOIR CHARACTERISTICS

MIDWAY SUNSET FIELD

Crude No.	Lease	Formation	Age	Gas-Oil Ratio	Prod. Zone Temp., °F
1	West Min (NCT-1)	Leutholtz	Upper Miocene	0	122
2	West Min (NCT-1)	Tulare	Pleistocene	0	101
3	Section 32 Fee	Lakeview	Upper Miocene	0	125
4	Section 32 Fee	Shallow (Hallmark)	Pliocene	0	90-95
5	Section 36 Fee	Etchegoin	Pliocene	0	114
6	Section 25 Fee	Tulare	Pleistocene	0	100
7	Section 36 Fee	Tulare	-	0	-
8	Section 12 Fee	S. J. Clay (Tar Sand)	Pliocene	0	±100
9	Section 25 Fee	Etchegoin	Pliocene	0	114

TABLE II

CRUDE OIL CHARACTERISTICS

Crude No.	Specific Gravity, °API	Viscosity,* cs At 100° F	Viscosity,* cs At 210° F	Pentane** Insolubles, % By Weight	Neutralization Numbers, (mg KOH/g Oil)
1	18.0	226	14.8	8.75	2.23
2	12.8	1584	36.5	9.18	4.69
3	20.4	61.6	7.82	9.99	2.18
4	15.1	320	17.3	9.09	4.37
5	17.5	116	9.84	9.75	3.27
6	12.1	3185	49.4	9.12	5.00
7	15.1	402	17.9	8.98	4.16
8	12.5	2761	47.6	10.73	4.57
9	16.9	182	12.6	5.24	2.48

*Kinematic viscosity determined on a Cannon-Fenske viscometer.

**Refer to the Experimental section, p. 40.

TABLE III

WATER CHARACTERISTICS

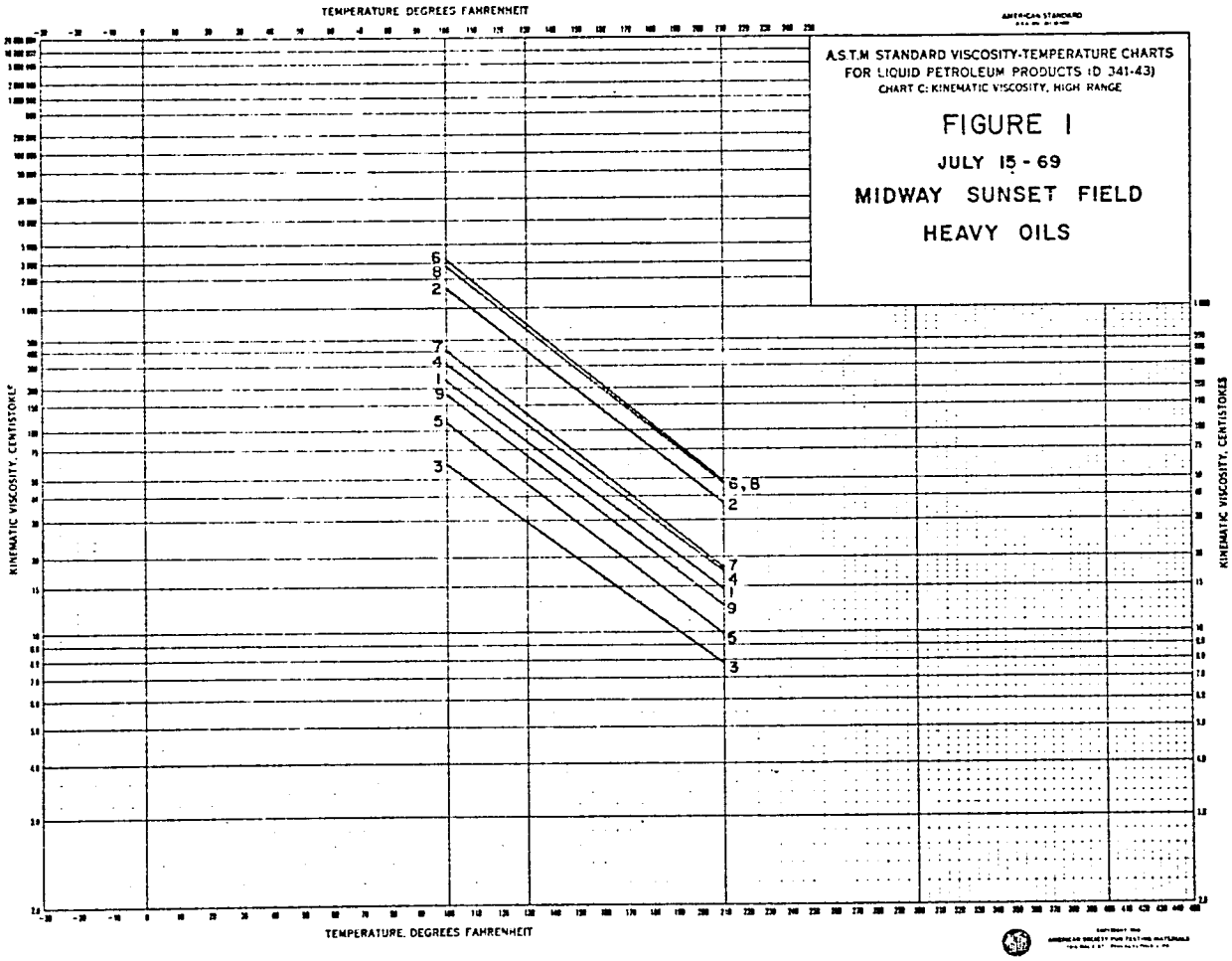
Crude No.	Water Content of Oil, Volume %: Emulsified Water	Water Content of Oil, Volume %: Free Water	Cl^- in ppm	pH
1	7.1	49.0	6,400	8.35
2	12.9	6.8	2,000	7.85
3	0.4	30.3	7,900	7.90
4	3.1	10.4	7,900	8.30
5	0.7	23.1	15,300	7.95
6	32.9	5.2	6,300	8.55
7	2.1	8.1	2,300	8.55
8	22.1	10.3	5,700	8.00
9	0.7	28.2	20,800	7.75

crude oil undergoes a maturation process leading from a heavy, naphthenic crude to a lighter, paraffinic crude. There is experimental evidence indicating the importance of temperature and the presence of natural catalysts in the maturation of crude oil.[30,31] The effect of temperature on the viscosity of the heavy oils is shown on Figure 1.

Samples of oil must come from the same producing interval in order to be comparable from a geological standpoint. The heavy oils that come from the same producing interval are #2 and #6; #5 and #9.

Pentane insolubles were obtained for all nine heavy oils and the values in % by weight range from 5.24 (for Heavy Oil #9) to 10.73 (for Heavy Oil #8). These values are an indication of the amount of high molecular weight materials present in the oils. The pentane insolubles data are tabulated in Table II.

Neutralization numbers (weight in milligrams of KOH required to neutralize 1 gram of sample) were also determined on all heavy oil samples (Table II). Heavy Oil #6 exhibits the highest neutralization number. It is interesting to note that it was more difficult to remove the water from this particular oil (#6) than from any of the other oils. A valid speculation would be that the amount of acid constituents in the oils is related to the presence of very stable oil-water emulsions.


TEMPERATURE DEGREES FAHRENHEIT
A.S.T.M STANDARD VISCOSITY-TEMPERATURE CHARTS
FOR LIQUID PETROLEUM PRODUCTS (D 341-43)
CHART C: KINEMATIC VISCOSITY, HIGH RANGE
FIGURE 1
JULY 15 - 69
MIDWAY SUNSET FIELD
HEAVY OILS
KINEMATIC VISCOSITY, CENTISTOKES
KINEMATIC VISCOSITY, CENTISTOKES
6
8
2
7
4
1
9
5
3
6,8
2
7
4
1
9
5
3
TEMPERATURE, DEGREES FAHRENHEIT

Samples from the nine heavy crude oils were analyzed by NMR spectroscopy in an effort to distinguish the crudes from each other by this technique. Characteristic resonance positions of the probable proton types are given in Table IV. The intensity of a resonance band in an NMR spectrum is directly proportional to the number of hydrogen atoms contributing to that band. The NMR spectra of the nine oils are shown as NMR Spectrum No. 1-1 through NMR Spectrum No. 9-9. Superimposed on the spectra are graphic records of the integrated NMR signal allowing the area under appropriate regions to be measured.

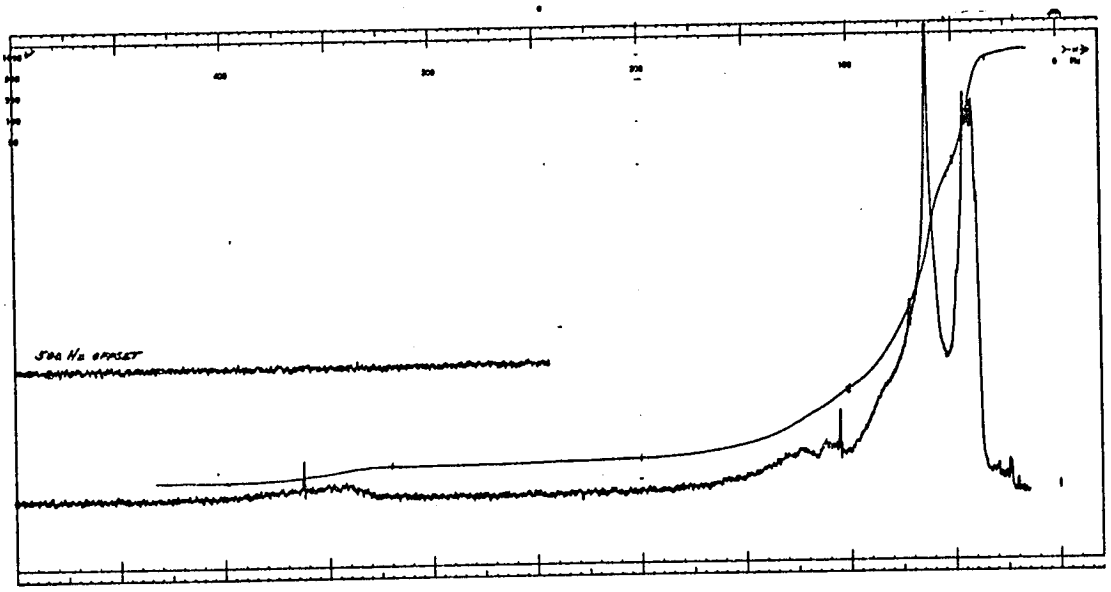
No peak corresponding to acidic protons could be detected in the NMR spectra. This could indicate either no acidic protons were present or that the band is too broad to be observed. A broad acidic band is not unusual in this type of material.

Relative proportions of other types of hydrogen were computed (Table V). For samples measured under the same conditions of resolution and other spectrometer operating parameters, the branchiness index (B.I. = branchiness index = H_γ/H_β) accurately measures their relative branchiness.[32] The branchiness index values for all nine heavy oils are clustered together, with oils #5 and #4 exhibiting the lowest and highest values, respectively (Table VI). Since the aromatic content is low in our heavy oil samples, a B.I. measurement may be used to determine the naphthenic

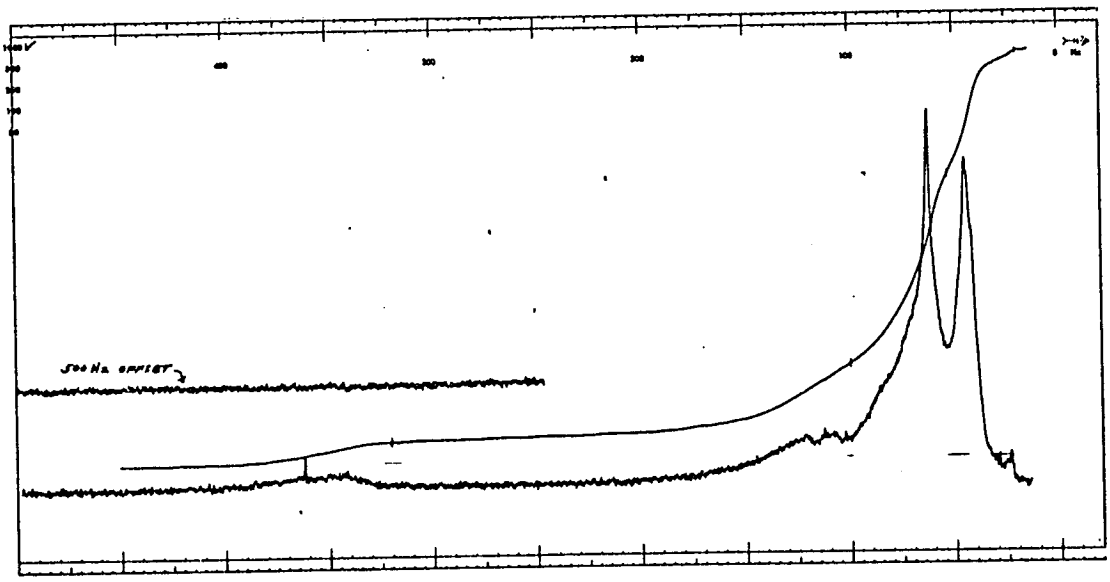
TABLE IV

ASSIGNMENT OF PROTON BANDS IN NMR SPECTRA OF HEAVY CRUDE OIL SAMPLES

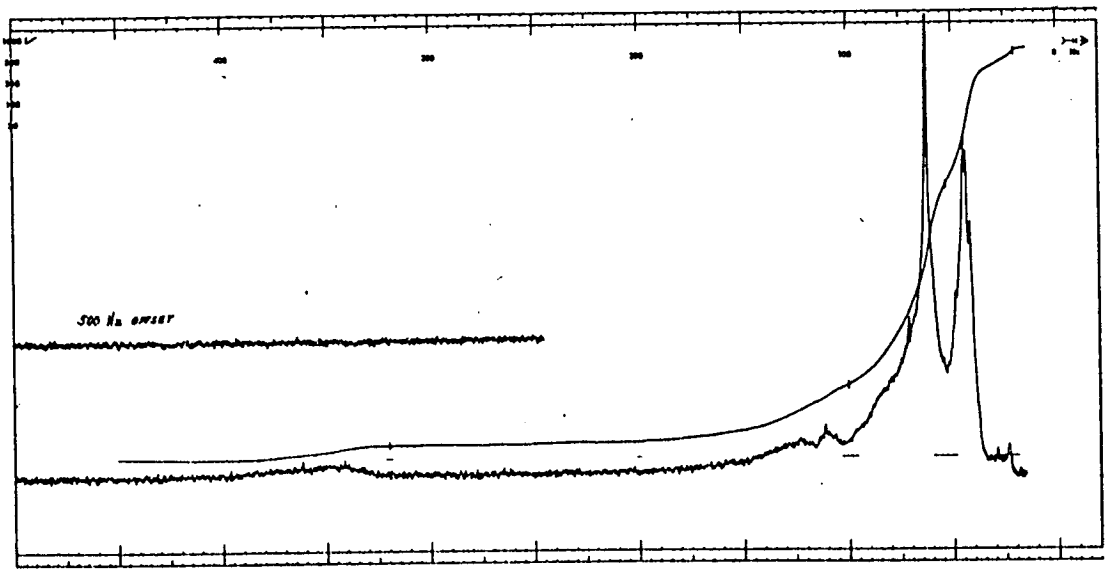
Range of Band Envelope Assignments (Hz from TMS)		Proton Types
40 - 106	H_{γ}	Paraffinic methyl and/or methyl gamma or further removed from aromatic rings.
106 - 200	H_{β}	Hydrogen of paraffinic methylene, methenyl, and naphthenes or methylene groups beta or further from an aromatic ring as well as methyl beta to an aromatic ring (the sharp portion of this peak is probably due to polymethylene structure).
200 - 400	H_{α}	Methyl, methylene, and methine alpha to an aromatic ring and possibly some methylene and methine beta to the ring. Also naphthenic groups alpha to an aromatic ring and methylene alpha to two aromatic rings. (This region also includes methyl and methylene groups alpha to nitrogen or sulfur.)
640 - 800	H_{A}	Aromatic protons (phenolic hydrogen if present).
900 -1000	H_{Other}	Acidic protons in this solvent.



NMR SPECTRUM NO. 1-1



NMR SPECTRUM NO. 2-2



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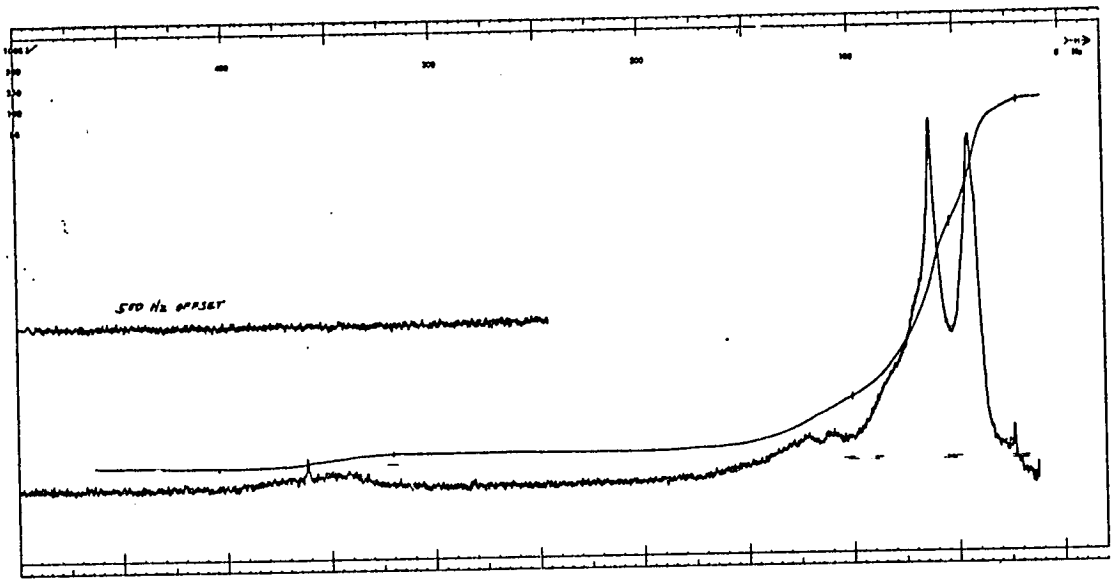

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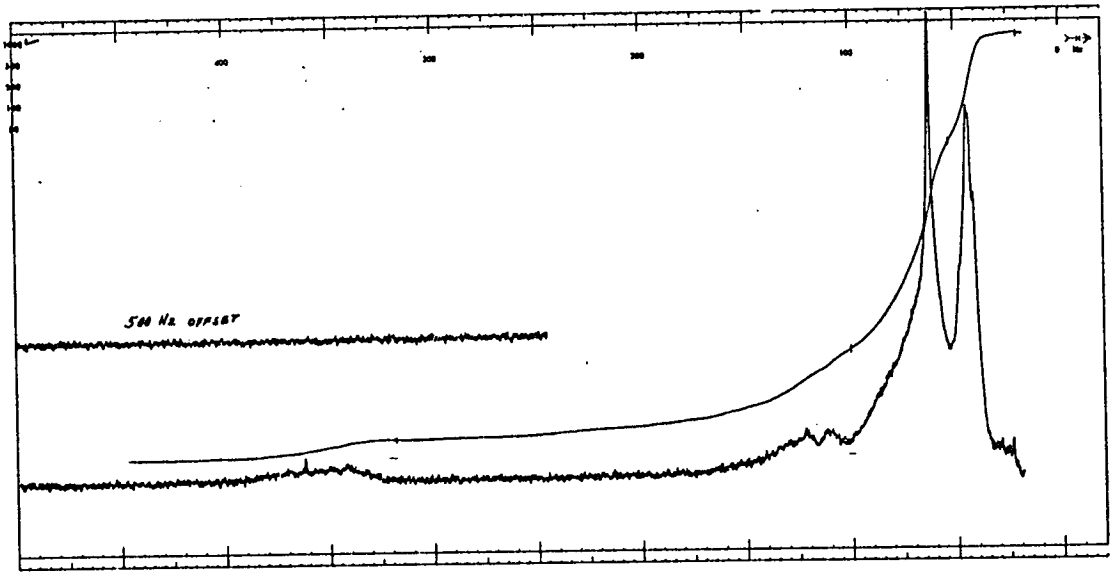
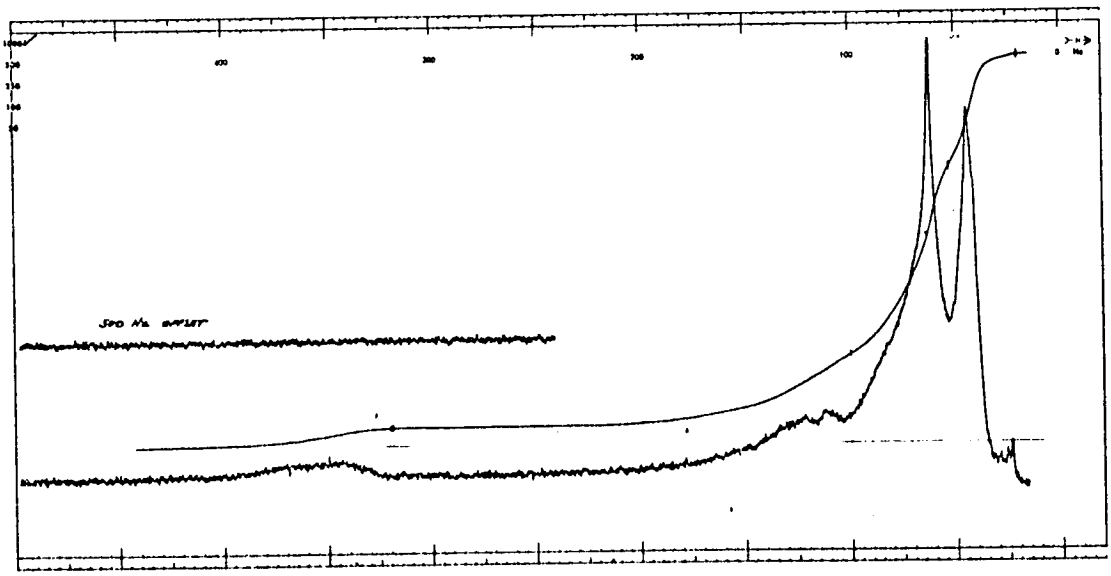


NMR SPECTRUM NO. 5-5



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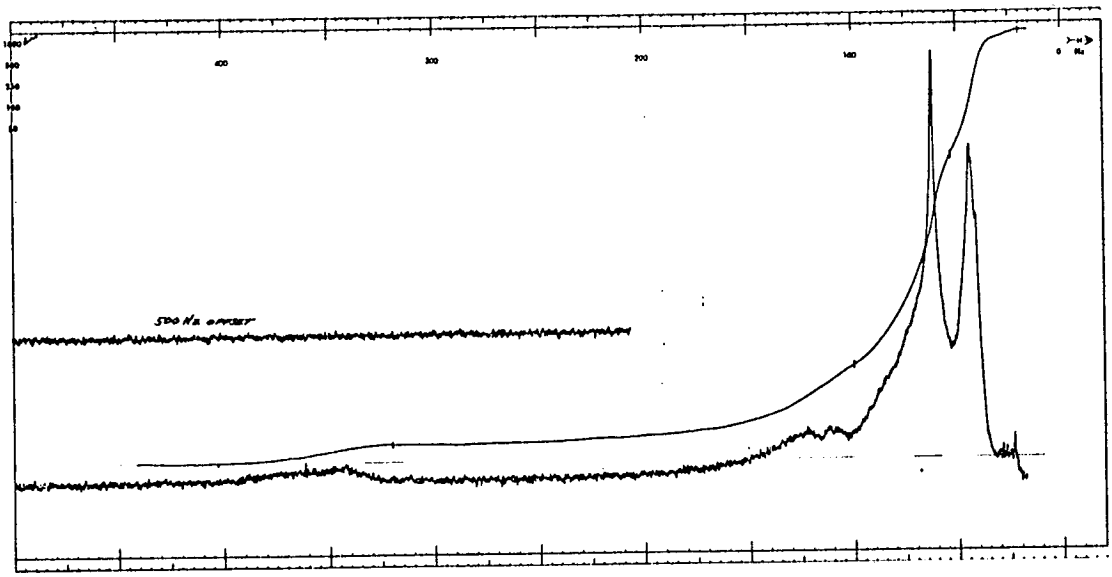

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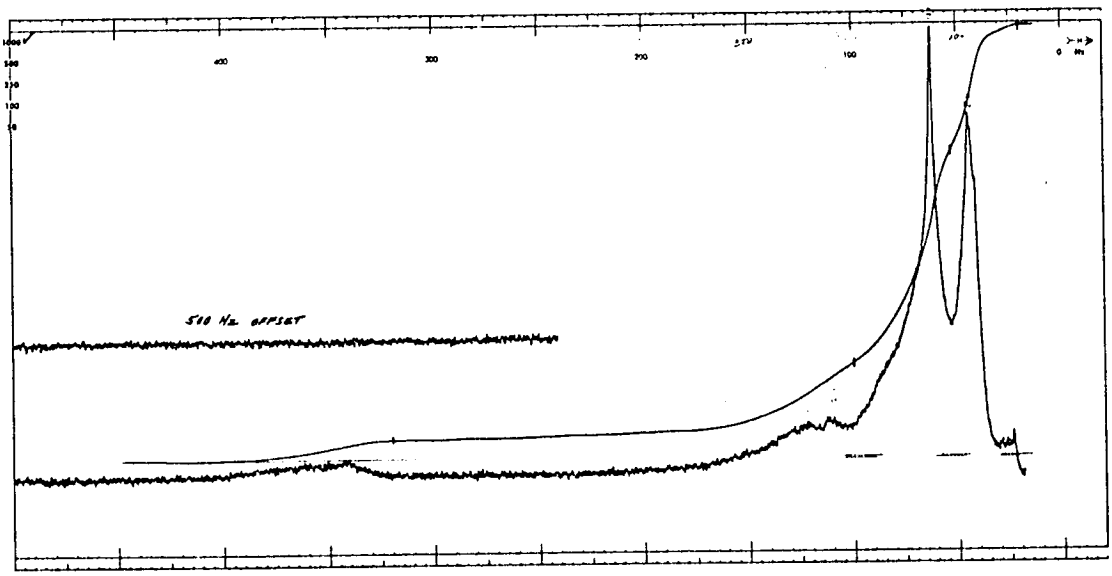
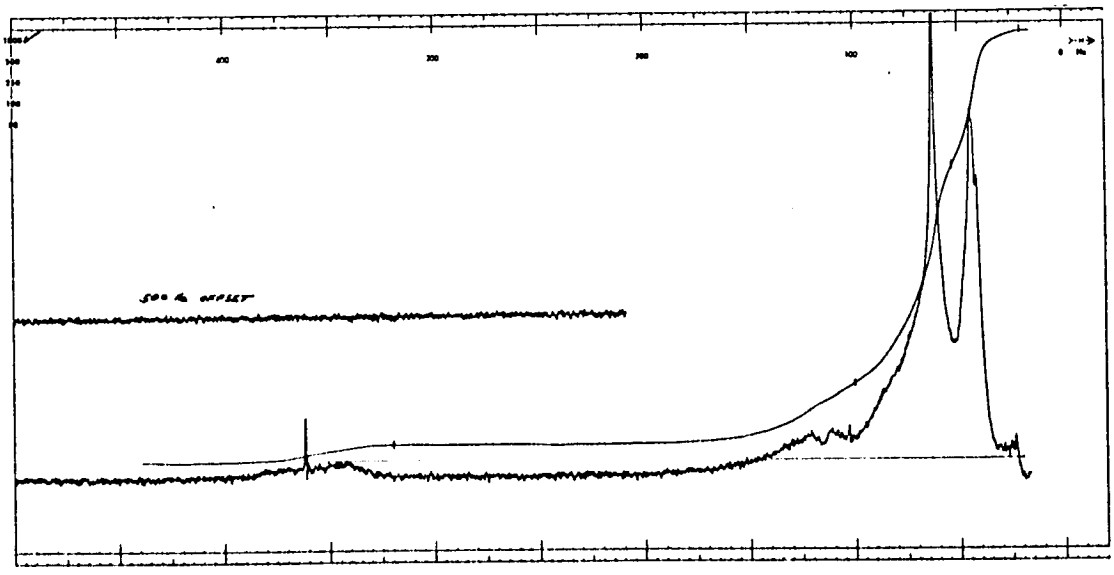


NMR SPECTRUM NO. 8-8



NMR SPECTRUM NO. 9-9

TABLE V

NMR VALUES FOR PROTON RATIOS OF HEAVY OILS

Crude No.	NMR Spectra No.	H_γ/H*	H_β/H	H_α/H	H_A/H
1	1-1	51.3	36.0	10.7	2.0
2	2-2	50.5	35.5	11.6	2.4
3	3-3	53.0	35.9	9.4	1.7
4	4-4	53.9	35.3	9.2	1.6
5	5-5	48.5	36.8	12.5	2.2
6	6-6	50.5	35.8	11.5	2.2
7	7-7	50.9	36.2	10.6	2.3
8	8-8	50.9	35.9	11.0	2.2
9	9-9	52.7	36.1	9.4	1.8

*Total integral

TABLE VI

NMR-DERIVED PARAMETERS FOR HEAVY OILS

Crude No.	NMR Spectra No.	B. I.*	% C_N
1	1-1	1.42	82.54
2	2-2	1.43	83.08
3	3-3	1.48	85.79
4	4-4	1.52	87.97
5	5-5	1.32	77.11
6	6-6	1.41	81.99
7	7-7	1.40	81.45
8	8-8	1.42	82.54
9	9-9	1.46	84.71

*B.I. = Branchiness index.

ring carbon content (% C_N). The nonaromatic carbon in naphthenic rings (% C_N) is obtained from the NMR data by the empirical formula of Williams:[33]

$$\% C_N = 54.3 (B.I. + 0.10)$$

The % C_N for all nine oils is very high with values ranging from 77.11 (Heavy Oil #5) to 87.97 (Heavy Oil #4). The % C_N is tabulated in Table VI.

The NMR-derived parameters for all nine heavy oils were quite similar in value; consequently, NMR spectroscopy could not be used to distinguish those oils. However, it was of interest to learn that these heavy oils show very high naphthenic content.

The heavy oils were associated with large amounts of water; the total water content ranged from 10.2% by volume for Heavy Oil #7 to 56.1% by volume for Heavy Oil #1 (Table III). The pH of the associated waters did not vary much from one oil to another; the pH range was from 7.75 - 8.55. It is interesting that the oils themselves were acidic as evidenced by the neutralization numbers given in Table II while the waters associated with the oils are slightly basic.

The water associated with each heavy oil was also analyzed for chlorides (Table III). The values for Cl^- in ppm cover a very wide range from 2,000 ppm for Heavy Oil #2 to 20,800 ppm for Heavy Oil #9. The two oils that exhibit unusually high chloride content are Heavy Oil #5 (15,300 ppm of Cl^-) and Heavy Oil #9.

The series of tests and analytical methods just described were applied to the heavy oils and their associated waters in an effort to learn as much as possible about their characteristics. Some of the tests, such as pentane insolubles determination and chloride determination, are routinely used in the oil industry for characterization of the oils and waters. That is not the case with some of the other methods, such as nuclear magnetic spectroscopy.

In general, the heavy oils have been found to have very high viscosities, to be acidic in nature, and to have a high naphthenic content. The heavy oils were associated with large amounts of water; in each case, the waters showed a basic pH.

B - Removal of Water from Heavy Oils

As received from the field, the heavy oil samples contained large amounts of water, ranging from 10% to 56% by volume. In order to use them for our studies, it was necessary to separate the oil from the water. Samples from the nine heavy oils were treated in a number of different ways in an attempt to separate entrained and/or emulsified water from the oil. Due to the nature of the oils it was a difficult task to remove the water. It appears that suspended water in high viscosity crude forms a very tight emulsion which is difficult to break without addition of chemicals. The different heavy oils vary considerably in their ability to release water from emulsions.

Shake-out tests to determine the percent of water present in the oil were performed periodically.

A description of the attempts to eliminate water follows, in the order in which they were carried out:

1. Centrifugation at room temperature at 1,000 rpm.

   At this point, the centrifuge could be operated only at room temperature. This approach was slow and tedious; besides that, some of the water could not be eliminated by the method even after many centrifugations.

2. Application of heat by heating mantles surrounding 500 ml separatory funnels.

   By itself, this method was unsatisfactory.

3. Alternate heating and centrifuging at room temperature.

   This method was successful with most of the samples except with those of highest viscosity.

4. Several types of distillation.

   Azeotropic distillation with benzene at 130° C: The main disadvantage of this distillation is that the oil becomes diluted with benzene rendering the sample useless for viscosity measurements.

   Atmospheric distillation at 170° C: The main drawback to this approach is that the high temperatures required could cause thermal alterations.

   Vacuum distillation at 86° C: This distillation is the most satisfactory one, although, due to the available glassware and to the large volumes of oil needed for our research, it would be very time consuming.

It was found that vacuum, atmospheric and azeotropic distillations successfully reduced water content in the oils to less than 1% by volume.

5. Molecular sieves.

The use of a molecular sieve column proved unsatisfactory. The flow of oil was extremely slow even though the column temperature went as high as 200° F. The flow of the oil through the sieves is in itself exothermic. Large volumes of heavy oils remain attached to the molecular sieves. Due to adsorption, some of the oil components are retained permanently by the molecular sieves.

6. Centrifugation with heat.

A large centrifuge was redesigned and modified for centrifugation with heat. The water could be removed by centrifuging at elevated temperatures (85 - 130° F). Shake-out tests gave negative results after repeated centrifuging for extended periods of time.

It was, therefore, concluded that repeated centrifugation at temperatures of 85° - 130° F and for extended periods of time was the best approach to water removal from heavy oils. This method was applied to all our heavy oils before subjecting them to any separation techniques.

C - Evaluation of Two Separation Methods and of Three Types of Distillations

The purpose of our research was the study of polar components of heavy oils. It was therefore necessary to select

a separation method that would isolate the polar fraction from the rest of the heavy oil. After previous screening, it was decided to test two separation methods: one consisted of silica gel chromatography; the other one was based on caustic extraction.

At the same time it occurred to us that it was of interest to try to find out whether various types of distillation, i.e. vacuum, atmospheric, and azeotropic, would thermally alter the polar fractions of heavy oils.

Heavy Oil #8, because of its high viscosity, was selected for the evaluation of the separation methods and of the distillation.

Silica gel chromatography[34] was performed on three samples obtained after subjecting Heavy Oil #8 in one case, to vacuum distillation, in another case to atmospheric distillation, and in still another one, to azeotropic distillation with benzene (Table VII). Petroleum acids and other polar compounds were eluted with ether from the silica gel column. The percent of polar components (ether fraction) basis original heavy oil are tabulated in Table VIII. The ether fraction, Fraction 8-1, was later split into two parts: Fraction 8-1a (1.32 g) and Fraction 8-1b (1.50 g).

Fraction 8-7, eluted with 5:5:20:70 water:acetone:acetic acid:isopropanol, was partially insoluble in benzene (Table VII). The insoluble material of white, flaky appearance was analyzed and it contained iron oxide, silica, carbonates and

TABLE VII

DETERMINATION OF THE EFFECTS OF VARIOUS DRYING PROCEDURES UPON THE PERCENTAGES OF POLAR FRACTION OBTAINED BY SILICA GEL CHROMATOGRAPHY

HEAVY OIL #8

	Dried By Azeotropic Distillation At 130° C			Dried By Vacuum Distillation At 86° C		Dried By Atmospheric Distillation At 170° C	
Eluent	Volume Of Eluent, liter	Recovered Fractions, Wt. % Of Crude Oil	Fraction No.	Volume Of Eluent, liter	Recovered Fractions, Wt. % Of Crude Oil	Volume Of Eluent, liter	Recovered Fractions, Wt. % Of Crude Oil
Benzene	46	Discarded	-	-	Discarded	14	Discarded
Ether	2	6.9	8-1	-	6.1	1	5.7
50/50 Methanol/Ether	1.5	3.2	8-5	-	-	-	-
Methanol	1	0.2	-	-	2.7	-	-
175/48 Chloroform/Methanol	3.8	1.6	8-6	-	-	-	1.1
Chloroform	1	0.1	-	-	-	-	-
50/50 Ethyl Acetate/Chloroform	1	-	-	-	-	-	-
Ethyl Acetate	1	-	-	-	0.7	-	-
50/50 Acetone/ Ethyl Acetate	1	-	-	-	-	-	-
Acetone	0.5	-	-	-	-	-	-
5/5/20/70 Water/ Acetone/Acetic Acid/Isopropanol	1	1.6	8-7	-	-	-	-

TABLE VIII

ANALYSIS OF ETHYL ETHER FRACTIONS AND OF MAJOR ACIDIC EXTRACT OF HEAVY OIL #8

Separation Methods	Distillation	Fraction No.	Wt. % Of Crude Oil	Melting* Point, °C	Molecular Weight**	IR Spectra No.	***$\varepsilon_{COOH}/\varepsilon_{CH_2}$
Ethyl ether fraction eluted from silica gel.	Azeotropic	8-1	6.9	72.5	1227	8-1	0.42
	Vacuum	8-2	6.1	73.5	1345	8-2	0.43
	Azeotropic	8-3	5.7	63.5	1304	8-3	0.41
Major Acidic Extract by caustic extraction.		8-4	2.6	27.5	573	8-4	0.45

*Determined on a Fisher-Johns Melting Point Apparatus.

**Number average molecular weight obtained by vapor-phase osmometry (Hitachi Perkin Elmer Model 115) in solutions in $CHCl_3$.

***Extinction coefficient ratio (infrared) of $\varepsilon_{1720\ cm^{-1}}/\varepsilon_{2950\ cm^{-1}}$.

unidentified organic matter. A spectrographic examination showed the presence of: Mg, 37%; Na, 35%; Ca, 20%, and Fe, 7%. This insoluble material could be impurities from the silica gel and from the solvents, or inorganic material present in the heavy oils or residue from evaporation of the water. The origin of this insoluble material remained in an unexplained basis.

The second separation method consisted of treating Heavy Oil #8 by caustic extraction (Table IX). This approach yielded a fraction labelled Major Acidic Extract[24] consisting mostly of acids and phenols.

Several measurements and analyses were performed on the Ether Fractions and on the Major Acidic Extract fractions to learn about their composition. Melting points and number average molecular weights were determined for the four fractions (Table VIII). The melting point values for all three ether fractions were fairly close (average value of 69.8° C) but the melting point of the Major Acidic Extract was significantly lower at 27.5° C. The number average molecular weights of the ethyl ether fractions range from 1,227 to 1,345; the value obtained for the Major Acidic Extract was considerably lower at 573. These two sets of measurements indicate that the ether fractions contain a large amount of high molecular weight materials such as asphaltenes. It is of interest to us to obtain a fraction as pure in carboxylic acid content as possible.

The polar fractions were further examined by NMR spectroscopy, infrared spectrometry and mass spectroscopy.

It was not possible to detect the acid protons by NMR (Tables VIII and IX). The spectra (NMR Spectrum No. 8-10 through 8-13) were nondescriptive and all four fractions had similar NMR spectra. The NMR-derived values (Table IX) were fairly close for all four fractions. NMR does not appear to be a good method for looking at mixtures of acids because very frequently the concentration of acid protons is too low (relative to the other types of protons) to contribute significantly to the NMR signal[35].

The IR spectra of all four polar fractions (IR Spectrum No. 8-1 through 8-4 and Table VIII) showed a carbonyl band at 1720 cm^{-1}, indicating the presence of carboxylic acids. The extinction coefficient ratios of $\epsilon_{1720\ cm^{-1}}/\epsilon_{2950\ cm^{-1}}$ are practically the same for all four fractions (Table VIII). Bands at 1380 cm^{-1} and at 1470 cm^{-1} are due to a combination of CH_2 and CH_3 bending vibrations. Bands at 3250 cm^{-1} are indicative of free hydroxyl. Strong C-H stretch appears at 2950 cm^{-1}. IR spectrum of the Major Acid Extract fraction, No. 8-4, shows a higher moisture content by the presence of a stronger band around 3300 cm^{-1}. All four IR spectra are quite similar. The primary difference is the absorption intensity in a band appearing around 790 cm^{-1}; this most likely due to solvent (CCl_4). Comparison of the most intense bands of the three ether fractions among themselves

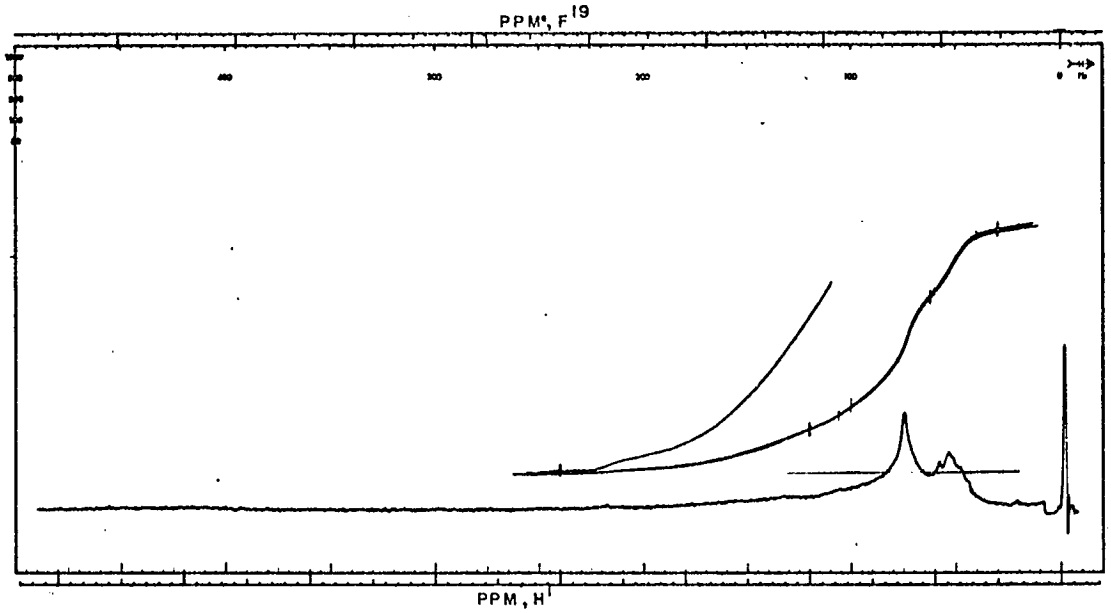
TABLE IX

VALUES DERIVED FROM NMR SPECTRA
OF POLAR FRACTIONS OF HEAVY OIL #8

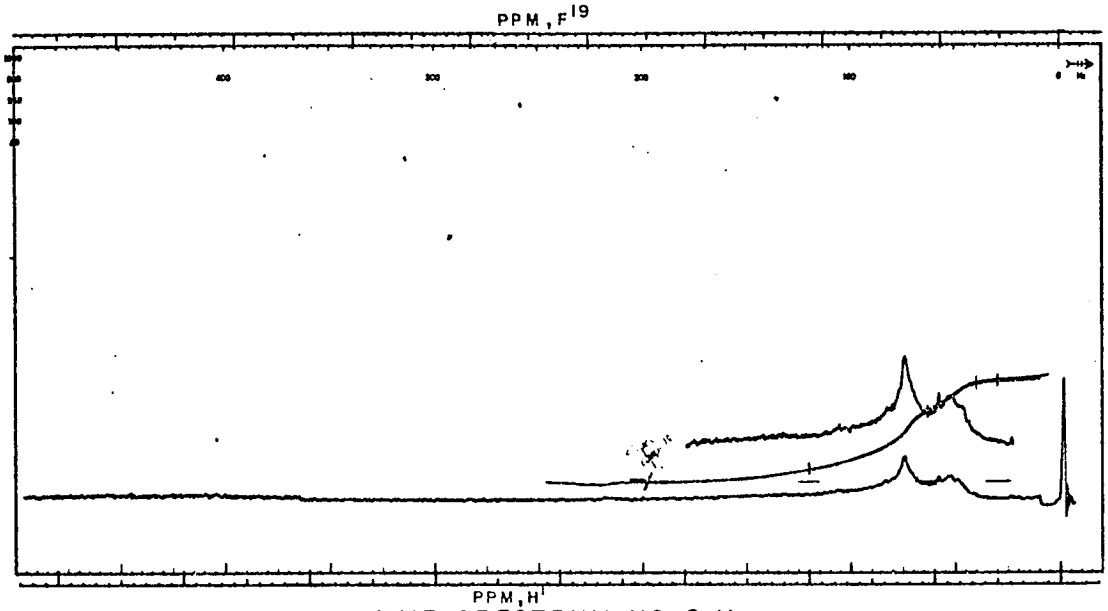
Fraction No.	NMR Spectra No.	H_γ/H*	H_β/H	H_α/H	H_A/H	B.I.**	% C_N
8-1	8-10	52.5	38.3	9.2	-	1.37	79.82
8-2	8-11	55.0	38.2	6.8	-	1.44	83.62
8-3	8-12	51.5	38.2	10.3	-	1.35	78.74
8-4	8-13	48.4	36.0	12.7	2.5	1.34	78.19

*Total integral

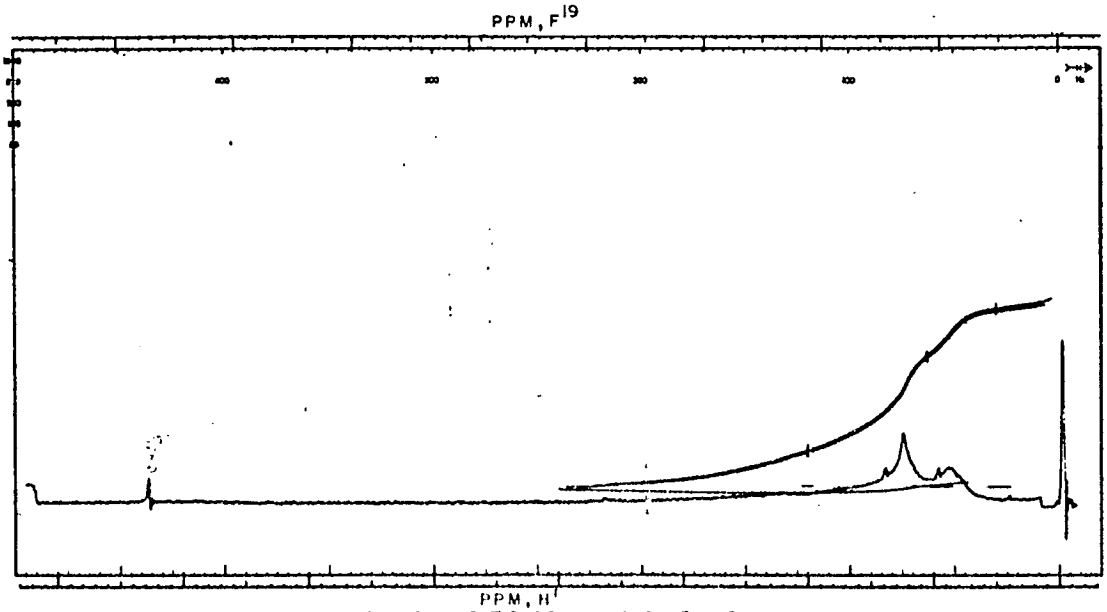
**B.I. = Branchiness index.



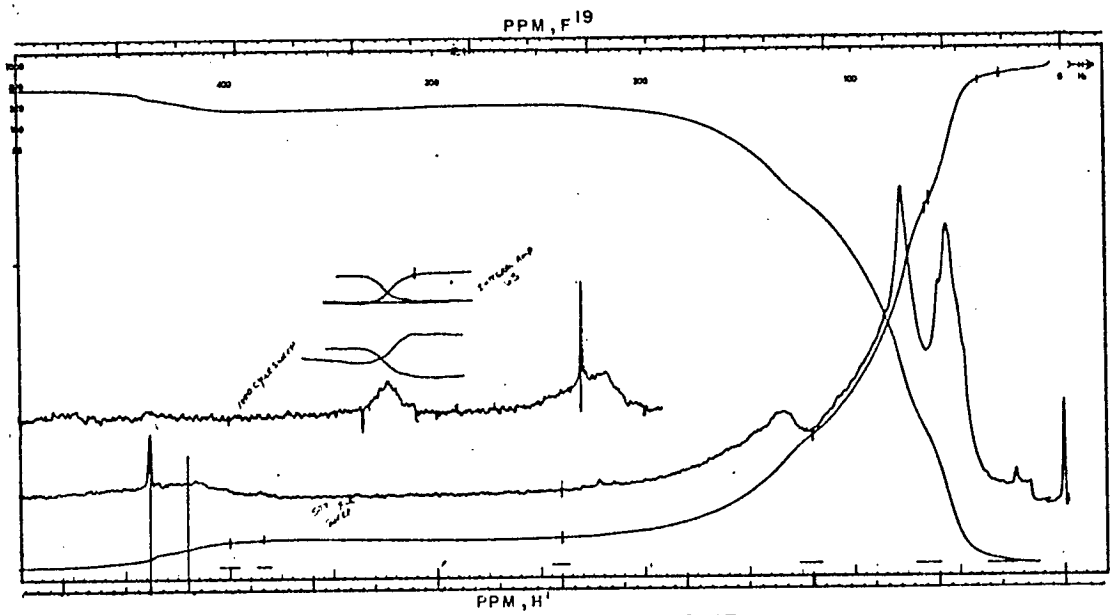
NMR SPECTRUM NO. 8-10



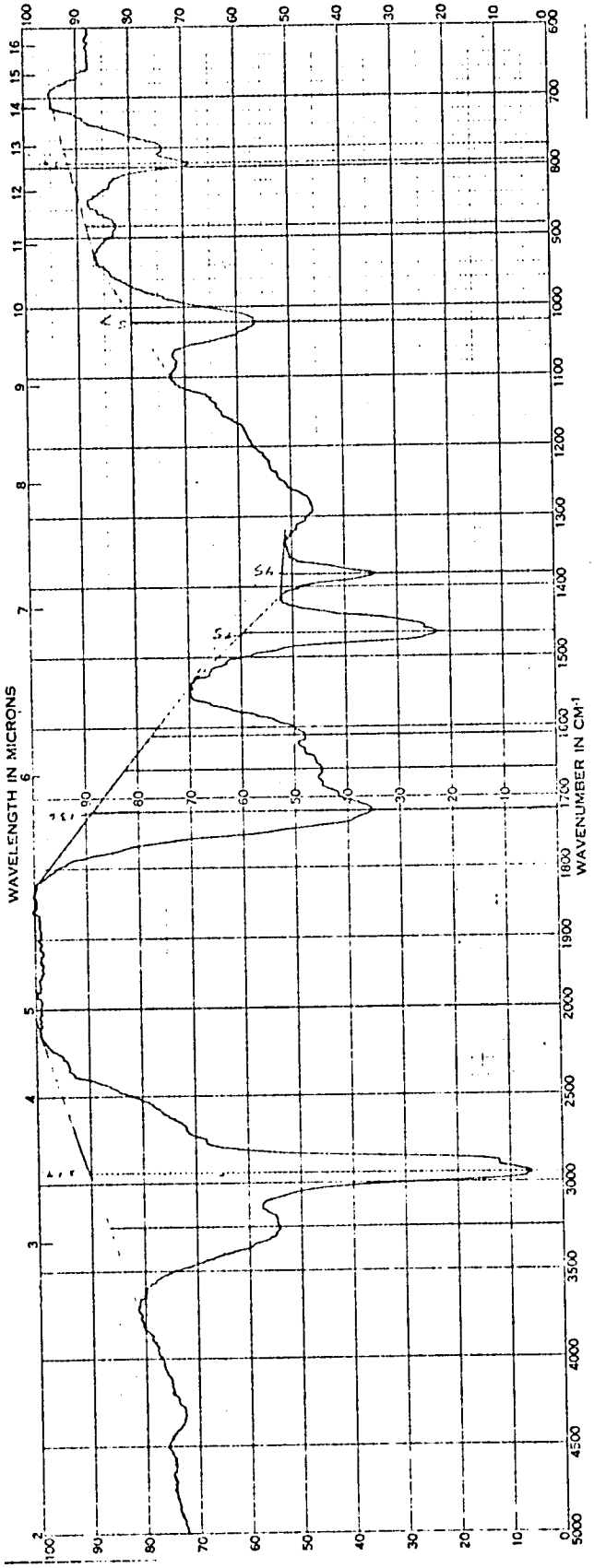
NMR SPECTRUM NO. 8-11



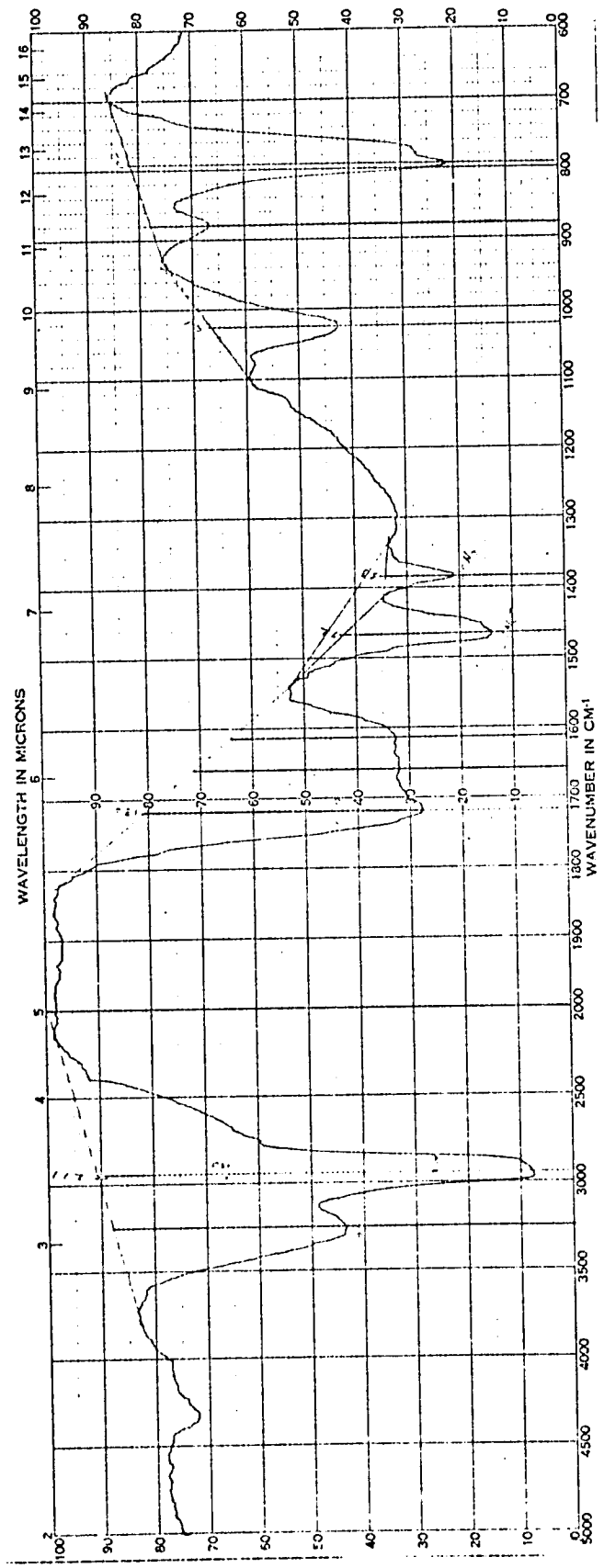
NMR SPECTRUM NO. 8-12



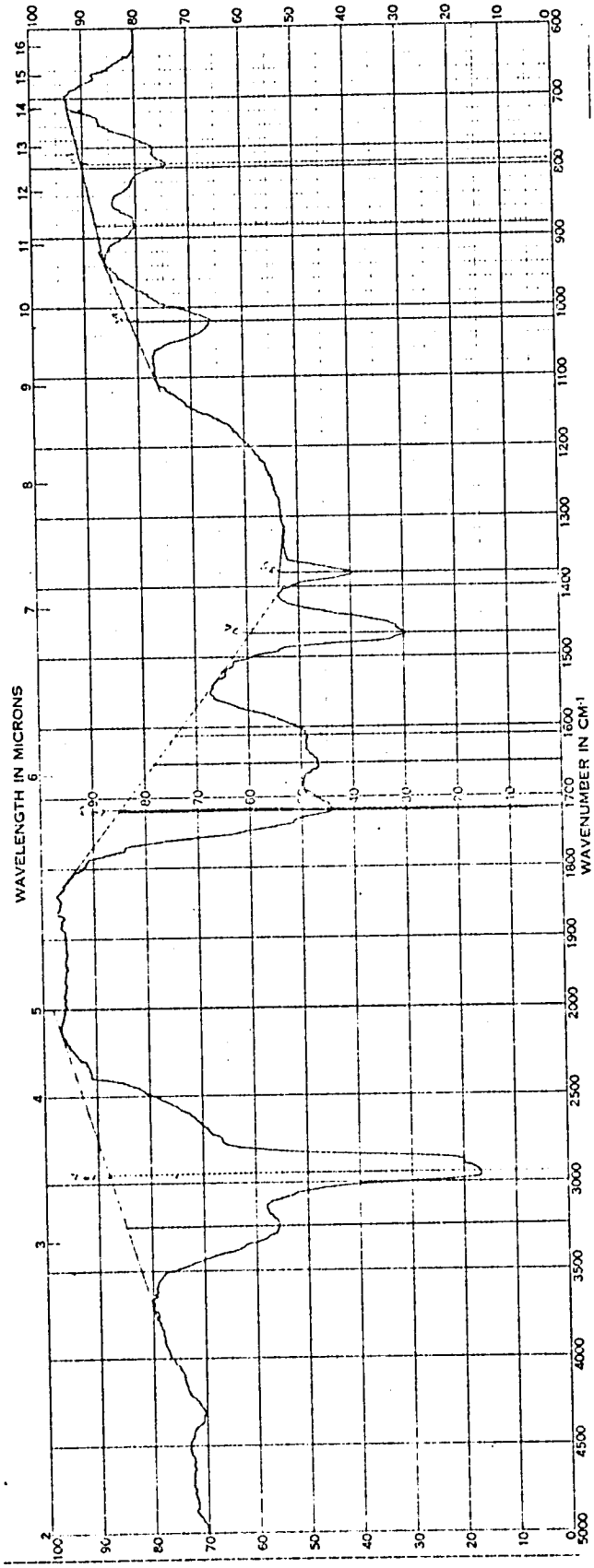
NMR SPECTRUM NO. 8-13



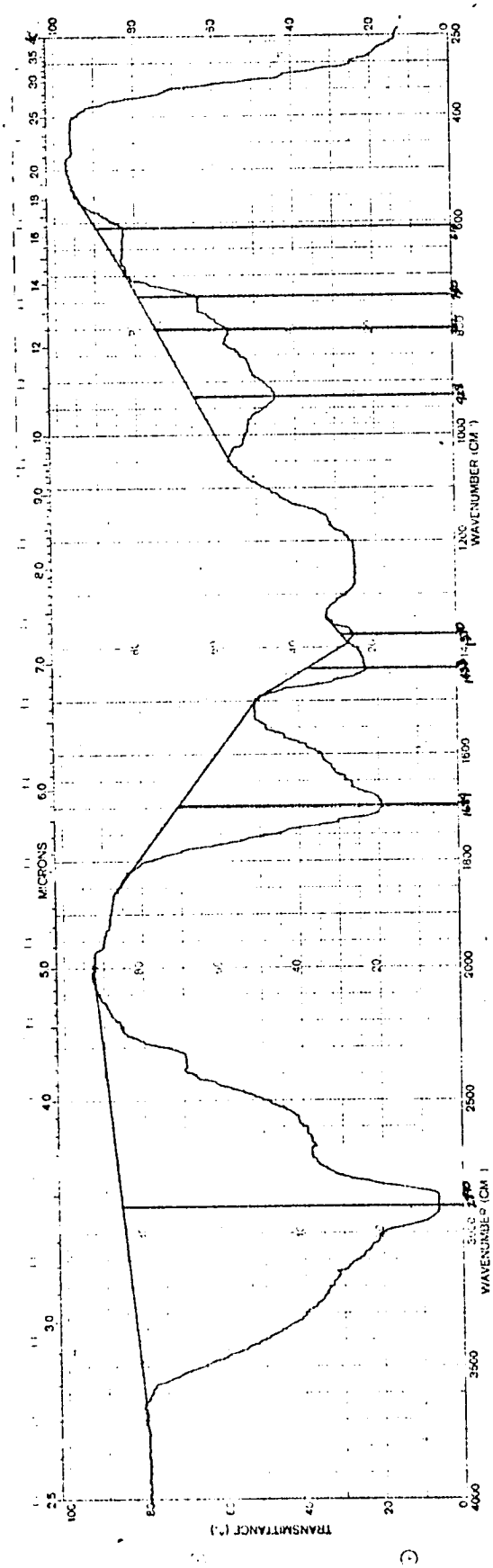
IR SPECTRUM NO. 8-1



IR SPECTRUM NO. 8-2



IR SPECTRUM NO. 8-3



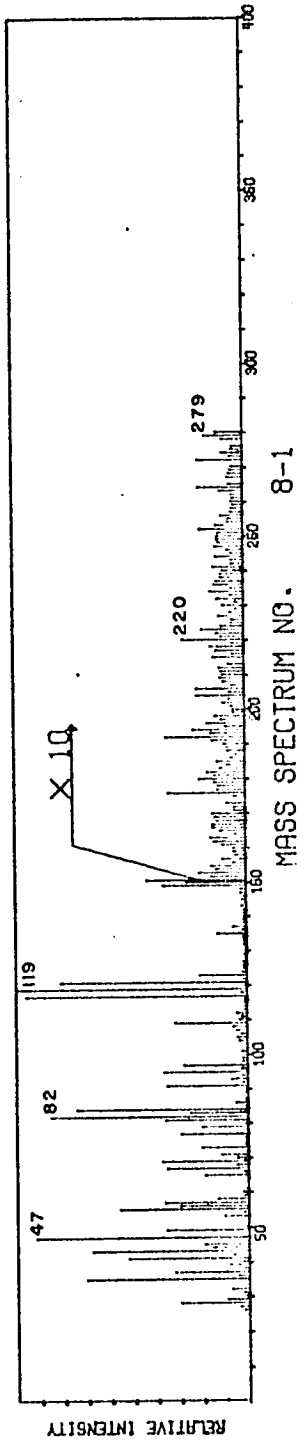
IR SPECTRUM NO. 8-4

and with the Major Acidic Extract fraction showed no significant differences. (The three ether fractions came from oil which had been distilled by various techniques; the Major Acidic Extract fraction was isolated from oil that had not been distilled.) No evidence of thermal alteration was thus observed by IR spectroscopy.

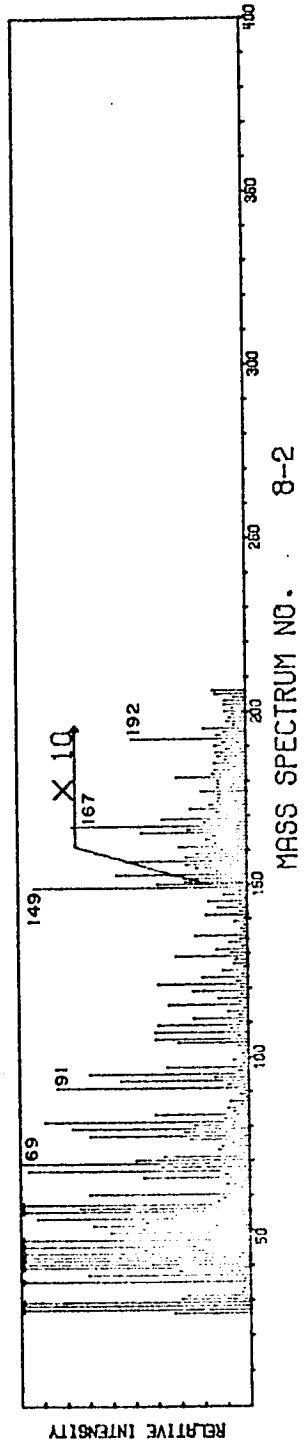
The mass spectra of two of the ether fractions and that of the Major Acidic Extract fraction (Mass Spectrum No. 8-1, No. 8-3 and No. 8-4) show strong contamination by CCl_4.[36] The mass spectrum of the ether fraction derived vacuum-distilled oil (Mass Spectrum No. 8-2) shows a molecular ion at m/e 192 and peaks of very high intensity at m/e 69 and at m/e 149 (Table X).

The mass spectra of the polar fractions were not very revealing. Based on molecular weight data, the spectra should have exhibited peaks at higher masses than m/e 280. Mass spectra were difficult to interpret on these samples because of solvent contamination.

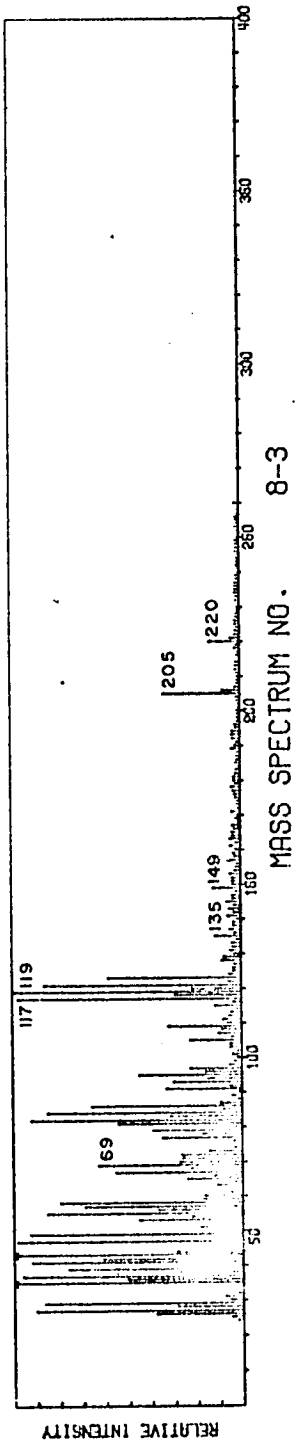
Summarizing the results obtained by the various techniques, it is obvious that the three ether fractions are fairly similar to each other and that no detectable decomposition occurred during the distillations. The Major Acidic Extraction fraction appears to be quite different from the ether fractions. Evidently, the Major Acidic Extract fraction is a narrower, more homogeneous fraction than the ether fraction. Hence, it seems more desirable to use a caustic extraction method for isolating the carboxylic acid fraction than silica gel chromatography.



MASS SPECTRUM NO. 8-1



MASS SPECTRUM NO. 8-2



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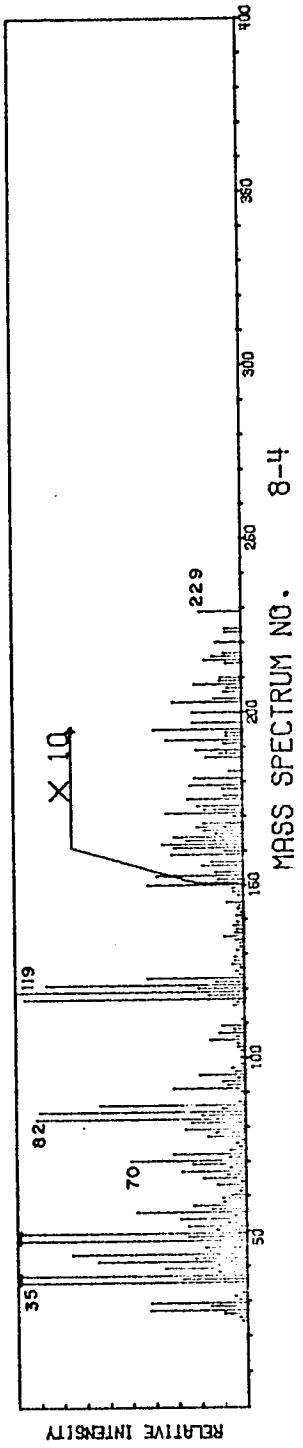


MASS SPECTRUM NO. 8-4

TABLE X

PARTIAL MASS SPECTRA OF POLAR FRACTIONS FROM HEAVY OIL #8

MS No.	Fraction No.	m/e	% Intensity	Remarks
8-1	8-1	69	37.5	
		119	100	Base peak due to contamination from CCl_4.
		135	12.7	
		149	36.6	
		205	0.7	(M-15)
		220	2.7	M, Molecular ion.
8-2	8-2	41	150.5	Base peak.
		69	100	
		135	22.9	
		149	93.1	
		177	2.0	(M-15)
		192	5.0	M, Molecular ion.
8-3	8-3	43	117.9	Base peak.
		69	62.9	
		135	11.2	
		149	12.0	
		205	33.4	(M-15)
		220	13.2	M, Molecular ion.
8-4	8-4	35	113.2	Base peak due to contamination from CCl_4.
		69	23.4	
		135	8.8	
		149	5.0	
		193	0.7	(M-15)
		208	2.1	M, Molecular ion.

Experimental

Pentane insolubles - The pentane insolubles were precipitated by addition of pentane to a weighed sample of heavy oil. They were then filtered, evaporated to dryness, and weighed.

Neutralization numbers - Neutralization numbers were determined on water-free samples of heavy oils by ASTM D974-64 method.[37] The sample size was limited to 2 g to minimize interference by the dark color.

NMR spectroscopy - Nine heavy crude oils were analyzed by NMR spectroscopy. Particular emphasis was placed on scanning for acidic protons. The spectra were obtained with a Varian Model HA-100D spectrometer operating at 100 mega Hertz. These NMR spectra were run at Texaco Research Laboratories in Beacon, New York.

Carbon tetrachloride was used as a solvent because it does not contain hydrogen atoms and does not yield interfering bands. Tetramethyl silane was used as internal reference. The spectral range covered was 1500 Hz from the zero reference; on the spectra this includes a 0 - 1000 Hz scan and a 500 Hz offset scan.

In an attempt to observe an acidic proton peak, a trace of trifluoroacetic acid was added to the solvent. The amount of acid added is insufficient by itself to give a peak. However, the added trace of acid should be enough to

increase the concentration of acidic protons in the sample so that a peak corresponding to acidic protons is detected. This technique is frequently successful, but in this case the results were negative. The trace of added acid could not be detected. Although NMR is not very sensitive, it should detect one part in ten thousand of a single, sharp proton type.

Proton ratios were calculated by comparing the integral of each proton band in a spectrum to the total proton integral, H.

Chloride determination - The water associated with each heavy oil was analyzed for chlorides using the following procedure: Pipette 1 ml of sample into a casserole and check alkalinity with phenolphthlein. If basic, neutralize with H_2SO_4 (0.02 N). Add 5% K_2CrO_4 indicator and titrate with $AgNO_3$ (1 M) to brick-red end point (Factor = 1,000).

Shake-out test - This test consists of thoroughly mixing equal volumes of oil and benzene followed by centrifugation for 30 minutes at high speed on a Precision Scientific Serial D centrifuge.

Centrifugation at room temperature - The heavy oils were centrifuged at 1,000 rpm in an International Centrifuge Model BE with 6-place horizontal head #267, trunnion carriers #392 and 125 ml Squibb separatory funnels. Separatory funnels were rinsed periodically with benzene and acetone; otherwise drops of water seem to adhere to the glass walls and eventually drop back into the oil.

Molecular sieves - A glass chromatographic column was packed with activated molecular sieves Linde type 3A. Sieves were activated by heating for 24 hours at 200° C in a vacuum oven. A sample of Heavy Oil #6 was loaded onto the column. A heating tape wrapped around the column provided heat and reduced the viscosity of the oil.

Silica gel chromatography - A solution of 46.5 g of Heavy Oil #8 in 50 ml of benzene was placed on a silica gel column (59 by 4.5 cm I.D.). Solvent mixtures of increasing polarity were employed for eluting various fraction (Table VII).

NMR spectroscopy - Four polar fractions isolated from Heavy Oil #8 were analyzed by NMR spectroscopy at Rice University, Houston, Texas. NMR spectra were obtained with a Varian A-56/60 spectrometer, at a constant oscillator frequency of 60 MC with CCl_4 as solvent and tetramethyl silane as internal standard.

Infrared spectrometry - Infrared spectra of the three ether fractions isolated from Heavy Oil #8 were recorded using a Beckman Model IR-4 infrared spectrophotometer. IR spectrum of Major Acidic Extract fraction was obtained with a Perkin Elmer Model 457. Spectra were obtained from dry neat films on AgCl discs. Carbon tetrachloride was used as the solvent.

Mass spectroscopy - Low resolution mass spectra for the four polar fractions isolated from Heavy Oil #8 were

determined on a C.E.C. mass spectrometer, Model 21-110b, with ionizing voltage of 70 ev. These spectra were run at Rice University, Houston, Texas. The samples were introduced by a direct inlet probe into the ion source.

Samples for mass spectra were prepared in capillary tubes. Difficulty was encountered in sample preparation because of the high viscosity of the fractions. The procedure followed was to dissolve the sample in CCl_4 and evaporate as much solvent as possible in a vacuum desiccator. The vacuum was then turned off and the capillary tube was allowed to fill completely with the sample. The capillary tube was placed in the rotary evaporator for several hours to eliminate the solvent.

Part II: Heat of Adsorption Studies

Discussion

A - Introduction

Liquid chromatography is an area that has been at a standstill for many years, mainly because of lack of a good detector to identify the fractions. At the time this research was initiated (August 1968) a new detector[38], the Micro-Adsorption Detector (M.A.D.) had just come onto the market and it seemed very promising. The M.A.D. is a simple, sensitive, and universal detector for liquid chromatography. It consists of two thermistors and a cell surrounding one of the thermistors that is filled with a suitable adsorbent material. Detection is based on the same process responsible for separation in a chromatographic column and, in principle, all compounds which can be separated can be detected. The detection thermistor can sense a temperature rise as small as 6×10^{-5} °C. Theoretically with this detector it is possible to separate fractions based on their different adsorption heats.

The M.A.D. appeared to have a tremendous potential as detector for liquid chromatography. An even more intriguing application was the possibility of relating the response of the detector (mainly due to adsorption heats) to one of the factors influencing thermal response in secondary recovery methods. This factor is the different adsorption strengths on sand grains of some of the chemical constituents of

heavy oils. The detector can be used to determine relative heats of adsorption for different sample-solvent-adsorbent combinations. Some experiments could be run using silica gel and other common adsorbents in the cell but one could also use some very finely divided sand in order to simulate field conditions more closely. This approach to measuring adsorption forces had not been previously reported but it appeared to be a significant area of research and potentially of great value to thermal recovery studies.

The initial research plans involved the use of the M.A.D. as an aid in the separation of polar fractions from the heavy oils by liquid chromatography. Also, the thermal detector response would be used in the study of relative heats of adsorption of the different isolated fractions.

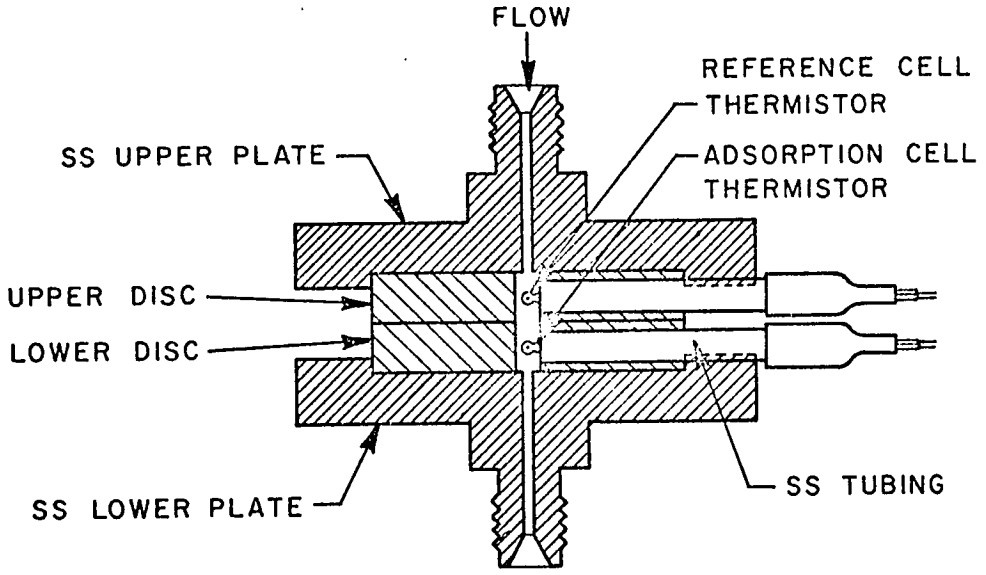
B - Instrumentation

The instrument selected was the Micro-Adsorption Detector, Varian Aerograph Model 4000. Varian's Model 4000 is a complete system that includes the detector, a constant temperature bath, a gas pressurized solvent reservoir[39], an injector, and a bridge power supply and amplifier.

As shown in Figure 2, the detector has two identical chambers. The glass encased thermistors are bonded into discs made of Teflon, and the discs are damped between stainless steel flanges. The lower chamber surrounding one thermistor is filled with a suitable sorbent material

FIGURE 2

CROSS-SECTIONAL VIEW
OF MICRO-ADSORPTION DETECTOR



and acts as the detection cell. Detection is based on measurement of an extremely small amount of heat evolved during sorption of the sample on this material. The upper chamber contains another thermistor and functions as a reference cell, thus minimizing noise and baseline drift. Both cells are 0.27 cm in diameter by 0.41 cm long and have an empty volume of approximately 20 μℓ. Dead volume is minimized by filling the reference cell with microscopic glass beads. The packings in the cells are held in place by three stainless steel screens. The maximum sensitivity obtained with this detector is in the order of 10^{-8} gm of substance, while the response is linear over a concentration range of at least 2 powers of 10. For example, the response was found to be linear over the ranges 0.5 - 20 μg and 0.05 - 3.0 μg.[38]

The peak shape produced by M.A.D. is dependent upon flow rate, type of adsorbent, nature of solute and nature of mobile liquid.[40] The signal of an ideal M.A.D. spectra would be the differential of a gaussian curve.

Our instrument was one of the first ten units of the Model 4000 to be manufactured. In the first seven months following delivery of the instrument our efforts to obtain satisfactory performance were futile. Serious problems encountered in the use of M.A.D. were random noise, unstable or drifting baseline, low sensitivity, and poor reproducibility.

Investigations indicated that M.A.D. was very sensitive to changes in solvent flow rates, to variations in sample injection techniques, and to localized fluctuations in the ambient temperature. Several modifications were introduced to correct each one of these problems. An SS-25A Nupro fine metering valve, an SS-2F-7 Nuprofilter and a Vernier handle for Nupro "S" were installed in the exit line from the solvent reservoir to achieve constancy of flow. A Hamilton Constant Rate CR-700 Syringe which delivers like quantities of similar liquids in like times was found necessary for reproducible sample injections. To minimize the effects of the temperature fluctuations, an atmospheric cabinet was built to enclose M.A.D. and related equipment.

Another problem encountered was a sudden increase in signal noise occurring after several hours of continuous operation. The cause was found to be the gas coming out of solution in the detector because of the pressure drop. Elimination of this type of noise is most efficiently accomplished by periodically emptying the solvent reservoir and refilling it with fresh solvent.

In some instances, a capillary restrictor was used with the M.A.D. unit instead of a column. A capillary restrictor is a stainless steel capillary tube with a piece of wire inside to restrict the flow. With the capillary restrictor, flow rates decreased after several hours of operation. Small particles seemed to plug up the capillary restrictor. Flow rate changes caused variations in the sensitivity of the instrument.

According to Varian[41], the accuracy of M.A.D. results varies: 3.5% for areas; 1.3% for total peak height; and 1.4% for half peak height. Our findings agree substantially with theirs. In this study we arbitrarily based our measurements on total peak height (positive plus negative components).

A M.A.D. chromatogram consists of a positive peak proportional to the heat and speed of adsorption and a negative peak due to the heat and speed of desorption. Basically, the process in the detector chamber starts with the adsorption of the solute in the lower chamber (i.e. the detection cell). The solute becomes energetically more stable in the bound state and a small amount of excess energy is released in the form of heat. This heat provides the signal for the positive peak in the chromatogram. After a finite residence time in the detector cell, the solute is eluted off the adsorbent and enters the flow stream again. Now the solute takes up energy (heat) from the surrounding environment for the desorption process and a negative signal is fed to the recorder.[40]

C - M.A.D. Studies of Known Compounds

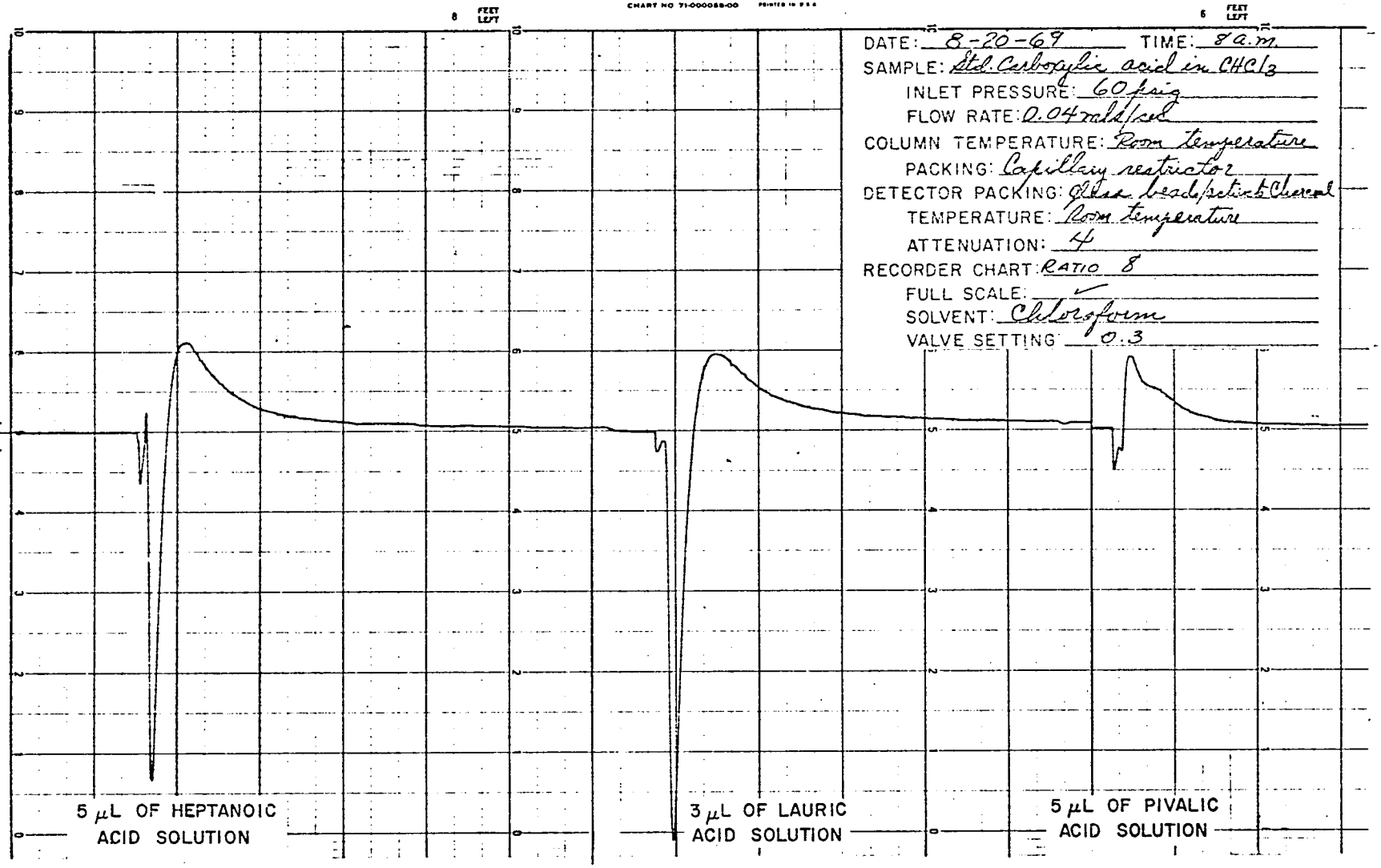
It was necessary to acquire some practical information about the performance of the M.A.D. unit before it was applied to the study of unknown materials. The response of the M.A.D. unit under various sets of conditions was studied. The relative heats of adsorption of standard acids and phenols were measured.

M.A.D. Chromatogram No. 1 illustrates the differences in total peak height for three carboxylic acids (lauric, heptanoic and pivalic acids) injected separately into M.A.D. There are large differences in the adsorption of these three acids on activated charcoal as shown in Table XI. The least strongly adsorbed of the three acids is pivalic acid giving a total peak height of only 14.0 chart units for 5 $\mu\ell$ of a 0.262 M/ℓ solution.

Peak shape and reproducibility of the Micro-Adsorption Detector response is illustrated by M.A.D. Chromatogram No. 2. This chromatogram is the result of three repetitive injections of 3 $\mu\ell$ of a 0.1 M solution of benzoic acid in chloroform.

Various carrier liquids were tested and information on their performance was acquired. Chloroform attacked the septa and gaskets requiring their frequent replacement. Solubility of helium (pressurizing medium) in diethyl ether was very high under pressure, causing bubbles to form. Benzene was found to be a satisfactory carrier liquid.

It was learned through experimentation that the presence of water in the solutions would give distorted values for peak heights in M.A.D. chromatograms. The peak height values were higher if moisture were present in the solution. It became necessary to start with water-free solutions and to maintain them water-free over a period of time.



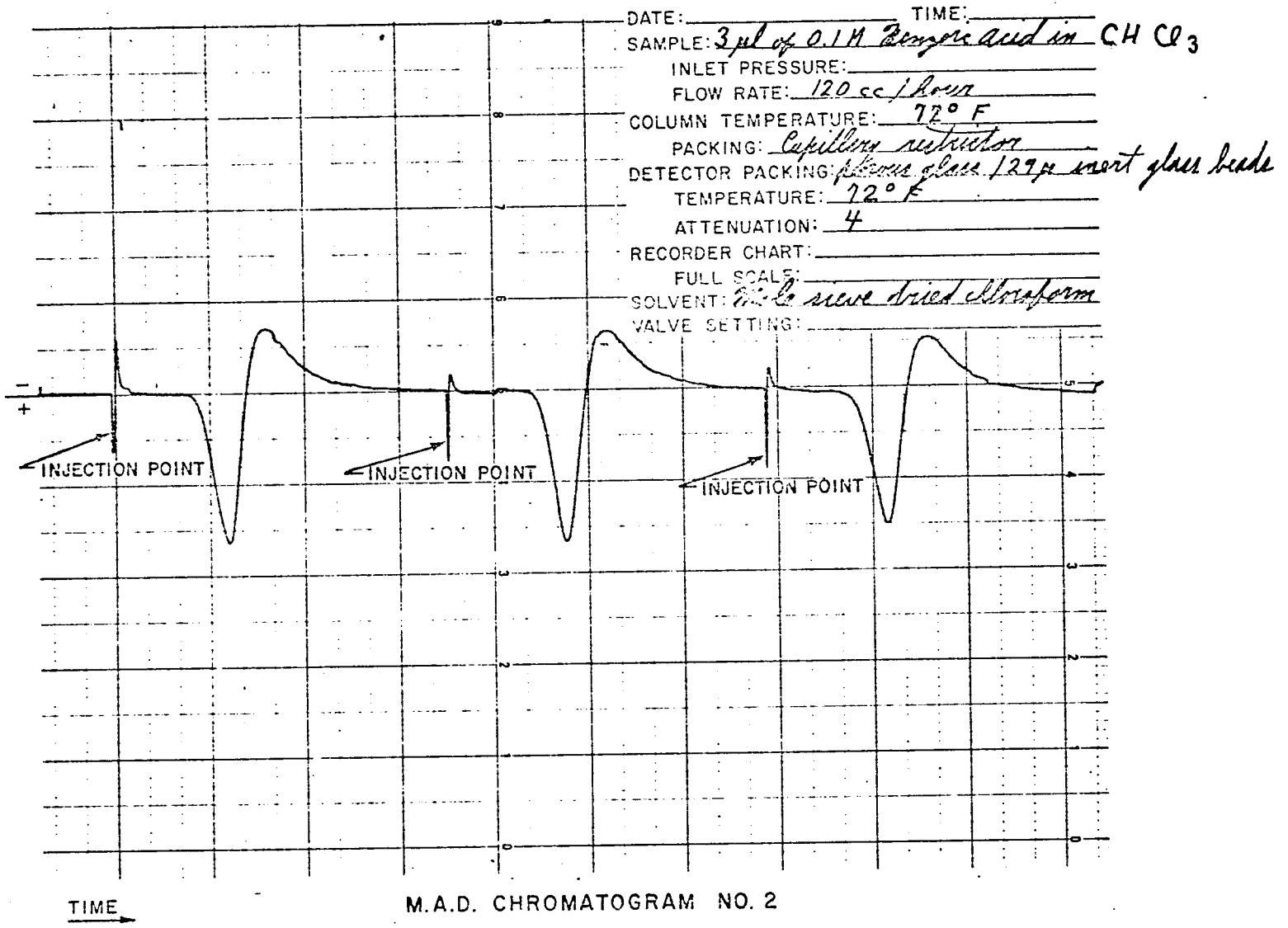
M. A. D. CHROMATOGRAM NO. I

TABLE XI

INVESTIGATION OF DIFFERENT SAMPLE RESPONSES

WITH VARIOUS CARBOXYLIC ACIDS

Material	Concentration in M/L in $CHCl_3$	Sample Size $\mu\ell$	Total Peak Height Chart Units
Lauric Acid	0.100	3	60.8
Heptanoic Acid	0.101	5	54.6
Pivalic Acid	0.262	5	14.0



M.A.D. CHROMATOGRAM NO. 2

A search for the best method for maintaining the solutions in a water-free state was undertaken. A series of solutions were stored under different conditions: in the presence or absence of molecular sieves, with or without rubber "serum stoppers", etc. Specific details of the storing conditions are given in Table XII. M.A.D. chromatograms were obtained for each solution type.

The M.A.D. peak height was less for those solutions to which molecular sieves* had been added, as shown in M.A.D. Chromatogram No. 3. This clearly indicates that the molecular sieves are adsorbing either acid or water from the solutions. To determine if any of the acid is being adsorbed by the molecular sieves, IR spectra of the different solutions were obtained (IR Spectra No. 5 through No. 12). The band at 1705 cm^{-1} due to carbonyl adsorption was selected as an indication of the amount of acid present. The IR spectra show that the sieves adsorb some of the acid, thereby reducing the total concentration of acid in the solutions (Table XII). The use of rubber stoppers does not seem to introduce any detrimental changes in the solutions as far as can be detected by IR spectrometry. However, it was decided to discontinue the use of rubber stoppers because with time the solvent vapors do attack the rubber and could eventually contaminate the solutions. The amount of acid adsorbed by the sieves was dependent on the total amount of sieves present (Table XII). It was concluded that molecular

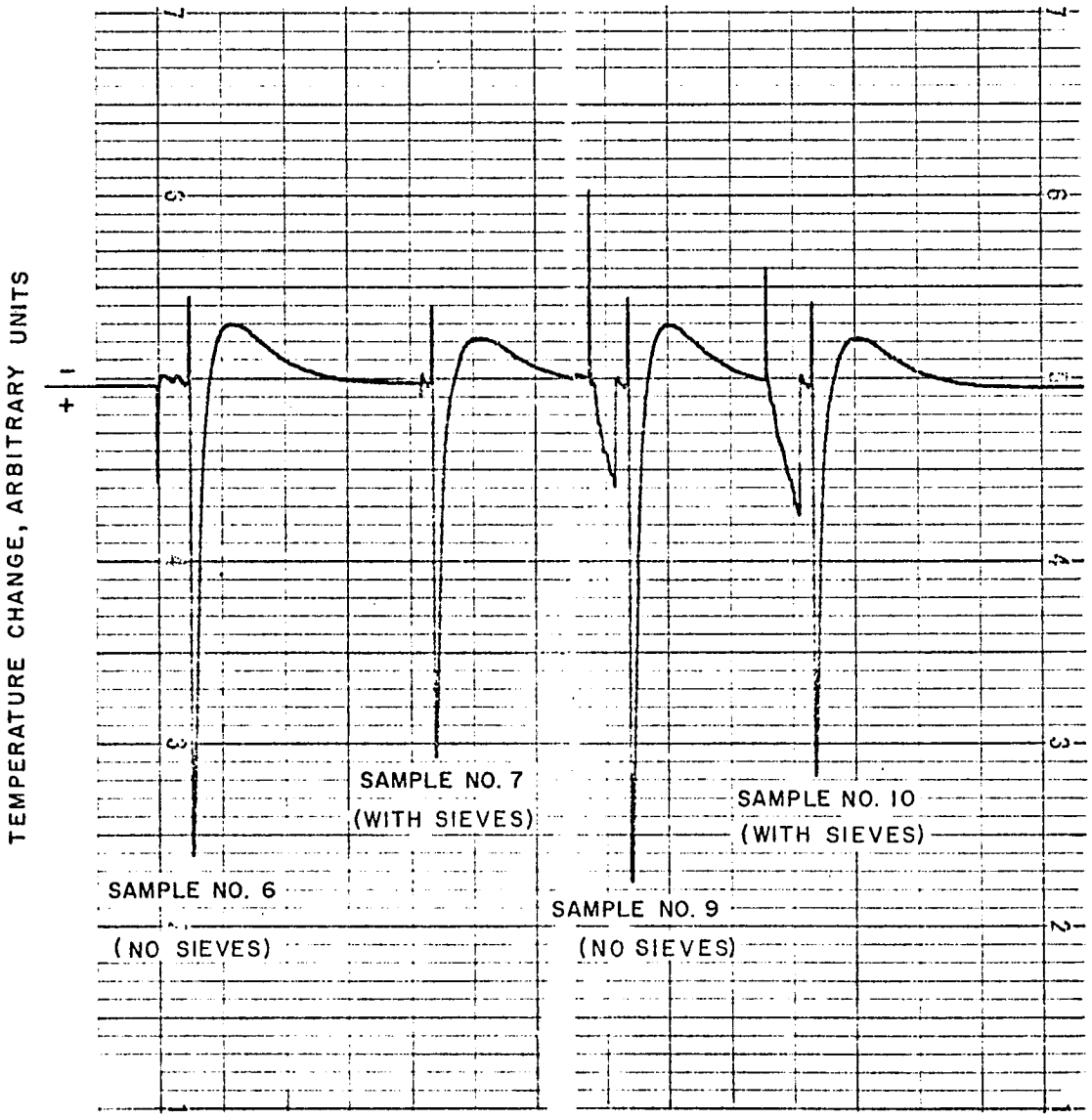
*Linde Type 3A molecular sieves are alkali-metal aluminosilicates with critical diameters up to 3 Å.

TABLE XII

STUDIES PERFORMED TO DETERMINE BEST DRYING METHOD FOR SOLUTIONS TO BE ANALYZED IN THE M.A.D. UNIT

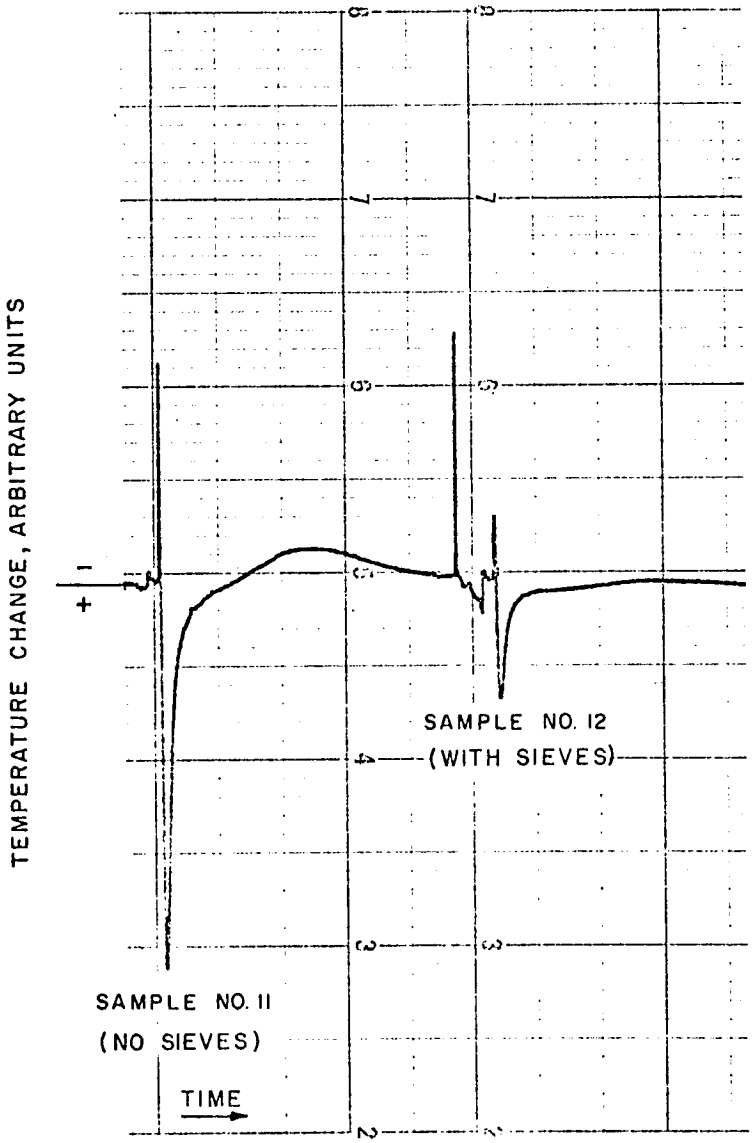
Sample Description	Sample No.	Total Peak Height (M.A.D.)	IR Spectrum No.	$[-\log_{10}T] \alpha C$, At 1705 cm^{-1} (IR)
Mole sieve dried benzene	5	-	IR-5	-
Cyclohexane carboxylic acid solution (0.11 M) in all-glass container	6	29.2	IR-6	0.62
Mole sieve dried cyclohexane carboxylic acid solution (0.11 M) in all-glass container	7	23.0	IR-7	0.51
Same solution as Sample No. 7 but with extra amount of molecular sieves added	8	-	IR-8	0.32
Cyclohexane carboxylic acid solution (0.11 M) in rubber stoppered vials	9	30.6	IR-9	0.69
Mole sieve dried cyclohexane carboxylic acid solution (0.11 M) in rubber stoppered vials	10	24.0	IR-10	0.60
Acetic acid solution (0.11 M) in all-glass containers	11	21.3	IR-11	0.61
Mole sieve dried acetic acid solution (0.11 M) in all-glass containers	12	6.8	IR-12	0.14

DATE: _____ TIME: _____
SAMPLE: cyclohexanecarboxylic Acid Solution
INLET PRESSURE: _____
FLOW RATE: 2.3 mls/minute
COLUMN TEMPERATURE: _____
PACKING: Inert glass beads
DETECTOR PACKING: Inert glass beads/paraffin bead
TEMPERATURE: _____
ATTENUATION: 32
RECORDER CHART: _____
FULL SCALE: _____
SOLVENT: Mole sieve dried benzene
VALVE SETTING: _____

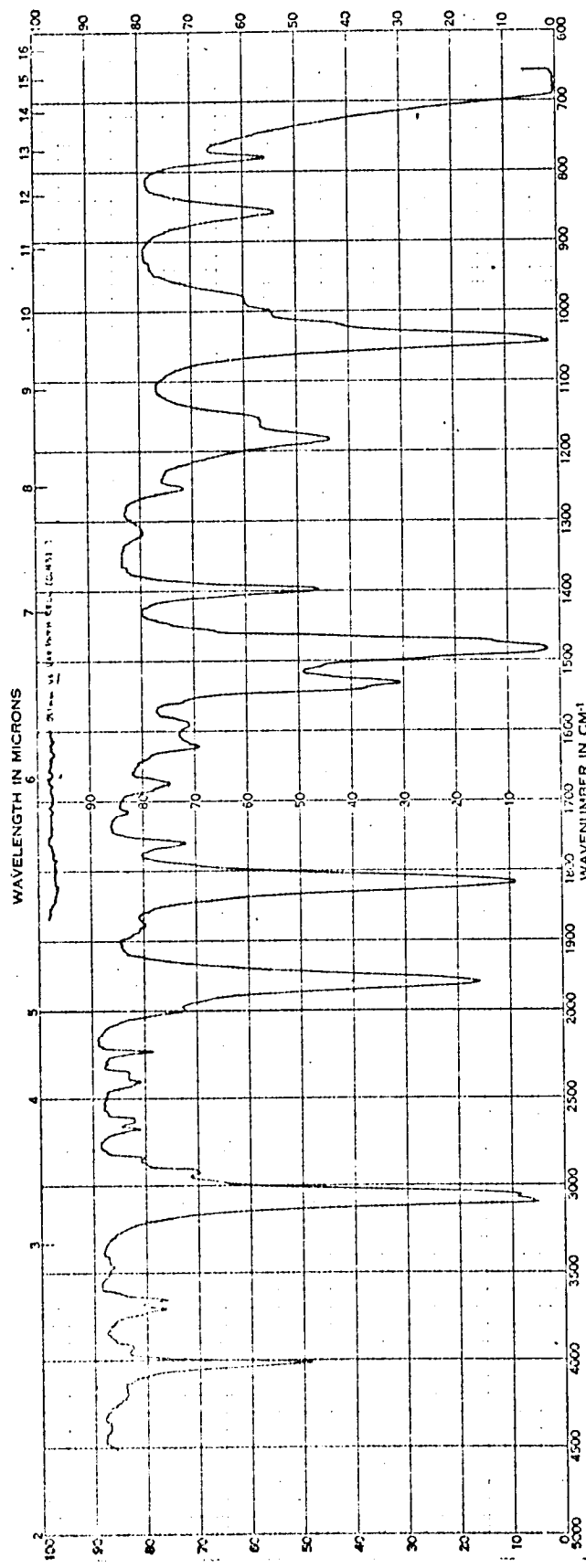


M. A. D. CHROMATOGRAM NO. 3

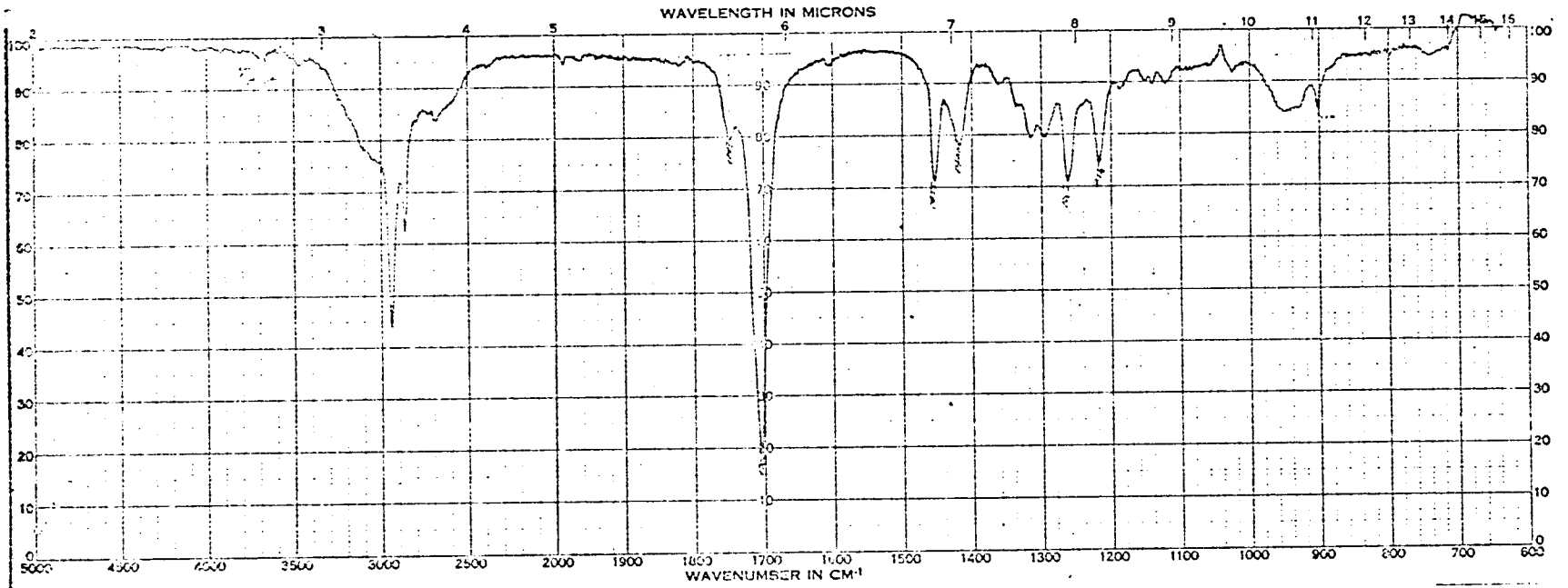
DATE: 11/13/69 TIME:
SAMPLE: 2 µl of 0.11M acetic acid solution
INLET PRESSURE: 80 PSIG
FLOW RATE: 2.3 ml/min.
COLUMN TEMPERATURE: 76°F
PACKING: Quartz glass beads
DETECTOR PACKING: Quartz glass beads/pyrex glass beads
TEMPERATURE:
ATTENUATION: 32
RECORDER CHART:
FULL SCALE:
SOLVENT: Mole sieve dried benzene
VALVE SETTING: 0.2



M.A.D. CHROMATOGRAM NO. 3(CONTINUED)



IR SPECTRUM NO. 5



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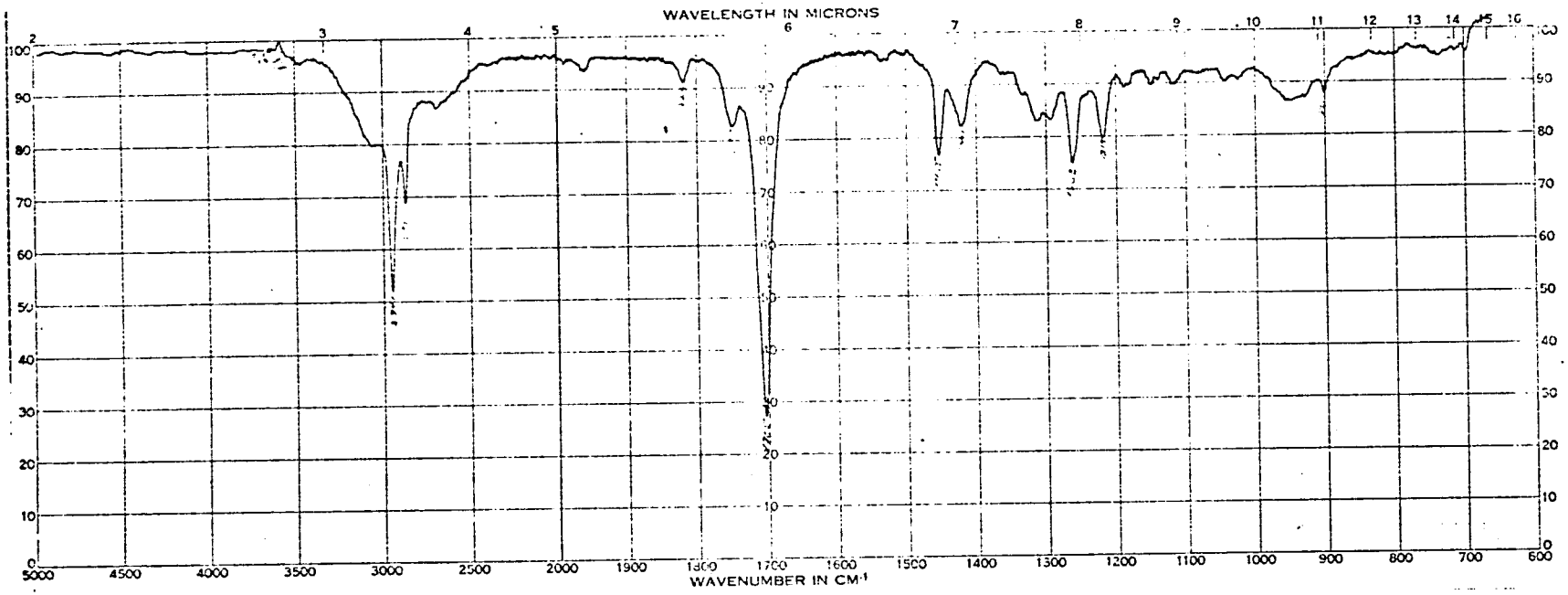

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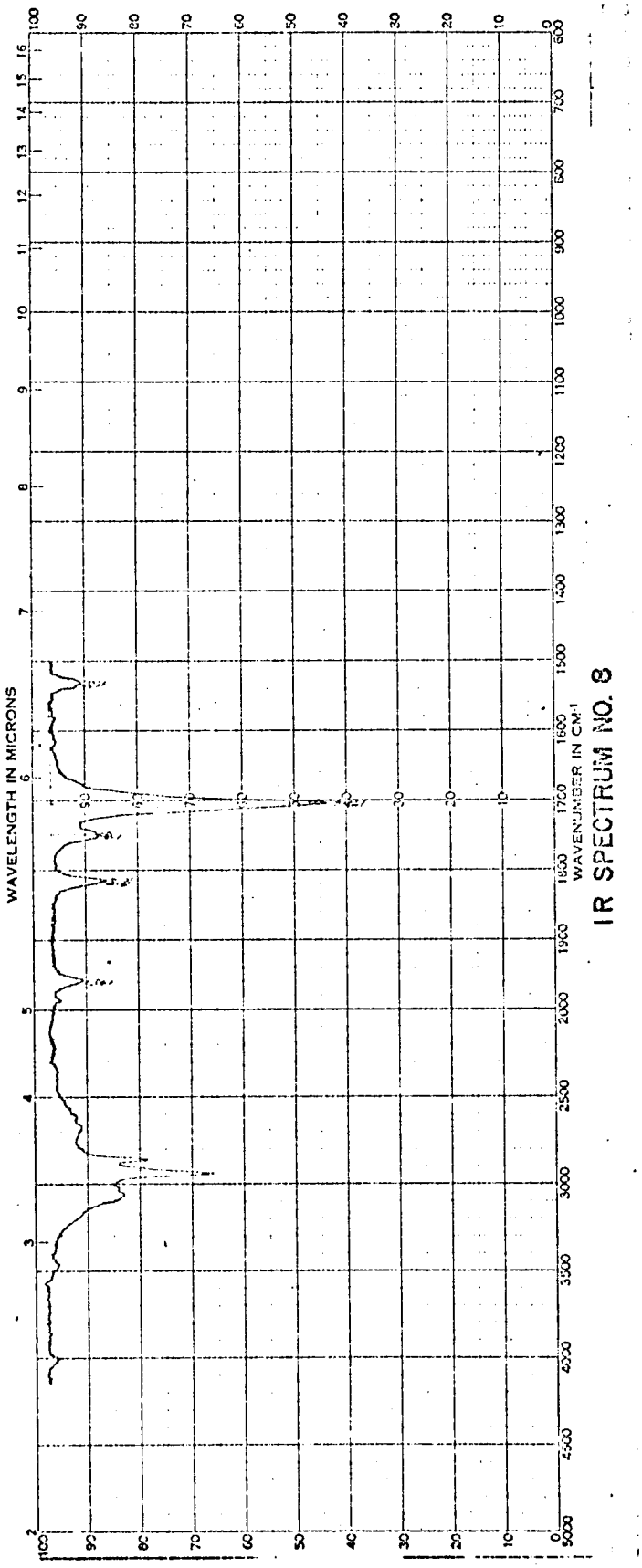
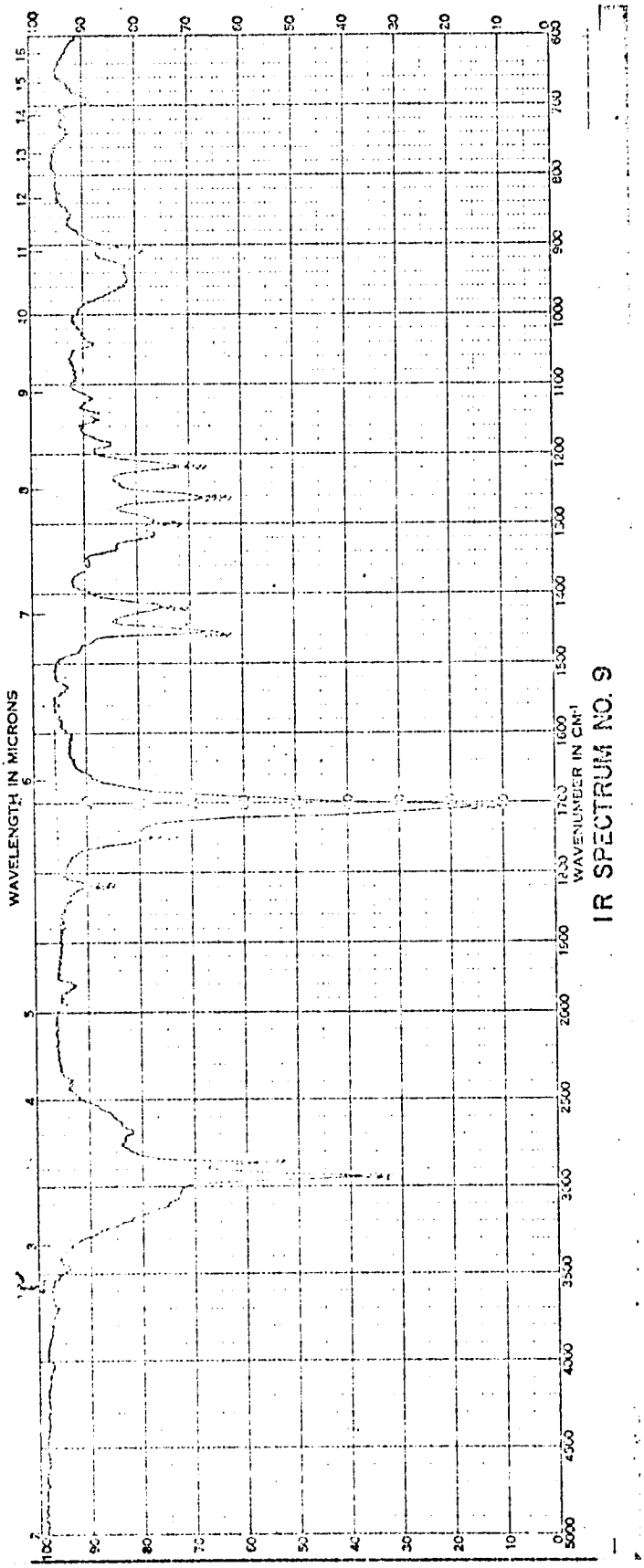
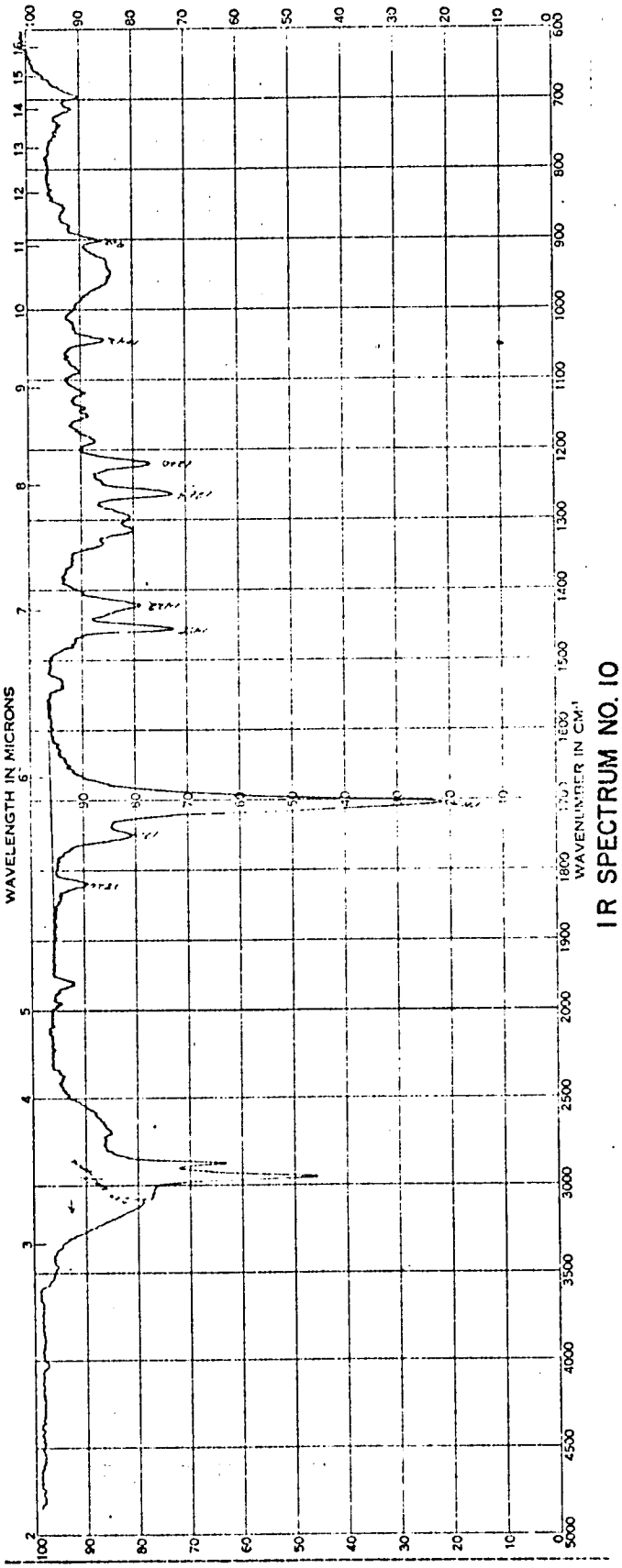


IR SPECTRUM NO. 8



IR SPECTRUM NO. 9



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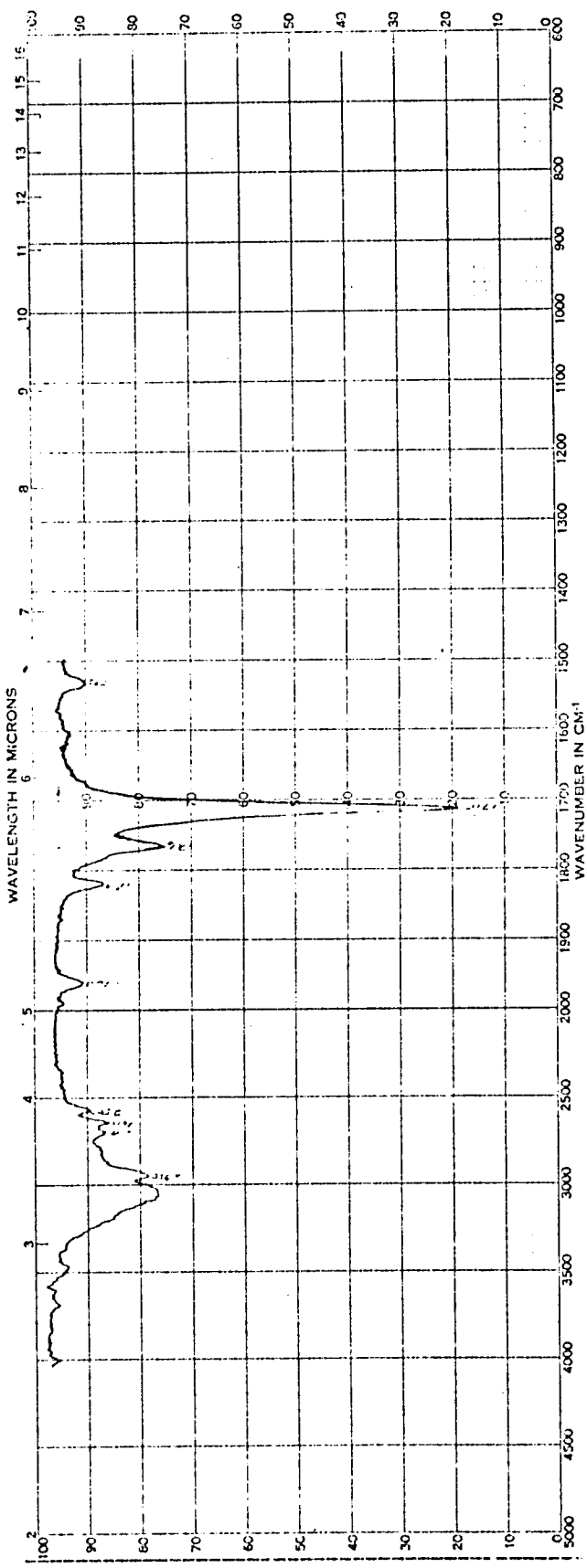
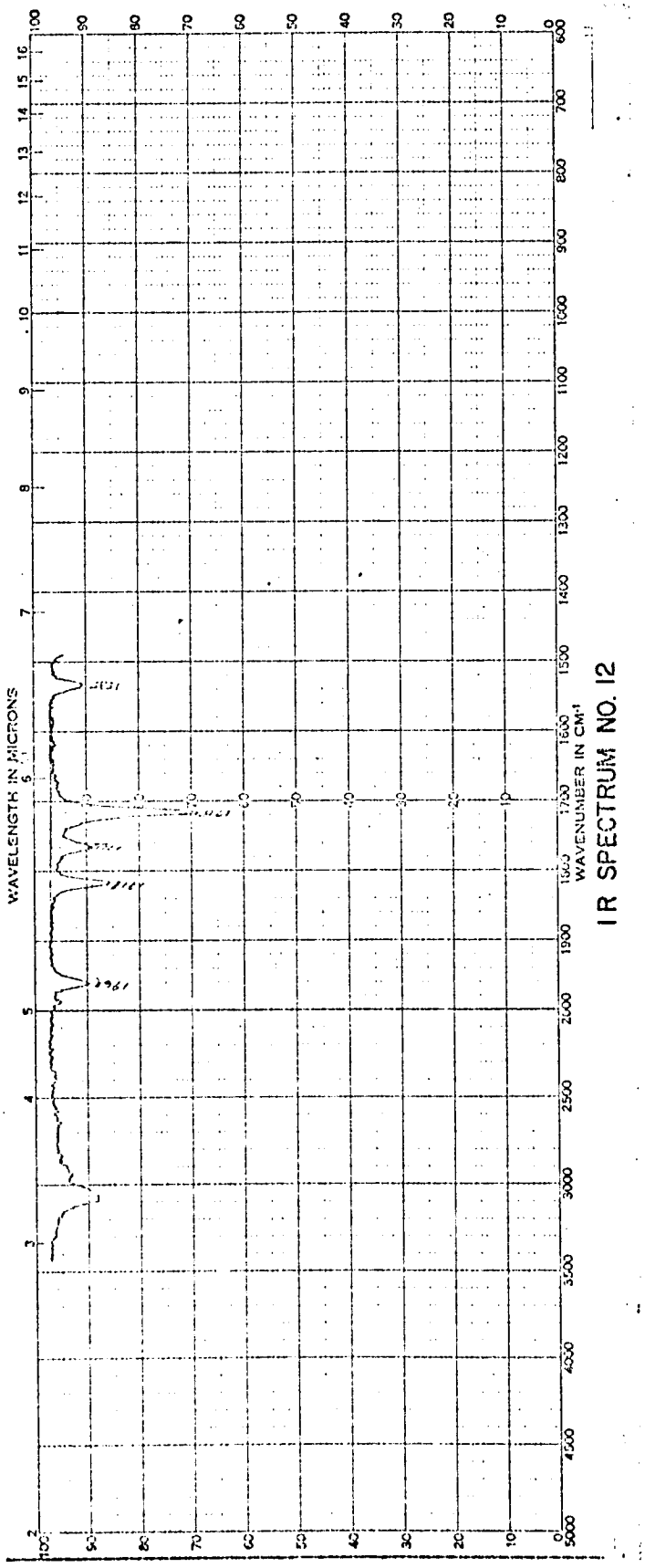


IR SPECTRUM NO. II



IR SPECTRUM NO. 12

sieves can be used to dry the pure solvent but should not be used to keep the acid solutions dry.

Acetic acid is adsorbed by the molecular sieves to a larger extent than cyclohexane carboxylic acid, as evidenced by the greater change in the intensity of the band at 1705 cm^{-1} in the acetic acid IR spectra (Table XII). Therefore, an important observation derived from this group of experiments is that different acids are adsorbed to various degrees by the same amount of molecular sieves.

Continued searching for a drying method resulted in a successful technique. It was found that traces of water could be removed by azeotroping with benzene and that, with special precautions, glass stoppers would effectively prevent water from getting into the solutions.

A series of commercially available carboxylic acids and phenols* were examined in the Micro-Adsorption Detector to provide background data prior to the study of polar components of heavy oils. The adsorption cell was packed with porous glass beads and the reference cell with smooth glass beads. The porous glass is a strong adsorbent and the smooth glass is very inert; this combination maximizes the difference in adsorptive ability, increasing the sensitivity of the detector.

Peak heights of the acid and phenol standards are indicative of the heats of adsorption of these materials on

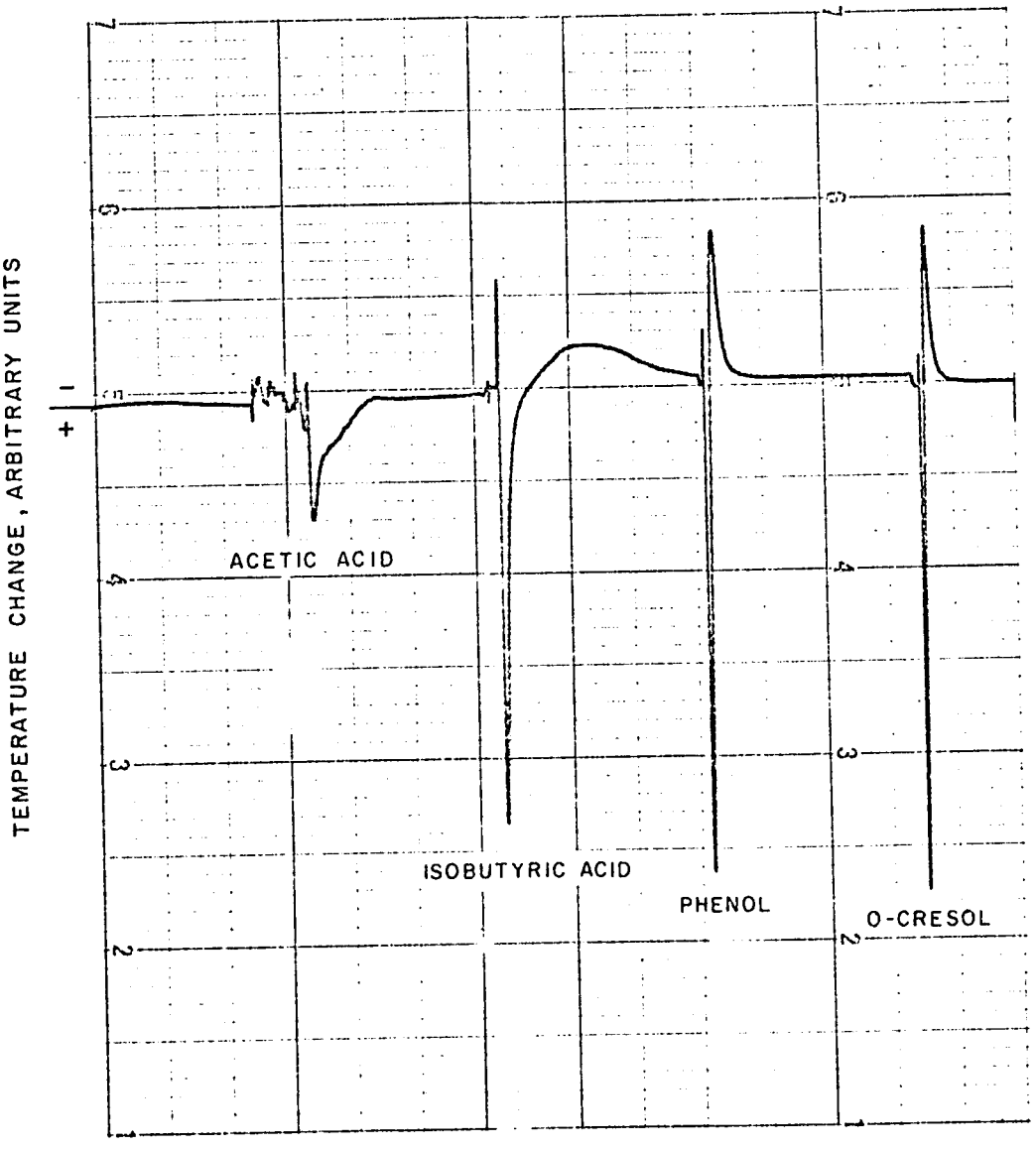
*Phenols are also found among the polar compounds in petroleum.

the porous glass. Peak heights were measured for five acids and five phenols from M.A.D. Chromatogram No. 4. The carboxylic acids exhibited total peak height values ranging from 7 for acetic acid to 28.4 for lauric acid (Table XIII). The phenols showed peak height values from 26.3 for phenol to 31.7 for 2,4-dimethylphenol. The values for all the phenols were fairly close together, while those for the carboxylic acids covered a wide range of values. A consistent linear relationship was not observed between peak height and molecular weight.

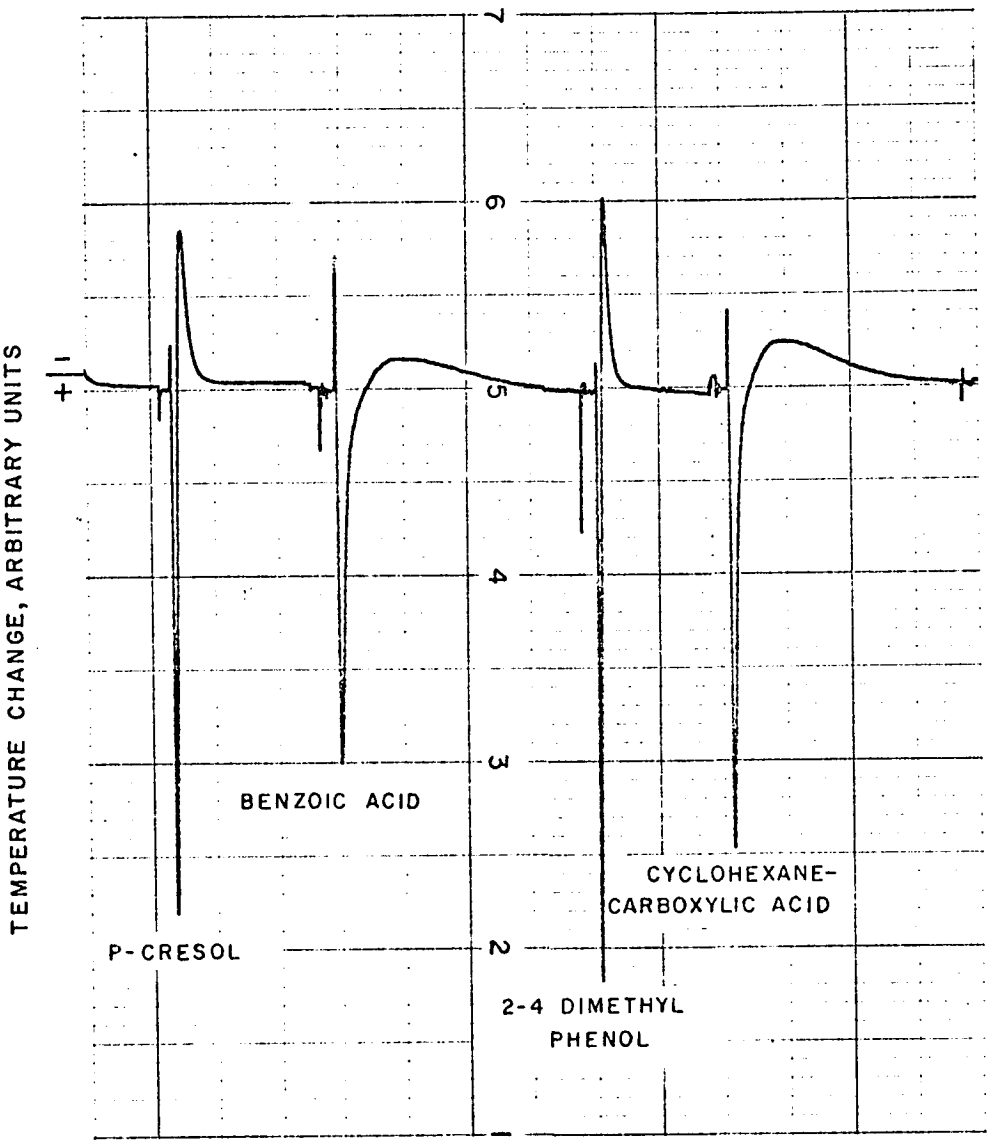
Melting points of the standard acid and phenols were also obtained, as a measure of their purity (Table XIII). Three of the compounds, phenol, o-cresol and p-cresol, showed melting points differing sufficiently from the literature values to require further purification.

Experimentation with known compounds thus revealed a number of important facts about M.A.D. The peak heights obtained for various acids and phenols in M.A.D. chromatograms depend on the particular chemical involved and on its concentration. Benzene is one of the most satisfactory carrier liquids for usage with M.A.D. The best method for keeping the solutions water-free consists of the removal of the water by azeotroping with benzene and storing the solution in glass containers with glass stoppers.

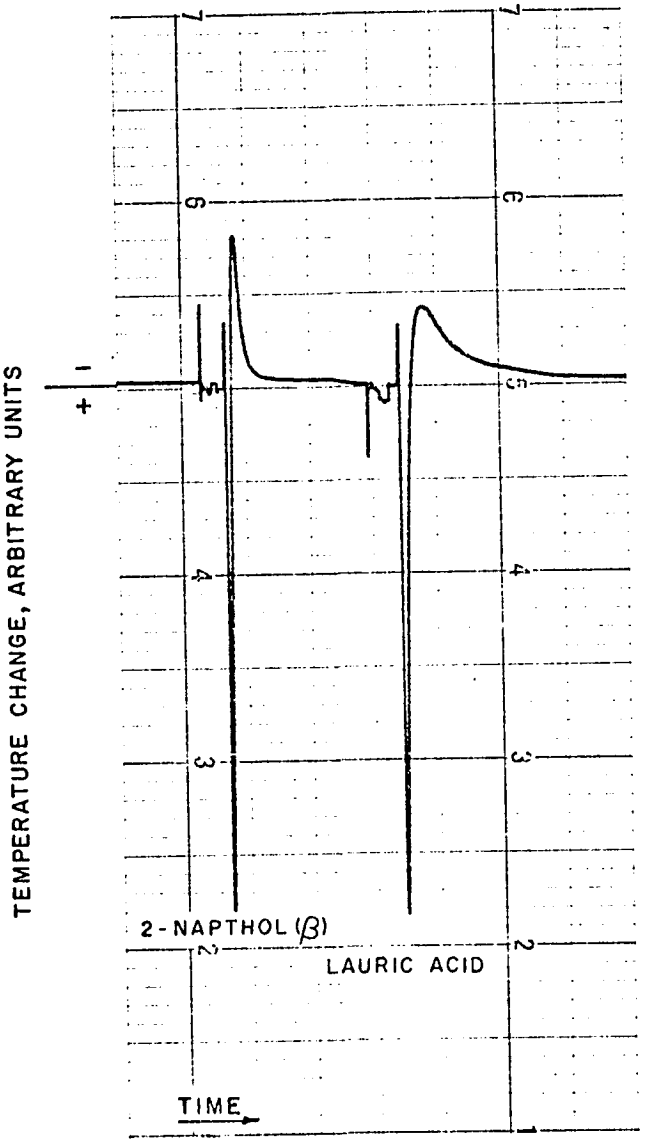
DATE: 12/12/69 TIME: 3:10 PM
SAMPLE: Std. carboxylic acids and phenols (2 µl each)
INLET PRESSURE: 80 PSIG
FLOW RATE: 2.2 ml/min.
COLUMN TEMPERATURE:
PACKING: Capillary restrictor
DETECTOR PACKING: bent glass beads / porous glass beads
TEMPERATURE:
ATTENUATION: 32
RECORDER CHART:
FULL SCALE:
SOLVENT: Mole sieve dried benzene
VALVE SETTING: 0.2



M.A.D. CHROMATOGRAM NO. 4



M.A.D. CHROMATOGRAM NO. 4 (CONTINUED)



M.A.D. CHROMATOGRAM NO. 4 (CONTINUED)

TABLE XIII

STANDARD CARBOXYLIC ACIDS AND PHENOLS

Sample	Molecular Weight*	Total Peak Height (M.A.D.) Chart Units**	Melting Points, °C Literature Value	Melting Points, °C Experimental Value
Acetic Acid	60.05	7.0	-	-
Isobutyric Acid	88.11	23.4	-	-
Phenol	94.11	26.3	40.9	40.0- 42.2
o-Cresol	108.13	27.2	30.9	28.0- 32.0
p-Cresol	108.14	28.0	34.8	33.0- 35.2
Benzoic Acid	122.12	20.1	122.4	121.5-122.5
2,4-Dimethylphenol	122.17	31.7	-	-
Cyclohexane Carboxylic Acid	128.17	24.8	29.5-30.5	29.8- 31.0
2-Naphthol (β)	144.17	28.0	122.0-123.0	120.5-121.5
Lauric Acid	200.32	28.6	44.1	44.8- 46.0

*Literature values.

**The measurements were made on 0.1 M benzene solutions of the acids and phenols.

D - Isolation of Polar Fractions from Heavy Oils by Two Different Techniques

A heavy oil sample (Heavy Oil #8) was separated to obtain several polar fractions by two different liquid chromatographic techniques. Relative heats of adsorption were determined on the polar fractions by the Micro-Adsorption Detector.

The separation schemes used for isolating the fractions were: (A) silica gel chromatography followed by ion exchange chromatography, and (B) sodium hydroxide-alcohol-water extraction followed by ion exchange chromatography.

Separation Scheme A: A dry ether fraction obtained by silica gel chromatography was subjected to ion exchange chromatography using both cation and anion exchange resins. Separation Scheme A is outlined in Figure 3. Silica gel chromatography was performed on Heavy Oil #8 (Table XIV). Petroleum acids and other polar compounds were eluted with ether from the silica gel column. The ether fraction, Fraction 8-1-2, was to be analyzed by M.A.D.

An ether fraction previously obtained, Fraction 8-1a (Part I, Section C), was subjected to cation exchange chromatography. The solvent mixtures employed for eluting Fractions 8-9 through 8-13 are shown in Figure 3 and in Table XV. The last three fractions eluted, Fractions 8-11 through 8-13, constitute the total basic fraction which was calculated to be 2.2% by weight of crude oil.

SEPARATION SCHEME A

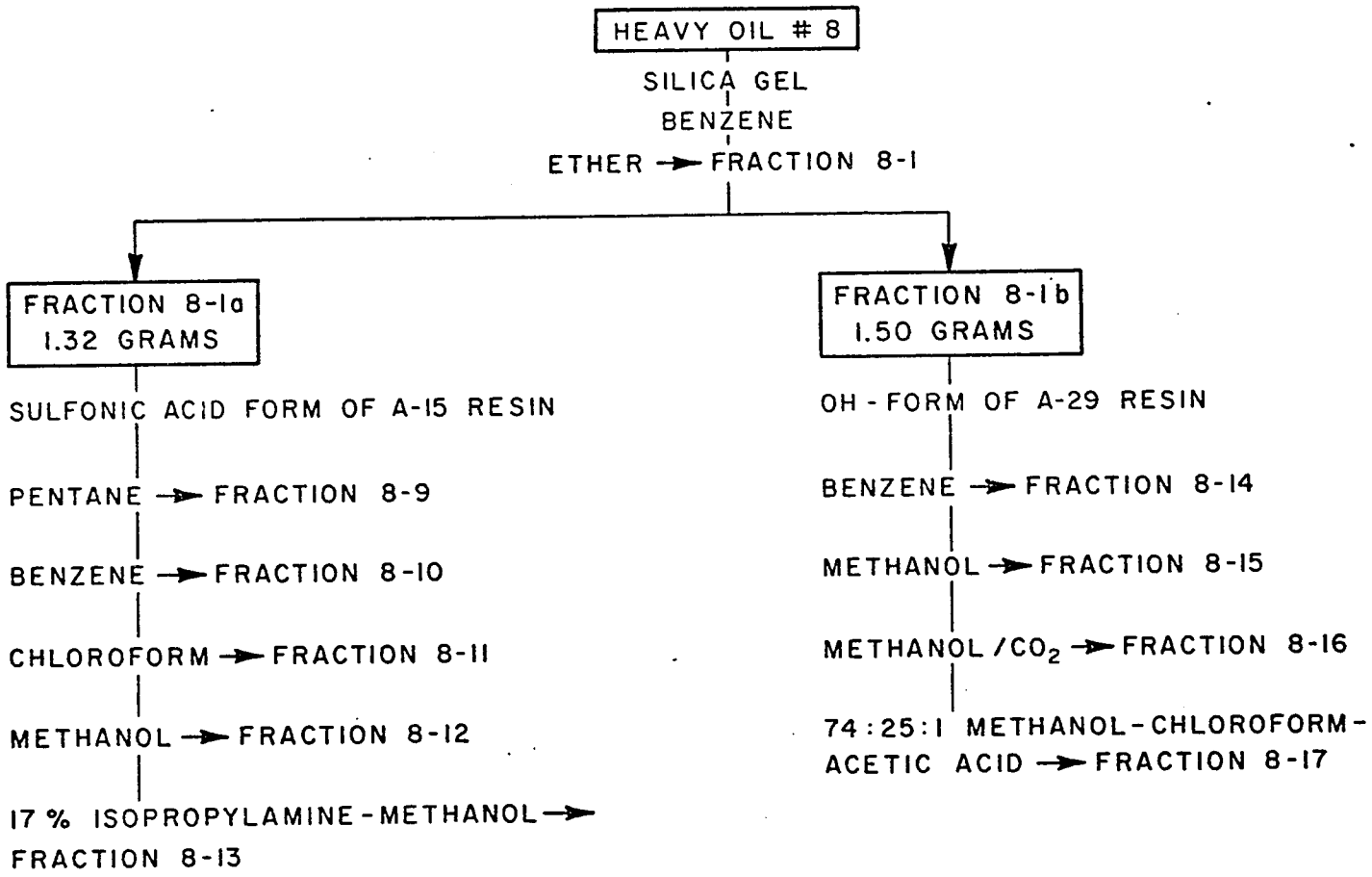


FIGURE 3

TABLE XIV

SILICA GEL CHROMATOGRAPHIC SEPARATION

OF HEAVY OIL #8

(RUN #2)

Eluent	Total Eluent Volume ℓ	Recovered Fractions, Wt. % Of Crude Oil
Benzene	40.0	84.5
Ether*	3.0	6.6
50/50 Methanol/Ether	1.5	3.2
Methanol	1.0	0.3
175/48 Chloroform/Methanol	6.6	2.1
5/5/20/70 Water/Acetone/ Acetic Acid/Isopropanol	1.0	1.7

*Fraction #8-1-2

TABLE XV

ANALYTICAL DATA ON ION EXCHANGE CHROMATOGRAPHY

ON FRACTIONS 8-1a AND 8-1b

Type of Resin	Eluent	Total Eluent Volume ℓ	Fraction #	Recovered Material Wt. % Of Material Injected To Resin	Wt. Of Fractions g
	Pentane	1.5	8-9	10.3	0.136
	Benzene	3.0	8-10	28.6	0.376
Amberlyst A-15:	Chloroform	3.0	8-11	17.4	0.229
	Methanol	1.0	8-12	2.5	0.0365
	17% Isopropylamine-Methanol	3.0	8-13	15.8	0.209
	Benzene	1.4	8-14	53.9	0.81
	Methanol	1.8	8-15	10.2	0.15
Amberlyst A-29:	Methanol/CO_2	2.0	8-16	18.0	0.27
	74/25/1 Methanol/ Chloroform/ Acetic Acid	1.9	8-17	10.9	0.16

Fraction 8-1b (Part I, Section C) was subjected to anion exchange chromatography. The solvent mixtures employed for eluting Fractions 8-14 through 8-17 are shown in Figure 3 and in Table XV. The last three fractions eluted give a total acid fraction (2.4% by weight of crude oil).

Separation Scheme B: Fractions A, B, C, and D were isolated from Heavy Oil #8 by extraction and ion chromatography procedures. A schematic diagram of the separation scheme is shown in Figure 4. The extraction and ion chromatography procedures are based on Seifert's articles.[24,42]

The fractions isolated by this separation scheme contain the following compounds:

Fraction A: phenols
Fraction B: phenols plus carboxylic acids
Fraction C: carboxylic acid plus derivatives
Fraction D: mostly carboxylic acids.[24]

Fraction D exhibits the highest acid number of all four fractions as shown in Table XVI. Fraction B exhibits an abnormally high value for its molecular weight (Table XVI). The reason for the high value is not evident; the presence of asphaltenes (high molecular weight materials) is a possibility.

Separation Scheme B was applied several times to three different oils and the results show good reproducibility (Table XVII). Heavy Oil #6 exhibits the highest percentage of Fraction D, i.e. the highest percentage of carboxylic acids.

SEPARATION SCHEME B

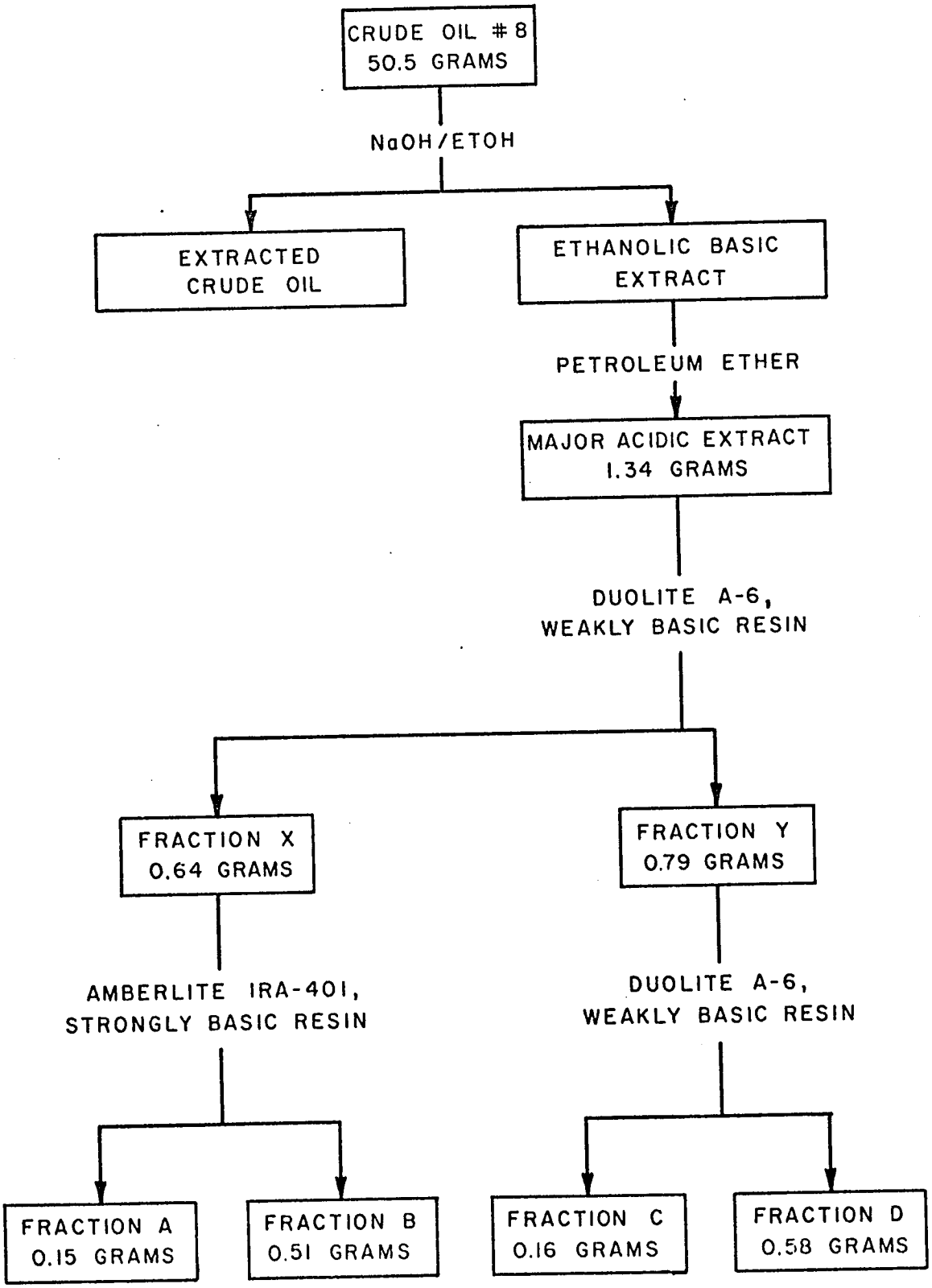


FIGURE 4

TABLE XVI

CAUSTIC EXTRACTION AND ION EXCHANGE CHROMATOGRAPHY

OF HEAVY OIL #8

(RUN #1)

Fraction #	Acid Number mg KOH/g*	Equivalent Weight**	Molecular Weight***	Number Of COOH Groups****
8-A-1	52.2	1076	467	0.4
8-B-1	109.0	513	827	1.6
8-C-1	79.7	704	585	0.8
8-D-1	136.0	412	568	1.4

*Acid numbers were determined by potentiometric titration in xylene:water:isopropanol solvent.

**Equivalent weight = $\frac{56.1}{\text{Acid Number} \times 10^{-3}}$.

***Number average molecular weight determined by vapor phase osmometry of the acid in $CHCl_3$.

****Number of COOH groups = $\frac{\text{Average Molecular Weight}}{\text{Equivalent Weight}}$.

TABLE XVII

COMPOSITION DATA OF HEAVY OILS AS DETERMINED FROM SEPARATION SCHEME B

	Weight Of Original Oil g	Major Acidic Extract Fraction % By Wt. Of Original Oil	Fraction X (% By Weight)	Fraction A (% By Weight)	Fraction B (% By Weight)	Fraction Y (% By Weight)	Pooled Y Fraction g	Fraction C (% By Weight)	Fraction D (% By Weight)	Fraction No.
HEAVY OIL #8:										
8-1	50.5	2.64	1.26	0.30	1.01	1.56	-	0.32	1.15	-
8-2	52.5	3.33	1.63	0.37	1.54	1.69	-	0.32	1.39	-
HEAVY OIL #2:										
2-1	55.02	4.18	2.08	-	-	1.48	-	0.47	1.41	-
2-2	54.02	3.95	1.80	-	-	1.40	-	0.43	1.49	-
HEAVY OIL #6:										
6-1	61.56	3.93	1.97	0.53	1.42	1.87	-	0.43	1.41	
6-2	62.20	4.27	1.85	0.60	1.77	1.73	-	0.30	1.34	-
6-3	47.09	3.88	1.74	-	-	1.89	-	0.15	1.70	
6-4	52.09	3.86	1.80	-	-	1.84	-	0.20	1.60	- $6\text{-}D_1$
6-7	52.89	3.98	2.11	-	-	1.73	-	0.12	1.58	
6-8	49.75	3.96	1.99	-	-	1.80	-	0.15	1.63	
6-5	48.92	4.35	2.30	-	-	1.89	-	0.26	1.64	
6-6	48.48	3.70	1.86	-	-	1.73	-	0.20	1.54	
6-9	60.23	4.17	2.52	-	-	1.57				
6-10	56.13	4.33	2.37	-	-	1.87	4.1383	0.06	1.82	$6\text{-}D_2$
6-11	53.46	4.22	2.18	-	-	2.02				
6-12	49.62	4.29	2.10	-	-	2.14				
6-13	55.36	4.18	2.16	-	-	1.93				
6-14	54.44	4.25	2.23	-	-	1.93	4.0204	0.07	1.75	$6\text{-}D_3$
6-15	56.23	4.19	2.44	-	-	1.69				
6-16	54.20	4.27	2.47	-	-	1.76				
6-17	52.86	4.32	2.26	-	-	1.92				
6-18	56.99	4.30	2.21	-	-	1.93	4.2858	0.06	1.86	$6\text{-}D_4$
6-19	49.48	4.50	2.23	-	-	2.11				
6-20	54.36	4.23	2.02	-	-	2.08				

This is in line with the neutralization numbers obtained for the total crude samples (Table II).

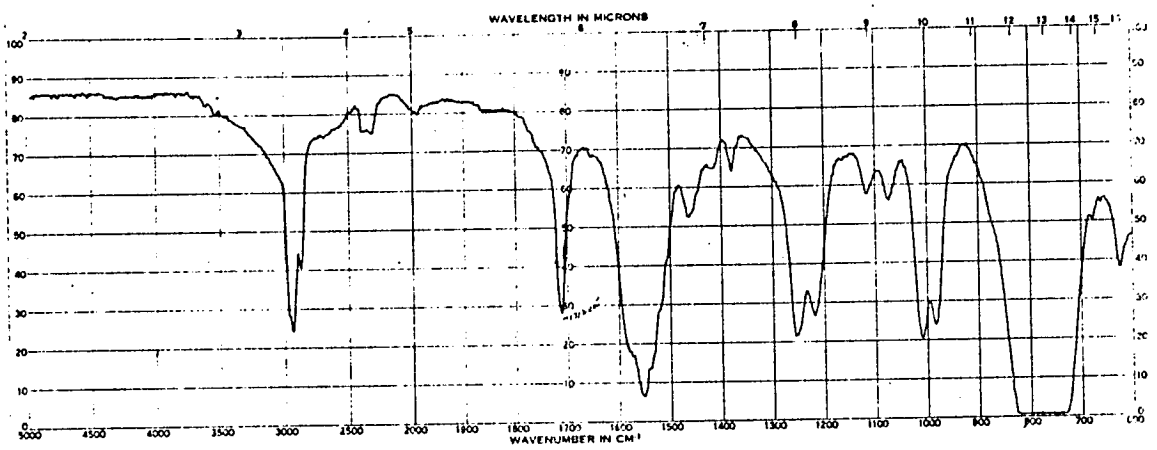
To confirm the isolation of carboxylic acids by Separation Scheme B, IR spectra of two of the fractions isolated from Heavy Oil #6 were obtained. The fractions selected, 6-Major Acidic Extract-3* and Fraction 6-D-2, exhibited carbonyl absorption bands at 1712-1713 cm^{-1}. At this particular wavelength, the percent transmittance was 27.2 for the Major Acidic Extract (IR Spectrum No. 13) and 16.2 for Fraction 6-D-2 (IR Spectrum No. 14). These spectra indicate that carboxylic acids are present in both fractions and that their concentration is much higher in Fraction D than in the Major Acidic Extract.

Separation Scheme A yielded a total of 10 polar fractions and Separation Scheme B, a total of 4 polar fractions. All fourteen polar fractions isolated from Heavy Oil #8 were analyzed with the Micro-Adsorption Detector.

E - Relative Heat of Adsorption Studies of Polar Fractions from Heavy Oils

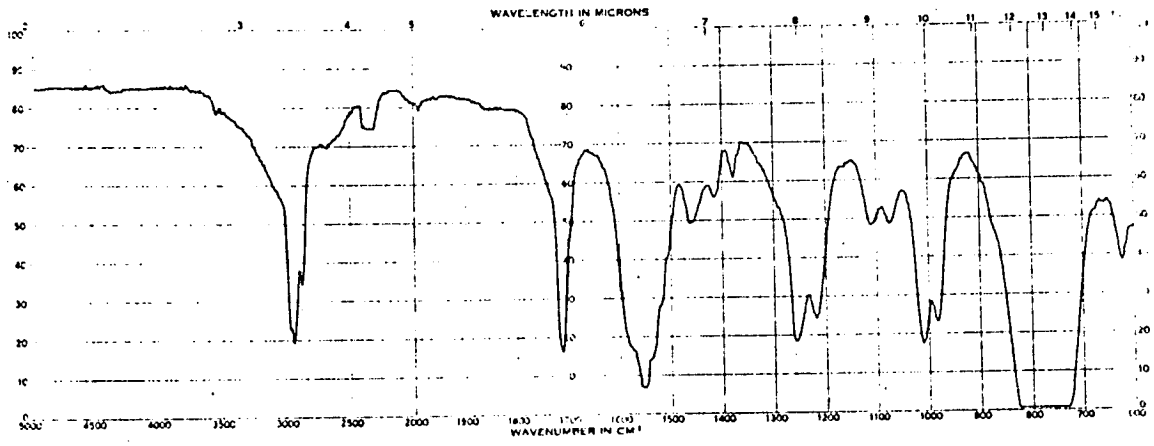
Relative heats of adsorption were determined on the polar fractions from Heavy Oil #8 by the Micro-Adsorption Detector. Number average molecular weights were also determined on these fractions by vapor phase osmometry. On the basis of the data thus accumulated, one of the fractions was selected for further studies.

*This notation implies that this is a Major Acidic Fraction from Heavy Oil #6 obtained during the third duplicate run.



IR SPECTRUM NO. 13

MAJOR ACIDIC EXTRACT OF OIL #6



IR SPECTRUM NO. 14

FRACTION 6-D-2

Polar fractions isolated from Heavy Oil #8 (Tables VII and XIV; Figures 3 and 4) were injected separately into the Micro-Adsorption Detector and peak height measurements taken from M.A.D. Chromatogram No. 5 were recorded in Table XVIII. Number average molecular weights* of the polar fractions were determined in chloroform by vapor phase osmometry and concentrations in M/ℓ were then calculated (Table XVIII). The peak heights calculated for a particular attenuation (A = 1) and concentration (C = 0.01 M/ℓ) are a measure of relative heat of adsorption values.

Those polar fractions exhibiting the higher molecular weight values consist of a very wide range of compounds and are probably contaminated with asphaltenes. Asphaltenes consist of graphite-like clusters of condensed aromatic rings joined together by alkyl groups.[43] Molecular weights of asphaltenes vary from 900 to several thousand.[44]

---

*Number average molecular weight is defined by the equation:

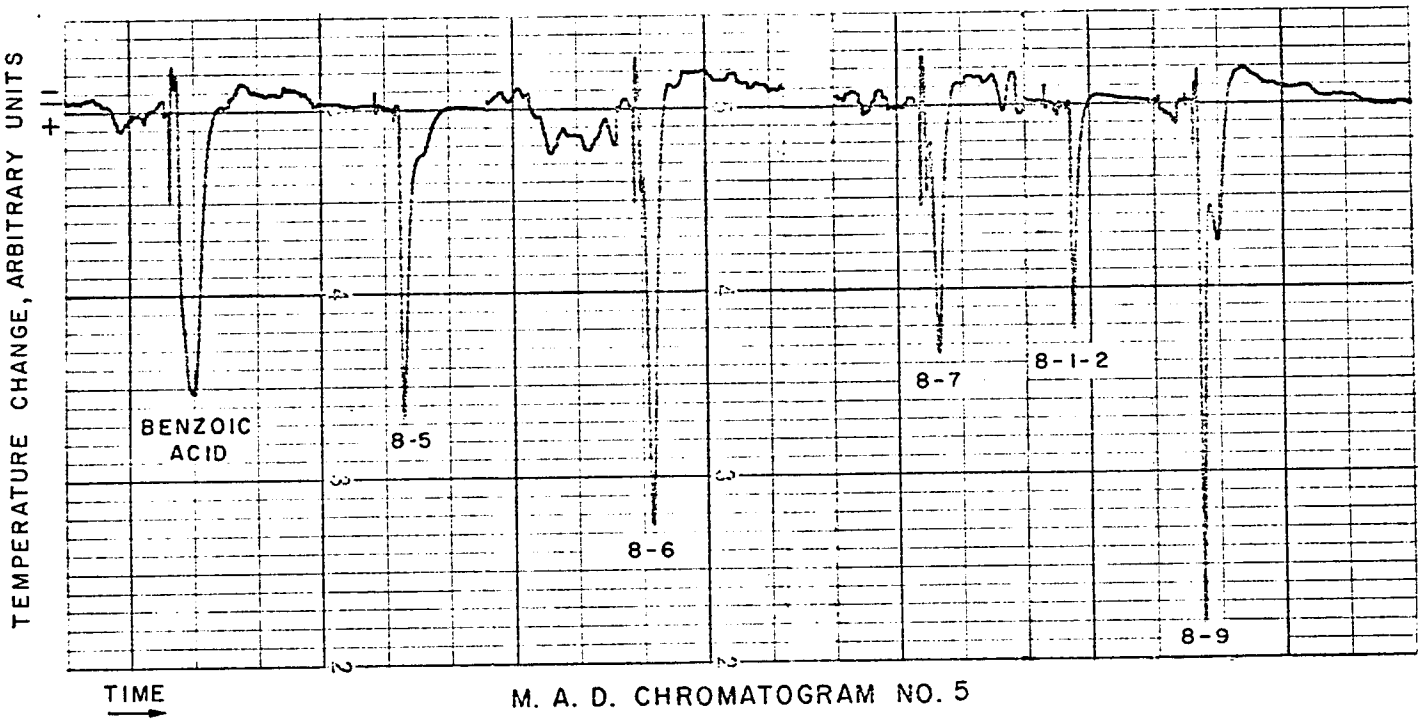
$$M_n = \frac{\Sigma W_i}{\Sigma(W_i/M_i)} ,$$

where: M_n = number average molecular weight,

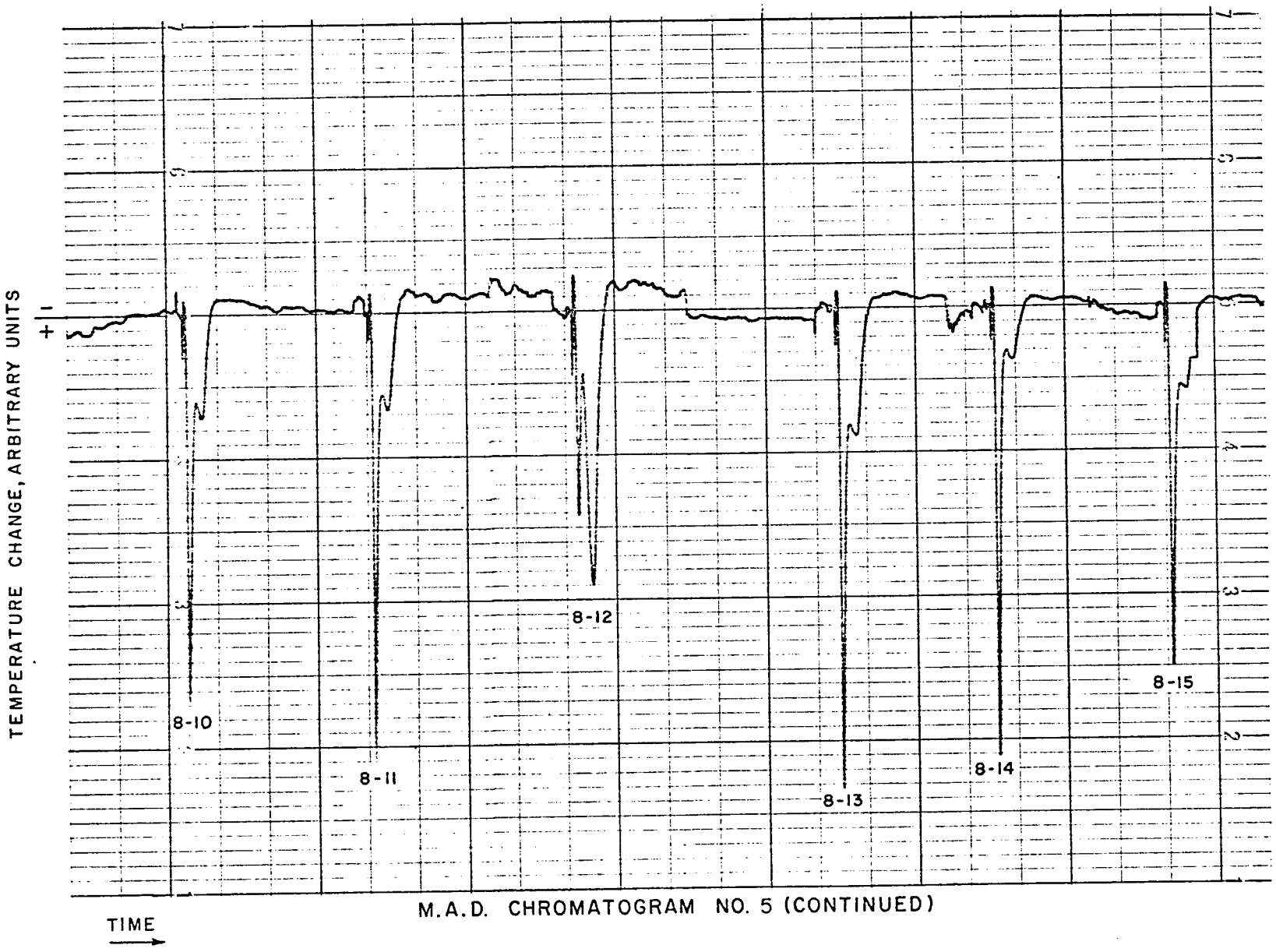
W_i = weight of each component in solution taken one at a time,

M_i = molecular weight of each compound taken one at a time.

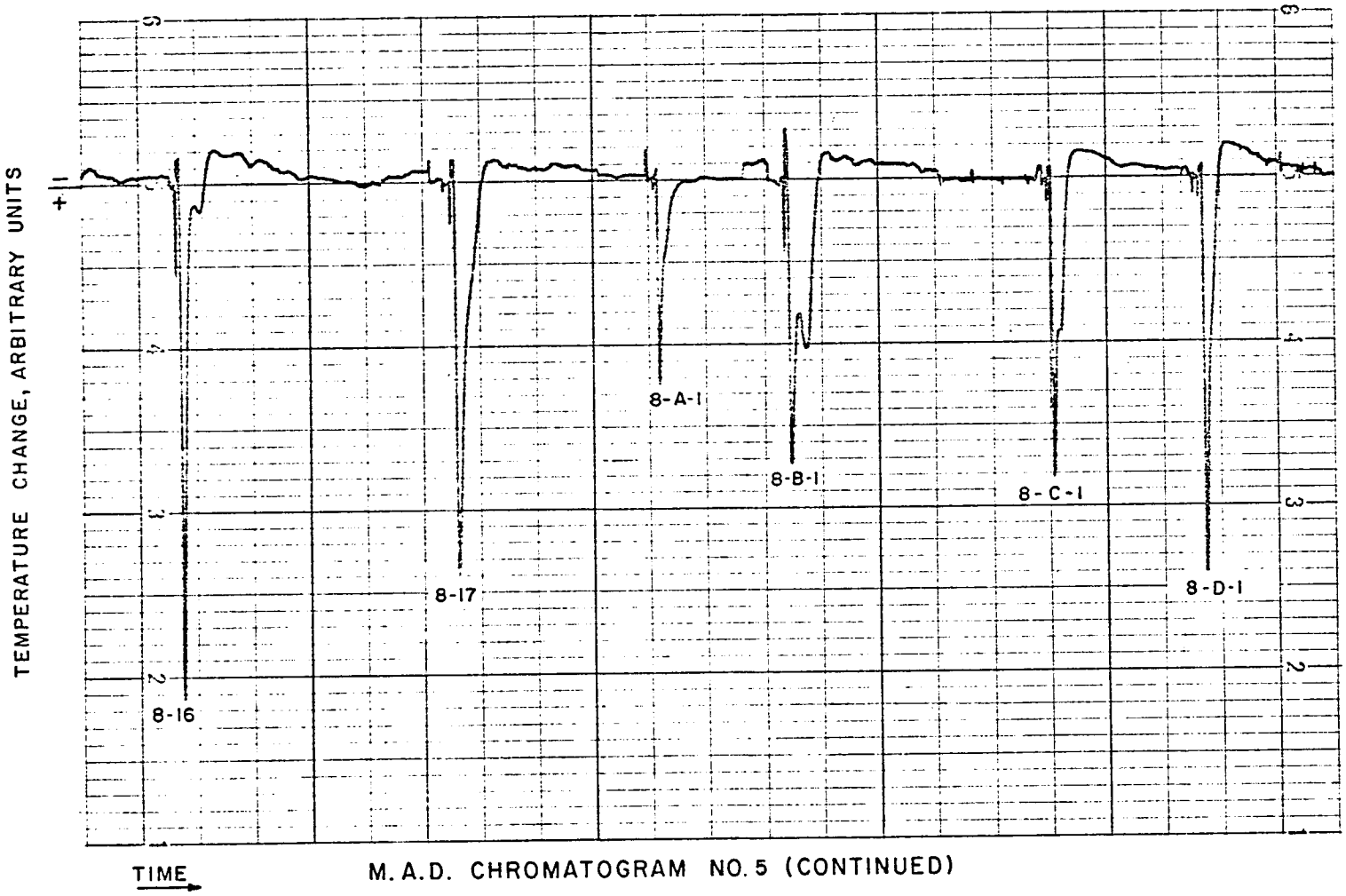
DATE: _____ TIME: _____
SAMPLE: 2 µl of each fraction
INLET PRESSURE: _____
FLOW RATE: 2.1 cc. / min.
COLUMN TEMPERATURE: 72° F
PACKING: inert glass beads
DETECTOR PACKING: pyrex glass / inert glass beads
TEMPERATURE: 72° F
ATTENUATION: variable
RECORDER CHART: _____
FULL SCALE: _____
SOLVENT: mole sieve dried benzene
VALVE SETTING: _____



M. A. D. CHROMATOGRAM NO. 5



M.A.D. CHROMATOGRAM NO. 5 (CONTINUED)



M. A. D. CHROMATOGRAM NO. 5 (CONTINUED)

TABLE XVIII

RELATIVE HEATS OF ADSORPTION STUDIES OF POLAR FRACTIONS FROM HEAVY OIL #8

Sample	A*	A x P**	Concentration In M/ In Benzene	M.W.***	Relative**** Heat Of Adsorption
Standard Benzoic Acid	1	15.2	2.00×10^{-3}	122	76.0
No. 8-5	8	34.0	12.6×10^{-3}	1023	107
8-6	1	22.0	0.950×10^{-3}	7406	231
8-7	1	13.8	1.24×10^{-3}	1206	133
8-1-2	16	200.0	24.3×10^{-3}	1098	82.4
8-1	2	56.6	12.2×10^{-3}	450	46.3
8-10	2	54.0	8.10×10^{-3}	1004	66.9
8-11	2	63.3	6.44×10^{-3}	804	98.3
8-12	1	18.9	1.60×10^{-3}	821	118
8-13	2	67.3	8.80×10^{-3}	907	76.9
8-14	2	62.4	8.55×10^{-3}	879	73.0
8-15	2	50.5	8.60×10^{-3}	803	59.0
8-16	2	63.6	10.0×10^{-3}	532	63.6
8-17	2	47.8	7.70×10^{-3}	808	62.1
8-A-1	8	101	13.1×10^{-3}	467	77.1
8-B-1	1	17.6	5.66×10^{-3}	827	31.1
8-C-1	4	74.9	10.7×10^{-3}	585	70.2
8-D-1	4	94.5	9.54×10^{-3}	568	99.0

*A = Attenuation factor.

**A x P = Attenuation factor times peak height from M.A.D. Chromatograms.

***Number average molecular weight determined by vapor phase osmometry (Hitachi Perkin Elmer Model 115) in solutions in $CHCl_3$.

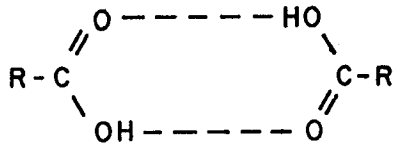
****Relative heat of adsorption expressed as (A x P) for a concentration of 0.01 M/ℓ and an attenuation of 1.

The Micro-Adsorption Detector response is a measure of relative strengths of adsorption and consequently of relative heats of adsorption. The heat of adsorption values are considered to be represented by (A x P), i.e., attenuation times M.A.D. peak height.

Fraction D was selected for further studies. The choice of Fraction 8-D-1 was made on the basis of highest relative heat of adsorption value (A x P = 99.0) and fairly low molecular weight (M.W. = 568) compared to all the major fractions isolated (Table XVIII). Fraction 8-D-1 was obtained by caustic extraction followed by ion exchange. It consists mostly of carboxylic acids.

The fraction with the highest heat of adsorption was chosen because it is the fraction most tenaciously adhered to the porous glass particles. Presumably this fraction would also adhere most tenaciously to sand grains, thereby interfering with secondary heavy oil recovery operations.

One point that must be kept in mind is that the determination of molecular weight of mixtures of carboxylic acids is complicated by the dimerization normally shown by acids, giving hydrogen-bonded structures of type (I):



(I)

In nonpolar solvents, such as carbon tetrachloride, acids exist as a mixture of monomers and dimers and perhaps even trimers.[45]

A molecular weight determination of lauric acid (in a 0.1 M solution in benzene) by the freezing point depression method gave a value of 294.5 for the molecular weight of lauric acid. The literature quotes a molecular weight of 200.32 for lauric acid. The discrepancy seems to indicate that at this particular concentration about half the lauric acid molecules are present in the dimeric form. If the molecular weight of an unknown acid is being determined by vapor phase osmometry or by freezing point depression, there is no way to determine what percentage of the molecules are dimers or trimers.

Summarizing, Fraction D was selected for further studies on the basis of its molecular weight and its relative heat of adsorption value. Fraction D required further fractionation for the ultimate aim of identifying some of its components.

F - Evaluation of Column Packing Materials Using M.A.D.

It was highly desirable to further fractionate Fraction D by liquid chromatography in order to be able to identify some of its components. Numerous packings were tested in a search for a suitable column for separating a mixture of carboxylic acids using the Micro-Adsorption Detector.

A mixture of 3.3×10^{-4} micromoles each of three carboxylic acids (lauric acid, benzoic acid, and acetic acid) in benzene was used as the test solution. The test solution contained 5.0×10^{-4} moles of each acid; the final volume was 15 ml. The objective was to obtain a M.A.D. spectra of the mixture of three carboxylic acids that would show three peaks indicating that the mixture had been resolved into its indicidual components. A list of some of the packings tested and comments on their performance is given in Table XIX. Whenever the carrier liquid was other than benzene, a peak appeared very shortly after injection due to the differences in heats of adsorption of carrier liquid and solvent. This type of peak is not mentioned or included in the comments of Table XIX. The combinations of packings and solvents tested were not successful in achieving the necessary resolution.

Another approach that was tested was to inject the standard test solution into a column of polyphenyl ether on Silicar connected to the Micro-Adsorption Detector. Fractions eluted from this liquid chromatography set-up were collected at regular intervals. These fractions were then analyzed individually by gas chromatography. Gas chromatography did not resolve these fractions and showed no indication that any of the fractions were pure compounds. The results from this attempt were thus quite discouraging.

TABLE XIX

EVALUATION OF COLUMN MATERIALS FOR SEPARATING CARBOXYLIC ACIDS

Column Packing	Dimensions	He Pressure psig	Carrier Liquid	Flow ml/min.	Comments
Silica Gel-H (Brinkmann)	24" x 1/8"	80	Benzene	-	Not enough resolution.
Corning 7235 Beads	18" x 1/8"	150	Benzene	-	Very slow flow through the column.
Corning 7235 beads	6" x 1/8"	150	Benzene	-	Very slow flow through the column.
Silica Gel, 200-325 Mesh	14" x 1/4"	82	Benzene	-	The carrier liquid did not seem to desorb the acid.
Mallinckrodt Silica Gel, 200-325 Mesh	14" x 1/4"	82	2-Methyl, 1-Propanol	-	Carrier liquid too viscous, causing flow rate to be very slow.
Mallinckrodt Silica Gel, 200-325 Mesh	14" x 1/4"	82	Methanol	-	Helium too soluble in this carrier liquid.
Corning CPG 10-240, Controlled Pore Glass Beads	12" x 1/8"	82	Benzene	-	Insufficient resolution; spectra gives 2 peaks only.

TABLE XIX (Cont'd.)

Column Packing	Dimensions	He Pressure psig	Carrier Liquid	Flow ml/min.	Comments
Corning CPG 10-240	12" x 1/8"	50	Sodium-dried Benzene	2.1	2 peaks only.
Corning CPG 10-240	12" x 1/8"	-	10% Ether in Benzene	-	2 peaks only.
Corning CPG-10-240	12" x 1/8"	50	Sodium-dried Benzene	2.1	2 peaks only.
Corning CPG 10-240	18" x 1/8"	50	10% Ether in Benzene	1.8	2 peaks only.
Corning CPG 10-240	18" x 1/8"	80	10% Ether in Benzene	2.8	2 peaks only.
Florisil 30/60 Å (i.e. magnesium trisilicate)	12" x 1/8"	12.5	10% Ether in Benzene	2.4	No resolution.
High Purity Silica Gel, 60-200 Mesh (Curtin Co.)	12" x 1/8"	-	10% Ether in Benzene	2.0	1 peak only.
High Purity Silica Gel, 60-200 Mesh	12" x 1/8"	40	Benzene	2.6	No resolution.
Silica Gel-H	6" x 1/8"	100	10% Ether in Benzene	1.6	1 peak; flow is too slow.

TABLE XIX (Cont'd.)

Column Packing	Dimensions	He Pressure psig	Carrier Liquid	Flow ml/min.	Comments
Silica Gel-H	6" x 1/8"	100	Sodium-dried Benzene	1.0	Flow is too slow; no resolution.
Silica Gel-H	6" x 1/8"	200	3% Ether in Benzene	2.0	1 peak; at this He pressure the instrument exhibits a lot of noise.
Corasil Type I	18" x 1/8"	50	3% Ether in Benzene	1.7	1 peak.
Corasil Type I	6" x 1/8"	-	3% Ether in Benzene	-	No resolution.
Corasil Type I	18" x 1/8"	100	Sodium-dried Benzene	3.0	2 peaks.
Celite #545	6" x 1/8"	43	Benzene	2.1	No resolution.
Activated Charcoal G-60	6" x 1/8"	238	10% Ether in Benzene	0.8	2 peaks; the column packing was very fine and it offered very high resistance.
Amberlite IRA 401-S (Ion Exchange Resin)	6" x 1/8"	240	2:2:1 Benzene/ Ethanol/Acetic Acid	2.1	No resolution.

The Micro-Adsorption Detector proved not suitable for detecting the separation of acids and was abandoned. Gas chromatography was selected as a more practical way of approaching the acid mixture separation.

In summary, a large number of column packings were tested in an attempt to resolve a carboxylic acid mixture with the aid of the Micro-Adsorption Detector. The required resolution was not achieved and thus liquid chromatography with M.A.D. was discarded in favor of gas chromatography.

Experimental

M.A.D. Chromatogram No. 1: The detector was packed with 29 μ glass beads and activated charcoal. Chloroform was the solvent (flow rate 144 ml/hr). The samples were injected into a capillary restrictor.

M.A.D. Chromatogram No. 3: This chromatogram resulted from six separate injections, 2 μℓ each, of cyclohexane carboxylic and acetic acid solutions stored under various conditions. A pre-column was connected in series to a capillary restrictor to prevent the plugging of the capillary restrictor. The pre-column was a 6" length of 1/8" stainless steel tubing, packed with inert glass microbeads from Microbeads Division of Cataphote Corporation. Benzene, dried by passing it over molecular sieves, was used as the mobile liquid, with a flow rate of 138 cc/hr.

Infrared Spectrometry: IR spectra of cyclohexane, carboxylic and acetic acid solutions stored under different conditions were obtained with a Beckman Spectrophotometer Model IR-4 using a 0.1 mm NaCl cell. To compensate for the solvent, the solutions were run against the solvent (benzene dried by passing it over molecular sieves) in a variable path length cell.

M.A.D. Chromatogram No. 4: This chromatogram shows the relative peak heights for five acids and five phenols of the best grade available. For this run the adsorption cell of the detector was packed with 60-74 μ porous glass beads (High Purity Porous Glass Adsorbent, 325 mesh; Catalog No. 7235, Corning Glass Works). The reference cell of the detector was packed with smooth glass beads (74-105 μ, Corning Glass Beads, Acid Washed). Conditions of this run were: sample size, 2 μl; attenuation, 32; carrier liquid, mole sieve dried benzene; capillary restrictor. The standards were 0.1 M benzene solutions of five acids and five phenols. Traces of water were removed from the solutions by azeotropic distillations with benzene.

Melting Points: Melting points of standard acids and phenols were determined by heating the samples in capillary tubes in a glycerin-filled beaker over a hot plate-stirrer.

Separation Scheme A - Silica gel chromatography followed by ion exchange chromatography.

1. Reagents - The silica gel (60-200 mesh) was obtained from W. H. Curtin & Company, Houston, Texas. The anion strong base exchange resin was Amberlyst A-29, and the strong acid cation exchange resin was Amberlyst A-15, both supplied by Rohm and Haas. The n-pentane was 98%, B.P. 35-36° C, from Matheson Coleman and Bell. All other chemicals were reagent grade and were used as received.

2. Preparation of Resins and Adsorbents

Anion-Exchange Resin - Amberlyst A-29 resin was washed twice with a 4 percent by weight aqueous sodium hydroxide solution and then rinsed with distilled water until the washings were neutral to litmus paper. Final preparation of the resin was made by rinsing with methanol followed by benzene.

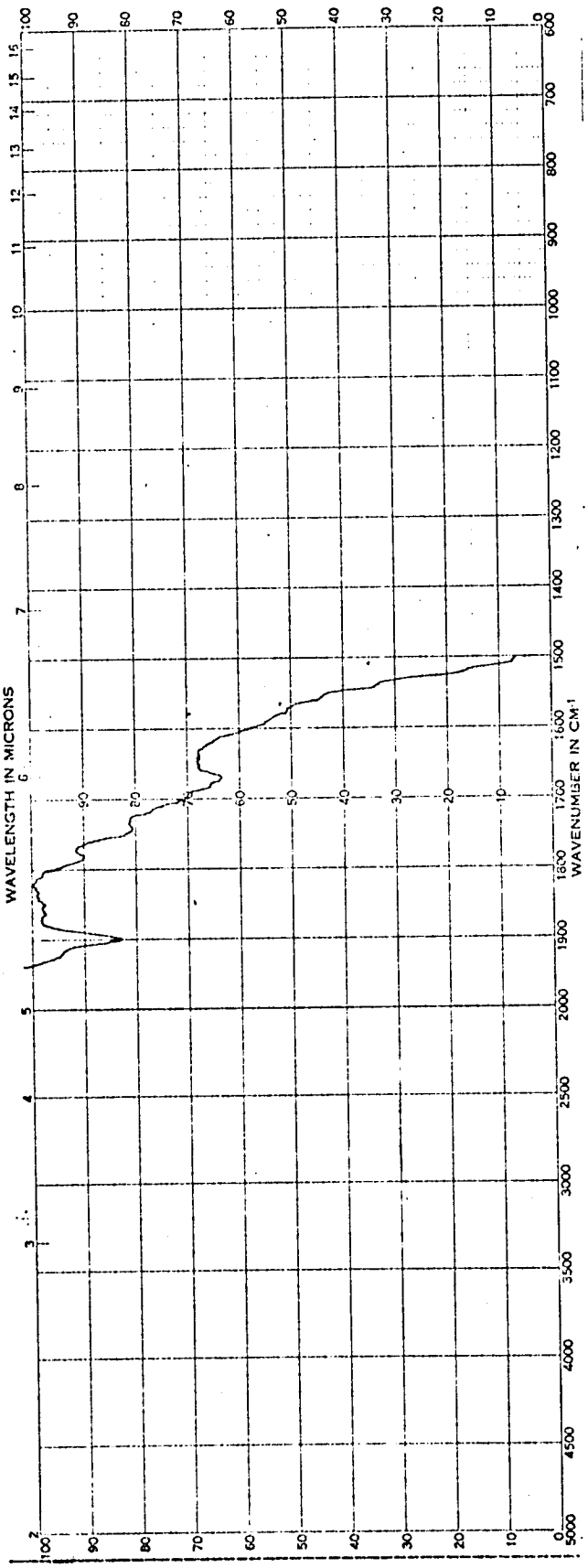
Cation-Exchange Resin - Amberlyst A-15 was washed with a 1.5 N aqueous sodium hydroxide solution and rinsed with distilled water. (Note: Exothermic reaction of the resin with water accompanied by swelling of the resin was observed at this point.) The resin was activated with a 4 percent by volume aqueous sulfuric acid solution. The resin was then washed with distilled water until the washings were neutral to litmus paper and dried for 72 hours at 40° C in a vacuum oven.[46]

Silica-Gel Adsorbent - The silica gel was activated by heating at a temperature of 110° C for 24 hours in a gravity convection oven and stored in a desiccator.

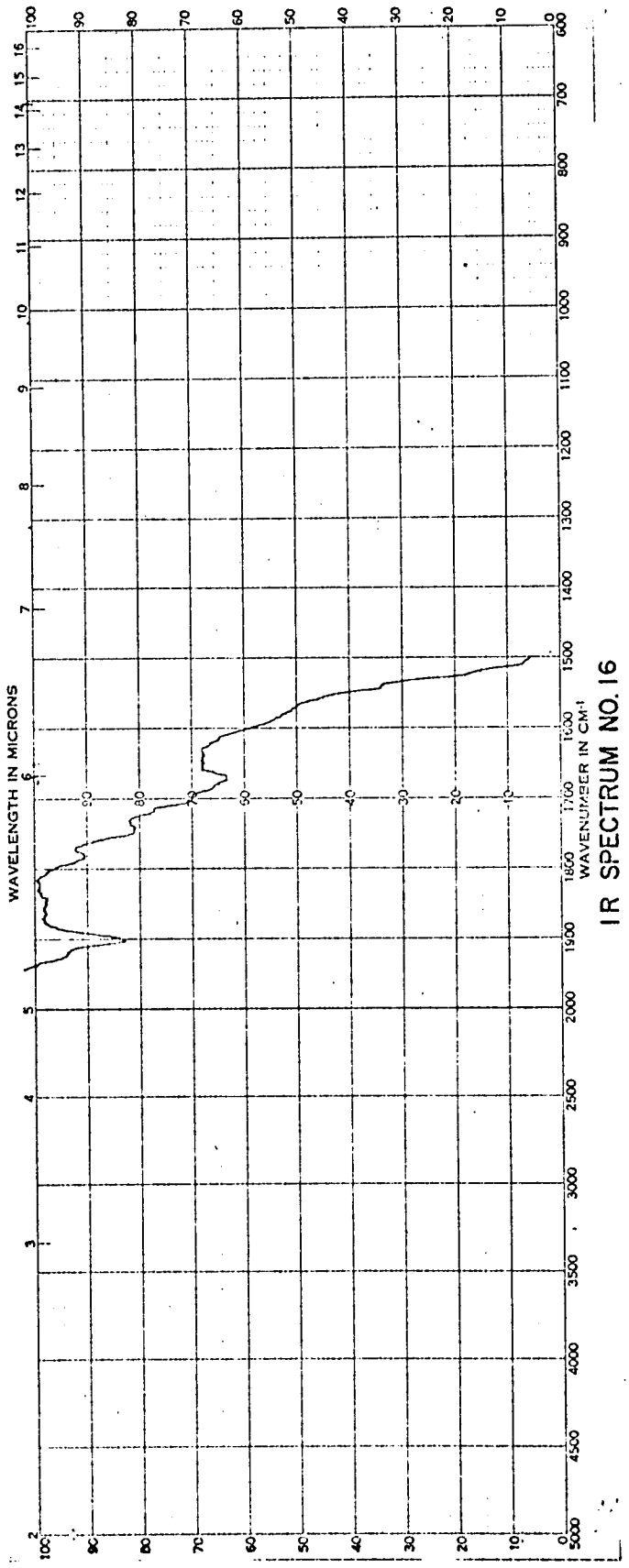
3. Silica-Gel Chromatography[24] - A 59 x 4.5 cm column was packed with 240 g of activated silica gel. A 46.5 g sample of Heavy Oil #8 was dissolved in benzene and charged to the column. The solvent mixtures employed for eluting the individual fractions are shown in Table XIV. Total recovery was 98.4 weight percent.

4. Cation-Exchange Chromatography - The cation-exchange resin was packed into a 40 x 2 cm column. Fraction 8-1a, 1.32 g, was added to n-pentane and charged to the resin (Figure 3). (Note: Fraction 8-1a was not very soluble in n-pentane.) Entrained, unreactive material was washed from the resin with pentane. An IR spectrum was run of the last volume of pentane through the column (IR Spectrum No. 15). Compared to a spectrum of fresh pentane (IR Spectrum No. 16), no differences can be observed around 1700 cm^{-1} (carboxyl region). Since this indicates that no more acidic material was being eluted by the pentane, the resin was then washed with benzene. IR Spectra No. 15 and No. 16 were run versus air using a 0.4 mm NaCl cell.

The reactive material (bases) was removed from the resin by successive elutions with chloroform, methanol, and 17 volume percent isopropyl amine in methanol. The bases were thus removed as three fractions (Table XV).



IR SPECTRUM NO. 15



IR SPECTRUM NO. 16

5. Anion-Exchange Chromatography - The anion-exchange resin was packed into a 40 x 2 cm column. Fraction 8-1b, 1.5 g, was dissolved in 30 cc of benzene and charged to the resin (Figure 3). Unreactive material was washed from the resin with benzene. The reactive material (acids) was then recovered by successive elutions with methanol, methanol saturated with CO_2, and 74:25:1 methanol:chloroform:acetic acid. These three solvent mixtures remove compounds of increasing acid strength and the combined fractions give the total acid fraction (Table XV).

All fractions were freed of solvent by vacuum stripping until a constant sample weight was obtained.

Separation Scheme B - A sodium hydroxide-alcohol-water extraction followed by ion-exchange chromatography.

The extraction and ion chromatography procedures described below are based on Seifert's articles.[24,42]

1. Sodium Hydroxide-Alcohol-Water Extraction - A water-free sample of Heavy Oil #8 (50.5 g) was dissolved in 100 ml of petroleum ether (B.P. = 60-110° C) and the solution extracted twelve times with 50 ml portions of 1% sodium hydroxide in 70% ethanol. All of the combined basic extracts were extracted ten times with 50 ml of petroleum ether. The petroleum ether extracts were reduced in volume to about 300 ml at 54° C and then back extracted with ethanolic sodium hydroxide. The back extract was combined with the main batch of alkaline extract and concentrated to 150 ml which, after acidification with HCl at 0° C, was extracted with diethyl ether until the extracts were colorless. The combined diethyl ether extracts were

washed with water to neutrality, dried with 2 g of magnesium sulfate, and filtered. The solvent was removed at room temperature. The last traces of water were removed by azeotropic distillation with benzene. Stripping to constant weight at 50° C gave 1.34 g of Major Acidic Extract (Table XVII, Heavy Oil 8-1).

2. Ion-Exchange Chromatography - A glass column 40 x 2 cm I.D. filled with deionized water was packed with Duolite A-6 resin to a height of 20 cm and successively washed with deionized water, 200 ml of 1.5 N aqueous sodium hydroxide and again with deionized water. Hydrochloric acid (2 N), 200 ml, was then passed through the column followed by deionized water. Finally, 250 ml of 1.5 N NaOH were added to the column and the resin was rinsed with deionized water until neutral. Ethanol, 550 ml, was then passed through the column, followed by 450 ml of a 2:1 mixture of benzene:ethanol. To this column was added 1.34 g of Major Acidic Extract dissolved in 10 ml of an 8:5 mixture of benzene:ethanol. Initial elution of the column was carried out with 1,000 ml of a 2:1 mixture of benzene:ethanol until the eluate became colorless. After solvent removal, 0.64 g of Fraction X was obtained. The column was then eluted with a 2:2:1 mixture of ethanol:benzene:acetic acid until the eluate became colorless. Traces of acetic acid were removed from the solvent mixture (1,000 ml) by azeotroping with n-heptane and 0.79 g of Fraction Y was obtained (Table XVII, Heavy Oil 8-1).

In each instance, solvent was removed using a rotary evaporator. Final pumping of fractions to constant weight was conducted in the vacuum oven at 50° C.

Fraction Y: A 40 x 2 cm I.D. column of Duolite A-6 ion exchange resin was prepared. To this resin was added 0.79 g of Fraction Y dissolved in 10 ml of 2:1 benzene:ethanol mixture. Elution of the column with 1 ℓ of a 2:1 mixture of benzene:ethanol afforded, after solvent stripping, 0.16 g of Fraction C. A further 0.58 g of Fraction D was eluted with 1.25 ℓ of a 2:2:1 mixture of ethanol:benzene:acetic acid (Table XVII, Heavy Oil 8-1).

Fraction X: A column 40 x 2 cm I.D. of Amberlite IRA-401S ion-exchange resin was prepared in the manner described above. A solution of 0.64 g of Fraction X in 2 ml of 2:1 benzene:abs. ethanol mixture was placed on the column and elution carried out initially with a 2:1 mixture of benzene:abs. ethanol. After removal of 1.3 ℓ of solvent, 0.15 g of Fraction A was obtained. Further elution with 2 ℓ of 2:2:1 mixture of abs. ethanol:benzene:acetic acid gave, after solvent stripping and use of n-heptane for azeotropic removal of acetic acid, 0.51 g of Fraction B (Table XVII, Heavy Oil 8-1).

Infrared Spectrometry - Infrared spectra of two of the polar fractions isolated from Heavy Oil #6 were obtained. The fractions analyzed were: 6-Major Acidic

Extract-3 and Fraction 6-D-2. Infrared spectra were obtained using a Beckman Model IR-4 infrared spectrophotometer.

Evaluation of Column Packing Materials - A mixture of three carboxylic acids was injected into a column connected to M.A.D. A number of columns were tested, each packed with a different material. The carrier gas was helium. The sample size was 10 $\mu\ell$. The detector was packed with porous glass beads and inert glass beads.

Part III: Characterization of Acid Fraction From Heavy Oils

Discussion

A - Introduction

Fraction D obtained thus far consisted mostly of a mixture of carboxylic acids. Since our research objective was to identify some of the polar components of heavy oils, it was necessary to further fractionate Fraction D. The carboxylic acids contained in Fraction D were converted to their methyl esters (methanol/boron trifluoride). This complex ester mixture was fractionated by liquid chromatography using urea and thiourea columns.

All of the fractions obtained from the urea and thiourea chromatographies were analyzed by gas chromatography (GC). The major peaks from the chromatograms were trapped in glass capillaries for mass spectral studies. Due to the small size of the samples collected from the GC, mass spectrometry was selected as the best analytical technique for determining compound structures present in the samples. Mass spectrometry requires only 20-50 μg of sample per analysis. The other available analytical tools such as NMR, IR and C-H-N analyses require approximately 20 to 100 mg.

B - Esterification Procedures

Ester derivatives of the carboxylic acids in Fraction D were prepared to facilitate the use of gas chromatography

to separate these compounds. Several esterification procedures were investigated and a satisfactory one was chosen. Methyl esters are more volatile and are less subject to thermal decomposition then the corresponding free acids[47], and they are therefore better suited for gas chromatographic separations and mass spectrometric structure determinations.[48]

Five different methods to prepare the esters were evaluated. Small portions of Fraction 8-D-1 were converted to trimethylsilyl esters by treatment with (1) Tri-Sil/BSA* in pyridine, (2) Trimethylsilyl reagent in dimethylformamide, and (3) Trimethylsilyl reagent in pyridine. The methyl esters of Fraction 8-D-1 were prepared with (1) BF_3 in methanol, and (2) BCl_3 in methanol.

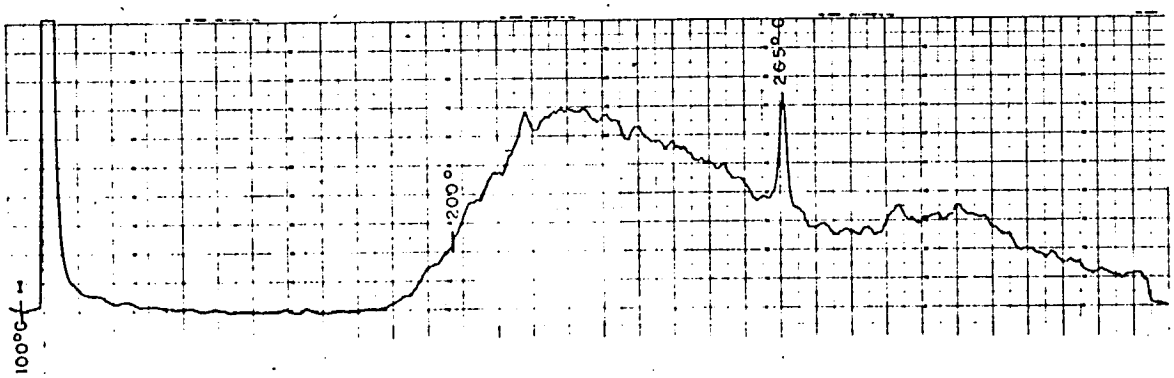
The ester mixtures resulting from the five different esterification techniques were examined by gas chromatography. From the GC spectra, no differences could be detected among the five ester mixtures. A large broad unresolved peak was observed from about 139 to 300° C. In some of the spectra a relatively sharp and distinct peak at 265° C, possibly due to contamination, was superimposed on the broad peak.

Gas Chromatogram No. 1 represents a gas chromatogram of the methyl esters of Fraction 6-D-1 on SE-30; it is a

*Tri-Sil/BSA is the trade name for N,O-bis-(trimethysilyl)-acetamide from Pierce Chemical Company, Rockford, Illinois.

GAS CHROMATOGRAM NO. I

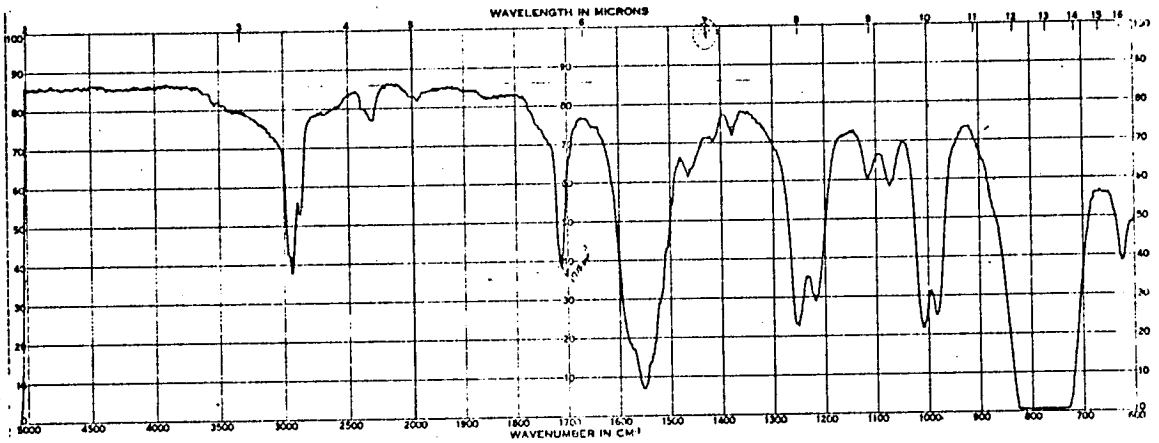
METHYL ESTERS DERIVED FROM
FRACTION 6-D-I BY BCl_3 - MeOH



typical example of the chromatograms obtained for the five ester mixtures. The GC spectra clearly indicate that the ester mixtures were still too complex for direct separation by gas chromatography. It became necessary to further fractionate the ester mixtures.

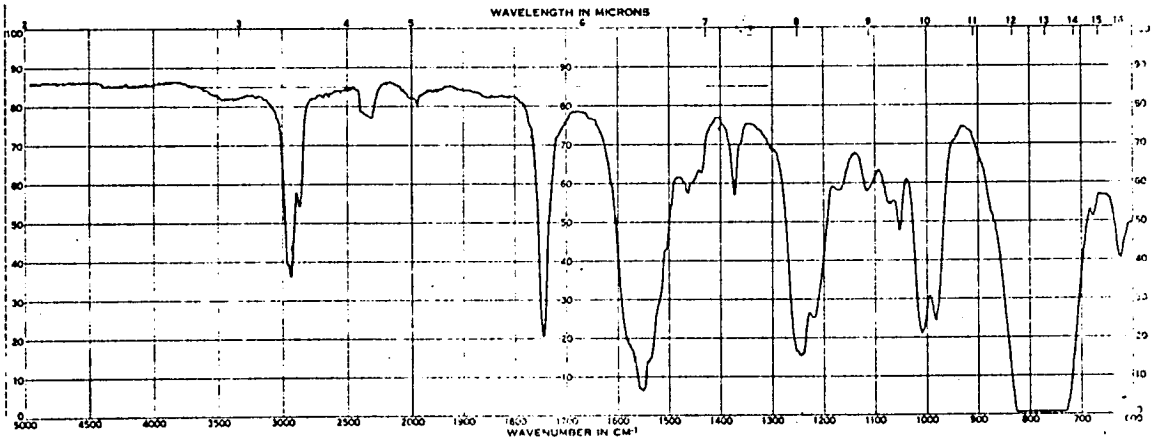
Methyl esters were chosen as the more desirable derivatives because the larger molecular weight of the silyl esters makes them harder to identify by mass spectrometry.[49]

To verify the success of the methylation procedures, IR spectra of the following samples were obtained: (1) Fraction 8-D-1, before esterification (IR Spectrum No. 17); (2) methyl esters derived from Fraction 8-D-1 by using BF_3/MeOH (IR Spectrum No. 18); and (3) methyl esters from Fraction 8-D-1 by BCl_3/MeOH (IR Spectrum No. 19). The IR spectra showed a shift from 1712 cm^{-1} to 1742-45 cm^{-1} when the carboxylic acids were converted to methyl esters.[50] There is a slight shoulder at 1710 cm^{-1} in the spectra of the BCl_3-methyl esters indicating the possibility of incomplete methylation. The extinction coefficient ratio of $\epsilon_{1712 \text{ or } 1745\ cm^{-1}}/\epsilon_{2950\ cm^{-1}}$ for the three samples is: (1) 1.52, (2) 1.51, and (3) 0.97, respectively. The absorption band at 3000 cm^{-1} is very broad in the spectrum of Fraction 8-D-1. In the ester spectra, this band is narrower, as expected. The results of these IR spectra indicate that BF_3 may be a more efficient esterification reagent for this particular carboxylic acid mixture.



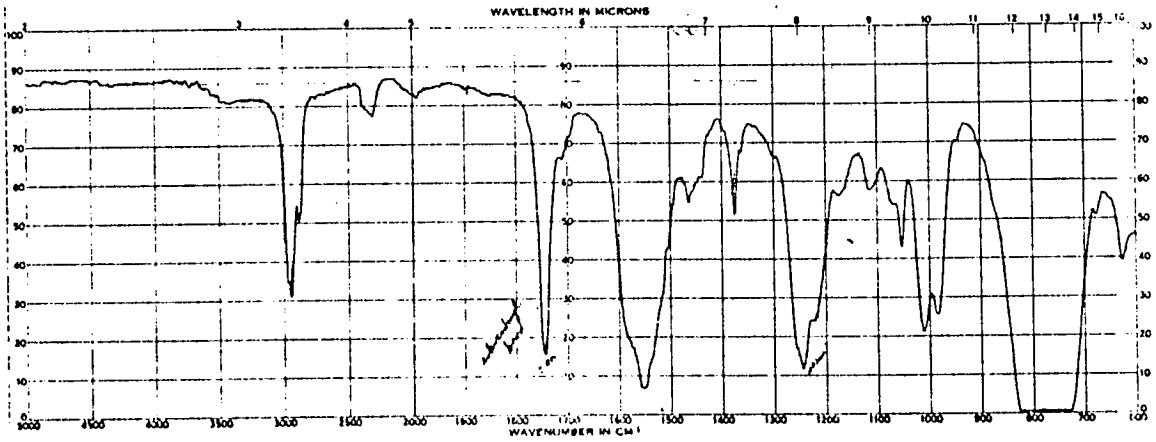
IR SPECTRUM NO. 17

FRACTION 8-D-1



IR SPECTRUM NO. 18

METHYL ESTERS OF FRACTION 8-D-1 BY BF_3-MeOH



IR SPECTRUM NO. 19

METHYL ESTERS OF FRACTION 8-D-1 BY BCl_3-MeOH

All methyl esters used in the rest of our research were prepared with BF_3/MeOH following an elaborate procedure of repeated back-washings. This procedure is described in the Experimental section.

C - Urea and Thiourea Chromatographies

Urea Chromatography - Normal esters can be separated from branched-chain esters by clathration with urea.[51] Urea inclusion compounds are formed more readily by straight-chain than by branched-chain compounds, thus giving a basis for separation.[52] Urea forms adducts with straight chain compounds (9-20 atoms) with iso-compounds and with alkylbenzenes. Also any aromatic or cyclic ester with a long alkyl chain attached to the ring would be adducted by urea.

For these reasons it was decided to fractionate the methyl esters of the acids in Fraction 6-D-2 by urea clathration. The column chromatographic method by Coles[53] was selected for the separation. Four fractions were obtained from the first urea treatment. The yield of non-clathrated materials, which were eluted with n-hexane, was 0.40 g (Fraction I). The yield of mixed compounds eluted with ether was 0.011 g (Fraction II). Fraction III, 4 mg, consisted of the clathrated material eluted with methanol. Fraction IV (0.002 g) consisted of the higher molecular weight clathrated materials that were

extracted with hot methanol-hexane solutions. The total recovery was 96%. Because of the poor yield of clathrated material, the nonclathrated material, i.e. Fraction I, was again subjected to the urea chromatography just described. The second urea treatment yielded the following four fractions: (1) Fraction VII, eluted with hexane; (2) Fraction VIII, eluted with ether; (3) Fraction IX, clathrated material eluted with methanol, 0.006 g (1.5% recovery); Fraction X, eluted with hot methanol-hexane solution. The recovery was 96.1% for the second urea treatment.

All the methyl ester mixtures derived from the various Fractions D were subjected to urea chromatography. The fractions resulting from the urea chromatography should be pure enough to be handled by gas chromatography.

Thiourea Chromatography - Thiourea also forms inclusion compounds, but the channel diameter is larger (about 6.5 Å) and the structural requirements of the guest molecule less specific than in the case of urea.[54] Consequently, thiourea adducts with straight-chain, branched and cyclic compounds, small ring compounds, isoprenoid compounds, and polycyclic hydrocarbons.

In order to obtain more fractions, the nonclathrated material obtained during the second urea treatment was subjected to thiourea chromatography. Thiourea chromatography was conducted following the procedure for urea

chromatography which has been described in detail in the Experimental section.

The fractions resulting from performing the thiourea treatment on Fraction VII-6-D-2 were: (1) Fraction XI, 0.374 g, eluted with hexane; (2) Fraction XII, eluted with ether; (3) Fraction XIII, 0.10 g, eluted with methanol; (4) Fraction XIV, eluted with hot methanol-hexane solution.

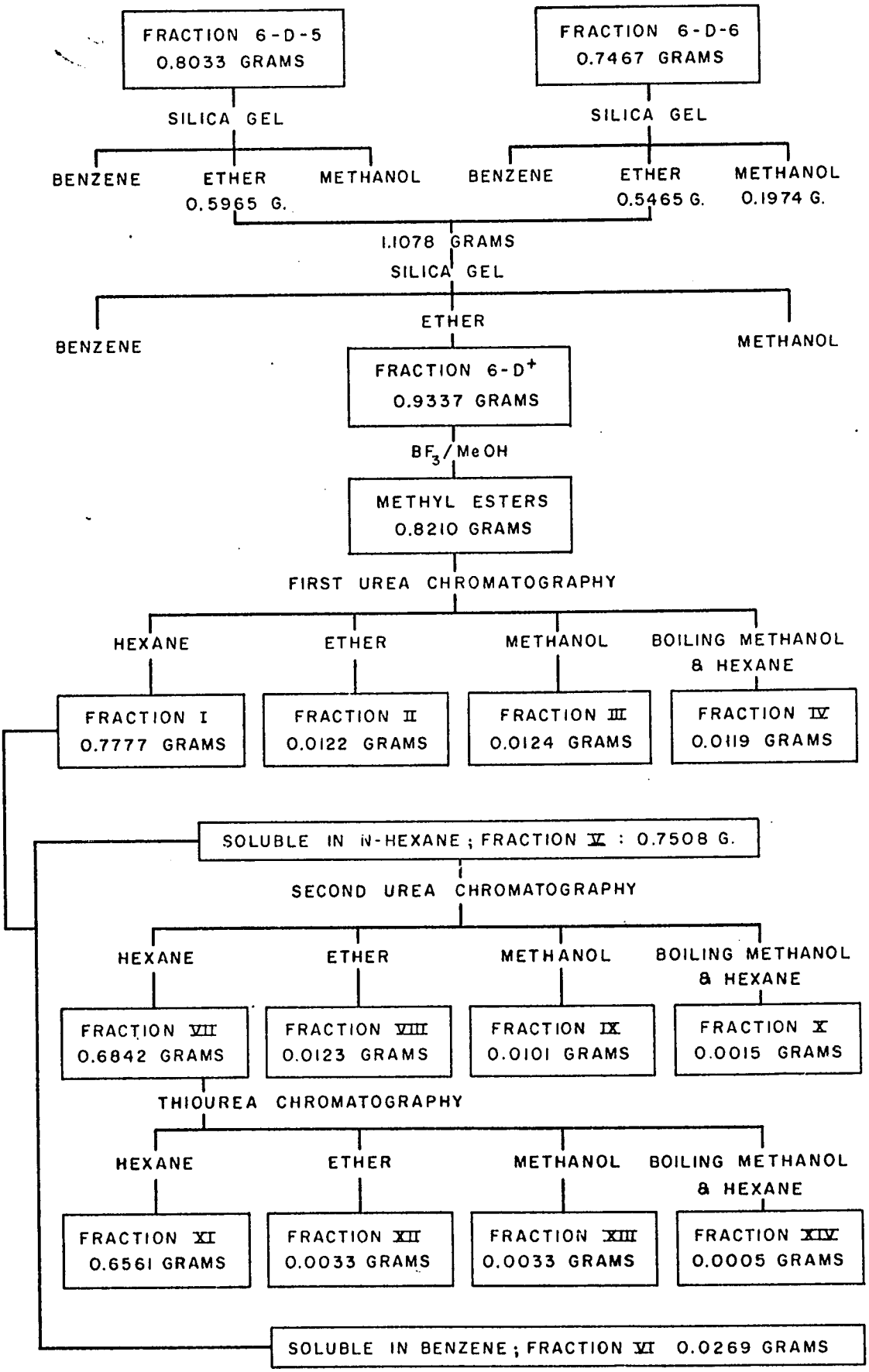
Use of thiourea adduction on Fraction VII yielded four more fractions through a clathration mechanism. Thiourea adduction was thus applied as one more step in the complex separation scheme intended to provide narrow fractions for gas chromatographic analysis. Several fractions underwent thiourea chromatography and reference to such fractions is made later in the text.

Thiourea Adduction with an Inducer - The approach described below was tested with one fraction only and was never repeated. In order to increase the adduction by thiourea in methanol, Schlenk[55] recommended the use of cyclohexane or iso-octane as an inducer. Because the yields of materials adducted by thiourea were very small, it was decided to try a thiourea adduction using cyclohexane as an inducer.

Fraction XI-6-D^{+} (Table XVII and Figure 5), too complex to be resolved directly by gas chromatography, was

FIGURE 5

SEPARATION SCHEME B (CONTINUED)



selected to test the effect of cyclohexane on thiourea adduction. Fraction XI-6-D^+ was subjected to thiourea adduction using cyclohexane as an inducer. Two fractions resulted from this treatment:

(1) Fraction XV-6-D^+, 0.015 g of adducted esters,

(2) Fraction XVI-6-D^+, 0.586 g of nonadducted esters.

A comparison of the results obtained by both types of thiourea adduction procedures shows that only 0.5% of the starting material was adducted, when thiourea was used by itself, while 2.47% of starting material was adducted by thiourea with cyclohexane as an inducer. A tremendous increase in the yield of adducted esters was obtained when cyclohexane was used as an additive. Unfortunately, this experiment was conducted near the end of our experimental program and no further work along these lines was conducted.

D - Gas Chromatography and Mass Spectrometry

The fractions obtained through the complex separation scheme were analyzed and separated by gas chromatography. Some of the fractions collected by gas chromatography were analyzed by mass spectrometry in an attempt to identify or characterize the individual components.

1. Instrumentation

Gas Chromatography - All the analytical gas chromatography was conducted at Texaco Research Laboratories in Bellaire, Texas on an Aerograph 204-B flame detector gas chromatograph.

Three major modifications were made to the gas chromatograph. The first was the introduction of a splitter-collector for collection of pure fractions for mass spectrometric analysis. Two commercially available splitter-collectors, one from Varian and the other one from Research Specialties Company, were tested but their performance proved unsatisfactory. The best results were obtained with a microsample splitter-collector that was designed*, built, and installed at our labs. An aluminum collar surrounding the collector tubing allowed enough heat transfer for collection of components having boiling points above 300° C (C_{18}). A septum, a stainless steel washer, and a compression nut arrangement were used to make sure the carrier gas passed through the capillary tube. Microsamples could be simultaneously detected and collected. Components above C_{11} were collected at room temperature. The homemade splitter provided approximately a 1:10 split ratio in favor of the collector; the split ratio is a function of temperature and of amount of sample injected. A melting point capillary (12" x 1.5 mm) was inserted into the effluent of the chromatograph when the peak of interest appeared. When the fraction was collected the tube was removed and other tubes inserted to collect other peaks. The tubes were cooled at one end and warmed at the other end to concentrate the sample, and immediately sealed for storage by melting the ends of the tube.[56]

*Splitter-collector was designed by Dr. B. G. Harnsberger.

The second modification introduced into the gas chromatograph consisted of special chromatographic inlets from Precision Sampling Corporation. These injectors permit high inlet block temperatures without heating the septum to the point of decomposition or "bleeding". Their design keeps the septum at 85-125° C, while bringing full heat of the block to within 1/4".

The third modification was the installation of additional silica gel traps between the flow controllers and the injectors. Their purpose is to remove any bleed of plasticizer, etc., from the rubber flow controller diaphragm and also to trap any flashback of chemicals from the injector should an excessive sample volume be injected.

The stationary phases used for the column packings were SE-30 or Dexsil 300 GC. Dexsil is a new polycarborane siloxane stationary phase recommended for efficient separations from 50° C to 500° C and good for high boiling samples. It is distributed by Analabs, Inc. Based only on the performance of Dexsil in our GC work, we found it to be highly satisfactory, showing signs of low bleed and providing good resolution. Fractions resulting from urea and thiourea chromatographies were resolved more efficiently with a 6' x 1/8" column of 15% Dexsil on 80-100 mesh Varoport-30 than with a 12' x 1/8" column of 5% SE-30 on 80-100 mesh Varoport-30.

The gas chromatography work was performed on an Aerograph 204-B flame detector gas chromatograph, modified to allow sample collection and prevent septum "bleeding". The stationary phases used were SE-30 and Dexsil 300 GC. On the basis of GC performance only, Dexsil 300 proved to be a superior stationary phase.

Mass Spectrometry - The collected samples resulting from gas chromatographic separations were analyzed by mass spectrometry. The mass spectra were run at three different locations on three different types of mass spectrometers. The locations were: (1) Chemistry Department, Rice University, Houston, Texas; (2) Baylor College of Medicine, Houston, Texas; (3) Beacon, New York. A description of the instruments and experimental conditions is covered in the Experimental section.

The mass spectrometers at Rice University and at the Texaco Laboratories at Beacon, New York, had a direct insertion probe. The availability of a solid inlet system, i.e., of a direct insertion probe, was essential because our samples collected into capillaries, could not be otherwise introduced into the mass spectrometer.

The gas chromatographic inlet system such as the one in the combined gas chromatograph-mass spectrometer (GC-MS) at Baylor College of Medicine was ideal for our work. Unfortunately, this combination was not available to us for many of the analyses.

Most of the mass spectra run at Rice University were supervised by Dr. P. A. Haug, to whom we are indebted for her cooperation and enlightening comments. All combined GC-MS spectra recorded for this research were run at Baylor College of Medicine, under the supervision of Dr. J. A. McCloskey.

A computer program was written for processing digitized mass spectral data. The program tabulated the spectra, after normalization, and the resulting relative peak intensity-mass number relationships were graphically displayed on the UCC 2000 drum plotter. Several variations were necessary to handle mass spec data from the three different sources.

2. GC Analysis of Fractions

The fractions isolated through the complex separation scheme (Figure 5) were analyzed by gas chromatography. Gas chromatography further resolved some of the fractions and these were collected and analyzed by mass spectrometry.

The purpose of the gas chromatographic analyses of the isolated fractions was to ascertain the complexity of the fractions and the approximate carbon number range of the components. Based on this information, the fractions adequately resolved by gas chromatography were selected for collection purposes.

The experimental conditions were not the same for all GC analyses. The early analyses were run using SE-30 columns. Dexsil GC 300 was used for later analyses because of its better resolution. The conditions for gas chromatography are specified in the individual cases described below and in the Experimental section.

All unknowns were compared to a series of pure methyl esters of fatty acids. The labelled peaks in all of the gas chromatograms indicate the retention time corresponding to a straight-chain methyl ester standard. The labelling must be taken only as an indication of approximate carbon number and does not indicate that a particular peak has been identified as a straight-chain ester. The labelling was possible because all the fractions were co-injected with pure methyl esters. Co-injection served to calibrate the mass scale for the gas chromatograms. Because of the complex nature of the gas chromatograms, no examples of the runs involving co-injections are included.

Those samples analyzed by gas chromatography which were not selected for collection purposes are described below.

1. Sample: Fraction VIII-6-D-2
   Column: SE-30 plus GC SE-30 (12' x 1/8")

The chromatogram obtained for Fraction VIII-6-D-2 exhibited three large and distinct peaks, the first was at a retention time equal to that of a n-C_{13} ester of a carboxylic acid.

2. Sample: Fraction III-6-D-2 (Gas Chromatogram No. 2)
Column: SE-30 plus GC SE-30 (12' x 1/8")

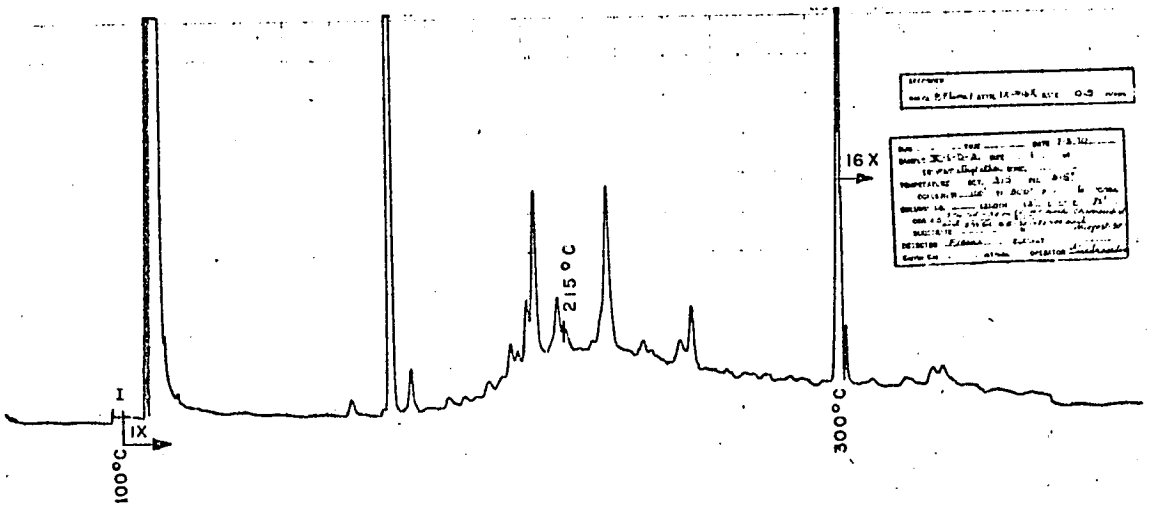
Injection of a sample of Fraction III-6-D-2 (Table XVII and Figure 5) on a silicone column showed presence of four major bands. The strongest peak appeared at approximately 300° C.

3. Sample: Fraction IX-6-D-2 (Gas Chromatogram No. 3)
Column: Dexsil (6' x 1/8")

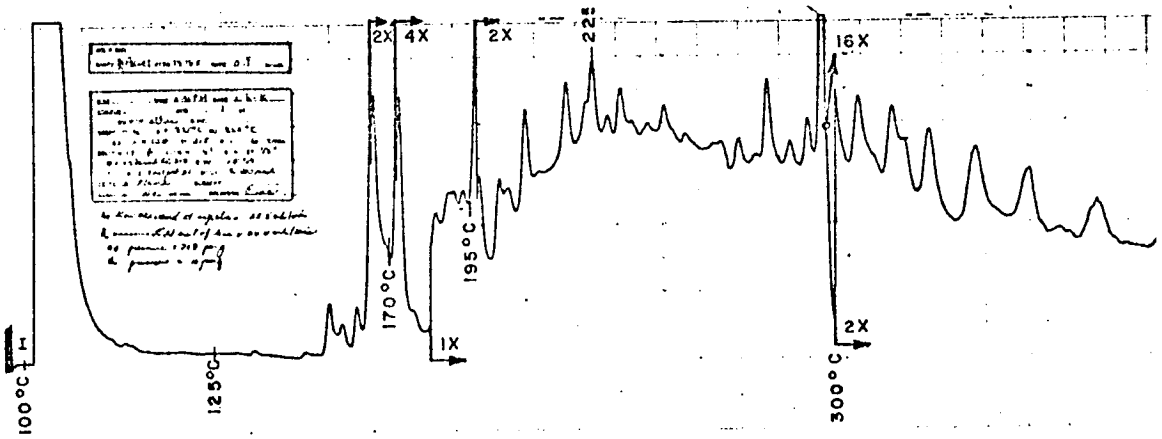
Gas Chromatogram No. 3 gave a series of distinct peaks; the strongest peak appeared near 300° C. Fraction IX-6-D-2 was the methanol eluate obtained after twice subjecting the methyl esters of Fraction 6-D-2 to urea chromatography (Table XVII and Figure 5).

4. Sample: Benzene eluate from silica-gel chromatography on Fraction 6-D-5 (Gas Chromatogram No. 4)
Column: Dexsil (6' x 1/8")

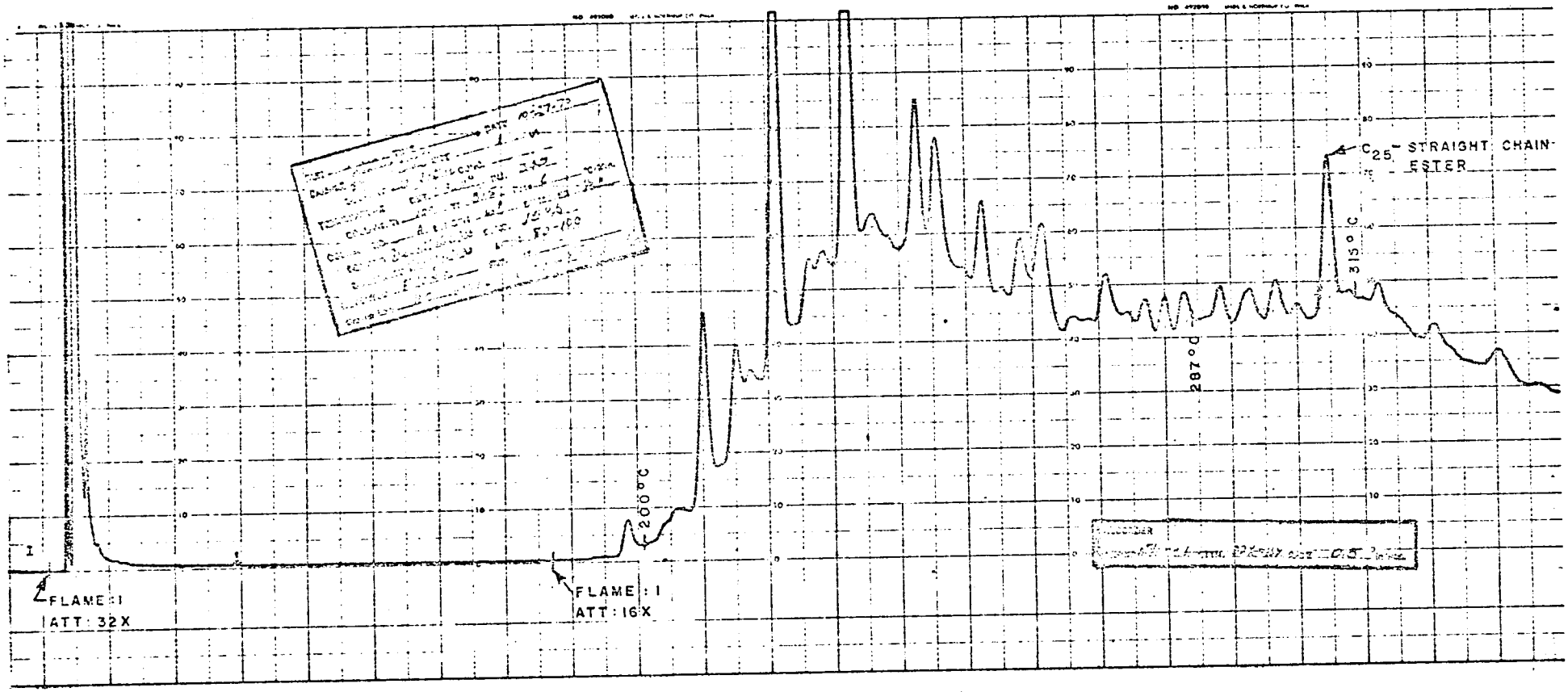
Gas Chromatogram No. 4 of this benzene eluate revealed the complexity of this fraction. This fraction should contain carboxylic acids only because it is part of Fraction D before esterification (Figure 5). The GC spectrum, however, did not show the characteristic tailing of acid peaks indicating the predominance of nonacidic components. A large peak appeared at the expected retention time for dioctyl phthalate or for a C_{25}-straight chain ester.



GAS CHROMATOGRAM NO. 2
FRACTION III-6-D-2



GAS CHROMATOGRAM NO. 3
FRACTION IX-6-D-2



GAS CHROMATOGRAM NO. 4

BENZENE ELUATE FROM SILICA-GEL

CHROMATOGRAPHY ON FRACTION 6-D-5

5. Sample: Pooled ether eluates resulting from first silica-gel chromatography on Fractions 6-D-5 and 6-D-6 (Gas Chromatogram No. 5)
Column: Dexsil (6' x 1/8")

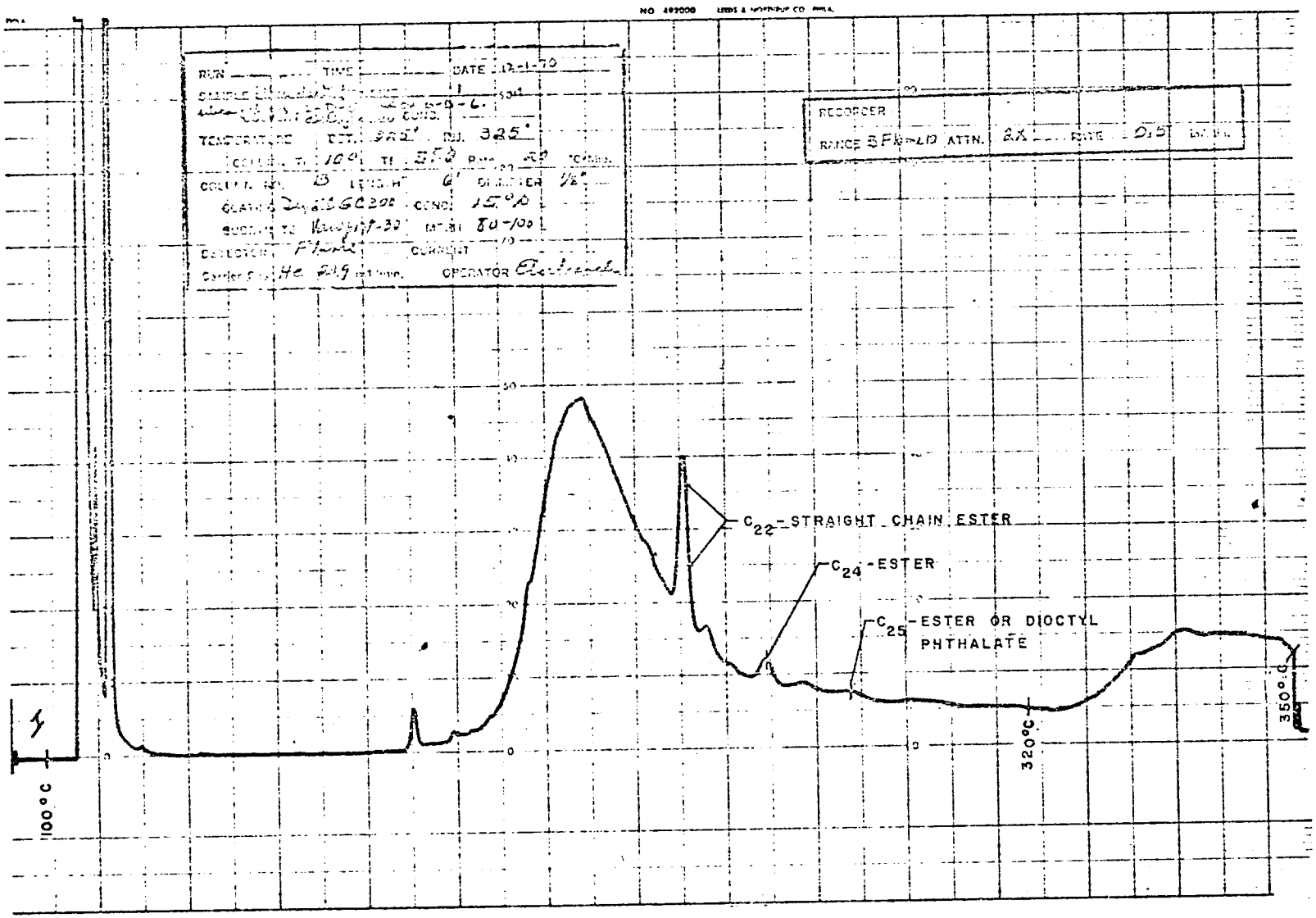
Gas Chromatogram No. 5 of these combined ether fractions gave a broad unresolved band, one distinct peak in the tracing projecting above the continuum. This outstanding peak had the same retention time as a n-C_{22} ester. No evidence of dioctyl phthalate could be detected in this spectrum.

6. Sample: Fraction III-6-D^+ (Gas Chromatogram No. 6)
Column: Dexsil (12' x 1/8")

Gas Chromatogram No. 6 of Fraction III-6-D^+ (Figure 5) exhibited a series of distinct peaks from C_{16}-C_{24}. Fraction III-6-D^+, eluted with methanol during the first urea chromatography, is the result of urea adduction of esters from Fraction 6-D^+. Fraction III-6-D^+ was expected to consist of straight-chain methyl esters of carboxylic acids. (Note: The sharp peak is due to reignition of the detector flame.)

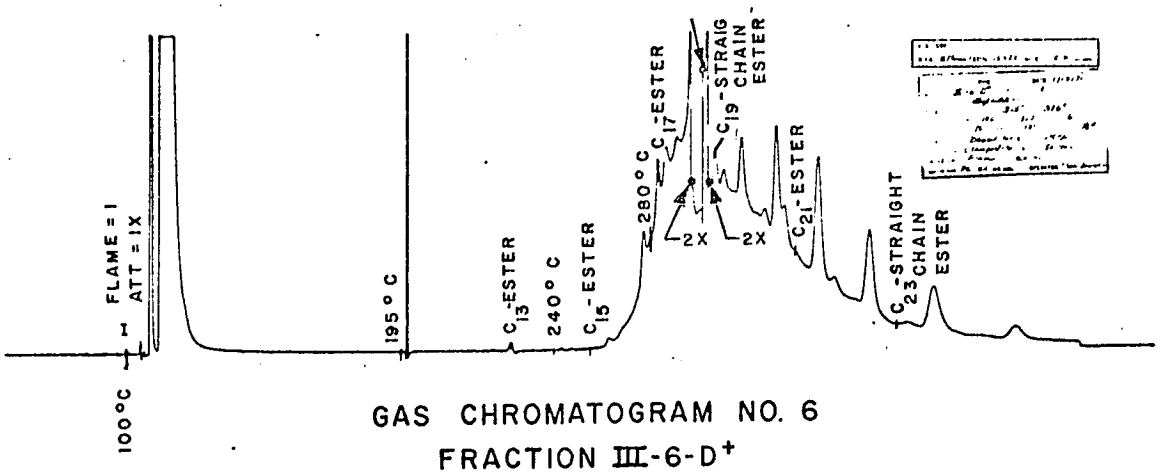
7. Sample: Fraction IV-6-D^+ (Gas Chromatogram No. 7)
Column: Dexsil (12' x 1/8")

Gas Chromatogram No. 7 of Fraction IV-6-D^+ presented a series of broad ill-defined peaks above C_{17}; no clean, narrow, distinct peaks were observed. (Note: The two sharp peaks are due to reignition of the flame.)

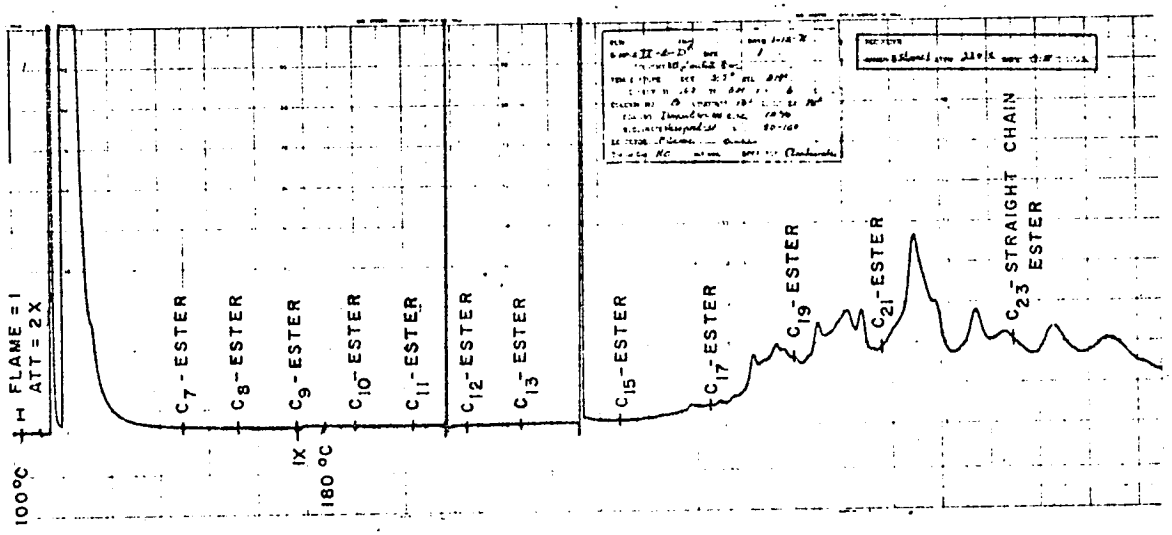


GAS CHROMATOGRAM NO. 5

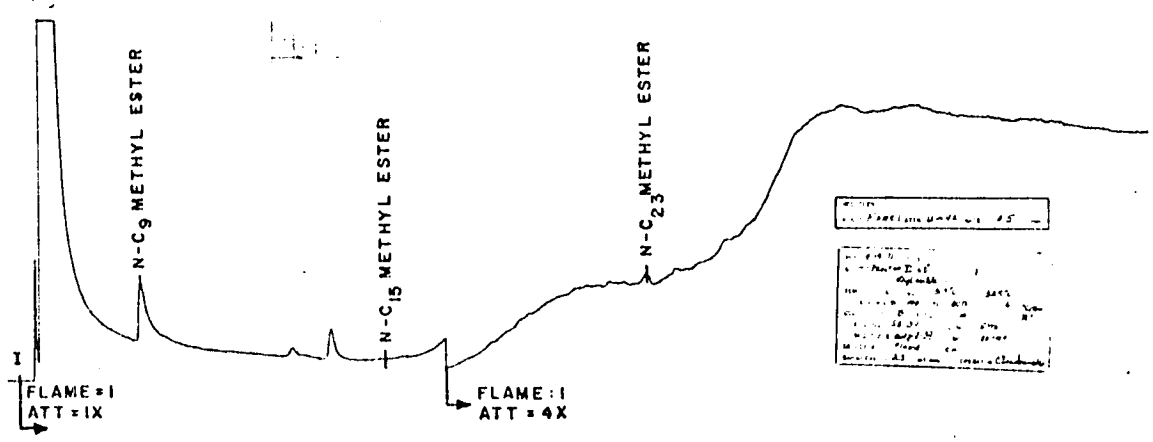
POOLED ETHER ELUATES RESULTING FROM
FIRST SILICA-GEL CHROMATOGRAPHY ON 6-D-5 AND 6-D-6



GAS CHROMATOGRAM NO. 6
FRACTION III-6-D+



GAS CHROMATOGRAM NO. 7
FRACTION IV-6-D+



GAS CHROMATOGRAM NO. 8
FRACTION VI-6-D+

8. Sample: Fraction VI-6-D^+ (Gas Chromatogram No. 8)
Column: SE-30 (10' x 1/8")

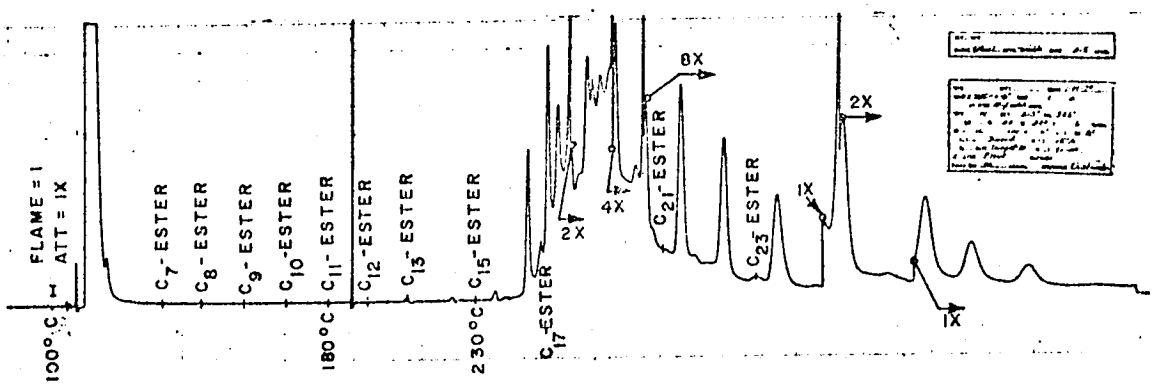
Gas chromatography of Fraction VI-6-D^+ consisted for the most part of a broad unresolved band, indicating a very complex mixture. However, there are three, very small but distinct, peaks at retention times equal to those of n-C_9 methyl ester, n-C_{12} methyl ester and n-C_{14} methyl ester. Fraction VI-6-D^+ consisted of that part of Fraction I-6-D^+ that is insoluble in hexane but soluble in benzene (Figure 5).

9. Sample: Fraction VIII-6-D^+ (Gas Chromatogram No.9)
Column: Dexsil (6' x 1/8")

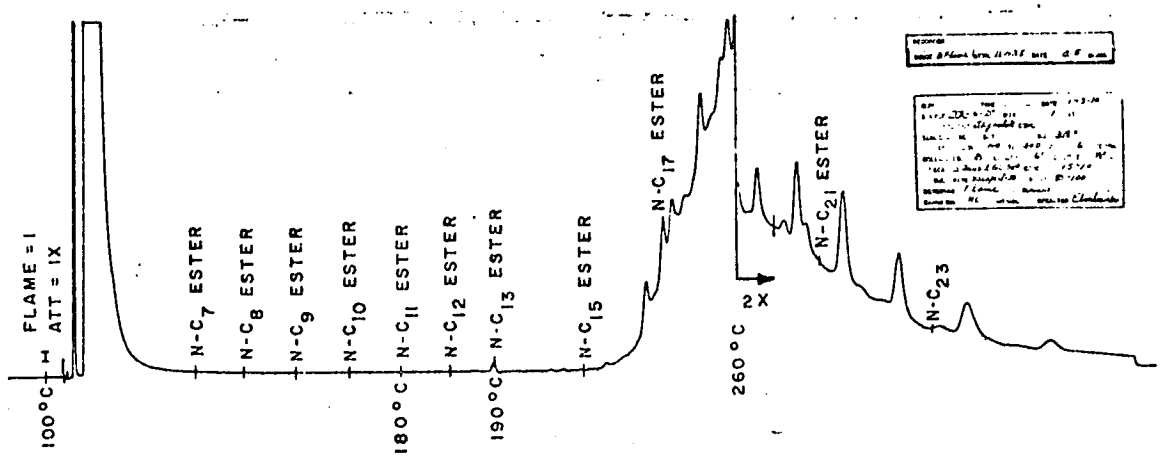
Gas Chromatogram No. 9, of Fraction VIII-6-D^+ (Figure 5), showed clear and distinct peaks from C_{16}-C_{28}. However, the peaks do not coincide with those from a mixture of standard straight-chain methyl esters. Fraction VIII-6-D^+ had been expected to contain a mixture of non-adducted (by urea) and some straight-chain esters. Based on the gas chromatographic data, no straight-chain esters were present.

10. Sample: Fraction IX-6-D^+ (Gas Chromatogram No. 10)
Column: Dexsil (6' x 1/8")

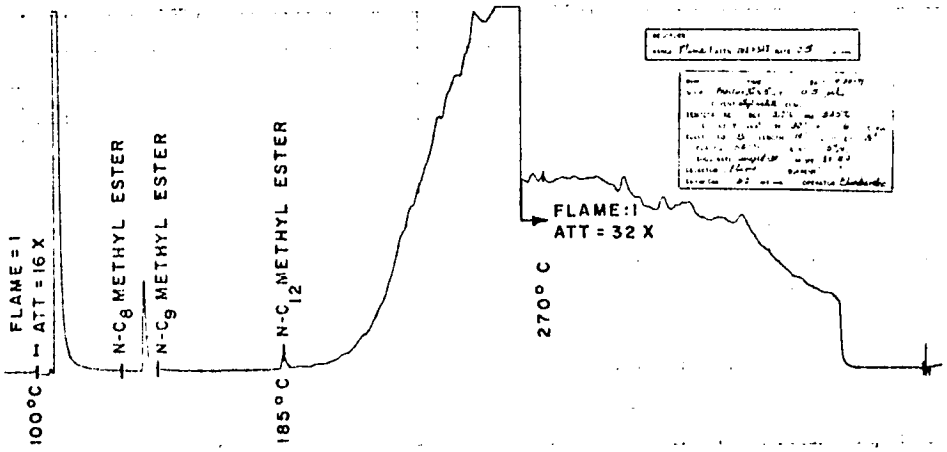
Gas Chromatogram No. 10, of Fraction IX-6-D^+, exhibited a series of distinct bands in the region representing esters with more than sixteen carbons.



GAS CHROMATOGRAM NO. 9
FRACTION VIII-6-D+



GAS CHROMATOGRAM NO. 10
FRACTION IX-6-D+



GAS CHROMATOGRAM NO. 11
FRACTION XI-6-D+

11. Sample: Fraction XI-6-D^+ (Gas Chromatogram No. 11)
Column: SE-30 (10' x 1/8")

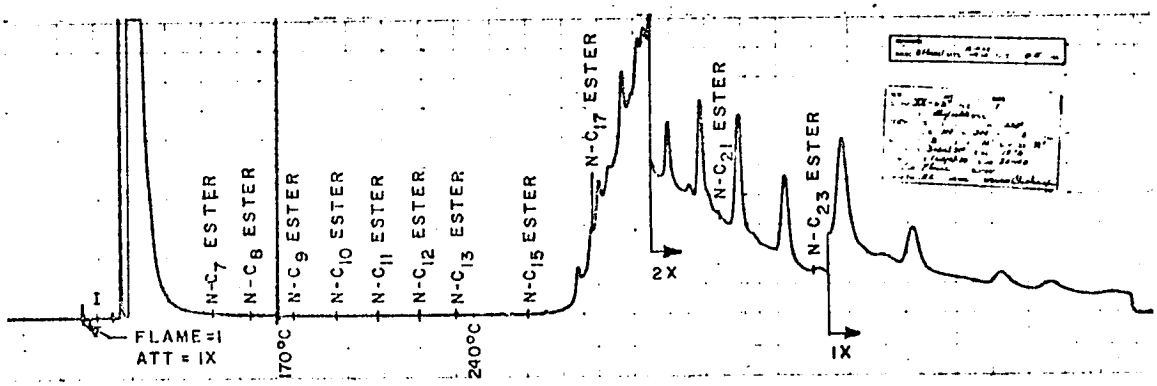
The complexity of Fraction XI-6-D^+ is illustrated by gas chromatography (Gas Chromatogram No. 11) of this fraction. The chromatogram consisted for the most part of a broad unresolved band with a small peak appearing between the retention time of a n-C_8 methyl ester and that of a n-C_9 methyl ester. Another small peak appears at the retention time for a n-C_{12} methyl ester.

12. Sample: Fraction XII-6-D^+ (Gas Chromatogram No. 12)
Column: Dexsil (12' x 1/8")

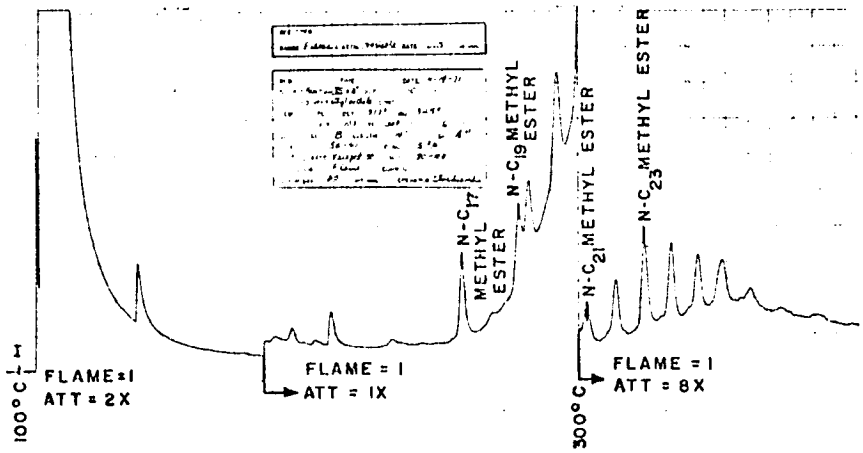
The major peaks in the Gas Chromatogram No. 12, of Fraction XII-6-D^+ (Figure 5), emerged from C_{12} to C_{25}. The peak that would correspond to a n-C_{13} ester is very small. For the most part, the peaks from Fraction XII-6-D^+ do not correspond to the peaks from n-methyl esters. Consequently, it is concluded that most of the compounds in Fraction XII-6-D^+ are not straight-chain methyl esters. This finding is not surprising since the previous two urea chromatographies had probably isolated all the straight-chain esters present (Figure 5).

13. Sample: Fraction XIII-6-D^+ (Gas Chromatogram No. 13)
Column: SE-30 (10' x 1/8")

Gas Chromatogram No. 13, of Fraction XIII-6-D^+, showed a series of distinct peaks from C_{17}-C_{26}. There was no indication of a n-C_{13} ester peak and the peak that would correspond to a n-C_{14} ester was quite small.



GAS CHROMATOGRAM NO. 12
FRACTION XII-6-D+



GAS CHROMATOGRAM NO. 13
FRACTION XIII-6-D+

14. Sample: Fraction XIV-6-D^+ (Gas Chromatogram No. 14)
Column: SE-30 (10' x 1/8")

Gas Chromatogram No. 14, of Fraction XIV-6-D^+, showed no peaks on the lower molecular weight region of the chromatogram. A series of distinct peaks appeared from C_{19}-C_{26}. There were no signs of a n-C_{13} ester peak. Fraction XIV-6-D^+ (Figure 5) was expected to contain higher molecular weight branched esters.

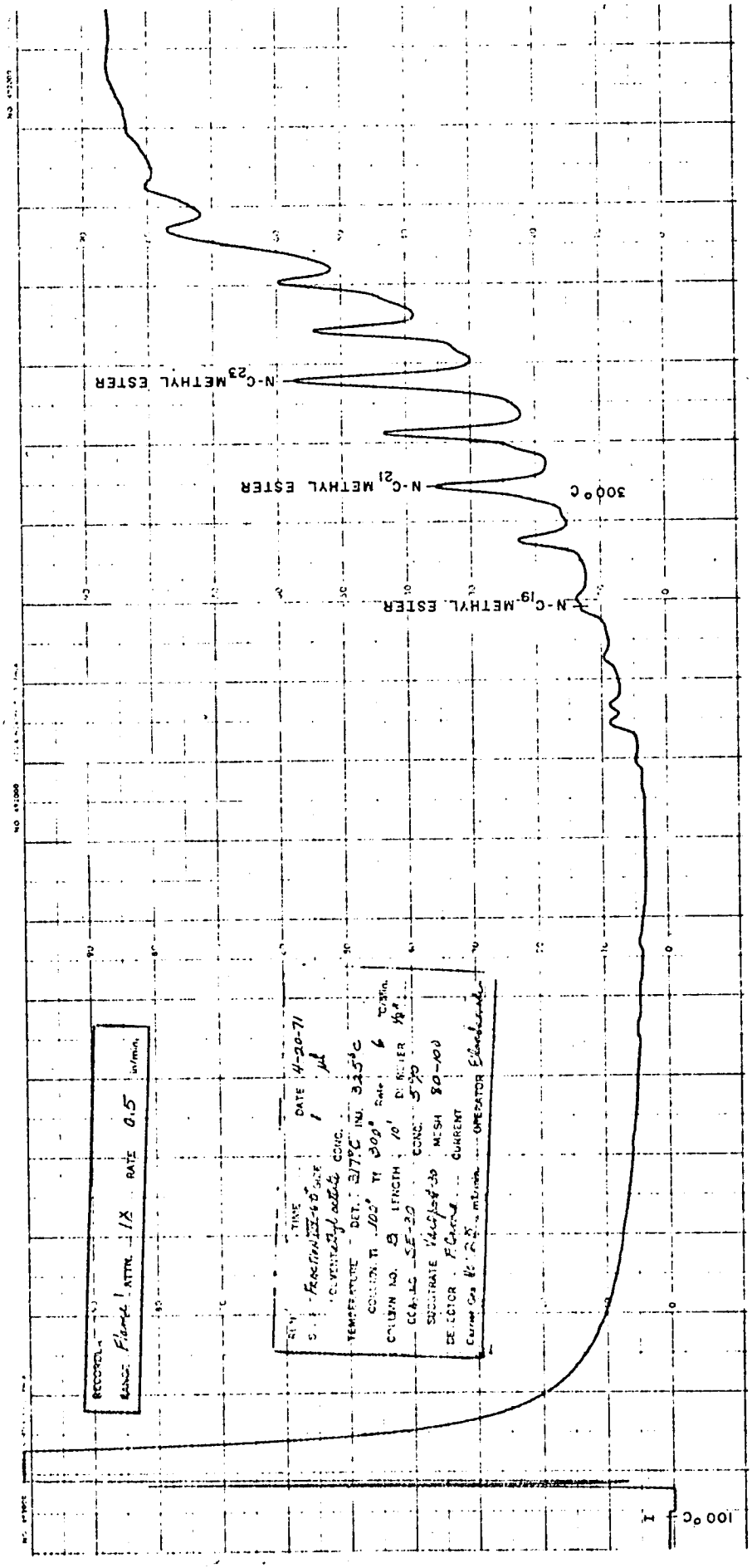
15. Sample: Fraction XVI-6-D^+ (Gas Chromatogram No. 15)
Column: SE-30 (10' x 1/8")

Gas Chromatogram No. 15, of Fraction XVI-6-D^+, featured for the most part a broad unresolved band. It also showed about four peaks from C_9-C_{13}. This fraction consisted of the unclathrated material remaining after thiourea adduction with an inducer of Fraction XI-6-D^+. Gas Chromatogram No. 15 shows that Fraction XVI-6-D^+ consisted of a mixture too complex to be resolved directly by gas chromatography.

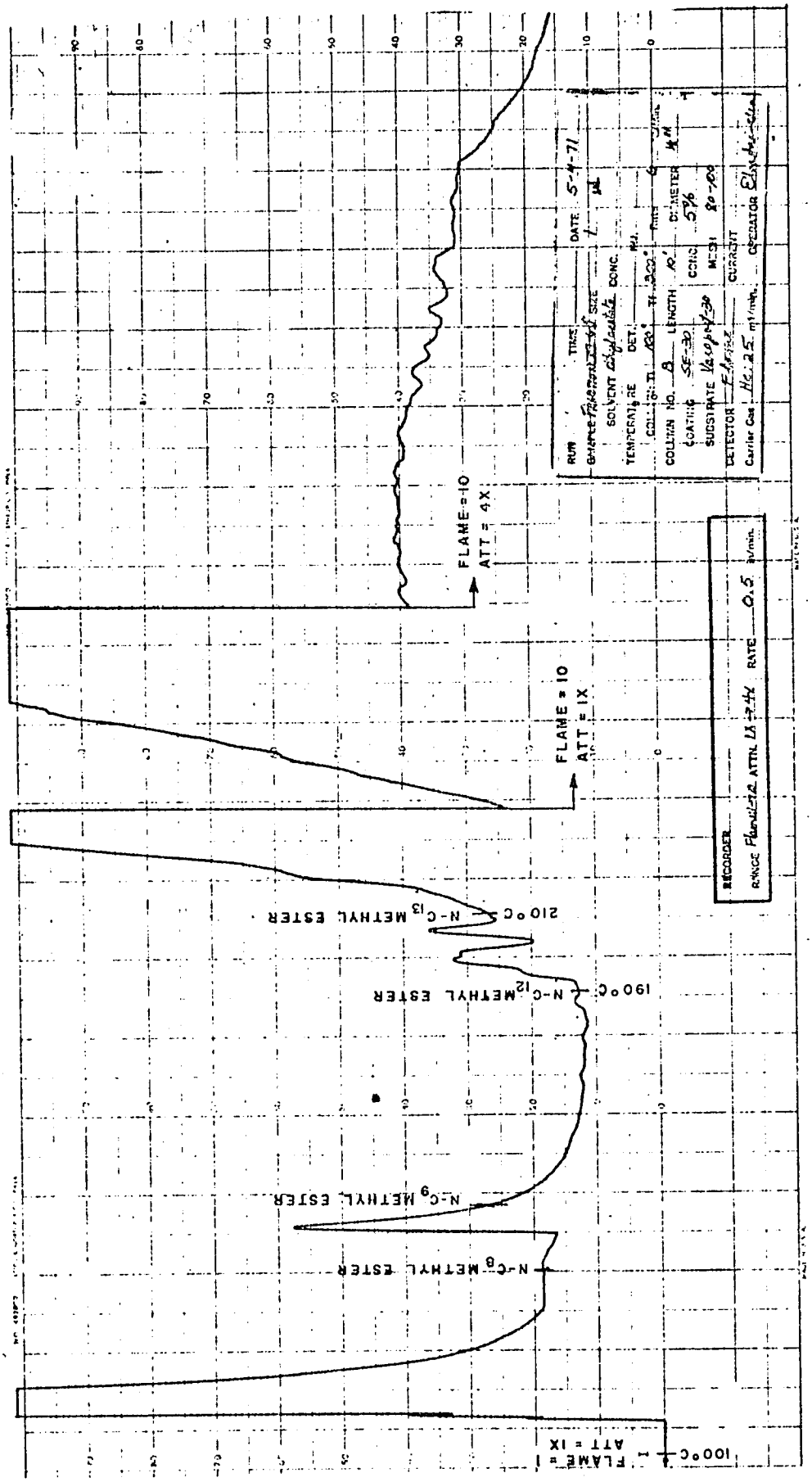
16. Sample: Methyl esters of Fraction 6-D^+
Column: SE-30 plus GC SE-30 (12' x 1/8")

The gas chromatogram of the methyl esters of Fraction 6-D^+ showed a broad band extending over most of the spectrum with one distinct peak projecting above the continuum. This chromatogram indicates that the methyl esters of Fraction 6-D^+ constitute a mixture too complex to be resolved by gas chromatography.

No evidence of dioctyl phthalate was observed in this chromatogram.



GAS CHROMATOGRAM NO. 14
FRACTION XIV-6-D+



GAS CHROMATOGRAM NO. 15
FRACTION XVI-6-D+

17. Sample: Fraction VII-6-D^+
Column: SE-30 plus GC SE-30 (12' x 1/8")

Gas chromatography of Fraction VII-6-D^+ gave a continuum, with two peaks projecting in the region around 180° C. Fraction VII-6-D^+ is the hexane fraction after the second urea chromatography (Figure 5). Its gas chromatogram clearly indicates that this nonadducted material is still an extremely complex mixture.

18. Sample: Fraction X-6-D^+
Column: SE-30 plus GC SE-30 (12' x 1/8")

A series of distinct peaks were observed in the gas chromatogram of this fraction.

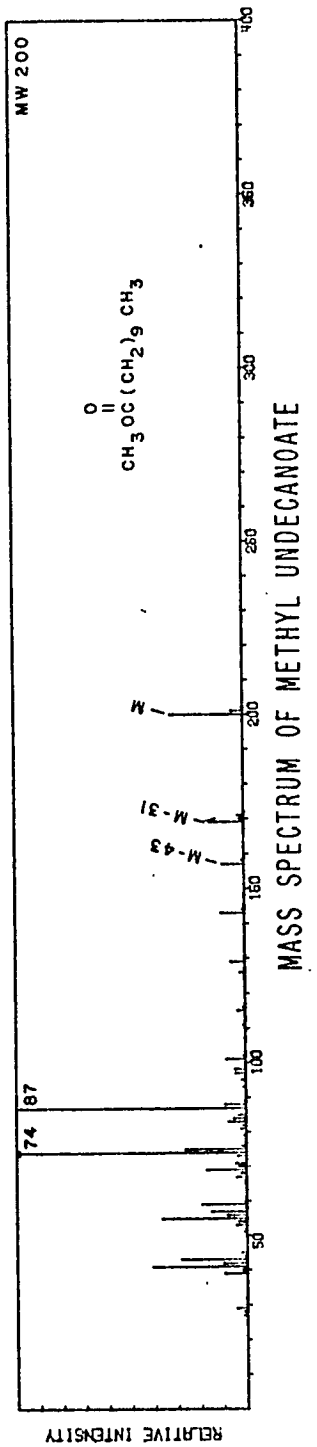
This ends the GC analysis of those fractions which were not selected for collection. The selection was based on the resolution of the fractions by gas chromatography and on whether the peaks were strong enough to justify collection. Identification of any of the components solely on the basis of gas chromatography was not attempted. In situations in which the possible structures of the unknowns being analyzed are infinite, it is not practical to identify compounds based solely on their GC retention times. Too many compounds have identical retention times; this could easily lead to erroneous identification.

3. Infrared (IR) and Mass Spectrometric (MS) Studies of Known Compounds

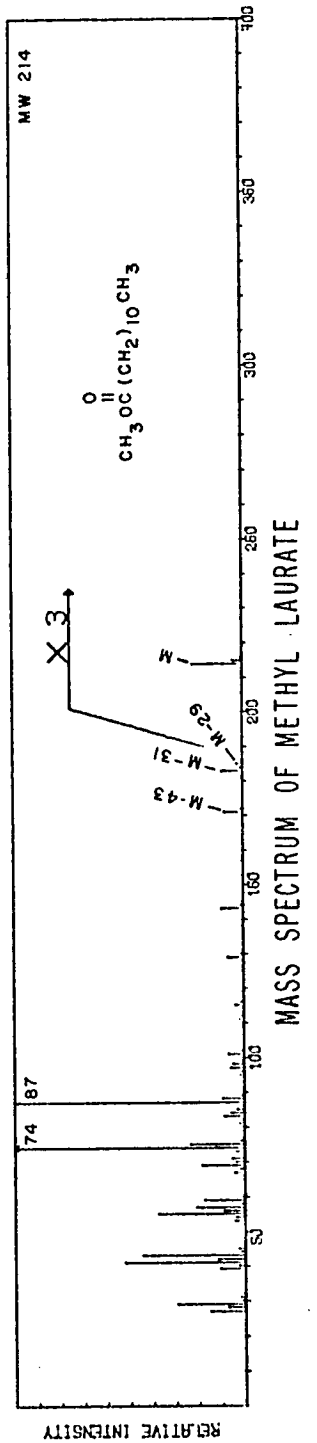
Before proceeding to identify esters and acids isolated from the heavy oils, a study was conducted of known compounds to acquire background information.

Straight-Chain Methyl Esters of Carboxylic Acids - Low resolution mass spectra of six methyl esters of saturated normal chain carboxylic acids were obtained on the analytical mass spectrometer Model 21-110B HRMS (CEC) at Rice University. The compounds analyzed were methyl undecanoate (n-C_{12} methyl ester); methyl laurate (n-C_{13} methyl ester); methyl tetradecanoate (n-C_{15} methyl ester); methyl palmitate (n-C_{17} methyl ester); methyl stearate (n-C_{19} methyl ester) and methyl behenate (n-C_{23} methyl ester). These straight-chain esters were analytical standards supplied by Poly Science Corporation. The spectral data are presented in Table XX.

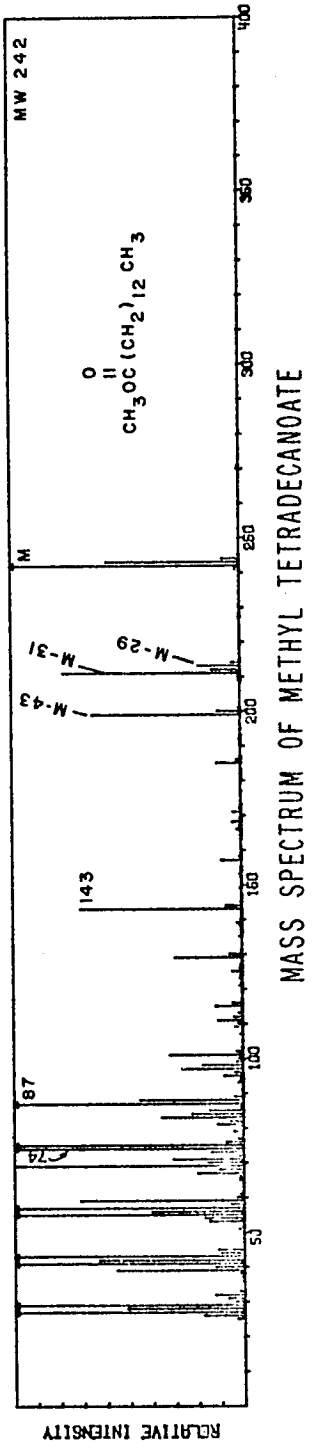
A molecular ion is usually observed for the straight-chain methyl esters. The base peak of the spectrum for the six straight-chain methyl esters was at m/e = 74. This peak is due to a rearranged ion of formula $-CH_3COOCH_3$. A moderately strong peak is present at m/e = 87 and it is due to the normal methoxycarbonyl type fragment $CH_3OOCCH_2CH_2^+$. In the higher mass range the highest peak is due to the molecular ion (m/e = M). The next highest peak observed in the high mass range of these esters is the ion of M-43, which is formed by a process involving



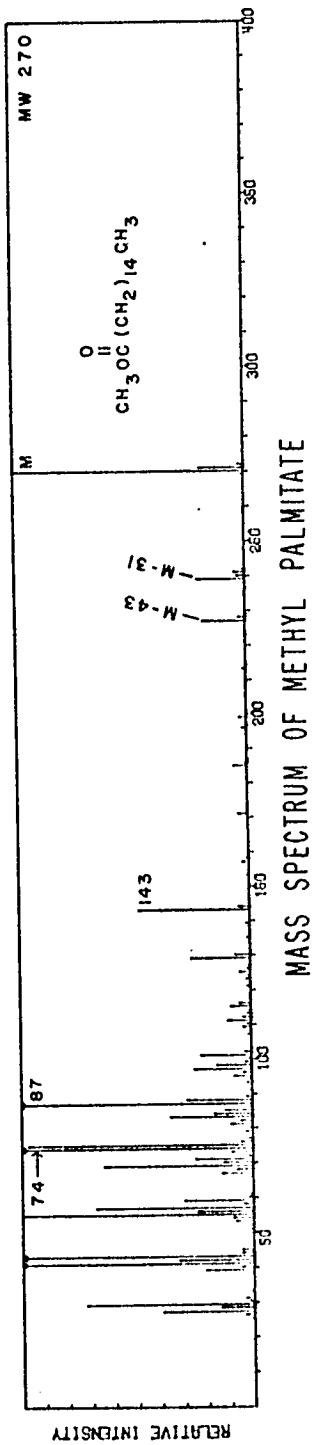
MASS SPECTRUM OF METHYL UNDECANOATE



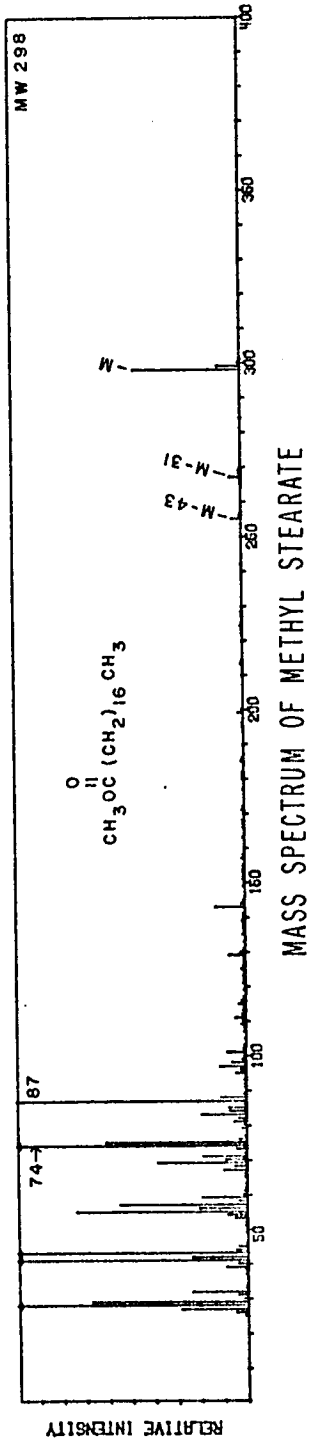
MASS SPECTRUM OF METHYL LAURATE



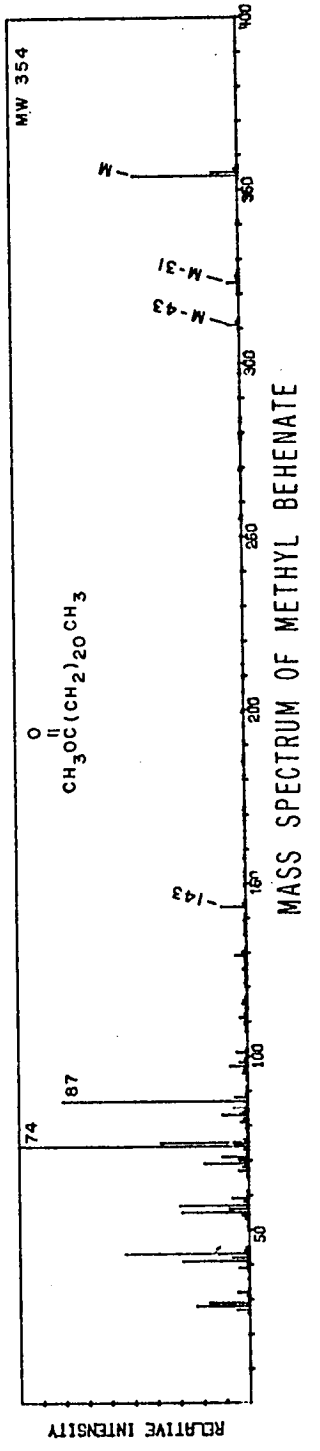
MASS SPECTRUM OF METHYL TETRADECANOATE



MASS SPECTRUM OF METHYL PALMITATE



MASS SPECTRUM OF METHYL STEARATE



MASS SPECTRUM OF METHYL BEHENATE

TABLE XX

MASS SPECTRAL DATA*

Mass Spectrum Identification No.	m/e Pk 1	I	m/e Pk 2	I	m/e Pk 3	I	m/e Pk 4	I	m/e Pk 5	I	m/e Pk 6	I
Methyl Undecanoate	74	OS**	87	100	200	32	75	27	169	15	143	11
Methyl Laurate	74	OS	87	100	75	24	143	10	88	10	183	9
Methyl Tetra-decanoate	74	OS	75	OS	87	OS	242	OS	69	100	211	78
Methyl Palmitate	74	OS	87	OS	270	100	75	97	143	48	83	36
Methyl Stearate	74	OS	87	100	75	61	298	47	83	19	143	13
Methyl Behenate	74	100	87	81	354	46	75	38	355	12	83	11
Methyl Cyclopentyl Propanoate	74	100	87	72	83	49	75	26	82	15	88	11
Methyl 2-cyclo-pentyl-n-hexanoate	87	100	130	48	142	34	141	14	101	10	113	10
4	57	100	93	65	91	23	67	22	77	20	108	16
8	69	100	80	52	119	28	131	22	91	16	77	13
9	149	100	167	23	150	13	279	10	113	9	104	7
83	71	100	69	90	85	57	81	54	83	47	95	43
84	81	100	95	92	83	67	151	67	109	63	69	58

TABLE XX (Cont'd.)

Mass Spectrum Identification No.	m/e Pk 1	I	m/e Pk 2	I	m/e Pk 3	I	m/e Pk 4	I	m/e Pk 5	I	m/e Pk 6	I
72-1	220	100	177	69	136	61	205	54	165	46	180	43
72-2	205	100	220	44	206	17	221	9	145	4	177	3
73	149	100	177	49	176	19	150	17	178	12	222	12
75-1	57	100	71	66	85	38	69	26	83	20	70	18
75-3	57	100	71	63	58	51	85	34	59	28	69	26
75-4	74	100	87	58	75	18	270	11	143	11	83	7
77-2	57	100	71	65	85	36	69	31	95	24	81	21
77-5	73	100	147	43	207	40	355	25	221	24	281	23
79-6	149	100	69	21	83	20	73	19	97	14	150	13
80-3	71	OS	69	197	83	158	99	103	81	97	97	91
81-2	69	OS	71	OS	85	OS	83	100	70	84	99	79
82-5	57	OS	71	OS	83	OS	85	OS	69	100	99	96
83-1	69	100	71	83	81	74	95	68	83	65	85	58
83-2	71	OS	85	OS	69	100	83	63	70	52	81	48
83-3	129	100	143	80	69	75	91	75	128	75	149	65

TABLE XX (Cont'd.)

Mass Spectrum Identification No.	m/e Pk 1	I	m/e Pk 2	I	m/e Pk 3	I	m/e Pk 4	I	m/e Pk 5	I	m/e Pk 6	I
84-1	69	100	81	83	95	72	83	61	97	53	85	50
85-4	69	100	71	88	81	72	95	69	83	63	85	56
88-3	71	OS	85	OS	69	100	83	70	97	69	99	62
131	74	100	87	70	85	38	81	25	83	24	77	24
132	69	100	81	74	95	61	83	60	135	54	77	49
135	74	100	81	84	95	79	87	54	83	51	109	50
136	69	100	81	82	95	69	71	67	74	60	83	51
137	69	100	71	85	81	77	95	64	83	58	85	51
138	69	100	71	84	81	61	83	54	95	53	85	52
139	69	100	71	92	81	64	95	58	83	52	85	52
140	69	100	71	80	81	63	83	60	95	57	85	51
141	69	100	71	100	81	69	83	63	95	60	85	51
169	41	100	43	79	55	76	74	44	57	43	69	35
173	55	100	57	72	69	49	81	45	67	40	95	39

*Peaks below m/e = 41 have not been included in this table.
**OS = Off scale.

removal of the methylene groups in positions 2, 3, and 4, together with one hydrogen atom, followed by recombination of the ester and hydrocarbon residues.[57] A fragment derived from the loss of a methoxy group (OCH_3) appears at M-31 as a constant feature of these straight-chain ester spectra.[48] Peaks due to oxygen-containing fragments of the general type

$$[CH_3 - O - \underset{\underset{O}{\|}}{C} - (CH_2)_n -]^+ \quad (n \geq 1)$$

are also characteristic of methyl esters.

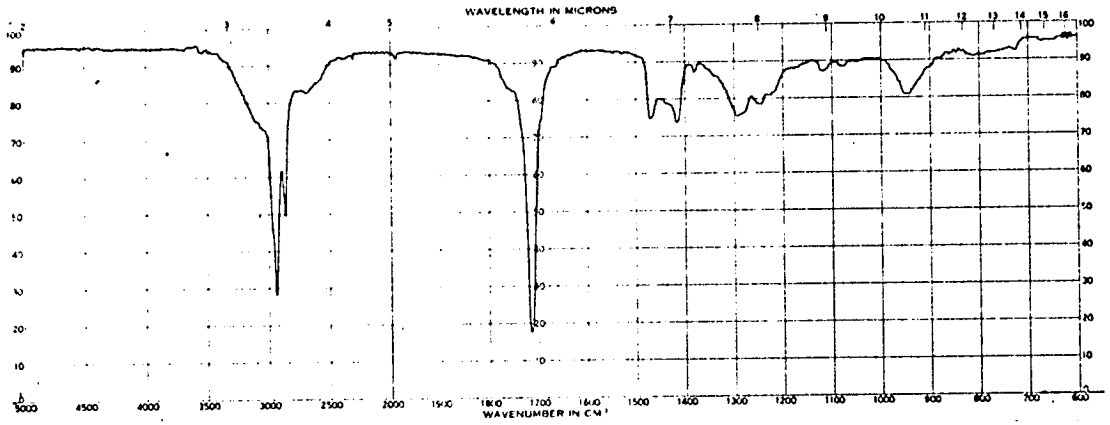
IR studies of various mixtures of undecanoic acid and methyl undecanoate were made to confirm the possibility of distinguishing the positions of individual carbonyl bands of acids and esters. The following samples were analyzed:

1. Undecanoic Acid (C_{11} acid)

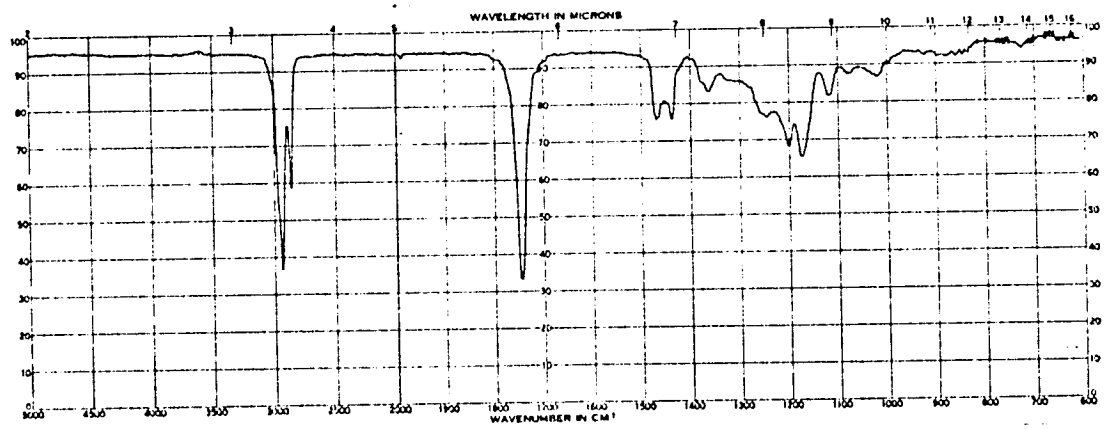
IR Spectrum No. 20 of standard undecanoic acid showed a carbonyl absorption of the acid dimer at 1715 cm^{-1} and a sloping shoulder from 3000-3500 cm^{-1}.

2. Methyl Undecanoate (n-C_{12} ester)

IR Spectrum No. 21 of standard methyl undecanoate showed an ester carbonyl band at 1745 cm^{-1} and C-H stretch at 2800-3000 cm^{-1}.



IR SPECTRUM NO. 20
UNDECANOIC ACID STANDARD



IR SPECTRUM NO. 21
METHYL UNDECANOATE STANDARD

3. Mixture of 1:1 Methyl Undecanoate:Undecanoic Acid

IR Spectrum No. 22 of the 1:1 mixture showed two carbonyl bands, one corresponding to the acid at 1715 cm^{-1} and one due to the ester at 1745 cm^{-1}. A sloping shoulder from 3000-3300 cm^{-1} is also observed for this mixture.

4. Mixture of 1:4 Methyl Undecanoate:Undecanoic Acid

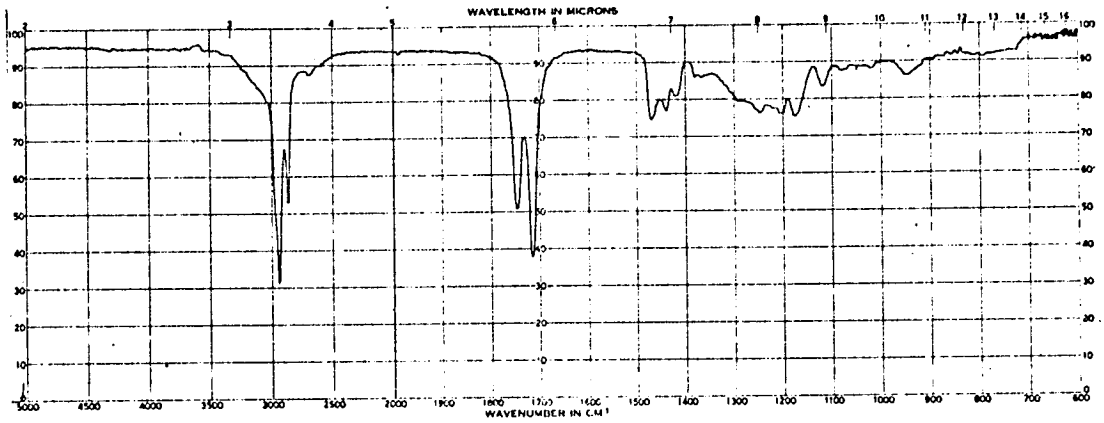
IR Spectrum No. 23 of this 1:4 mixture showed sloping shoulders from 3000-3300 cm^{-1}, a weak ester carbonyl band at 1745 cm^{-1} and a very strong acid carbonyl band at 1715 cm^{-1}.

5. Mixture of 4:1 Methyl Undecanoate:Undecanoic Acid

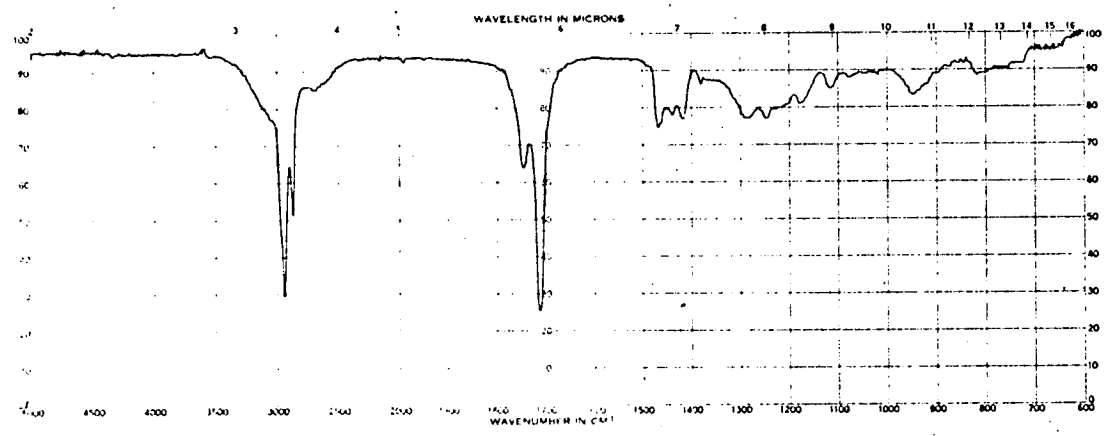
IR Spectrum No. 24 of this 4:1 mixture showed two distinct bands in the carbonyl region, one at 1715 cm^{-1} (due to acids) and one at 1745 cm^{-1} (due to esters).

These five IR spectra prove that in the case of mixtures of pure acids and esters one can observe very distinctly two bands in the carbonyl region of their IR spectra.

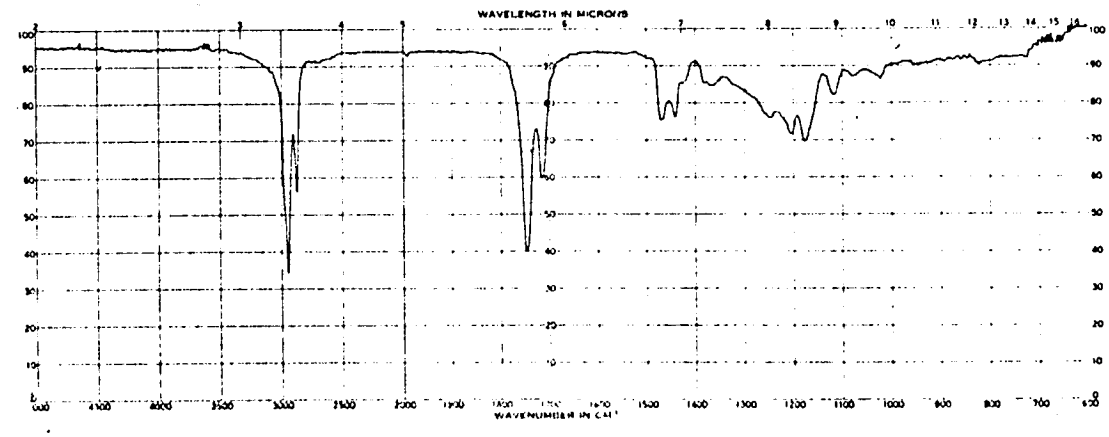
Aromatic Acids and Their Methyl Esters - Benzoic acid was used as a model compound to study esterification of aromatic acids. It was possible that Fraction D isolated from the heavy oils would contain aromatic acids. If the methylation reaction would not be effective on aromatic acids, the acids could undergo decarboxylation during their exposure to high temperatures in the GC and MS. This could be a factor in complicating the mass spectra. IR spectra of benzoic acid and of its methyl ester were obtained.



IR SPECTRUM NO. 22
1:1 METHYL UNDECANOATE - UNDECANOIC ACID



IR SPECTRUM NO. 23
1:4 METHYL UNDECANOATE - UNDECANOIC ACID



IR SPECTRUM NO. 24
4:1 METHYL UNDECANOATE - UNDECANOIC ACID

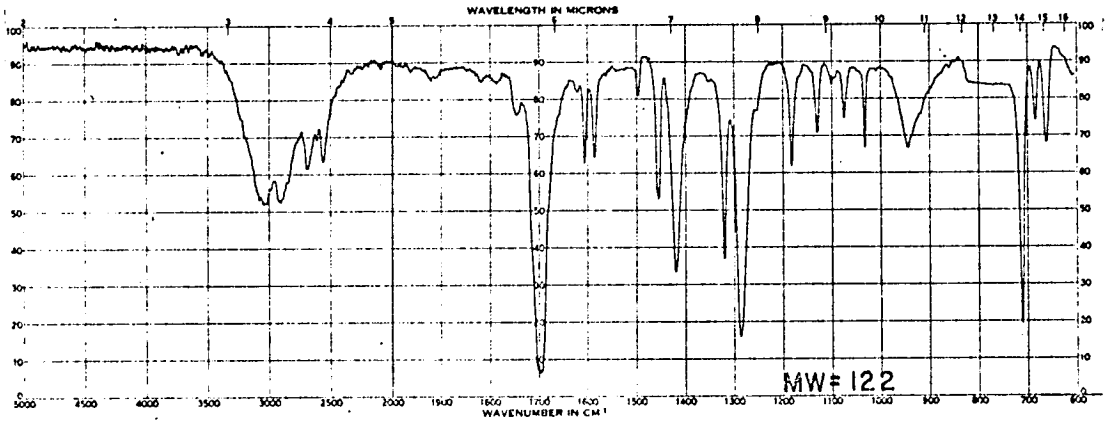
IR Spectrum No. 25 of benzoic acid (Primary Standard from Baker) showed a strong acid carbonyl band at 1695 cm^{-1} and broad absorption between 2500-3100 cm^{-1} indicative of aromaticity and of its acid character. Bands at 1285 cm^{-1}, 1320 cm^{-1}, 1420 cm^{-1} and 1455 cm^{-1} are characteristic of the IR spectrum of benzoic acid in CCl_4.[58]

Methyl benzoate was synthesized in our laboratories by methylating standard benzoic acid with BF_3-MeOH. IR Spectrum No. 26 of methyl benzoate showed an ester carbonyl band at 1730 cm^{-1}. In methylation, the carbonyl band has shifted from 1695 cm^{-1} to 1730 cm^{-1}. There is a series of bands from 2800-3100 cm^{-1}, indicative of aromaticity.

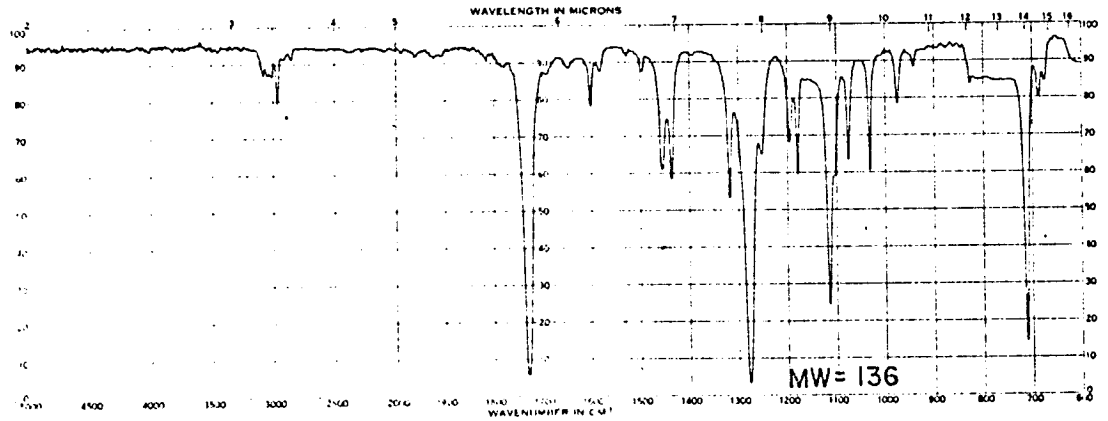
These IR spectra confirmed that the boron trifluoride methylation technique works for aromatic acids.

Saturated Cyclic Acids and Their Methyl Esters - IR spectra of two saturated cyclic acids and of their corresponding methyl esters were recorded. The starting materials used were: cyclopentylpropionic acid, manufactured by Chemical Procurement Labs and 2-cyclopentyl-n-hexanoic acid, manufactured by Aldrich Chemical Co. Both of these chemicals were very pure. The methyl esters were prepared in our laboratories from the acids by means of fresh methanol/boron trifluoride reagent.

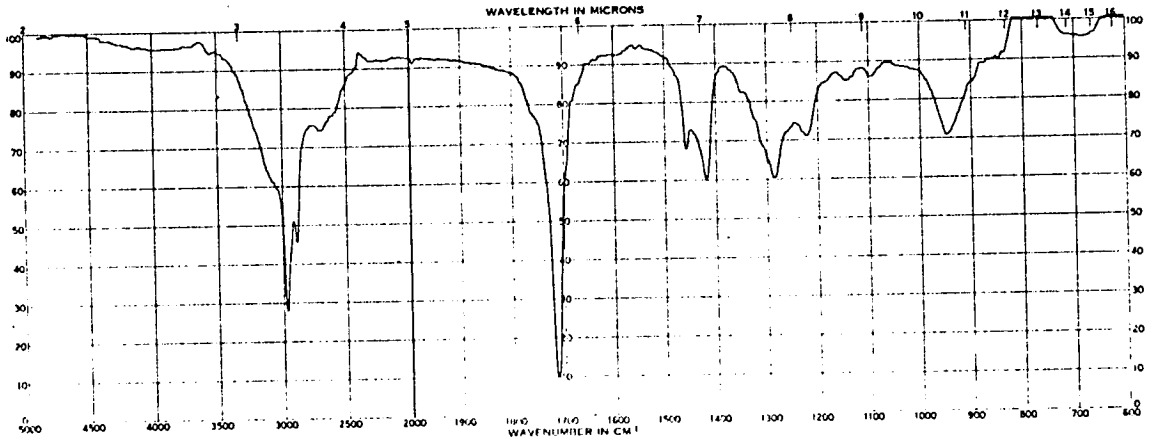
IR Spectrum No. 27 of cyclopentylpropionic acid showed a carbonyl band at 1715 cm^{-1}. IR Spectrum No. 28 of methyl cyclopentylpropanoate showed the shift of the carbonyl band, appearing now at 1750 cm^{-1}.



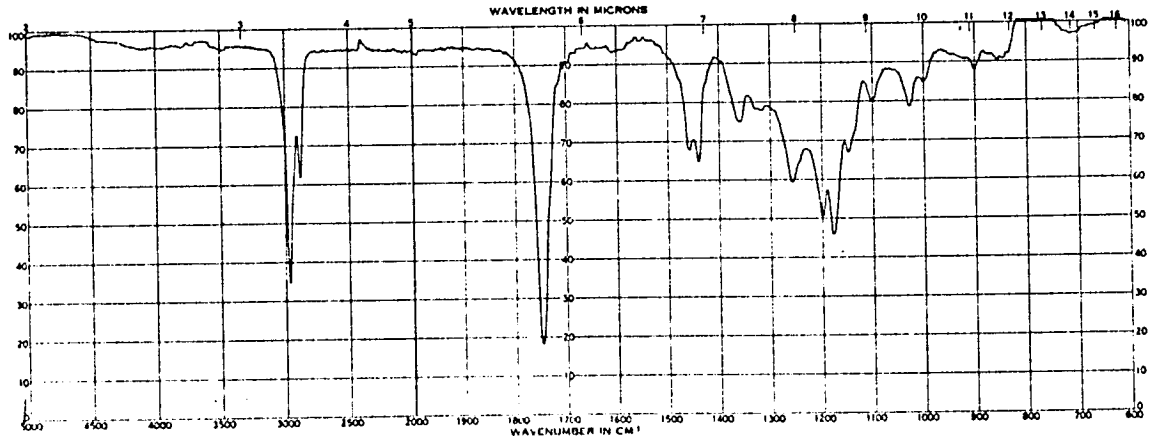
IR SPECTRUM NO. 25
BENZOIC ACID STANDARD



IR SPECTRUM NO. 26
METHYL BENZOATE



IR SPECTRUM NO. 27
CYCLOPENTYLPROPIONIC ACID



IR SPECTRUM NO. 28
METHYL ESTER OF CYCLOPENTYLPROPIONIC ACID

IR Spectrum No. 29 of 2-cyclopentyl-n-hexanoic acid showed a band at 1715 cm^{-1} as expected. In the IR Spectrum No. 30 of methyl 2-cyclopentyl-n-hexanoate, a very intense band appeared at 1745 cm^{-1}.

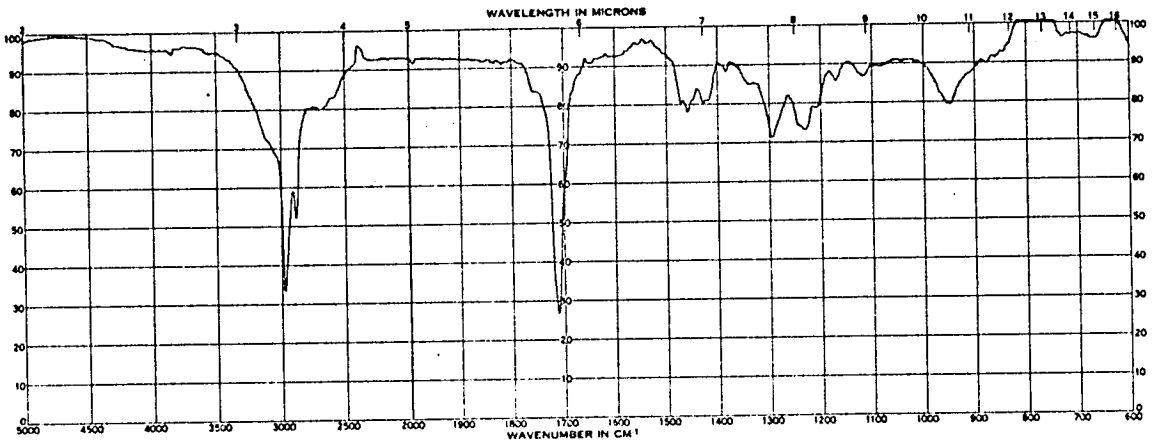
In addition, the methyl esters of the two cyclic acids, i.e. methylcyclopentylpropanoate and methyl 2-cyclopentyl-n-hexanoate, were analyzed by low resolution mass spectroscopy on a CEC Model 21-110B HRMS Mass Spectrometer. The mass spectra of these two cyclic esters have not been described in the literature to the best of my knowledge.

The mass spectrum of methylcyclopentylpropanoate shows a base peak at m/e = 74 and a molecular ion (M) at m/e = 156. Other peaks of interest appear at m/e = 113 (M-43) and at m/e = 125 (M-31).

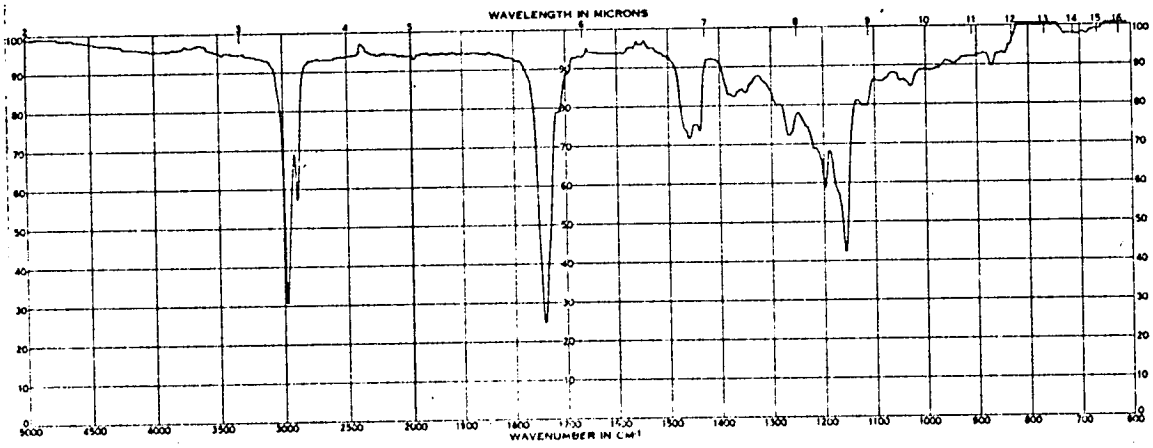
The mass spectrum of methyl 2-cyclopentyl-n-hexanoate has m/e = 87 as its base peak and shows its molecular ion (M) at m/e = 198. Other outstanding peaks appear at: m/e = 142 (M-56); m/e = 155 (M-43); m/e = 167 (M-31); and m/e = 169 (M-29).

The six strongest peaks in the mass spectrum of these two cyclic esters are listed in Table XX.

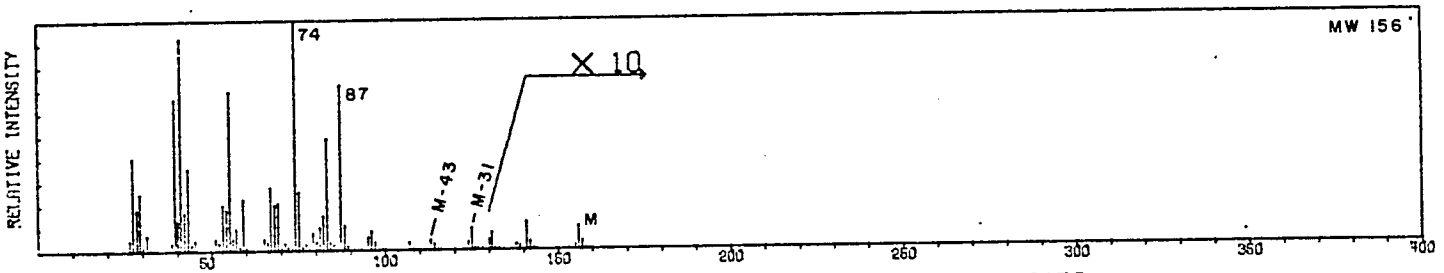
The two cyclic esters of known structure that we analyzed are of fairly simple structure. The cyclopentyl esters that we identified from petroleum have MW ranging from 240 to 394. Consequently, the molecules are much larger and complex, and the fragmentation pattern quite



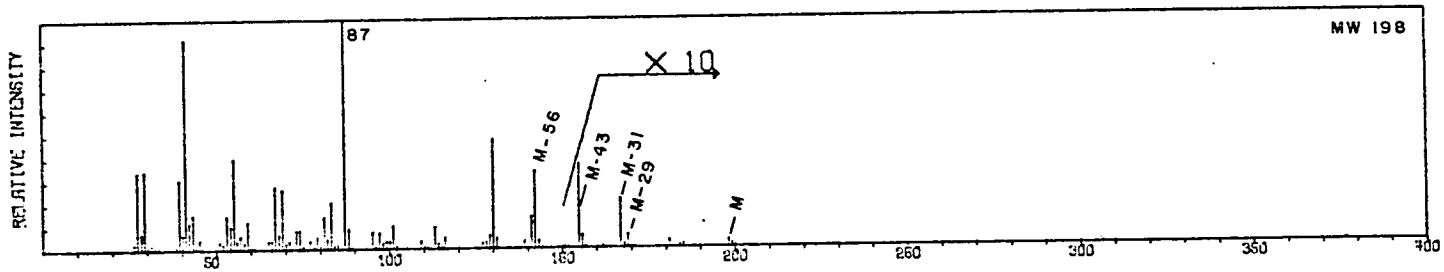
IR SPECTRUM NO. 29
2-CYCLOPENTYL-n-HEXANOIC ACID



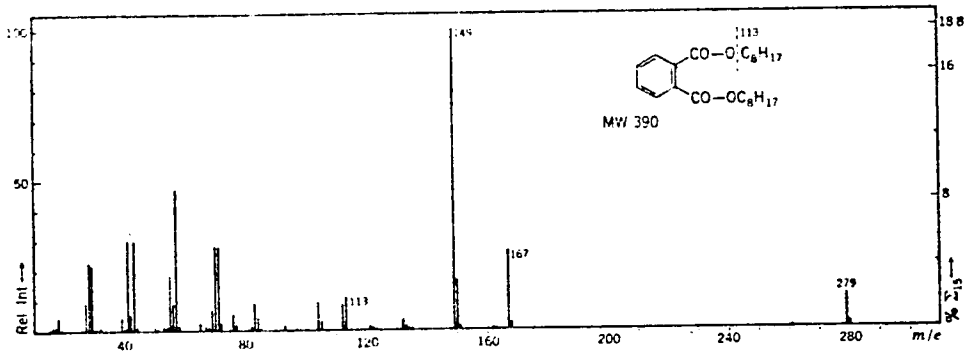
IR SPECTRUM NO. 30
METHYL ESTER OF 2-CYCLOPENTYL-n-HEXANOIC ACID



MASS SPECTRUM OF METHYL CYCLOPENTYLPROPANOATE

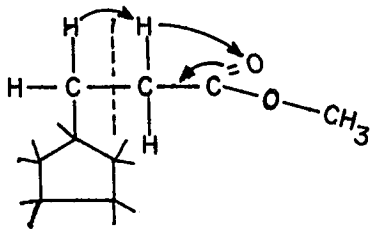


MASS SPECTRUM OF METHYL 2-CYCLOPENTYL-N-HEXANOATE

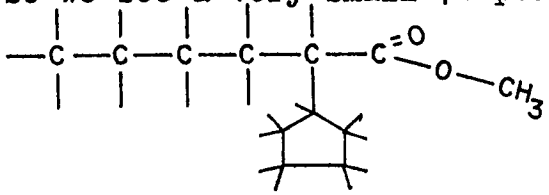


MASS SPECTRUM OF DIOCTYLPHTHALATE

different from the simple standards available. In the case of methylcyclopentylpropanoate we see 74 as the base peak because the structure allows this type of rearranged ion:

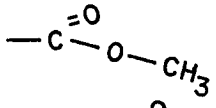
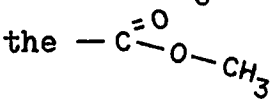


In the case of methyl 2-cyclopentyl-n-hexanoate, the cyclopentyl ring is too close to the methyl ester to allow such rearrangement, so we see a very small 74 peak:



High Molecular Weight Cyclopentyl Esters - These high molecular weight esters usually exhibit a series of peaks at m/e = 69, 83, 97, 111, etc., corresponding to the formula C_nH_{2n-1}; this series is characteristic of naphthenic and/or olefinic fragments. However, a very large m/e = 69 ($C_5H_9^+$) peak strongly indicates a cyclopentyl ring.

A series of peaks at m/e = 57, 71, 85, etc., corresponding to C_nH_{2n+1} is also usually observed due to fragmentation of the hydrocarbon tail of the high molecular weight cyclopentyl esters.

A peak at m/e = 74 is characteristic of the cyclopentyl methyl esters, if the ring is not close to the  group. However, if the ring is adjacent to the  , the 74 peak is usually small. The rearrangement peak at 74 is due to the $-CH_2\ COOCH_3$ component of the structure.

A M-15 peak can be expected for some of the cyclopentyl methyl esters. This indicates methyl branched somewhere in the long hydrocarbon chain attached to the cyclic structure.

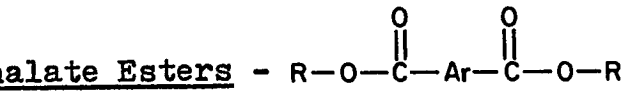
Characteristic ions for normal carboxylic methyl esters such as M-43 and M-31 are also observed for some of the cyclopentyl methyl esters. The M-31 peak is due to loss of methoxyl ($\cdot OCH_3$) radical by cleavage.

Both cyclopentyl and cyclohexyl compounds exhibit peaks at m/e = 69 and at m/e = 83. However, cyclopentyl compounds exhibit 69 as the larger peak while cyclohexyl compounds exhibit 83 as the largest of the two peaks.

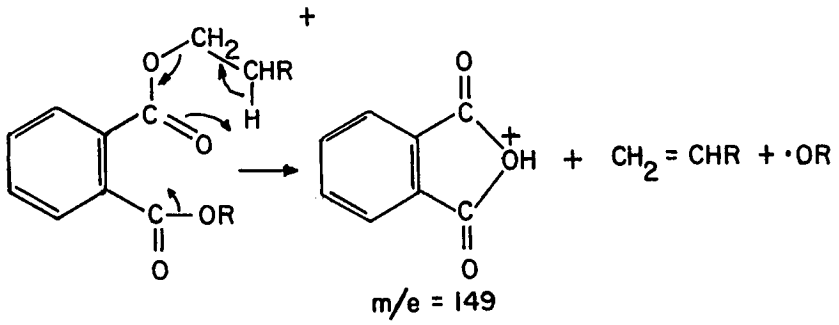
The cyclopentyl methyl esters correspond to the chemical formula $C_nH_{2n-1}COOCH_3$.

Unsaturated or Olefinic Esters - "Assignment to the class of unsaturated esters is based primarily on the molecular weight; differentiation from the cyclic esters is based on the strong loss of 32 mass units ($HOCH_3$). Such fragmentation is characteristic for unsaturated esters and

would not be expected for monocyclic esters. Major ions at 55, 69, 74 and 87 are observed in the spectra of unsaturated esters."[59]

Phthalate Esters - 

Phthalate esters (except allyl and nonyl) give m/e 149 as the base peak of the spectrum.[60] The ease of formation and high ion stability suggest the following mechanism:

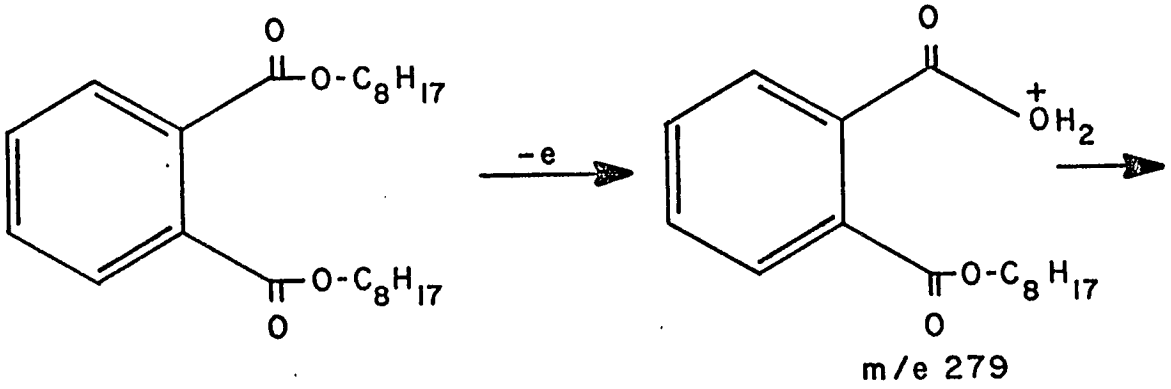


The mass spectrum of dioctylphthalate is included (p.147). The presence of the base peak at m/e = 149 accompanied by other significant peaks at m/e = 167 and at m/e = 279 are the main characteristic point of this spectrum.[61] In general, the mass of the molecular ion represents the molecular weight of the compound. However, dioctylphthalate ($C_{24}O_4H_{38}$) has a MW of 390 but a molecular ion is not usually observed for this compound. The fragmentation pattern is outlined in Figure 6.

The spectrum of ethyl phthalate is shown in Table XXI. The base peak appears at m/e = 149 and the molecular ion at m/e = 222. The fragmentation process is similar to that described in Figure 6.

The spectrum of butyl phthalate is shown in Table XXI. The base peak appears at m/e = 149 and the parent peak at m/e = 278.

FIGURE 6
FRAGMENTATION SEQUENCE



DIOCTYLPHTHALATE
MW 390

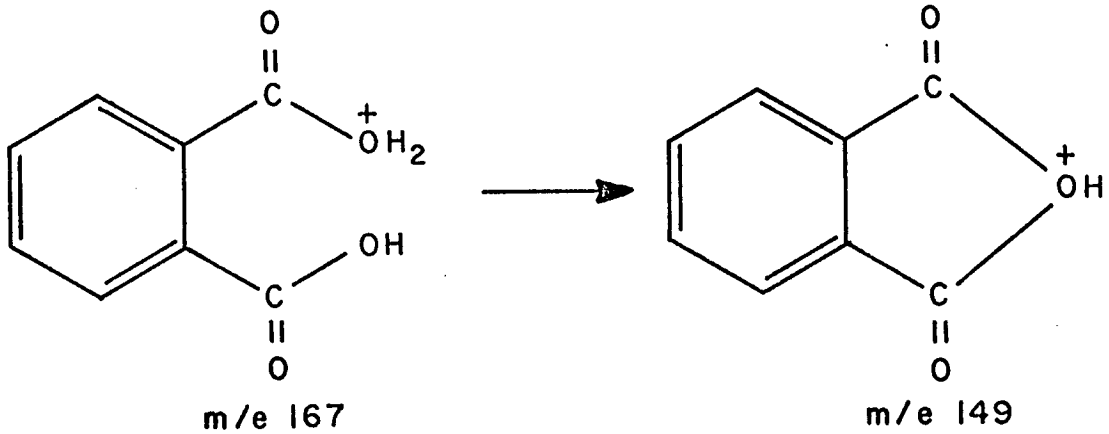


TABLE XXI

MASS SPECTRA OF PHTHALATE ESTERS*

m/e	Ethyl Phthalate % Intensity	Butyl Phthalate % Intensity	m/e	Ethyl Phthalate % Intensity	Butyl Phthalate % Intensity
41	0.58	15.00	104	9.09	6.24
42	--	1.07	105	9.39	3.28
43	1.96	2.86	106	1.74	--
44	--	0.75	121	4.65	1.80
50	11.90	3.96	122	3.74	1.75
51	5.20	1.52	123	--	1.07
52	1.66	--	132	1.54	0.66
53	1.23	0.68	135	0.63	--
55	--	2.38	148	0.77	--
56	--	5.98	149	100.00	100.00
57	--	6.94	150	12.50	9.18
63	0.87	--	151	1.32	0.99
64	0.82	--	176	5.13	--
65	14.80	4.20	177	26.00	--
66	4.58	0.57	178	5.75	--
74	2.26	0.74	179	0.61	--
75	3.00	1.18	205	--	4.23
76	14.70	7.26	206	--	0.70
77	6.68	2.14	222	4.53**	--
78	0.77	--	223	0.58	4.98
91	0.57	--	224	--	0.66
92	0.84	--	278	--	0.78**
93	5.80	2.42			

*Spectra were obtained on a CEC Model 21-103C Mass Spectrometer.[62]

**Molecular ion.

Straight-Chain Hydrocarbons - The formula for straight-chain hydrocarbons is C_nH_{2n+2}. The mass spectra of straight-chain hydrocarbons are characterized by a smooth envelope of peaks fourteen mass units apart (m/e 43, 57, 71, 85, etc.) which maximize at C_4 and decrease in intensity with increasing mass.[59] The peak at m/e = 57 is due to the fragment $C_4H_9^+$. The alkyl series is represented by the formula C_nH_{2n+1}. Mass spectral data of two high molecular weight hydrocarbons[63,64], n-tetracosane and n-pentacosane, are shown in Table XXII.

A molecular ion is usually observed for the normal hydrocarbons. A peak at M-15 (M = molecular ion) is usually due to loss of a methyl group, suggesting the presence of methyl branching.

4. Collection of Samples by Gas Chromatography (GC) and Mass Spectrometric (MS) Analysis of Such Fractions

Samples from selected fractions were collected from the analytical gas chromatograph and subsequently analyzed by mass spectrometry. The fractions selected for collection purposes were: Fraction XIII-6-D-2, Fraction II-6-D^+, Fraction Urea Clathrate-6-D^+, and Fraction XV-6-D^+. The gas chromatograms of these fractions and the mass spectra of the collected samples are discussed in the following pages. Table XXIII is a compilation of data derived from the gas chromatograms of the selected fractions. The spectral data presented in Table XX consists of the six

TABLE XXII

MASS SPECTRAL DATA OF NORMAL HYDROCARBONS

m/e	n-Tetracosane Relative Intensities*	n-Pentacosane Relative Intensities**
41	32.7	--
42	10.5	--
43	85.7	--
44	2.85	--
50	0.10	--
51	0.21	--
52	0.10	--
53	1.07	--
54	3.02	--
55	26.6	--
56	16.4	--
57	100.0	--
58	4.46	--
65	0.20	0.60
66	0.20	0.50
67	2.86	5.68
68	3.07	5.15
69	16.2	28.64
70	11.7	18.60
71	57.8	100.00
72	3.17	5.53
79	0.29	0.76

TABLE XXII (Cont'd.)

m/e	n-Tetracosane Relative Intensities*	n-Pentacosane Relative Intensities**
80	0.12	0.30
81	1.09	2.27
82	2.56	3.80
83	11.50	19.19
84	7.16	10.93
85	40.1	69.14
86	2.65	4.61
95	0.51	1.01
96	1.10	1.79
97	7.46	11.97
98	5.11	7.06
99	10.00	15.53
100	0.77	1.28
109	0.19	0.38
110	0.58	0.87
111	3.37	5.09
112	3.68	4.94
113	6.85	10.27
114	0.58	0.96
124	0.31	0.46
125	1.48	2.21
126	3.00	3.78

TABLE XXII (Cont'd.)

m/e	n-Tetracosane Relative Intensities*	n-Pentacosane Relative Intensities**
127	5.29	7.70
128	0.52	0.81
138	0.18	0.30
139	0.59	0.87
140	2.48	2.97
141	4.40	6.04
142	0.46	0.69
153	0.30	0.48
154	2.11	2.43
155	3.78	4.90
156	0.46	0.61
167	0.20	0.32
168	1.86	2.00
169	3.33	4.14
170	0.41	0.55
181	0.16	0.22
182	1.55	1.69
183	3.01	3.59
184	0.41	0.53
195	0.09	0.14
196	1.33	1.40
197	2.71	3.14
198	0.41	0.49

TABLE XXII (Cont'd.)

m/e	n-Tetracosane Relative Intensities*	n-Pentacosane Relative Intensities**
210	1.13	1.19
211	2.52	2.79
212	0.39	0.45
223	0.04	0.07
224	0.94	1.01
225	2.33	2.52
226	0.40	0.47
227	0.03	0.06
237	0.02	0.05
238	0.81	0.85
239	2.17	2.29
240	0.38	0.43
252	0.68	0.73
253	2.01	2.09
254	0.38	0.41
266	0.56	0.60
267	1.84	1.90
268	0.37	0.40
280	0.48	0.51
281	1.61	1.74
282	0.34	0.39
293	0.04	0.02

TABLE XXII (Cont'd.)

m/e	n-Tetracosane Relative Intensities*	n-Pentacosane Relative Intensities**
294	0.25	0.41
295	1.12	1.51
296	0.22	0.36
308	0.16	0.24
309	0.61	1.00
310	0.13	0.25
323	0.03	0.62
336	0.19	--
337	0.10	0.06
338	7.36***	0.01
339	1.95	--
340	0.24	--
352	--	1.67***

*Mass spectrum was obtained on a CEC Model 21-102 (Modified)[63]

**Mass spectrum was obtained on a CEC Model 21-103 (Modified)[64]

***Molecular ion.

TABLE XXIII

COLLECTION OF ESTERS BY GAS CHROMATOGRAPHY

Collected Sample No.	GC Elution Temp. °C	Carbon No. Range	MW Range	Comments
		Fraction XIII-6-D-2 - Gas Chromatogram No. 16		
GC-1	163-180	C_{10}-C_{11}	172-186	--
GC-2	182	C_{11}	186	--
GC-3	190	C_{12}-C_{13}	200-214	Second largest peak in the chromatogram.
GC-4	208	C_{13}-C_{14}	214-228	--
GC-6	218	C_{14}	228	Third largest peak in the chromatogram.
GC-5	232	C_{15}	242	--
GC-7	245-285	C_{17}	270	--
GC-8	285-315	C_{19}-C_{23}	298-354	--
GC-9	315	C_{25}	382	Largest peak in the chromatogram.

TABLE XXIII (Cont'd.)

Collected Sample No.	GC Elution Temp. °C	Carbon No. Range	MW Range	Comments
		Fraction II-6-D^+ - Gas Chromatogram No. 19		
GC-72	230	C_{13}	214	This peak coincides exactly with a n-C_{13} ester.
GC-73	243	C_{14}	228	--
GC-75	268	C_{15}	242	This fraction consisted of two peaks.
GC-77	279	C_{16}	256	It looks like a very clean peak.
GC-78	290	C_{17}	270	It includes at least two peaks.
GC-79	300	C_{18}-C_{19}	284-298	It includes several peaks.
GC-80	300	C_{19}-C_{20}	298-312	--
GC-81	>300	C_{20}	312	It includes several peaks.
GC-83	>300	C_{21}	326	It is a very clean peak.
GC-84	>300	C_{22}	340	It is a very clean peak.
GC-82	>300	C_{23}	354	It consists of two peaks.
GC-85	>300	C_{24}	368	It is a very clean peak.
GC-86	>300	C_{25}	382	It is a small, broad peak.
GC-87	>300	C_{26}	396	--
GC-88	>300	C_{26}-C_{27}	396-410	--

TABLE XXIII (Cont'd.)

Collected Sample No.	GC Elution Temp. °C	Carbon No. Range	MW Range	Comments
		Urea Clathrate-6-D^+ - Gas Chromatogram No. 20		
GC-130	130-186	C_{10}-C_{13}	172-214	No significant peaks were observed.
GC-131	190	C_{13}	214	Clean peak of low intensity.
GC-132	190-230	C_{14}-C_{17}	228-270	No significant peaks were observed.
GC-133	235	C_{18}	284	No significant peaks were observed.
GC-134	245	C_{19}	298	One peak was recorded.
GC-135	255	C_{20}	312	Several peaks are included in this fraction.
GC-136	267	C_{21}	326	Several peaks constitute this fraction.
GC-137	277	C_{22}	340	Two superimposed peaks are observed.
GC-138	287	C_{23}	354	Clean and strong peak.
GC-139	297	C_{24}	368	Most intense peak of the chromatogram.
GC-140	>300	C_{25}	382	Clean peak.
GC-141	>300	C_{26}	396	Clean peak but somewhat broad.
GC-142	>300	C_{27}	410	Broad, single peak.

TABLE XXIII (Cont'd.)

Collected Sample No.	GC Elution Temp. °C	Carbon No. Range	MW Range	Comments
		Fraction XV-6-D^+ - Gas Chromatogram No. 21		
GC-162	--	C_{12}-C_{13}	200-214	Two peaks are observed.
GC-163	--	C_{13}	214	One clean, distinct peak.
GC-165	--	C_{15}	242	One small peak.
GC-166	--	C_{15}-C_{16}	242-256	Several peaks are included in this sample.
GC-167	--	C_{16}-C_{17}	256-270	One major peak with considerable tailing.
GC-168	--	C_{17}-C_{18}	270-284	Two peaks.
GC-169	--	C_{18}-C_{19}	284-298	Two peaks.
GC-170	--	C_{20}	312	Fairly clean peak.
GC-171	--	C_{21}	326	Two peaks superimposed on each other.
GC-172	--	C_{22}	340	Clean peak.
GC-173	--	C_{23}	354	Clean peak.
GC-174	--	C_{24}	368	Clean peak.
GC-175	--	C_{25}	382	Two peaks.
GC-176	--	C_{26}	396	This is the strongest peak in the chromatogram.

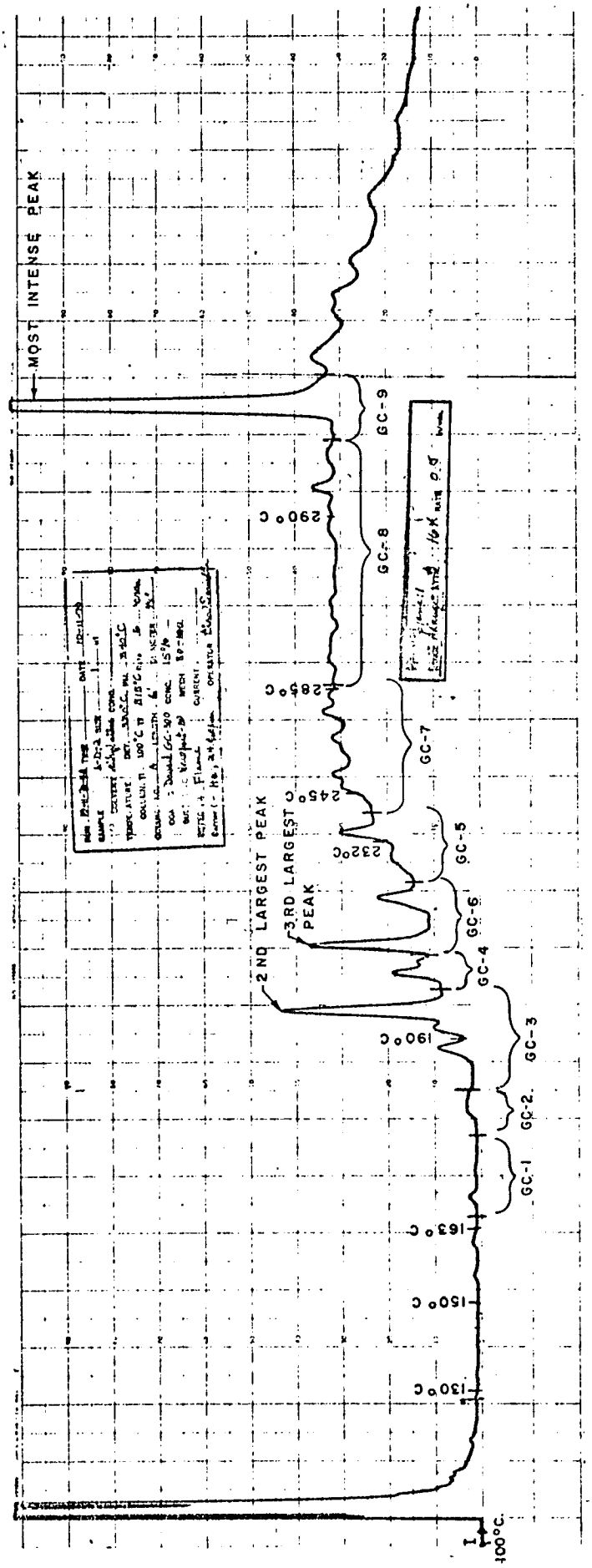
strongest peaks in each spectrum and their relative intensities, along with the mass spectrum identification. The peaks at m/e = 28 and at m/e = 32 should be neglected due to atmospheric nitrogen and oxygen. Line drawings of the mass spectra, composed of m/e values on the abscissa and relative intensity on the ordinate, are also included in the text.

Fraction XIII-6-D-2

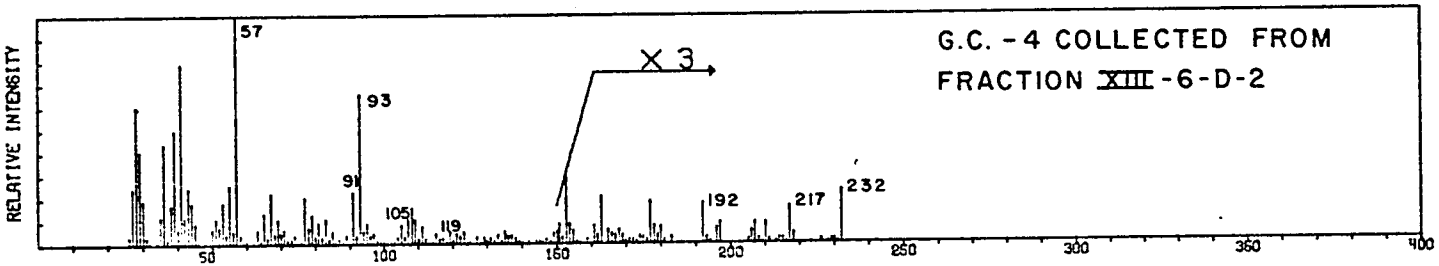
GC Fractionation of Fraction XIII-6-D-2 - Fraction XIII-6-D-2, eluted from a thiourea column with methanol, was further fractionated by gas chromatography (Gas Chromatogram No. 16). The resulting nine fractions were collected into melting point capillaries. Retention times and co-injection of standard straight-chain methyl esters of carboxylic acids were used to determine approximate carbon numbers and molecular weights of the collected fractions (Table XXIII).

Mass Spectrometric Analysis of Collected Samples from Fraction XIII-6-D-2 - Low resolution mass spectra were run at Rice University of Samples GC-4, GC-8 and GC-9.

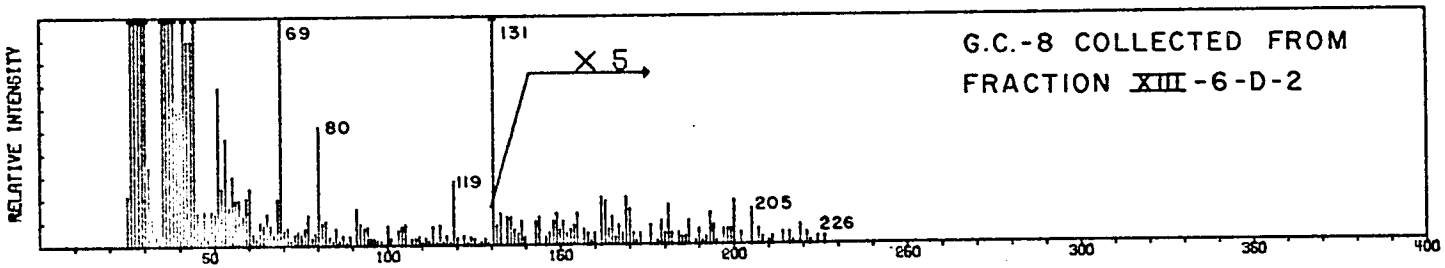
Mass Spectrum No. 4 of Fraction GC-4 shows a molecular ion (M) at m/e = 232 and the base peak at m/e = 57. The molecular weight range for Sample GC-4 was estimated from GC data to be 214-228; so the molecular weight of 232 suggested by MS analysis falls within a reasonable range.



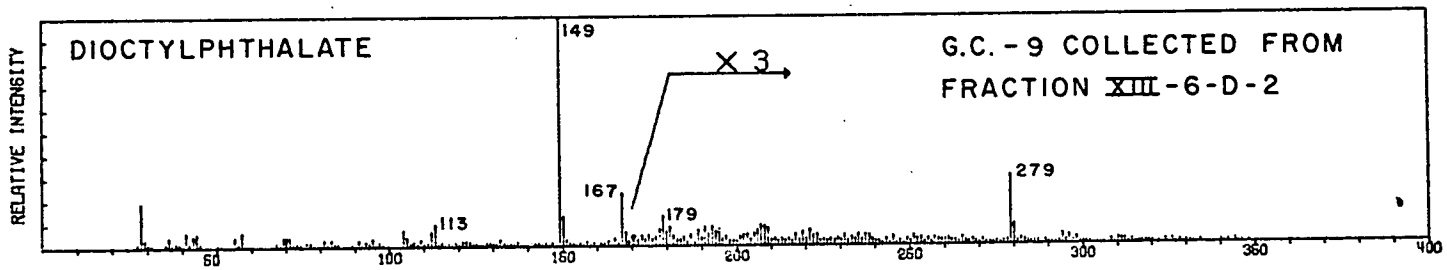
GAS CHROMATOGRAM NO.16
FRACTION XIII-6-D-2



MASS SPECTRUM NO. 4

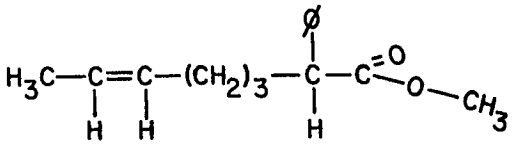


MASS SPECTRUM NO. 8



MASS SPECTRUM NO. 9

This mass spectrum exhibits a small but significant peak at m/e = 217 (M-15) (p. 165). A proposed structure for this compound is:



The formula is C_7H_{12}-Ø——$COOCH_3$. In order to satisfy the molecular weight of 232, the methyl ester must have an aromatic ring plus a double bond somewhere in the chain. Since the peak at m/e = 74 is of low intensity, the methyl ester group must be either on the aromatic ring or on the terminal carbon adjacent to the one with the aromatic ring. The aromatic ion (at m/e 91, 105, 119, etc., for a hydrocarbon aromatic nucleus) is an important feature of this spectrum. The proposed structure includes an aromatic ring on one side of the aliphatic chain, and methyl branching somewhere on the structure.

Mass Spectrum No. 8 of Fraction GC-8 is a very weak spectrum with peaks appearing up to m/e = 226 only. The molecular weight range of Fraction GC-8 based on GC data was expected to be 298-354. According to this information, the mass spectrum should have shown peaks at masses higher than m/e = 226. The most significant peaks in the spectrum show up at m/e of 69, 80, 119, 131, 205 and 226. The parent peak at m/e = 226 could correspond to the formula $C_{12}H_{23}COOCH_3$ but the other numbers of the C_nH_{2n-1} series are not observed.

Sample GC-9 was positively identified to be dioctylphthalate from its Mass Spectrum No. 9. The mass spectrum of standard dioctylphthalate is given in Part III, Section D-3. It is of interest to note that Sample GC-9 gave the most intense peak in the GC chromatogram, indicating that this compound is the major component of Fraction XIII-6-D-2. It is unlikely that a contaminant would give the strongest peak in the chromatogram.

The finding of dioctylphthalate in a crude oil provoked many speculations. It was debatable whether the phthalate ester was present originally in the oil, whether it came from contact with plasticizers used polyvinyl chloride (PVC) materials or whether the crude had oxidized by long standing.[50]

If the crude oils were ever in contact with PVC (plasticizer) materials, e.g. Tygon tubing, the possibility of contamination by a dialkyl phthalate existed. "Octoil", which is a trade name for dioctylphthalate, is commonly used in the oil diffusion pumps of the inlet systems of mass spectrometers and could stream back into the reservoir, contaminating the samples.[61]

A very detailed study to investigate the possible introduction of phthalate contamination into our samples was conducted. The vacuum pump oils were checked first but they had to be discarded as sources of contamination. The vacuum pump oil used with the mass spectrometer at Rice

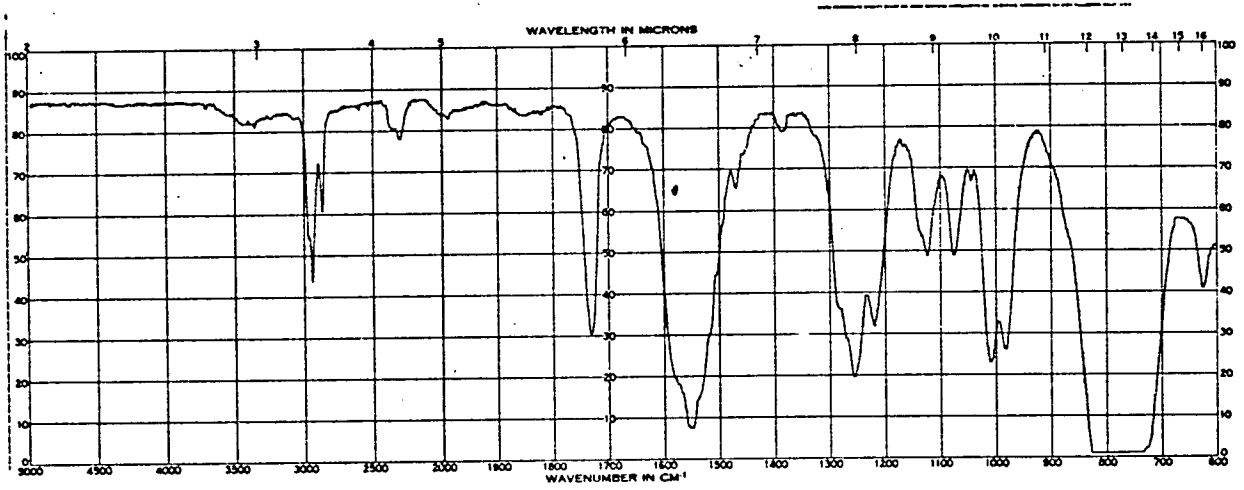
University was "Conblex 10", a polyphenyl ether and not dioctylphthalate. The house vacuum at the Texaco Laboratories in Bellaire operated from a water pump; so no oil contamination could possibly be coming from the house vacuum.

The following samples were analyzed by IR spectrometry and by gas chromatography in an effort to find possible sources of contamination by phthalates:

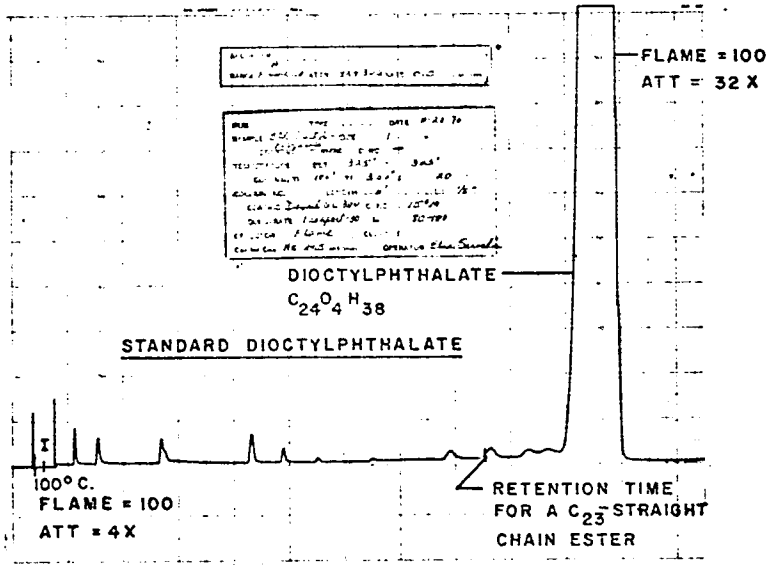
1. Commercial sample of pure dioctylphthalate - This standard was run to provide background data for interpretation of GC and IR spectra. IR Spectrum No. 31 of the phthalate standard showed a definite band around 1730 cm^{-1}, characteristic of esters. The triplet at 7.8, 8.95, and 9.35 μ is quite typical of dioctylphthalate.[65] Gas Chromatogram No. 17 of standard dioctylphthalate presented a peak at a temperature >300° C, with a retention time equal to that of n-C_{25} methyl ester.

2. Sample of Welch Duo-Seal Pump Oil used for the vacuum pump connected to the vacuum oven at Texaco Research Laboratories in Bellaire - IR Spectrum No. 32 of this pump oil indicated no evidence of phthalates, i.e. no band was observed at 1730 cm^{-1}. The IR spectrum indicated that this pump oil is a saturated hydrocarbon. The gas chromatogram of this sample gave no indication of the presence of dioctylphthalate.

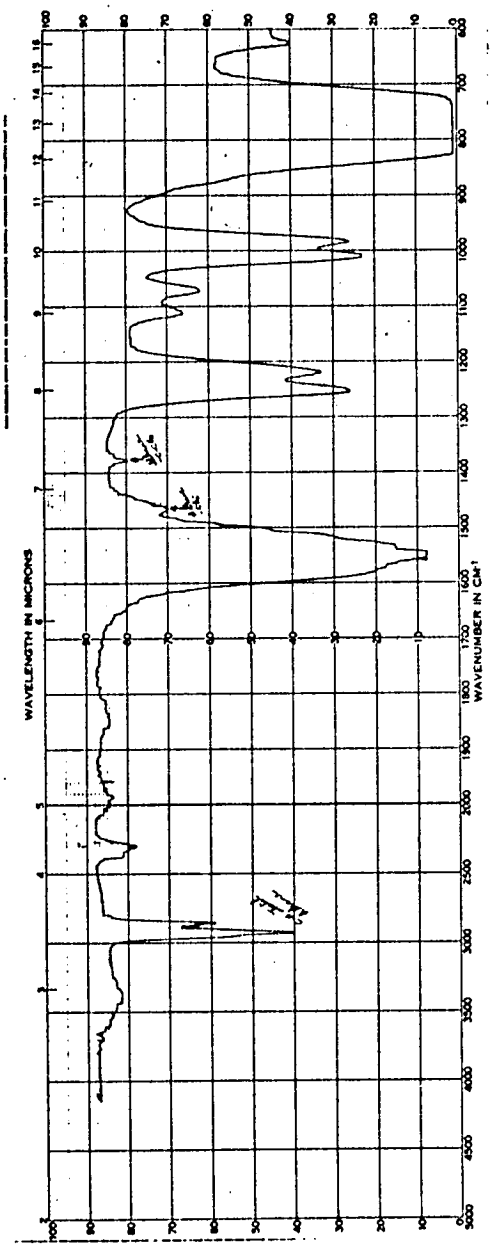
3. Sample of residue found in dry ice trap connected to vacuum pump - IR Spectrum No. 33 of residue showed no indication of dioctylphthalate.



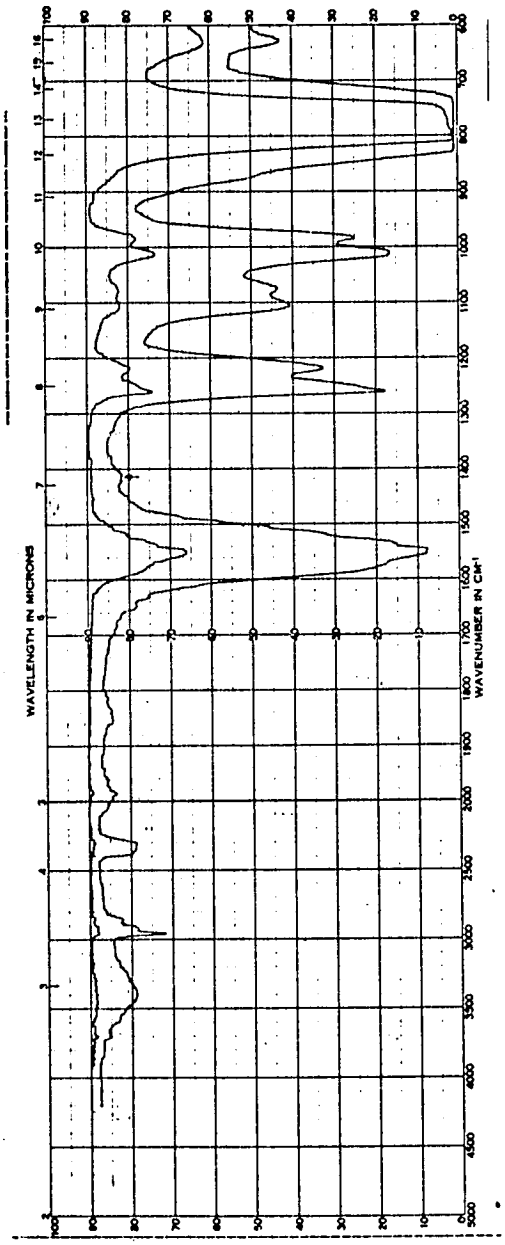
IR SPECTRUM NO. 31



GAS CHROMATOGRAM NO. 17



IR SPECTRUM NO. 32



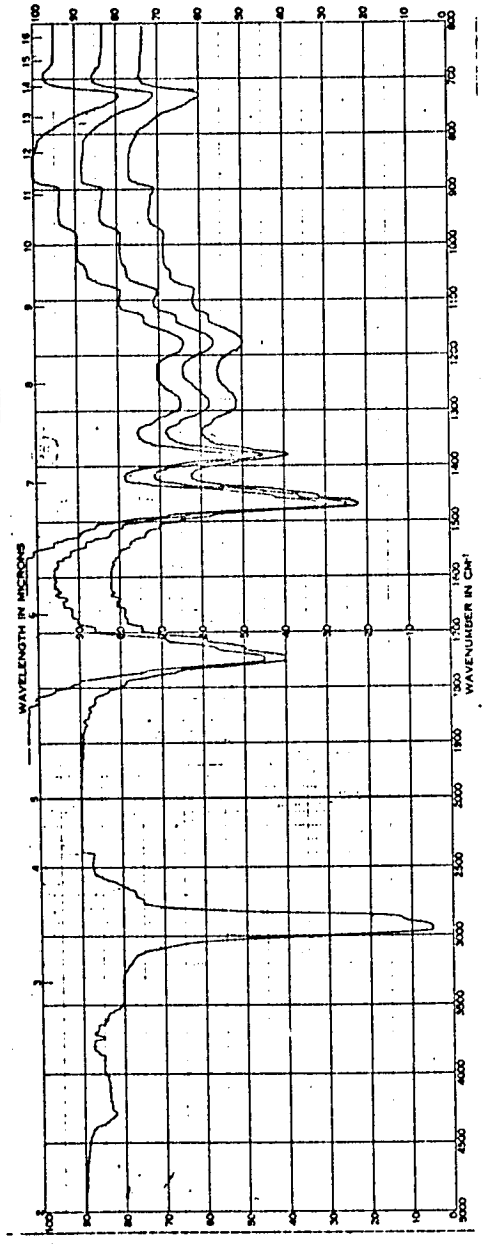
IR SPECTRUM NO. 33

4. Sample of residue remaining after evaporation of 500 ml of benzene which had been stored in polyethylene bottles with polypropylene screw caps - IR Spectrum No. 34 of benzene residue showed a strong C-H stretch at $\approx$2950 cm^{-1}; an absorption band at 1380 cm^{-1}, indicative of aromatic esters; and a carbonyl band at $\approx$1745 cm^{-1}, characteristic of carboxylic esters. IR Spectrum No. 34 is similar to the spectra of a dialkyl phthalate in gas oil, published by Jenkins.[50] Gas Chromatogram No. 18 of the benzene residue showed a series of peaks ranging from C_{12}-C_{22} but no indication of dioctylphthalate. Contamination by other types of phthalates could come from benzene stored in plastic wash bottles but this was not the source of dioctylphthalate.

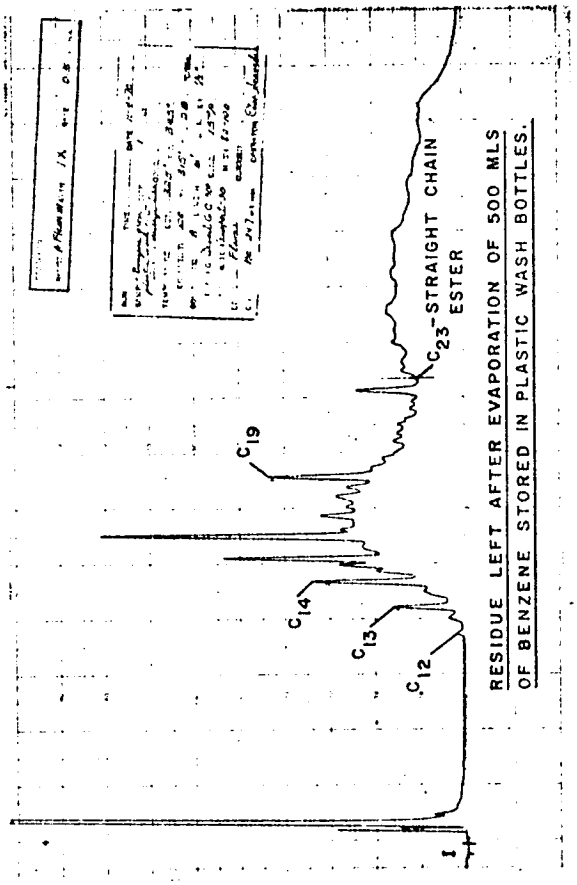
5. Sample of concentrated extract obtained by boiling pieces of silicone tubing with acetone - The gas chromatogram of this extract exhibited a series of peaks starting around C_{11} and continuing on through. No conclusive evidence for the presence of dioctylphthalate could be obtained from this chromatogram.

All our efforts to trace the source of dioctylphthalate were fruitless. Based on the GC and IR work, it was concluded that dioctylphthalate was indeed present in the total virgin crude oil and that it was not introduced as a contaminant.

A number of investigators have reported occurrences of phthalates in crude petroleum. Phillips and Breger[65]



IR SPECTRUM NO. 34



GAS CHROMATOGRAM NO. 18

identified a dioctylphthalate in a crude oil from Duchesne County, Utah. Dioctylphthalate has also been identified in an extract of the surface active constituents from a crude oil from Panhandle Field, Texas and in oils associated with mineralized rocks from Seven Rivers formation, Eddy County, New Mexico.[66] The presence of esters in petroleum has been regarded as due to the persistence of waxes or wax-hydrolysis products from the original source materials.[67] Breger has speculated that the phthalate esters could be preserved metabolic products or that they might have been formed during formation of the oil.[65]

In another instance, published infrared spectra of fractions from Italian crudes showed that aromatic esters, possibly dialkyl phthalates, were present. However, the author assigned the ester absorption to oxidation.[68] The possibility of contamination from PVC plasticizers was not discussed in most of the reports of esters in petroleum.[65,67,68] Jenkins[50] has also reported on the presence of carboxylic esters in petroleum.

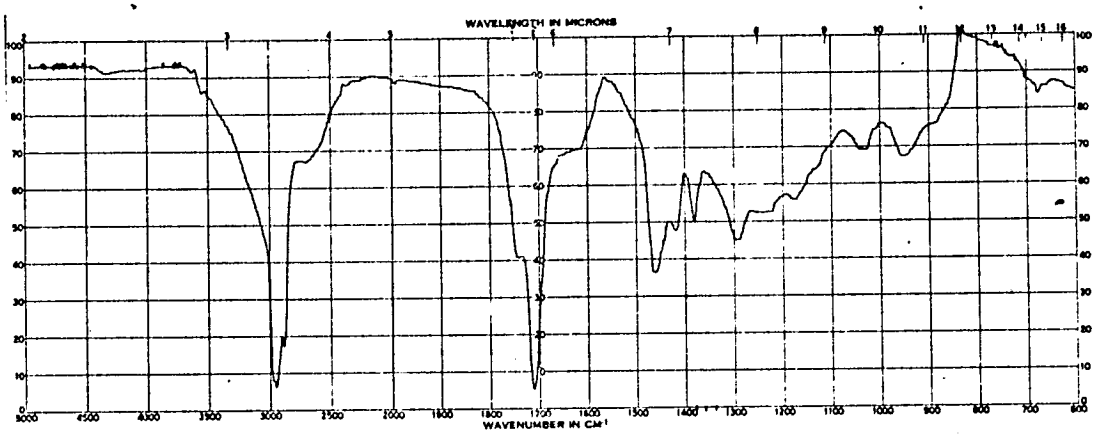
Even though it was quite obvious that the dioctylphthalate was not introduced as a contaminant, from this point on, every possible precaution to avoid contamination of any sort was taken. One of the changes introduced was to pass Fraction D, before esterification, through silica gel in an attempt to further purify Fraction D. The ether fractions eluted from the silica gel columns were expected to be free from hydrocarbons and other extraneous materials (Figure 5 and Experimental section).

The other measures introduced to prevent contamination were to discontinue the use of plastic wash bottles and to create a deliberate air leak in the lines connected to the vacuum desiccator and to the vacuum oven (preventing any backflow from the vacuum lines).

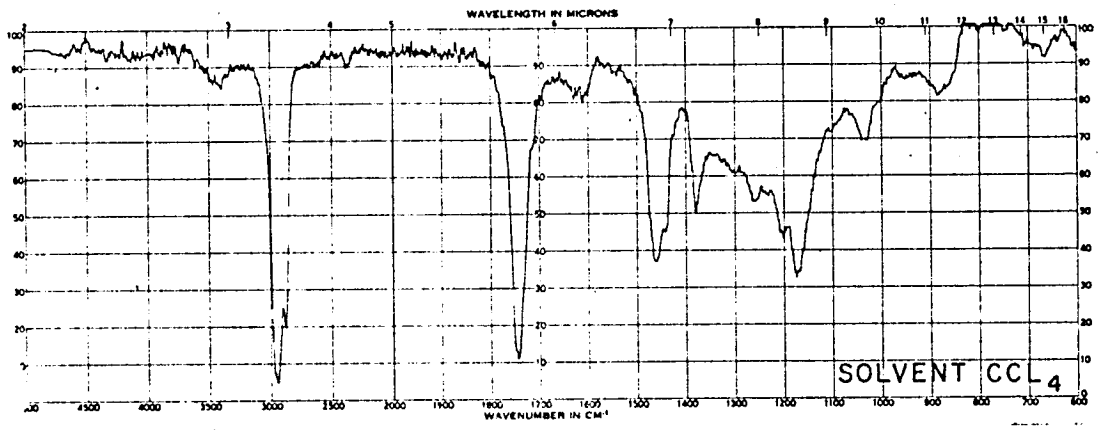
Various fractions obtained using all possible precautions to avoid contamination were then examined by infrared spectroscopy to determine the presence of esters and/or acids in fractions obtained at various steps in the heavy oil separation procedure. The IR of the selected fractions is described below.

1. Fraction 6-D_3 (Table XVII)

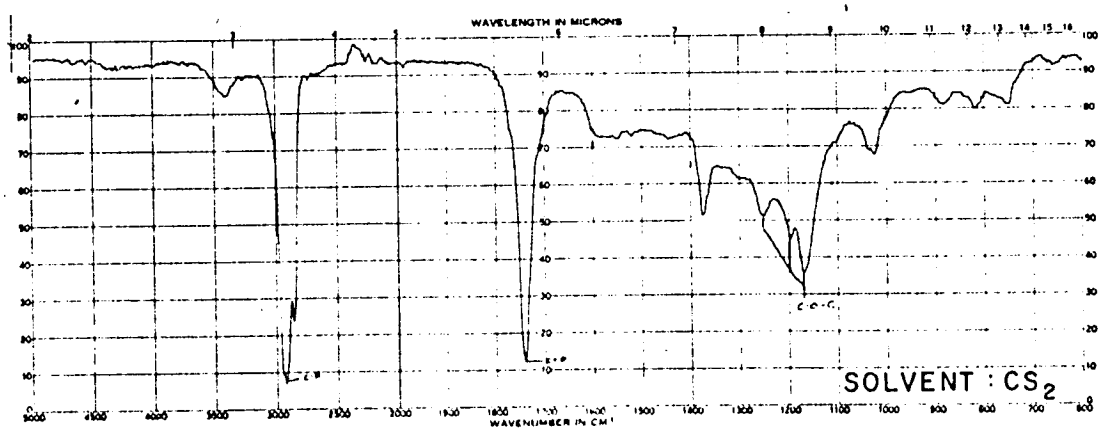
Fraction 6-D_3 showed a strong carbonyl[69] band at 1710 cm^{-1} and a definite shoulder at 1740 cm^{-1} in its IR Spectrum No. 35. The fact that the carbonyl band appears higher than 1700 cm^{-1} indicates aliphatic acids.[34] There is also a sloping shoulder between 3000 and 3500 cm^{-1} characteristic of acids. The IR spectrum suggested that Fraction 6-D_3, which is the result of pooling four Fraction D's (Table XVII), consists mostly of carboxylic acids. However, the presence of esters though in a smaller concentration than the acids is indicated by the band at 1740 cm^{-1}. Fraction 6-D_3 has not been passed through silica gel.



IR SPECTRUM NO. 35
FRACTION 6-D_3



IR SPECTRUM NO. 36
FRACTION 6-D_3-BENZENE



IR SPECTRUM NO. 37
FRACTION 6-D_3-BENZENE

2. Fraction 6-D_3-Benzene

Fraction 6-D_3-Benzene is the benzene eluate after passing Fraction 6-D_3 through silica gel. IR spectra of this fraction were run in two different solvents. IR Spectrum No. 36 was run using CCl_4 as a solvent and IR Spectrum No. 37, using CS_2 as a solvent. IR No. 36 shows an ester carbonyl band at 1745 cm^{-1} and a series of three bands at 1165 cm^{-1}, 1200 cm^{-1} and 1260 cm^{-1}, typical of methyl esters of fatty acids. IR Spectrum No. 37 also presents an ester carbonyl band at 1742 cm^{-1}.

Both of these spectra show that Fraction 6-D_3-Benzene consists of a mixture of esters. Up to now, it was assumed that Fraction D (before esterification) had consisted only of carboxylic acids. Since Fraction 6-D_3-Benzene is indeed a part of Fraction D, it is a fact that both acids and esters were present in Fraction D. Having avoided any possible contamination sources, the esters in Fraction 6-D_3-Benzene had to be indigenous to Heavy Oil #6.

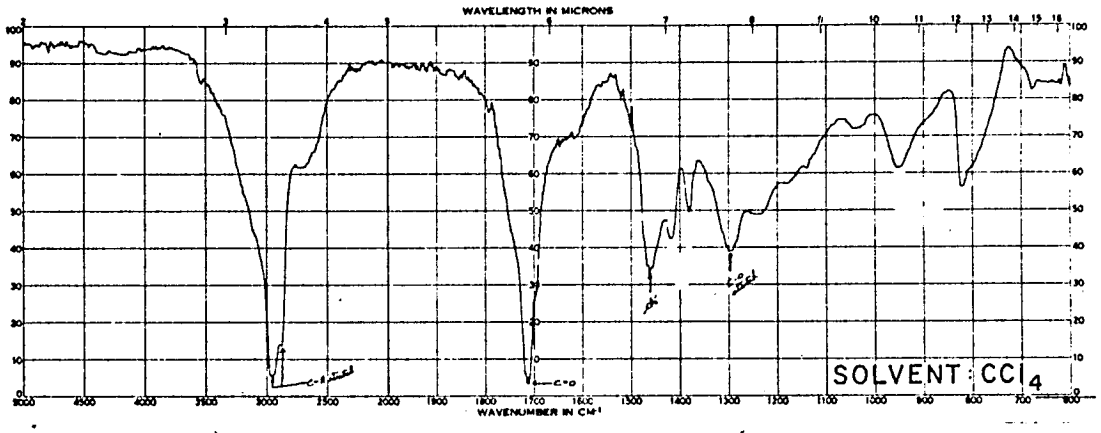
3. Fraction 6-D_3-Ether

Fraction 6-D_3-Ether is the ether eluate after Fraction 6-D_3 is passed through silica gel. We hoped to concentrate the carboxylic acids in Fraction 6-D_3-Ether by the silica gel chromatography. Otherwise, identification of compounds in Fraction 6-D_3-Ether would be even more complex and confusing. Fraction 6-D_3-Ether was selected for

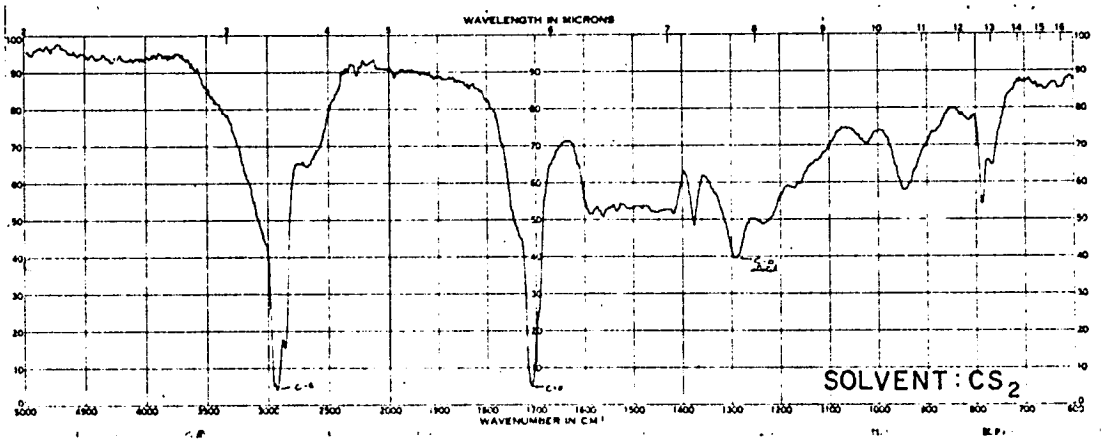
further separation. IR Spectrum No. 38 of Fraction 6-D_3-Ether in CCl_4 was characteristic of an acid, with carbonyl absorption at 1715 cm^{-1} and sloping shoulders from 3000-3300 cm^{-1}, due to a C-H stretching vibration. IR Spectrum No. 39 of this fraction in CS_2 shows the same characteristic bands. It is interesting to compare these two spectra with IR Spectrum No. 35 of Fraction 6-D_3. The definite shoulder at 1740 cm^{-1} due to esters in Fraction 6-D_3 is not as pronounced in Fraction 6-D_3-Ether. Fraction 6-D_3-Ether shows a sloping shoulder around 1740 cm^{-1}. It is obvious that we have removed some of the esters from Fraction 6-D_3, but from IR No. 38 and No. 39, there is indication that some of the esters could still be present.

4. Methyl esters of Fraction 6-D_3-Ether

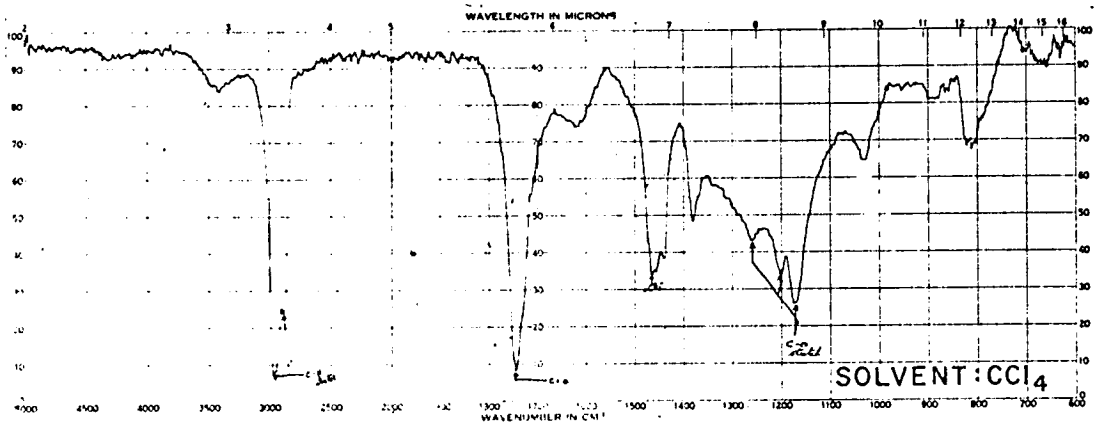
IR Spectrum No. 40 of the methyl esters of Fraction 6-D_3-Ether in CCl_4 shows a band characteristic of the ester group at 1745 cm^{-1}. IR Spectrum No. 41 of the methyl esters in CS_2 shows a strong carbonyl band at 1740 cm^{-1} and three bands at 1170 cm^{-1}, 1195 cm^{-1} and 1255 cm^{-1} indicative of methyl esters of long chain fatty acids. Aromatic bands at 1600 cm^{-1} and at 750 cm^{-1} indicate about 5% aromaticity in this fraction, probably caused by some impurity. These two IR spectra show that the methyl esters of Fraction 6-D_3-Ether were successfully prepared.



IR SPECTRUM NO. 38
FRACTION 6-D_3-ETHER

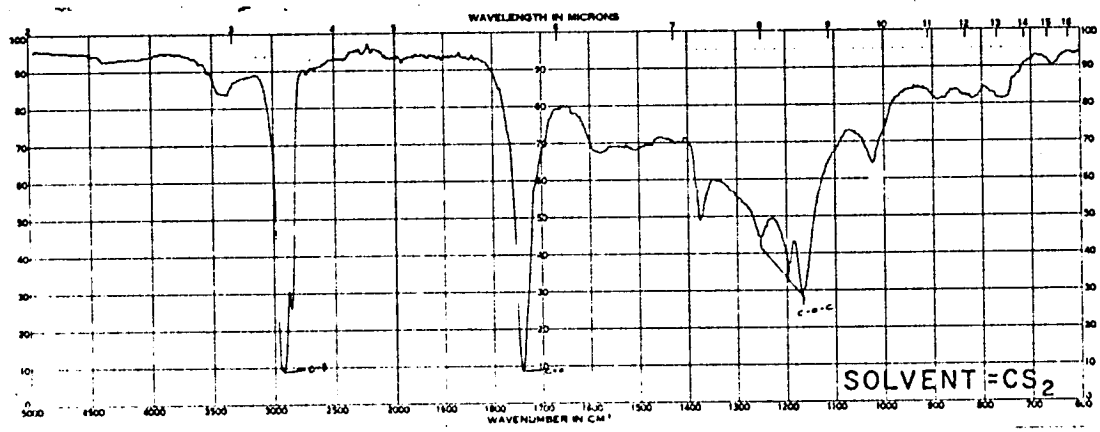


IR SPECTRUM NO. 39
FRACTION 6-D_3- ETHER



IR SPECTRUM NO. 40

METHYL ESTERS OF FRACTION 6-D_3-ETHER



IR SPECTRUM NO. 41

METHYL ESTERS OF FRACTION 6-D_3-ETHER

5. Fraction 6-D^+ (Figure 5)

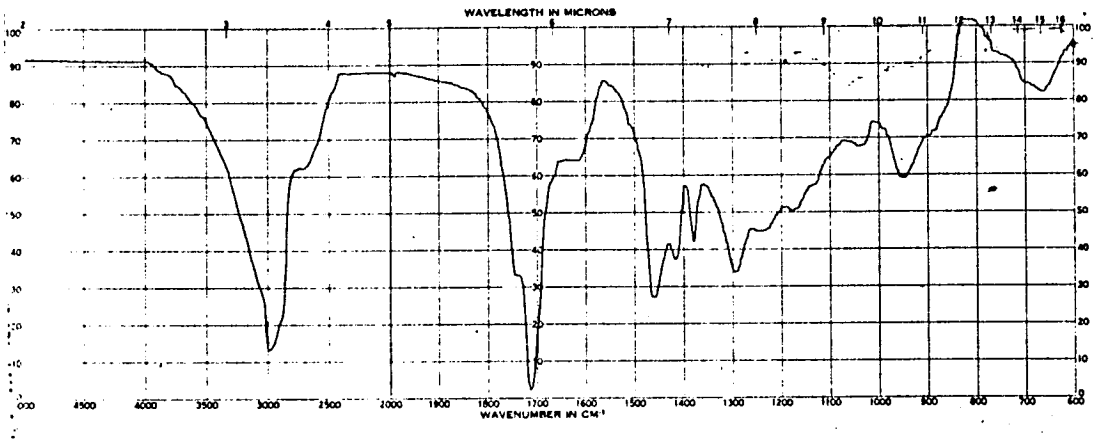
IR Spectrum No. 42 of Fraction 6-D^+ showed a very strong carbonyl absorption at 1710 cm^{-1}, due to carboxylic acids, a sloping shoulder from 3000-4000 cm^{-1} and a very strong C-H absorption. A definite shoulder at 1742 cm^{-1} indicates the presence of esters. Fraction 6-D^+ is the ether eluate after passing Fraction D twice through silica gel. It consists mostly of acids but it does contain some esters.

6. Fraction II-6-D^+ (Figure 5)

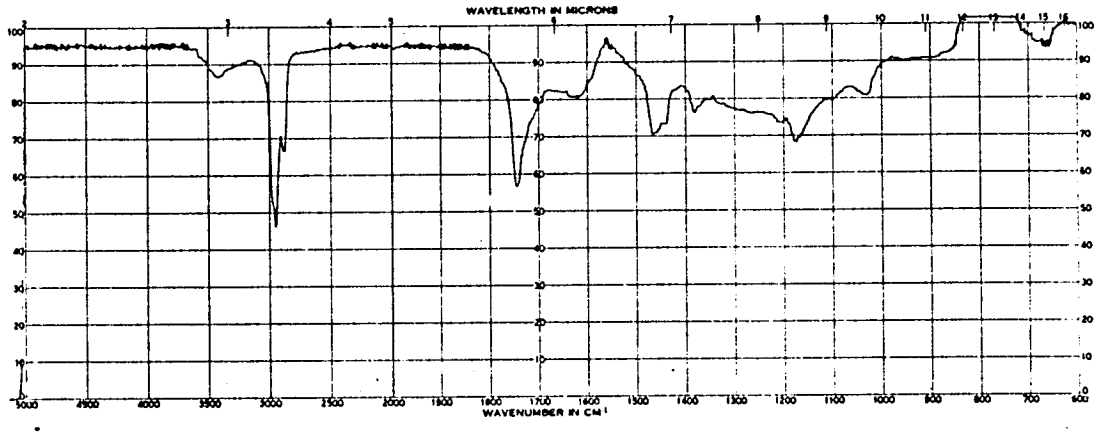
IR Spectrum No. 43 of Fraction II-6-D^+ exhibits a definite band at 1742 cm^{-1} confirming the presence of methyl esters. Fraction II-6-D^+ consisted of those methyl esters of Fraction 6-D^+ eluted with ether during the first urea chromatography. No evidence of acids is seen on this spectrum. The Experimental section contains the details of the methylation and further chromatography on Fraction 6-D^+.

7. Fraction Urea Clathrate-6-D^+

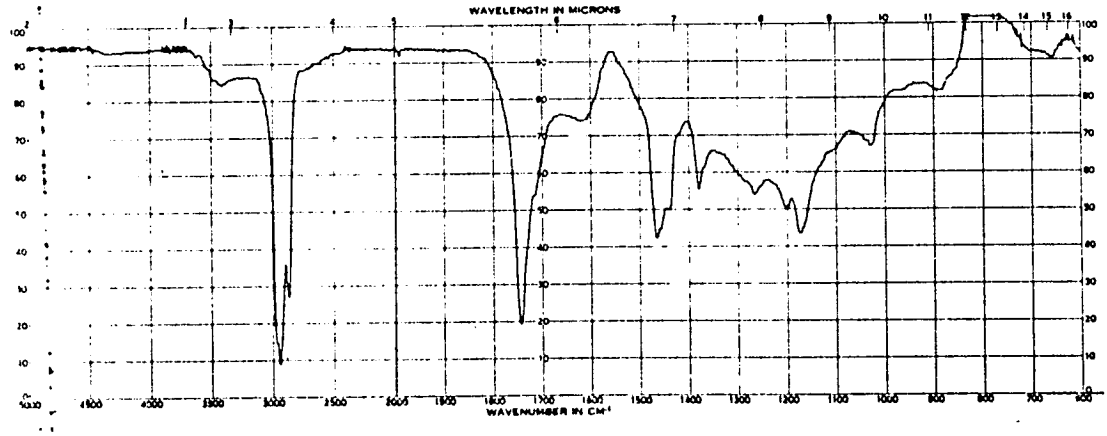
Fraction Urea Clathrate-6-D^+ was the result of combining several fractions: Fraction III-6-D^+, IV-6-D^+, IX-6-D^+ and X-6-D^+ (Figure 5). Their respective gas chromatograms showed that all four fractions were fairly similar. IR Spectrum No. 44 of Fraction Urea Clathrate-6-D^+ shows a large carbonyl peak at 1745 cm^{-1} indicating a high



IR SPECTRUM NO. 42
FRACTION 6-D$^+$



IR SPECTRUM NO. 43
FRACTION II-6-D$^+$



IR SPECTRUM NO. 44
FRACTION UREA CLATHRATE-6-D$^+$

concentration of esters, and a band at 1440 cm^{-1} that could be due to an alicyclic ring. Fraction Urea Clathrate-6-D^+ consists of a mixture of esters.

The conclusions derived from these series of IR spectra can be summarized as follows:

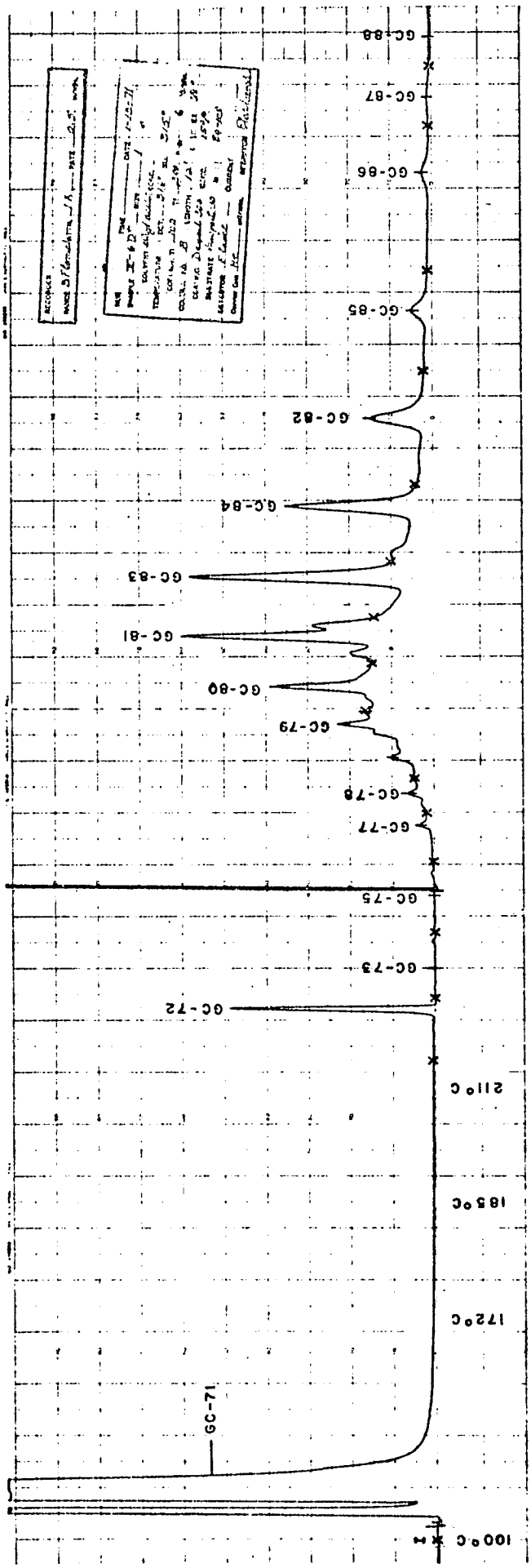
1. Fraction 6-D_3 contained both carboxylic acids and esters.
2. Fraction 6-D_3-Benzene consisted of carboxylic esters from petroleum.
3. Fraction 6-D_3-Ether and Fraction 6-D^+ consisted mostly of carboxylic acids but esters were present in small amounts.
4. Methyl esters of Fraction 6-D_3-Ether: Methylating procedure was effective and no acid remained in this fraction.
5. Fraction II-6-D^+ consisted of a mixture of esters of carboxylic acid.
6. Fraction Urea Clathrate-6-D^+ consisted of a mixture of esters of carboxylic acids.

The other samples collected from Fraction XIII-6-D-2 by gas chromatography were not analyzed by mass spectrometry because the mass spectrometer operator claimed, based on visual observation, lack of material for such analyses. The contents of these remaining capillaries were reinjected into the GC and strong GC spectra were obtained. However, since the sensitivities of the GC and the MS are quite different, it was impossible to prove by this experiment that there would have been sufficient material in those capillaries for MS analyses. Nevertheless, this first experience taught us that visual observation is a poor criterion for judging the amount of material required for MS analysis.

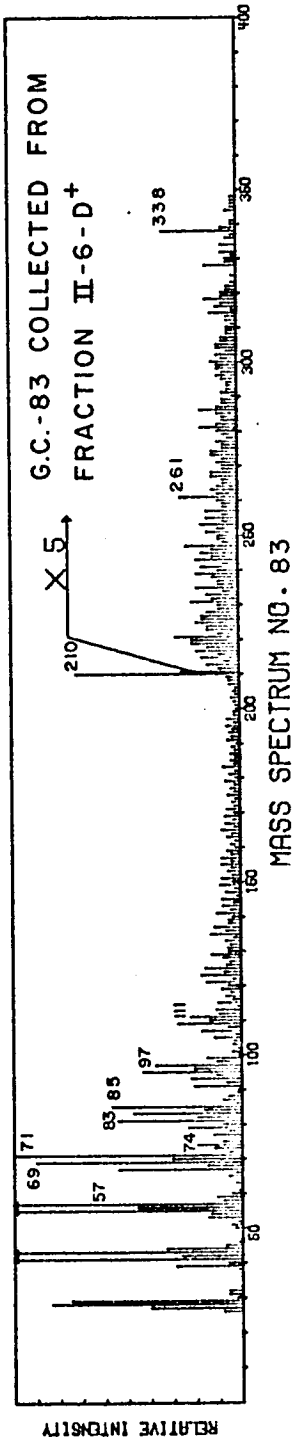
Fraction II-6-D^+

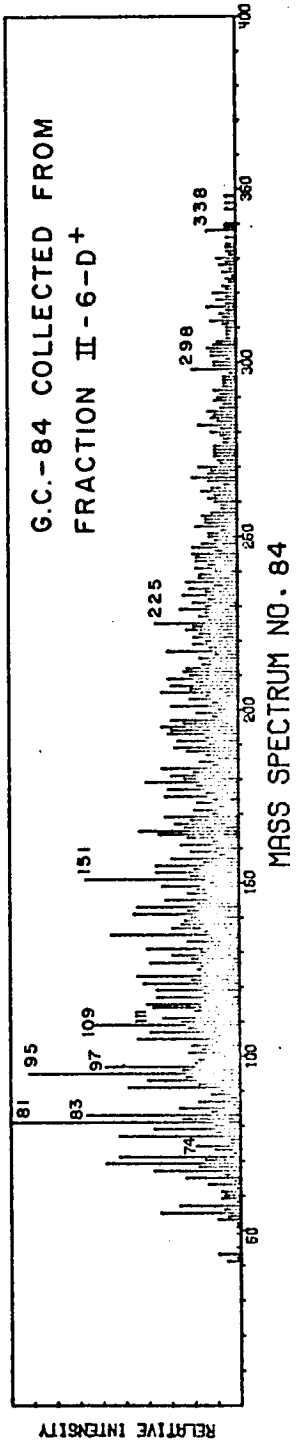
GC Fractionation of Fraction II-6-D^+ - The fraction eluted from a urea column with ether, Fraction II-6-D^+, was further fractionated by gas chromatography. Gas Chromatogram No. 19 shows the separation of Fraction II-6-D^+ into 15 components with carbon numbers ranging from C_{13}-C_{27} (Table XXIII). A list of the 15 fractions collected into melting point capillaries in order of decreasing GC peak intensities follows: GC-83, GC-81, GC-84, GC-72, GC-80, GC-82, GC-79, GC-78, GC-77, GC-85, GC-86, GC-87, GC-88, GC-75, and GC-73.

Mass Spectrometric Analysis of Collected Samples from Fraction II-6-D^+ - Two of the fractions, GC-83 and GC-84 were analyzed by low resolution mass spectrometry at Rice University and Mass Spectrum No. 83 and No. 84 were thus obtained. Mass Spectrum No. 83 of Fraction GC-83 exhibits a molecular ion at m/e = 338 and a base peak at m/e = 71. The gas chromatographic retention time of GC-83 had indicated a MW of 326 for this fraction. Some of the other significant peaks in the spectrum appear at m/e = 69, 83, 85, 210, and 261. From the molecular ion we can speculate that fraction GC-83 consists of a C_{22} methyl ester with one degree of unsaturation, i.e. $C_{20}H_{39}COOCH_3$ and MW = 338. The series of peaks at m/e = 69, 83, 97, 111 etc. and a small peak at m/e = 74 are observed in the spectrum. A very large m/e = 69 peak is an important characteristic of



GAS CHROMATOGRAM NO. 19
FRACTION II-6-D+


G.C.-83 COLLECTED FROM
FRACTION II-6-D+
X 5
MASS SPECTRUM NO. 83
RELATIVE INTENSITY


G.C.-84 COLLECTED FROM
FRACTION II-6-D+
MASS SPECTRUM NO. 84
RELATIVE INTENSITY

this spectrum, as well as the series of peaks at m/e = 57, 71, 85, etc. Fraction GC-83 consists of a cyclopentyl methyl ester of MW= 338.

The mass spectrum of GC-84, i.e. Mass Spectrum No. 84, indicated a molecular ion at m/e = 338 and a base peak at m/e = 81. The dominant series in this Mass Spectrum No. 84 is 81, 95, 109, etc. Based on its molecular ion only, Fraction GC-84 could also be interpreted as a C_{22} cyclopentyl methyl ester with a molecular weight of 338. The series 83, 97, 111, etc., is also present in this spectrum. The presence of an unidentified dominant series indicates that this fraction must be a mixture. So about all we can say about Fraction GC-84 is that it probably contains a cyclopentyl methyl ester (C_{22}) as one of its components. The peak at m/e = 74 is not very intense, so the cyclopentyl ring again must be near the methyl ester group.

All 15 samples collected from Fraction II-6-D^+ were examined further by combined gas chromatography-mass spectrometry at Baylor College of Medicine. A mass spectrum was recorded at several points on a GC peak. A total of 62 mass spectra were recorded for the 15 samples. An example of the labelling selected for these mass spectra follows: for Fraction GC-72, Mass Spectrum No. 72-1 and No. 72-2 were recorded. Table XX lists the strongest peaks of some of the mass spectra. Comments on selected spectra are covered on the next few pages.

Mass Spectrum (MS) No. 72-1 exhibits a molecular ion (M) at m/e = 220 suggesting a molecular weight of 220. This value is within a reasonable range of the proposed molecular weight of 214 from GC spectra (Table XX). Fraction GC-72 had exactly the same GC retention time as methyl dodecanoate (n-C_{13} ester) but it is obvious from the mass spectrum that Fraction GC-72 is not the n-C_{13} ester. Significant peaks in MS No. 72-1 appear at m/e = 205 (M-15) and at m/e = 177 (M-43). The molecular weight of 220 corresponds to the formula $C_{12}H_{17}COOCH_3$. The $C_{12}H_{17}$ part has to contain a benzene ring; the structure is probably the following:

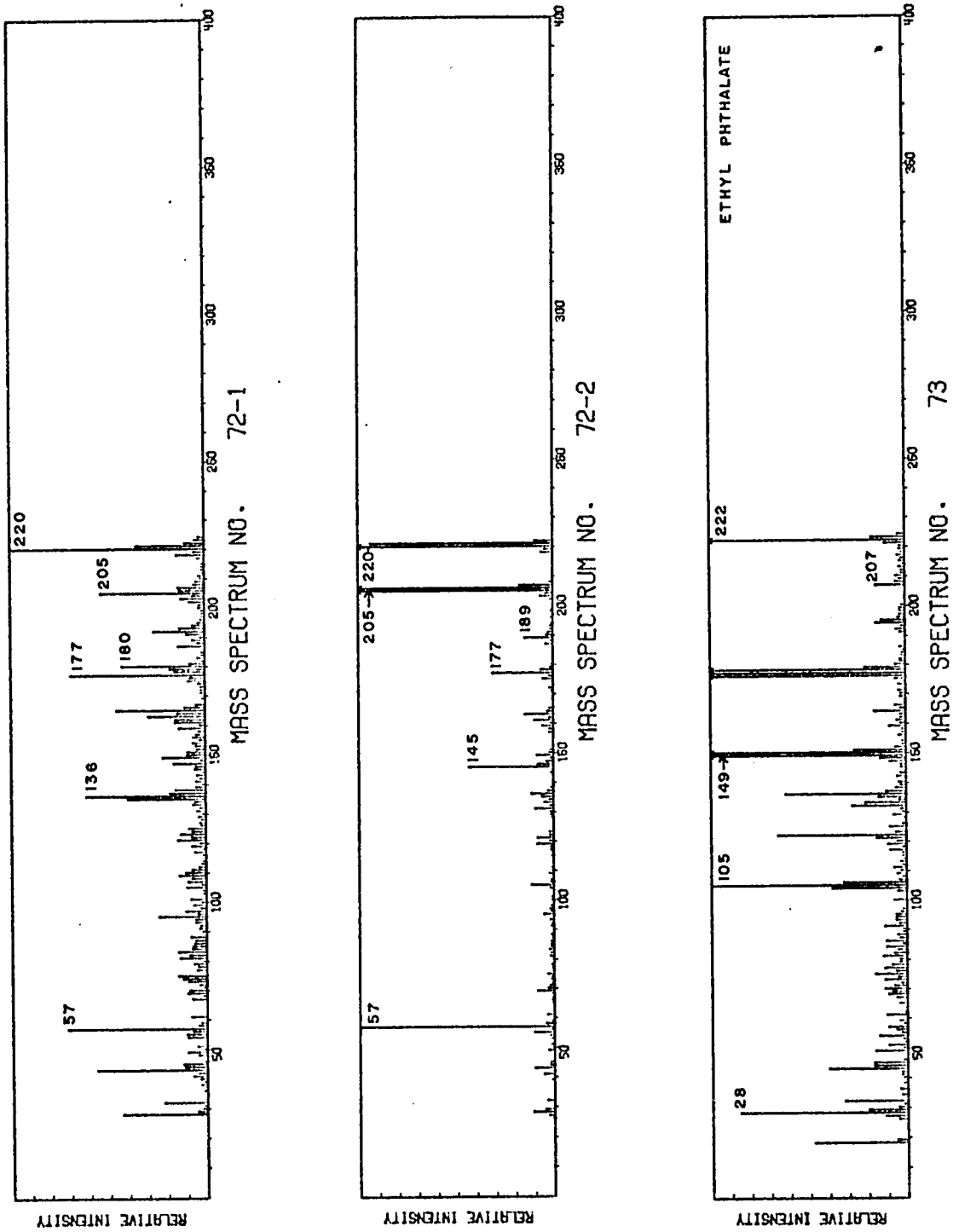
$$C_6H_{13}-\phi-CO_2CH_3$$

The M-15 peaks indicate methyl branching,[57] somewhere in the long hydrocarbon chain, possibly:

$$C-C-\underset{\underset{C}{|}}{C}-C-C-\phi-CO_2-CH_3$$

The peak at m/e = 136 is probably due to $C_6H_5-CO_2CH_3$, indicating that the methyl ester group is either on the benzene ring or next to it.

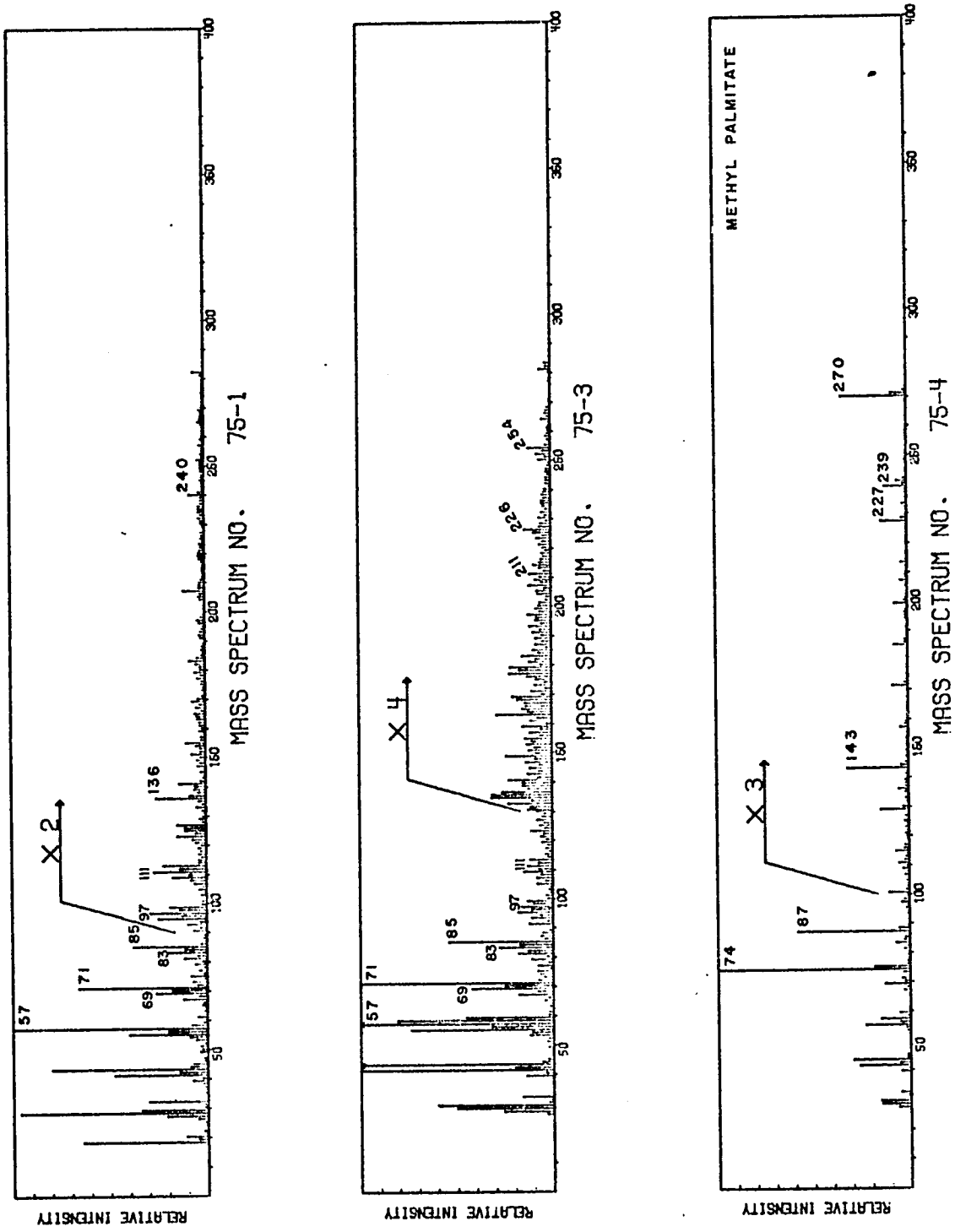
MS No. 72-2 also exhibits the molecular ion at m/e = 220 and a very intense (M-15) peak. MS No. 72-2 shows both the (M-43) fragment at m/e = 177 and the (M-31) fragment at m/e = 189. The base peak at m/e = 57 indicates a long hydrocarbon chain.


MASS SPECTRUM NO. 72-1
RELATIVE INTENSITY
220
205
180
177
136
57
MASS SPECTRUM NO. 72-2
RELATIVE INTENSITY
220
205
189
177
145
57
MASS SPECTRUM NO. 73
ETHYL PHTHALATE
RELATIVE INTENSITY
222
207
149
105
28

Mass Spectrum No. 73 has been positively identified as ethyl phthalate. The identification has been confirmed by direct comparison with published spectra (Part III, Section D-3). The molecular weight for Fraction GC-73 is 222. This is the second instance of phthalate identification among our unknowns.

Diethyl, dioctyl and dibutyl phthalates are all used as plasticizers,[70] however dioctylphthalate is the most common one. At this stage of our research great care was taken to avoid contamination of any sort and there is no doubt in our minds that the phthalatesidentified were indigenous to our heavy oil samples. Dioctylphthalate was the only phthalate from petroleum that had been previously reported in the literature.

Three mass spectra were selected from Fraction GC-75, i.e. MS 75-1, MS 75-3 and MS 75-4. The base peak for MS 75-1 and 75-3 appears at m/e = 57. Taking into consideration the GC data previously obtained (Table XXIII), a molecular ion was selected for each spectrum. A molecular ion at m/e = 240 was chosen for MS No. 75-1 and one at m/e = 254 for MS No. 75-3. MS No. 75-1 corresponds to $C_{13}H_{25}COOCH_3$ with a molecular weight of 240 and represents a cyclopentyl methyl ester. MS No. 75-3 corresponds to the formula $C_{14}H_{27}COOCH_3$ with a molecular weight of 254 and it is also a cyclopentyl methyl ester. The dominant series for both MS 75-1 and 75-3 is 57, 71, 85, etc., up

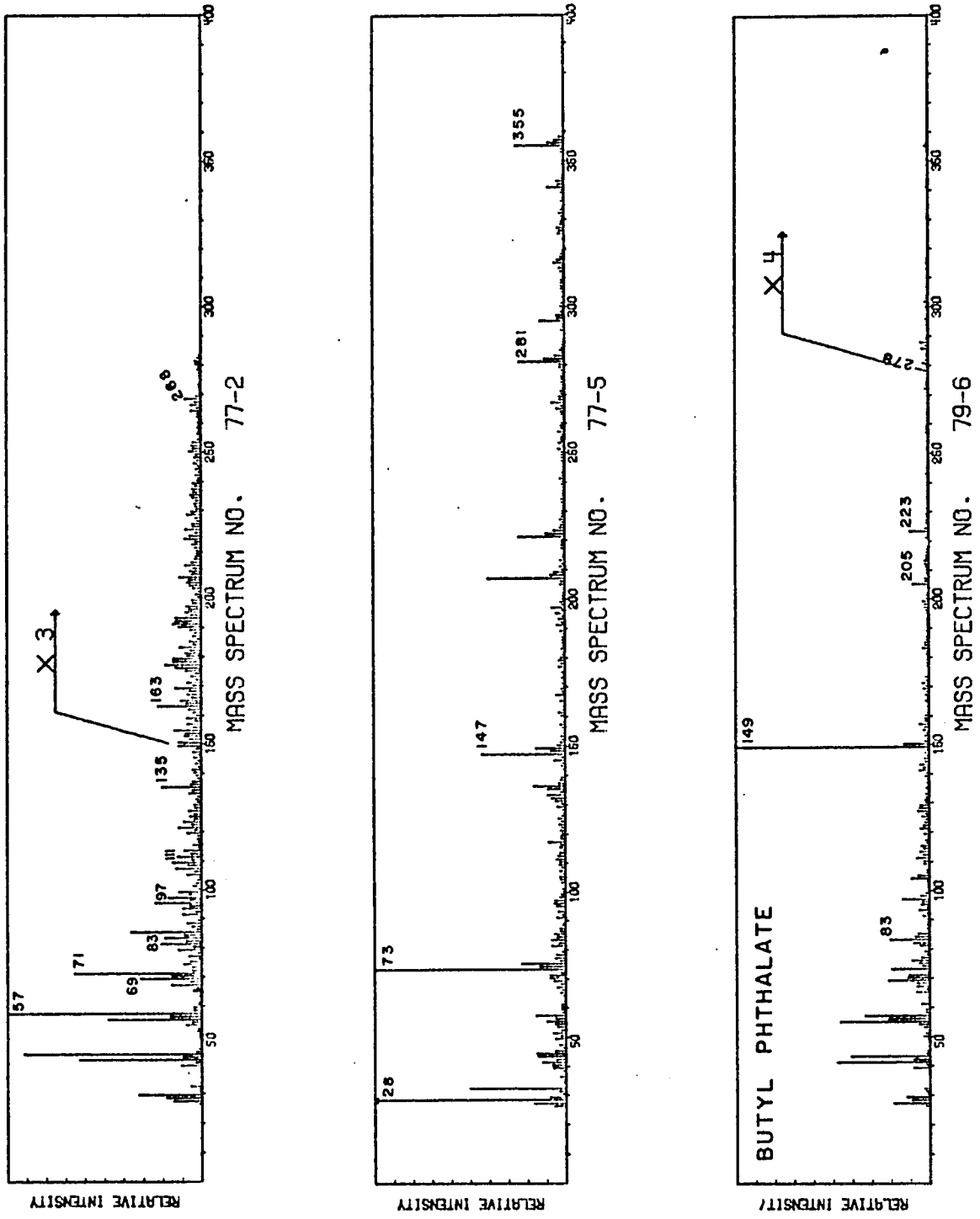

METHYL PALMITATE
RELATIVE INTENSITY
MASS SPECTRUM NO. 75-1
MASS SPECTRUM NO. 75-3
MASS SPECTRUM NO. 75-4
X 2
X 4
X 3
57
71
69
83
85
97
111
136
240
74
87
143
227
239
270
254
226
211

to m/e = 127, suggesting a long alkyl chain. However, the series 83, 97, 111, etc., also appears in both spectra. Tentatively, we can identify MS 75-1 and 75-3 as the mass spectra of cyclopentyl methyl esters with formulas $C_{13}H_{25}CO_2CH_3$ and $C_{14}H_{27}CO_2CH_3$, respectively. In each case the peak at m/e = 69 is larger than the one at m/e = 83.

MS No. 75-4 was identified as methyl palmitate, i.e. a n-C_{17} methyl ester (Part III, Section D-3). This spectrum shows a molecular ion at m/e = 270 and a base peak at m/e = 74. A fragment is observed at m/e = 239 (M-31). It is interesting to note that the most abundant normal fatty acid in sediments is C_{16}, i.e. palmitic acid.[71]

Two scans were selected from the mass spectra obtained for Fraction GC-77: MS 77-2 and MS 77-5. The base peak for MS 77-2 is at m/e = 57 ($C_4H_9^+$) and the one for MS 77-5 is at m/e = 73. It is logical to assume that MS No. 77-5 corresponds to bleed since some of its distinguishing features, such as the ion at m/e = 355, appear again in many of the other spectra. MS No. 77-2 could represent a cyclopentyl methyl ester of molecular weight 268 and formula $C_{15}H_{29}COOCH_3$. The series 69, 83, 97, 111, etc., is observed in MS 77-2. We have found some methyl esters in the literature such as methyl 10, 14, 18, 22-tetramethyltricosanoate which have 57 as the base peak.

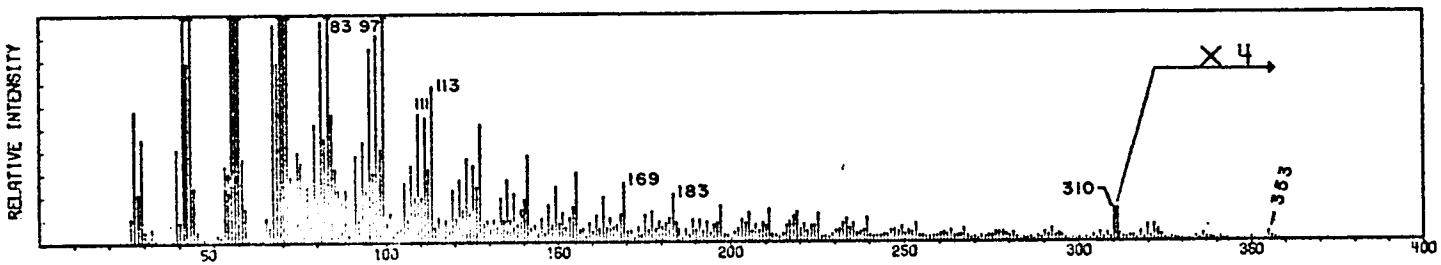
MS No. 79-6 was identical to that of butyl phthalate with a base peak at m/e = 149 and a parent peak at m/e = 278[62] (Part III, Section D-3).


BUTYL PHTHALATE
MASS SPECTRUM NO. 77-2
MASS SPECTRUM NO. 77-5
MASS SPECTRUM NO. 79-6
RELATIVE INTENSITY
X 3
X 4
57
71
69
83
97
111
135
163
260
73
28
147
281
355
149
205
223
278

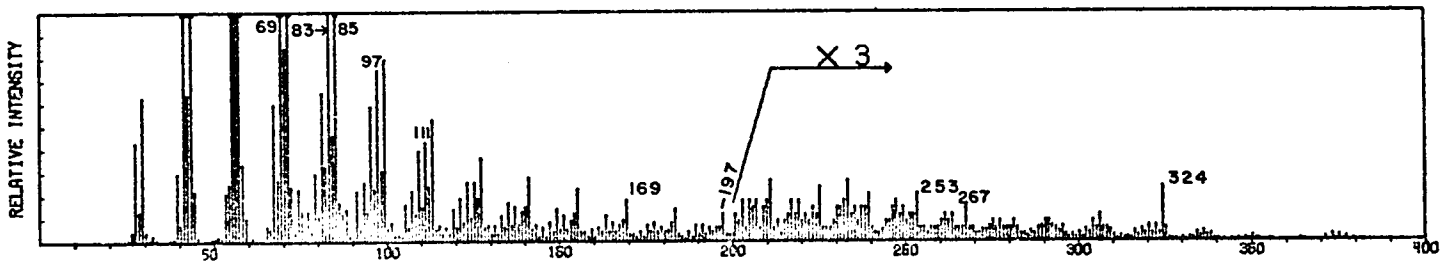
MS No. 80-3 shows a molecular ion at m/e = 310. This could indicate that this compound has a molecular weight of 310. The mass spectrum indicates the presence of a long saturated hydrocarbon; this compound corresponds to the formula $C_{22}H_{46}$. The series 83, 97, 111, etc., for olefinic and/or naphthenic fragments is observed in this spectrum but it could be due to impurities. A characteristic feature of this spectrum is the occurrence of a high peak at m/e = 113; this is part of the saturated hydrocarbon series.

MS No. 81-2 shows a very distinct molecular ion at m/e = 324 and a series of peaks at 69, 83, 97, 111, etc. The peak at m/e = 83 is quite intense. This spectrum is tentatively identified as that of a cyclopentyl methyl ester with a molecular weight of 324 and formula corresponding to $C_{19}H_{37}CO_2CH_3$. The strong peaks at m/e = 253 and at m/e = 267 are part of the series $C_nH_{2n-2}CO_2CH_3$. The continuity of this series indicates that the cyclopentyl ring is near to the methyl ester group. The series of peaks at 57, 71, 85, etc., indicates a long paraffin chain. Sample GC-81 was the strongest peak in the gas chromatogram recorded during its collection.

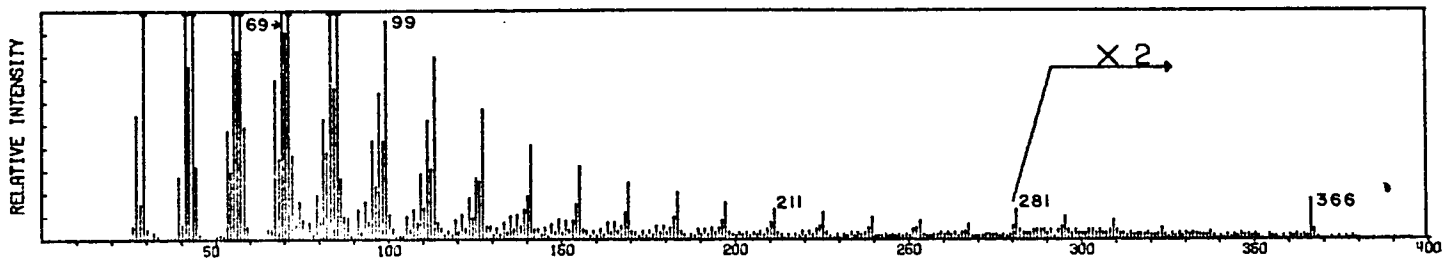
MS No. 82-5 looks like the mass spectrum of a straight-chain saturated hydrocarbon. The hydrocarbon in question is probably $C_{26}H_{54}$, a straight-chain hydrocarbon of molecular weight 366.



MASS SPECTRUM NO. 80-3



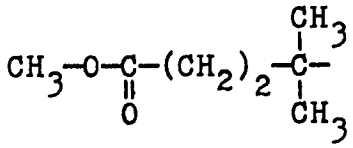
MASS SPECTRUM NO. 81-2



MASS SPECTRUM NO. 82-5

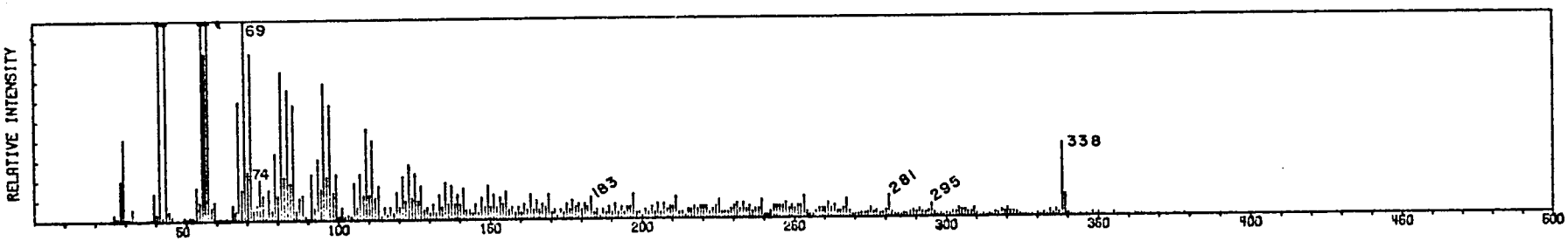
Both MS No. 83-1 and 83-2 show similar characteristics but MS No. 83-1 is the more intense of the two. The molecular ion of MS No. 83-1 appears at m/e = 338. The base peak at m/e = 69 suggests that this spectrum corresponds to that of a cyclopentyl methyl ester of molecular weight 338 and corresponding to the formula $C_{20}H_{39}CO_2CH_3$. A fragment corresponding to M-43 appears at m/e = 295. The series for C_nH_{2n-1} is observed in this spectrum as well as a small m/e = 74 peak.

The most interesting feature of MS No. 83-3 is the base peak at m/e = 129. This peak could be due to the methoxycarbonyl fragment formed with 4,5-cleavage:

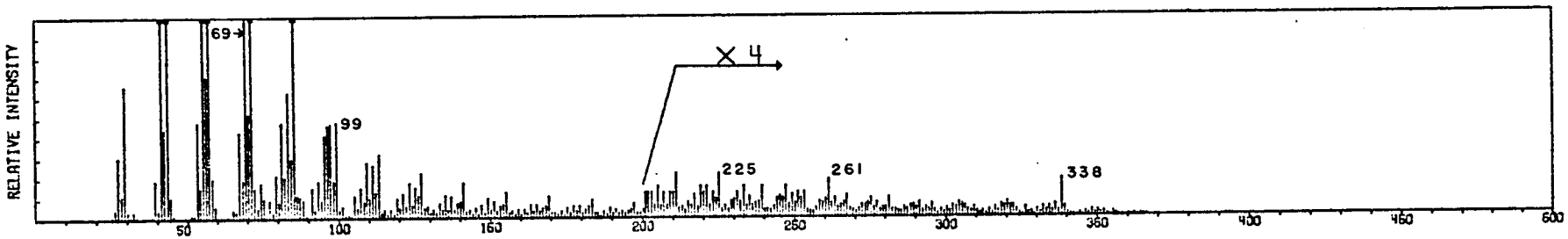


m/e 129

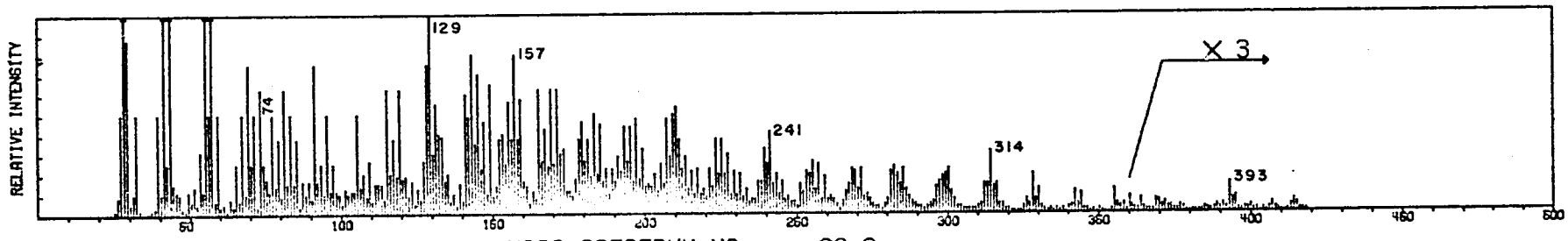
A strong tendency to cleavage of the bonds to the quaternary carbon atom has been repeatedly observed for methyl esters of acids containing a quaternary carbon atom.[72] The peak at m/e = 74 is not very large. This is due to the fact that since no hydrogen atoms are available on carbon atom 4, rearrangement with formation of ions of m/e = 74 does not occur to a significant extent.[72] The peaks at m/e = 74 and at m/e = 129 clearly indicate that



MASS SPECTRUM NO. 83-1



MASS SPECTRUM NO. 83-2



MASS SPECTRUM NO. 83-3

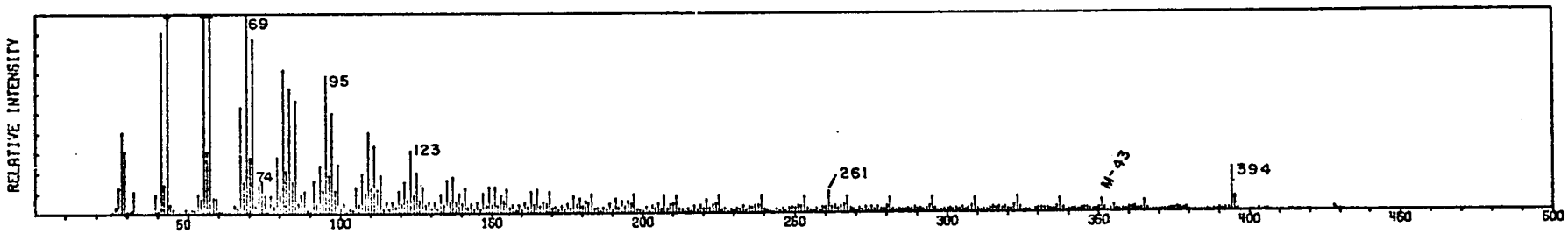
the compound is a methyl ester. However, no more details of the structure can be given because the rest of the spectrum is obscured by contamination of unknown nature.

MS No. 84-1 exhibited a molecular ion at m/e = 352 and a base peak at m/e = 69. This spectrum suggests cyclopentyl methyl ester of molecular weight 352 and corresponding to the formula $C_{21}H_{41}CO_2CH_3$. Other interesting feature in this spectrum is the series 69, 83, 97, 111, etc., and the ion at m/e = 309 (M-43). Most of the predominant ions in the lower mass region correspond to the series C_nH_{2n-1}. The cyclopentyl ring and the methyl ester group are most likely near the end of the hydrocarbon tail since the peak at m/e = 74 is fairly small.

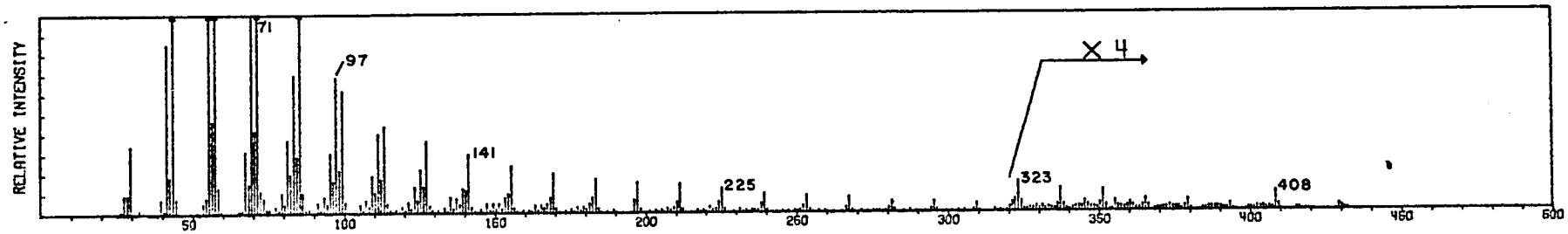
MS No. 85-4 had a molecular ion at m/e = 394 and a base peak at m/e = 69. This compound can be interpreted to be a cyclopentyl methyl ester with a molecular weight of 394 and corresponding to the formula $C_{24}H_{47}CO_2CH_3$. The (M-43) fragment is observed at m/e = 351. A small peak appears at m/e = 74. This spectrum also indicates a long chain paraffin as part of the methyl ester structure.

MS 88-3 exhibits a molecular ion at m/e = 408 and a base peak at m/e = 71. This compound appears to be a straight-chain hydrocarbon with formula $C_{29}H_{60}$. The spectrum exhibits a strong alkyl sequence all the way up to m/e = 408.

MASS SPECTRUM NO. 84-1



MASS SPECTRUM NO. 85-4



MASS SPECTRUM NO. 88-3

It is of interest to compare Mass Spectrum No. 83 (p. 185) with Mass Spectrum No. 83-1 (p. 196). Both were obtained from Fraction GC-83 but under two completely different sets of conditions, i.e. MS No. 83 was recorded using low resolution mass spectrometry and MS No. 83-1 was obtained by direct combination of GC-MS. Both show the molecular ion at m/e = 338 but MS No. 83 has 71 as its base peak while MS No. 83-1 showed 69 as its base peak.

Most of the spectra obtained by direct combination of GC-MS are not included in the text. The reason for discarding these mass spectra was that they were very poor either because (1) there was insufficient amount of sample in the capillaries for a good mass spectral analysis or, (2) the samples were contaminated by material derived from the stationary phase and by other unidentified extraneous materials.[73] The insufficient amount of sample could have been caused by poor collection efficiency. Apparently, collection efficiency from analytical gas chromatographs is usually not very high, and can be as low as 1%. In order to supply enough material for mass spectral analysis, repetitive collections of each sample were conducted hereafter.

Our GC spectra had given no indication that Dexsil was bleeding. From the mass spectral data, we conclude that Dexsil must have been bleeding at all temperatures. The samples analyzed by MS had been collected over a range

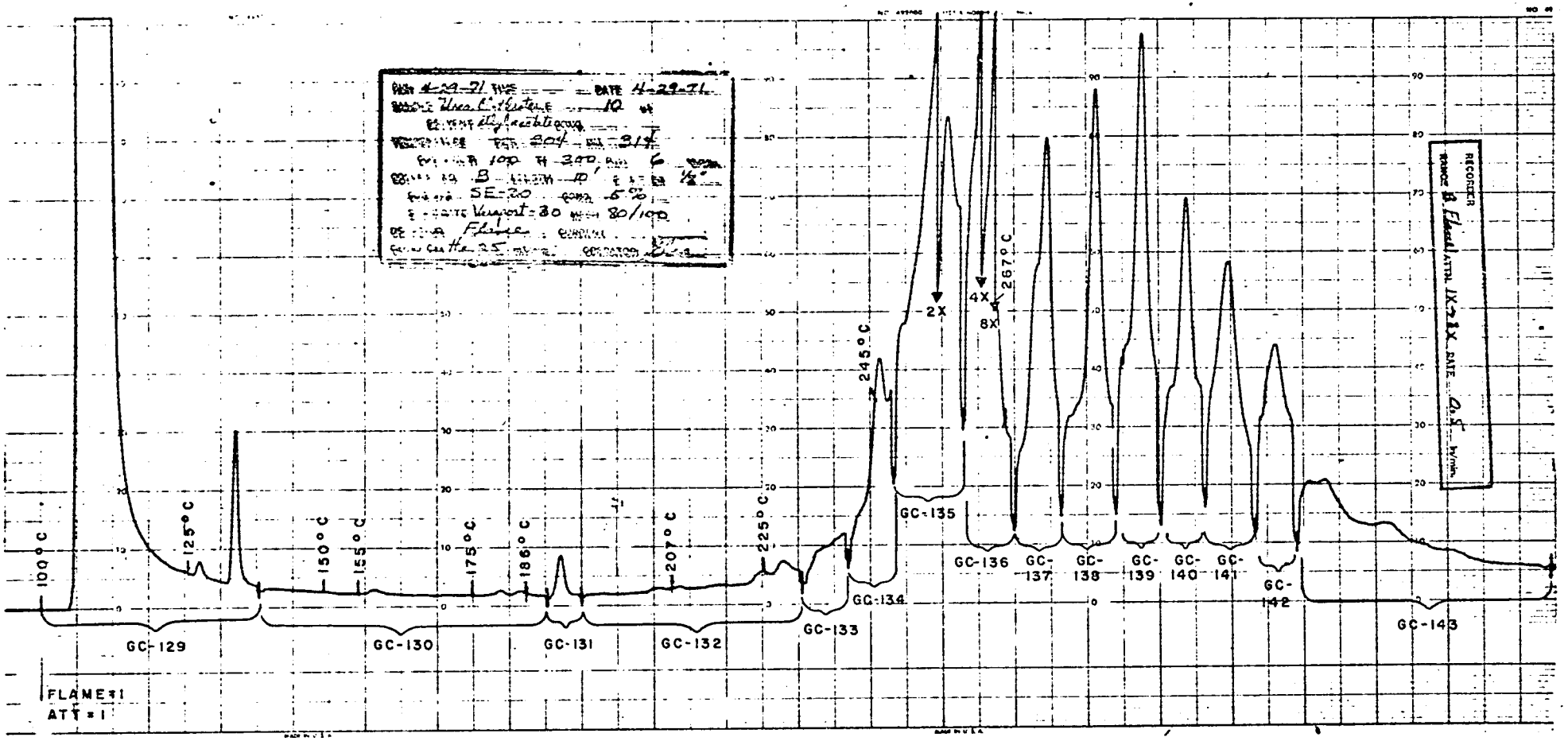
of temperatures but they all show signs of bleed (MS No. 77-5). Consequently, the use of Dexsil columns was discontinued. Since then it has been reported in the literature[74] that bleed problems make the liquid phase, Dexsil 300 GC, impractical for use in coupled gas chromatography-mass spectrometry. SE-30 was selected as a more desirable liquid phase for the GC separation of esters.

Fraction Urea Clathrate-6-D^+

GC Fractionation of Fraction Urea Clathrate-6-D^+

Fraction Urea Clathrate-6-D^+ - Fraction Urea Clathrate-6-D^+ is the result of pooling the fractions eluted with methanol and with hot hexane and methanol during the first and second urea chromatography, i.e. Fractions III, IV, IX and X (Figure 5). This step was justified on the basis that the gas chromatograms of these four fractions look very similar and that the amount of each individual fraction was so small that collection for mass spectrometric analysis would not have been feasible.

Final separation and purification of Fraction Urea Clathrate-6-D^+ was carried out by gas chromatography. Each fraction (GC-130 - GC-143) shown in Gas Chromatogram No. 20 was collected (Table XXIII). Fractions were collected as indicated by parentheses on the tracing of the gas chromatogram.



GAS CHROMATOGRAM NO. 20
FRACTION UREA CLATHRATE-6-D+

Mass Spectrometric Analysis of Collected Samples from Urea Clathrate-6-D^+ - Twelve samples collected from Urea Clathrate-6-D^+ were analyzed by mass spectrometry at Rice University.

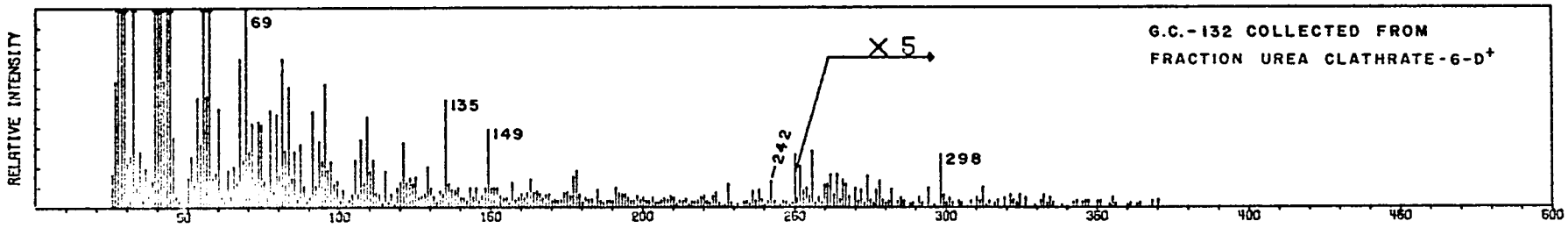
The mass spectrum of fraction GC-131 was identified[75] as methyl behenate, n-$C_{21}H_{43}CO_2CH_3$ (Part III, Section D-3). It shows its molecular ion at m/e = 354 and its base peak at m/e = 74.

The mass spectrum of fraction GC-132 exhibits a molecular ion at m/e = 298. The most important peaks were at m/e = 69, m/e = 135, and m/e = 149. This fraction contains as one of its constituents methyl stearate of formula $C_{19}H_{38}O_2$ and a molecular weight of 298 (Part III, Section D-3).

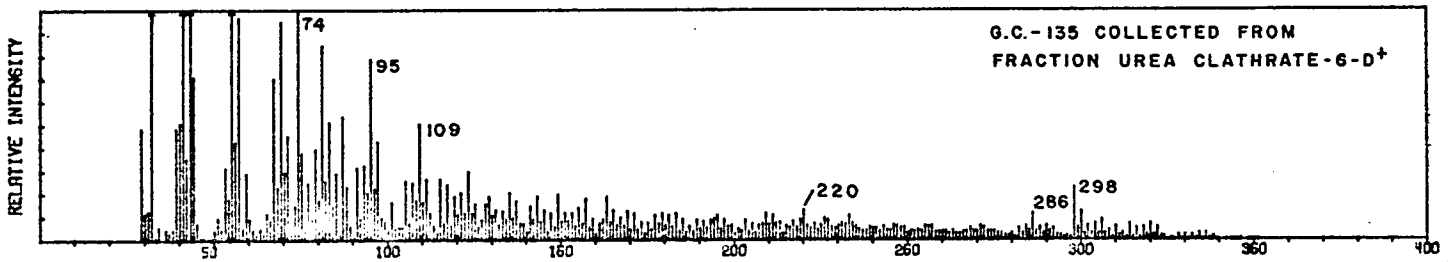
The mass spectrum of fraction GC-135 indicates that the main component of this fraction is methyl stearate, n-C_{19} ester (Part III, Section D-3). The molecular ion appears at m/e = 298 and the base peak at m/e = 74 (disregarding the peaks below m/e = 56).

The mass spectrum of fraction GC-136 exhibits two molecular ions, one at m/e = 298 and the other at m/e = 324. Based on the mass spectrum, fraction GC-136 consists of at least two components, one of which is methyl stearate as evidenced by the peak at m/e = 74 and the molecular ion at m/e = 298. The other component of fraction GC-136 is a cyclopentyl methyl ester with a molecular weight of

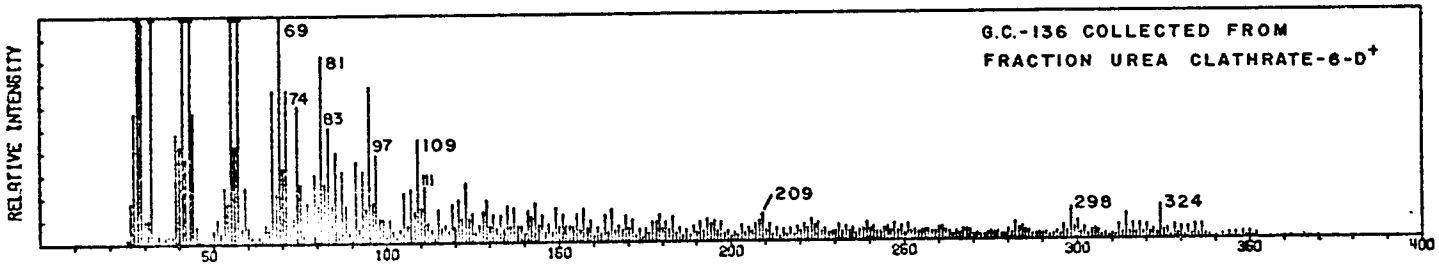
MASS SPECTRUM NO. 131



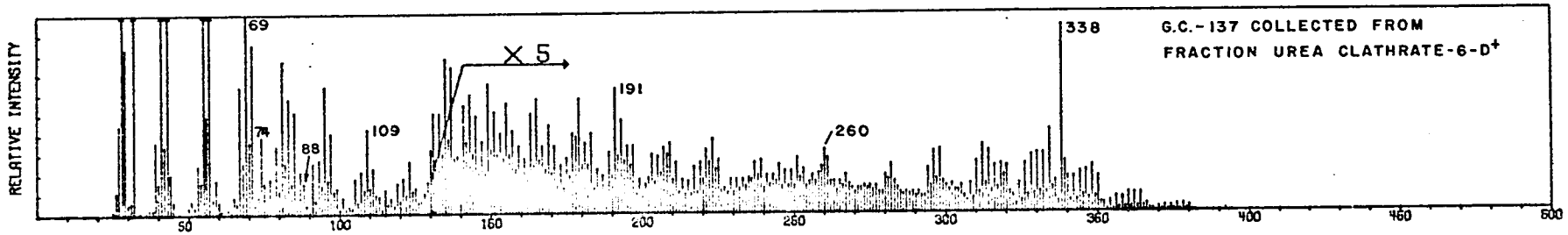
MASS SPECTRUM NO. 132



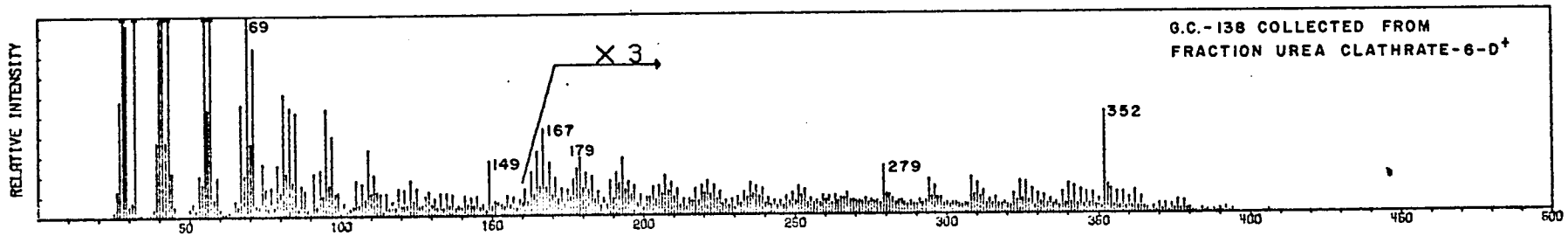
MASS SPECTRUM NO. 135



MASS SPECTRUM NO. 136



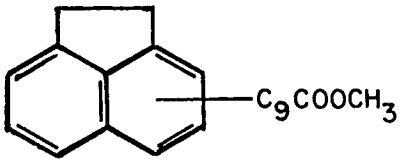
MASS SPECTRUM NO. 137



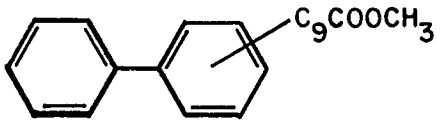
MASS SPECTRUM NO. 138

324 and formula $C_{21}H_{40}O_2$. The presence of a saturated cyclic ester is evidenced by the base peak of the spectrum at m/e = 69, its molecular ion at m/e = 324 and the series of naphthenic fragments at m/e = 83, 97, 111, etc.

The mass spectrum of fraction GC-137 shows a molecular ion at m/e = 338. The presence of impurities complicates this mass spectrum. The long list of odd-mass fragments running up to m/e = 123 is a good indication of a low concentration of ester in this sample. The peak at m/e = 74 is the most intense even-mass peak at the low end of the spectrum. The presence of a fairly large m/e = 88 suggests some $-CH(CH_3)COOCH_3$, too.[76] The base peak of the spectrum is at m/e = 69. The molecular weight of 338 also satisfies the formulas for the following acenaphthene (I) or biphenyl (II) methyl esters:



I



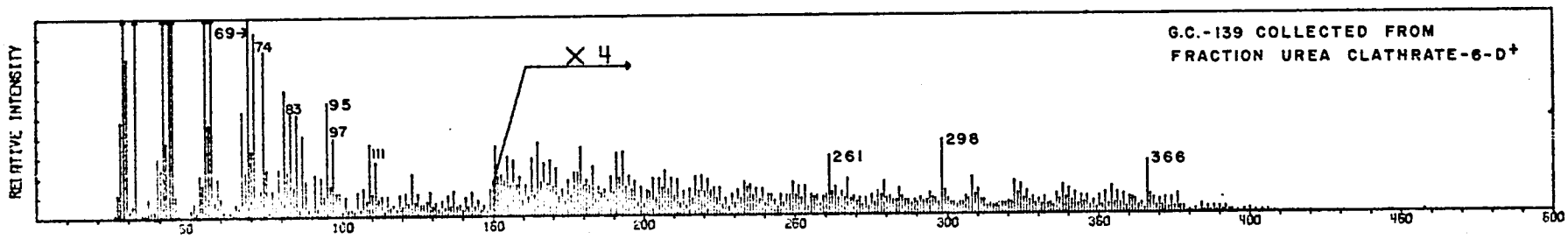
II

Either I or II, however, should show respectable peaks at m/e = 153 or 167, depending on the structure.[76] They are small; so the compound in question is a cyclopentyl methyl ester of carboxylic acid of formula $C_{20}H_{39}CO_2CH_3$ and molecular weight 338.

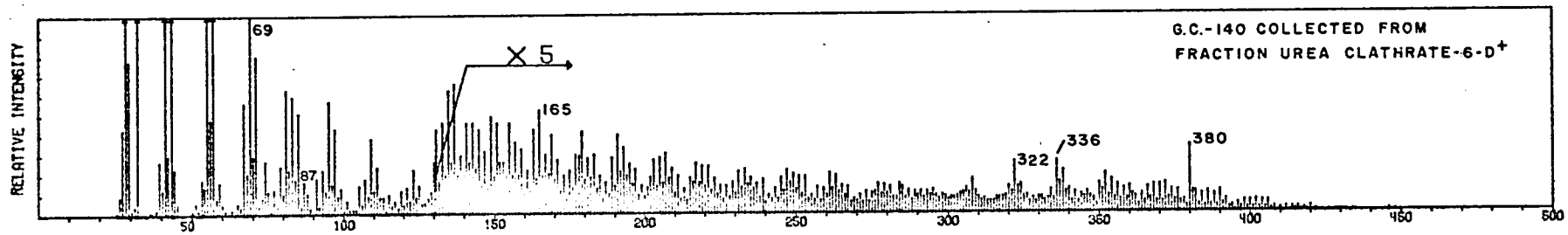
The mass spectrum of fraction GC-138 indicates the presence of two components, one of which is dioctylphthalate as evidenced by the fragments at m/e 149, 167, 179 and 279 (Part III, Section D-3). Dioctylphthalate must be a minor constituent based on the relative intensity of its fragment peaks. If dioctylphthalate were the major component, the base peak in the spectrum would be at m/e 149. The other component is characterized by its base peak at m/e = 69 and molecular ion at m/e = 352. This second component is probably the cyclopentyl methyl ester of molecular weight 352 and formula $C_{23}H_{44}O_2$.

The mass spectrum of fraction GC-139 shows the presence of a rearrangement peak at m/e 74. This rearrangement peak and the molecular ion at m/e = 298 are evidence for methyl stearate. This spectrum is very complex and other components are also present. A second component is characterized by a molecular ion at m/e = 366, base peak at m/e = 69 ($C_5H_9^+$) and the series of naphthenic fragments at 69, 83, 97, 111, etc. This second component is expected to be a cyclopentyl methyl ester of molecular weight 366 and formula $C_{24}H_{46}O_2$. Fraction GC-139 had given the strongest peak in the gas chromatogram recorded during the collection runs.

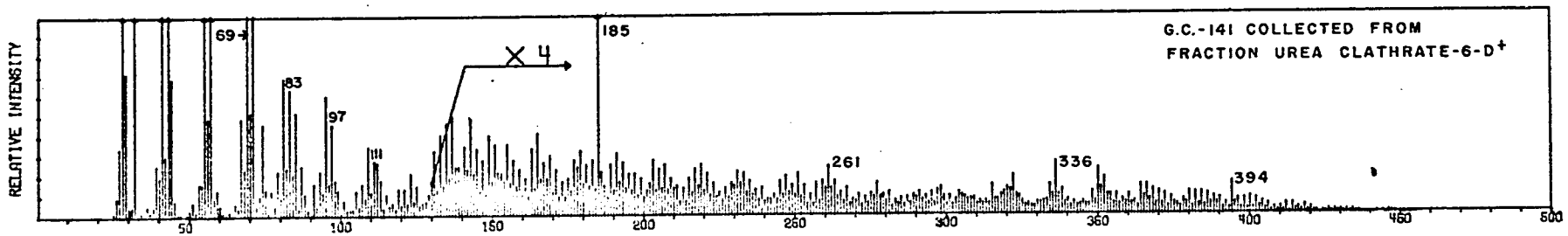
The mass spectrum of GC-140 presented a molecular ion at m/e = 380, base peak at m/e = 69 and fragmentation in the C_nH_{2n-1} series. This compound is probably a cyclopentyl methyl ester of molecular weight 380 and formula $C_{25}H_{48}O_2$.



MASS SPECTRUM NO. 139



MASS SPECTRUM NO. 140



MASS SPECTRUM NO. 141

The mass spectrum of fraction GC-141 exhibits a base peak at m/e = 69, and a molecular ion at m/e = 394. The very strong peak at m/e 185, is a member of the $C_nH_{2n-1}COOCH_3$ series, and corresponds probably to a branch at a position 9 carbon atoms removed from the ester group. The series of naphthenic fragments at m/e = 83, 97, 111, etc., is also observed in this spectrum. This compound is expected to be a cyclopentyl methyl ester of molecular weight 394 and formula $C_{26}H_{50}O_2$. Another feature of this mass spectrum is fragmentation at m/e 73, 87, 101, etc.

At this point we have identified in Fraction Urea Clathrate-6-D^+ the following compounds: methyl stearate, methyl docosanoate, dioctylphthalate and a series of six saturated cyclic methyl esters with molecular weights as low as 324 and differing by 14 mass units. Lack of reference mass spectra of related cyclopentyl methyl esters of carboxylic acids made positive identification difficult. Commercially available compounds of similar structure in this molecular weight range (324-394) have not been located to date. "The correct interpretation of mass spectral data from alicyclic acids from petroleum is quite difficult. Ring size or nature and number of substituents on the ring are difficult to deduce from the mass spectrum alone of a cyclic ester. A further complication arises from the observation that stereochemical detail of substitution on the ring seems to have a pronounced effect in the intensity of certain peaks."[77]

In the interpretation of some of the mass spectra described above, the question of whether the compounds were unsaturated or cyclics was considered. It is very unlikely that unsaturated or olefinic carboxylic acids would be originally present in petroleum. Olefinic structures are rarely found in petroleum. It is probable that even if they were present at one time in petroleum, such acids would have become unstable and decomposed. The spectra are thus proposed to correspond to saturated cyclic (5 membered rings) methyl esters of carboxylic acids. The cyclopentyl structure is favored because cyclohexyl compounds usually have very large peaks at m/e = 83; in most cases m/e = 83 is the base peak. In our spectra the base peak is usually at m/e = 69.[78] The peak at m/e = 69 represents an unsubstituted cyclopentyl group.[79]

Because n-paraffin parent peaks give the same nominal masses as saturated cyclic methyl esters, two of our spectra were compared to those of straight-chain hydrocarbons. The mass spectrum of fraction GC-137 was not the same as that of the normal hydrocarbon, n-tetracosane (molecular weight 338)[63] (Part III, Section D-3). The mass spectrum of fraction GC-138 does not correspond to that of n-pentacosane, molecular weight 352[64] (Part III, Section D-3).

The aliphatic methyl esters of long chain fatty acids with a naphthenic structure in the chain that we have identified are in the $C_nH_{2n-1}COOCH_3$ series, corresponding to

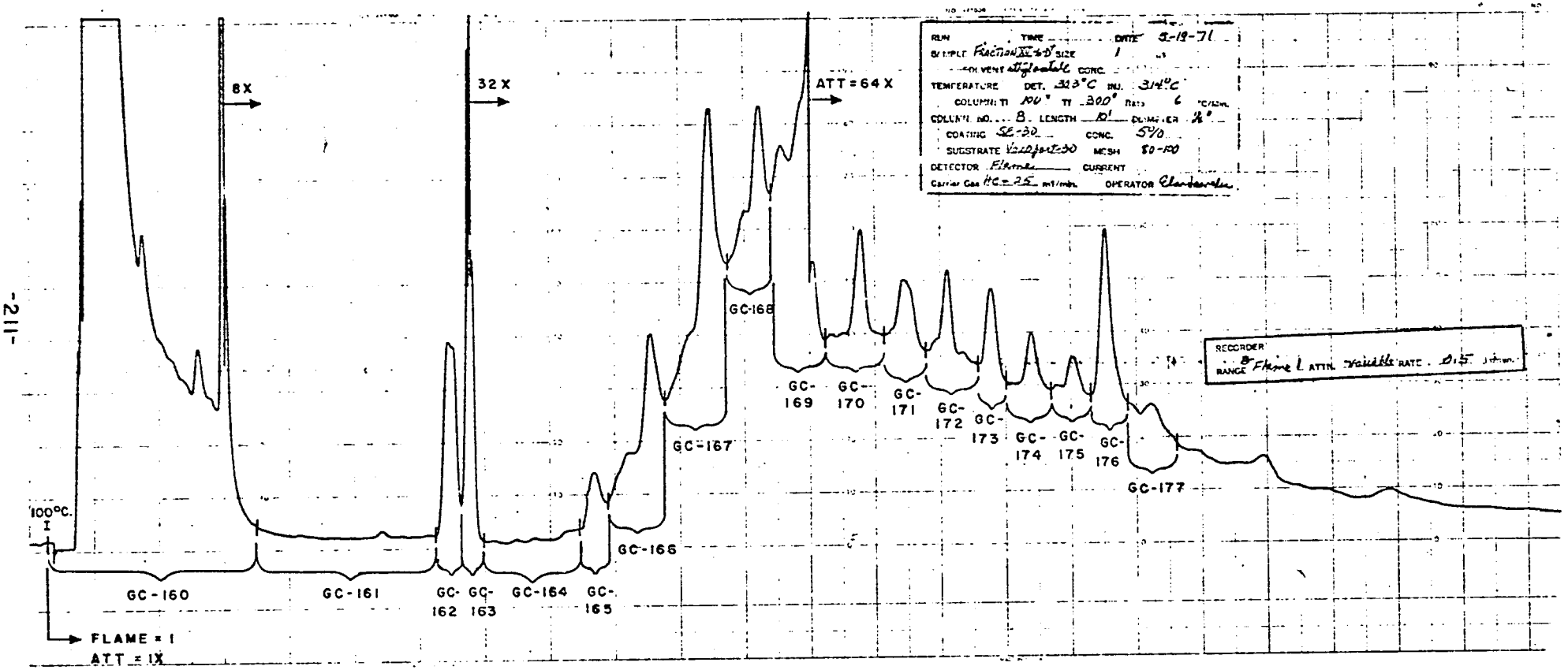
n equal 19 through 24. "It must be noted that n-paraffin parent peaks give the same nominal masses. Absence of major paraffinic fragmentation peaks indicates, however, that these parent peaks are not due to paraffins."[80] The more detailed interpretation of the mass spectra were conducted for the fractions 137, 139, 140 and 141. To avoid repetition, the other analyses were briefer.

Fraction XV-6-D^+

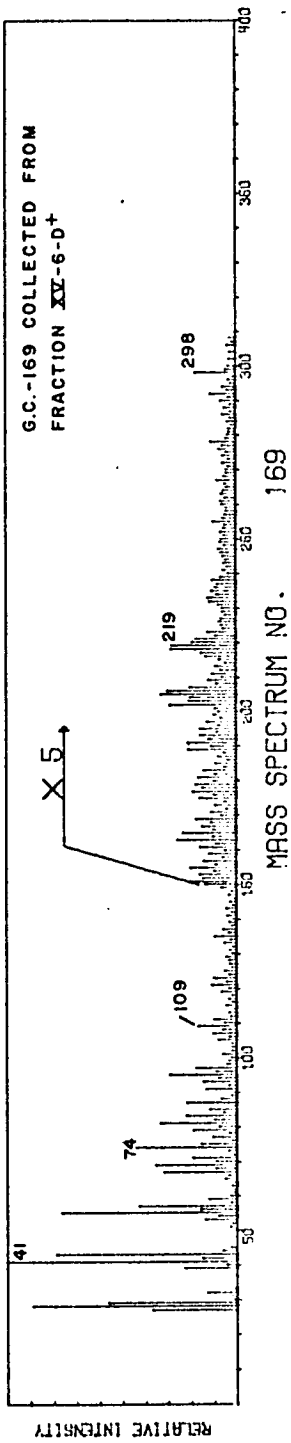
GC Fractionation of Fraction XV-6-D^+ - Fraction XV-6-D^+ is the clathrated material resulting from performing a thiourea adduction on Fraction XI-6-D^+ in the presence of cyclohexane. Fraction XV-6-D^+ was subjected to gas chromatography and 14 samples, numbered GC-162 - GC-176, were collected. The collected samples had carbon numbers ranging from C_{12}-C_{26}. Gas Chromatogram No. 21 showed the successful resolution of Fraction XV-6-D^+. The background was high for this particular fraction. Data based on the interpretation of Gas Chromatogram No. 21 are presented in Table XXIII.

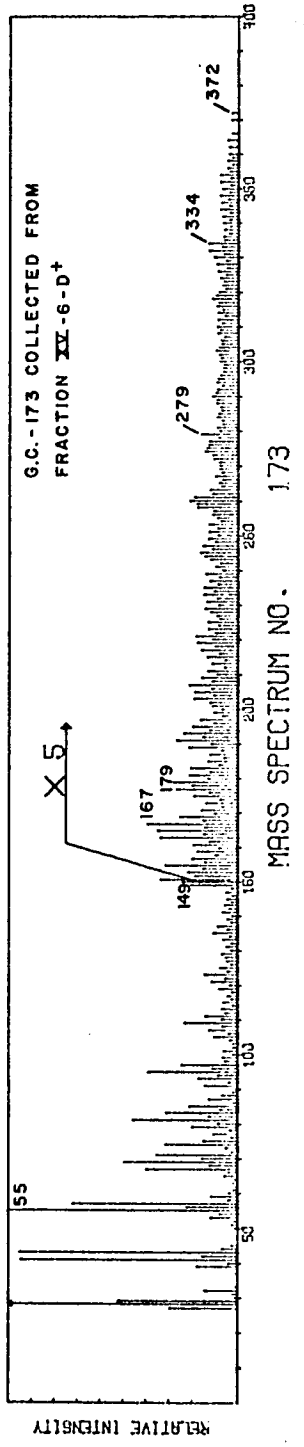
Mass Spectrometric Analysis of Collected Samples from Fraction XV-6-D^+ - The major peaks were collected and their low resolution mass spectra were determined at Beacon Research Laboratories using the solid inlet system.

The mass spectrum of fraction GC-169 shows a molecular ion at m/e = 298 and a fairly large peak at m/e = 74. The major component of this fraction is methyl stearate (Part III, Section D-3).



GAS CHROMATOGRAM NO. 21
FRACTION XV-6-D+


G.C.-169 COLLECTED FROM
FRACTION XV-6-D+
X 5
41
74
109
219
298
RELATIVE INTENSITY
MASS SPECTRUM NO. 169


G.C.-173 COLLECTED FROM
FRACTION XV-6-D+
X 5
55
149
167
179
279
334
372
RELATIVE INTENSITY
MASS SPECTRUM NO. 173

The mass spectrum of fraction GC-173 shows a series of ions at m/e = 149, 167, 179 and 279 which indicates that dioctylphthalate is a minor constituent of this mixture.

The rest of this group of samples analyzed by mass spectrometry exhibited very complex spectra, suggesting that the samples were mixtures. These spectra are not included because it was not possible to identify the components of the mixtures.

Conclusions

In summary, the number of compounds identified from each fraction were: Fraction XIII-6-D-2, 2 compounds; Fraction II-6-D^+, 11 compounds; Fraction Urea Clathrate-6-D^+, 8 compounds; and Fraction XV-6-D^+, 2 compounds. Fraction II-6-D^+ (Figure 5, p. 111) yielded the largest number of identifiable compounds. Any future work should be carried out on Fraction II. Fraction II was eluted with ether during the first urea chromatography. A list of the compounds identified from each fraction follows:

Fraction No.	Compounds Identified
XIII-6-D-2	Aromatic methyl ester with double bond in the chain, MW = 232; dioctylphthalate.
II-6-D^+	Seven cyclopentyl methyl esters, MW = 240-394; ethyl phthalate; butyl phthalate; methyl palmitate; aromatic methyl ester, MW = 220.
Urea Clathrate-6-D^+	Six cyclopentyl methyl esters, MW = 324-394; dioctylphthalate; methyl docosanoate; methyl stearate.
XV-6-D^+	Dioctylphthalate; methyl stearate.

Experimental

Silylation Procedure - Silyl esters were prepared by adding 1.0 ml of Tri Sil/BSA in pyridine (or 0.5 ml of Trimethylsilyl reagent) and 0.5 ml of solvent (either dimethylformamide or pyridine) to 10 mg of the acids in Fraction 8-D-1. The reaction mixture was vigorously agitated and stoppered.

Methylation of Fraction 8-D-1 - Methyl esters of Fraction 8-D-1 were prepared by adding about 10 mg of the acid mixture to 1 ml of reagent (either BCl_3 in methanol or BF_3 in methanol) allowing the reaction mixture to boil for two minutes. About 10 ml of water were then added to the reaction mixture. The mixture was extracted with 15 ml of petroleum ether. The separatory funnel was shaken vigorously and the layers allowed to separate. The solvent was removed from the petroleum ether layer under reduced pressure at about 60° C.

Gas Chromatography - The ester mixtures from five different esterification techniques were examined by gas chromatography. The instrument used was a Varian Model 204-1B flame detector gas chromatograph. A 5-foot column, 1/8" O.D., packed with 5% SE-30 on Chromosorb W, 80-100 mesh, was used. The carrier gas (helium) had a flow rate of 25.5 ml/min. throughout. The runs were temperature programmed at 10° C/min. between 50 and 300° C.

Infrared Spectrometry - IR spectra of Fraction 8-D-1 and of methyl esters derived from Fraction 8-D-1 were obtained in CCl_4. The spectra were recorded on a Beckman Spectrophotometer Model IR-4 using 0.4 mm NaCl cells.

Methylation Procedure Using BF_3/MeOH - All methyl esters used in the rest of our work were prepared by the method described below.

A 0.762 g sample of Fraction 6-D-2, dissolved in a small amount of benzene, was treated with 15 ml of BF_3/MeOH for 3 to 4 hours.[59] The solution was then concentrated, 100 ml of water added, and extracted four times with ether. The combined ether extracts were reduced in volume, washed with dilute sodium hydroxide and then with water. The water wash and the sodium hydroxide wash were each extracted 5 times with ether. The ether was washed with water, then dried over anhydrous calcium sulfate. Removal of solvent at reduced pressure yielded 0.439 g of methyl esters.

Urea Chromatography[53] - A typical procedure followed for urea chromatography is described. The methyl esters of Fraction 6-D-2 were separated into several fractions by urea chromatography. A urea-methanol clathrate was prepared by dissolving 2 g of urea in 50 ml of methanol and then adding 0.439 g of the methyl esters of the acids in Fraction 6-D-2. A small amount of ether was also required

to dissolve the sample. The esters were shaken with 4 g of Celite 545 and very slowly evaporated to dryness on a rotary evaporator. The resulting solid was powdered and packed dry into a chromatographic column. N-hexane was slowly added until the column was saturated. Elution with the same solvent yielded the nonclathrated material (0.404 g). Because of the limited miscibility of n-hexane and methanol, ether was used as an intermediate solvent, and eluted small quantities of mixed compounds (0.010 g). Elution with methanol yielded the clathrated material (0.004 g). However, urea was also eluted by the ether and the methanol. Therefore these fractions were concentrated, poured into water and extracted three times with ether and with hexane. The combined hexane-ether layers were washed with water, dried and evaporated. Finally, the columns were extruded and boiled with methanol and hexane. After filtration, the hot methanol-hexane solution was treated to remove any urea present.

Thiourea Adduction Using Cyclohexane as an Inducer - One of the fractions resulting from thiourea chromatography, Fraction XI-6-D^+ was subjected to thiourea adduction using cyclohexane as an inducer. Fraction XI-6-D^+ (0.6 g) was added to 10 ml methanol saturated with 1.6 g thiourea, then 1 ml cyclohexane was added and mixed well. After standing at room temperature for 24 hours, the needle-shaped crystals of the adduct were filtered. The crystals were washed

carefully with two 20-ml portions of cyclohexane, and dried. The adducted esters were liberated by addition of 250 ml of warm water. The liberated esters were extracted with three 75-ml portions of ether. Washing and drying of the extract, followed by removal of solvent, left 0.015 g (2.5% recovery) of adducted esters.[21] The adducted esters were labelled Fraction XV-6-D^+.

The nonadducted esters, obtained as a filtrate from collection of the adducted, were washed with 250 ml of warm water. The nonadducted esters were extracted with 75-ml portions of ether and the extract was washed with water, then dried. Removal of solvent from extract left 586 mg of nonadducted esters (Fraction XVI-6-D^+).

Mass Spectrometry at Rice University - Some of the fractions derived from heavy oils were analyzed by mass spectrometry at Rice University. Low resolution mass spectra were obtained on the analytical mass spectrometer Model 21-110B H.R.M.S. (Consolidated Electrodynamics Corp.), using photographic recording. All samples were introduced into the mass spectrometer via a direct insertion probe at ion source temperatures from 83° to 320° C. The energy of the electrons was 70 volts; acceleration potential, 8 Kv; multiplier reading, 180.

Gas Chromatography Combined with Low Resolution Mass Spectrometry at Baylor College of Medicine - A number of fractions collected from the GC and consisting of derivatives of heavy oils were examined by combined GC-MS at Baylor College of Medicine. All combined GC-MS spectra included in this text were recorded at Baylor College of Medicine. Low resolution mass spectra were obtained on LKB 9000 combination gas chromatograph-mass spectrometer. Electron energy was 70 eV for all spectra except for MS 72-1, 72-2 and 73 in which cases it was 20 eV; and the ionizing current was 60 μA. The gas chromatographic inlet system was equipped with a 6 ft.by 0.25 in. column packed with 1% OV-17; injection temperature, 100-130° C. Fractions trapped from 15 percent Dexsil on Varoport-30 were reinjected into the gas chromatography-mass spectrometry apparatus.

Mass Spectrometry at Texaco Research Laboratories - Low resolution mass spectra of the rest of the fractions derived from the heavy oils were run at Texaco Research Laboratories, Beacon, New York. Spectra were obtained on a Consolidated Model 21-104 mass spectrometer equipped with a solid inlet system for sample introduction.

Gas Chromatography - All the analytical gas chromatography was conducted on a modified Aerograph 204-B flame detector gas chromatograph. The analytical standards used for comparison purposes were obtained from Poly Science Corp.

The standards consisted of the following compounds: methyl caproate, methyl heptanoate, methyl caprylate, methyl pelargonate, methyl caprate, methyl undecanoate, methyl laurate, methyl myristate, methyl palmitate, methyl stearate, methyl arachidate, methyl behenate, methyl undecylenate, methyl oleate, and methyl linoleate.

The samples were dissolved in ethyl acetate or in ether and injected directly into the gas chromatograph. Four different types of columns were used: (1) a 12 ft. x 1/8 in. column, packed with 5 percent SE-30 on 80-100 mesh Chromosorb W and 5 percent GC SE-30 on 80-100 mesh Varoport-30; (2) a 10 ft. x 1/8 in. column, packed with 5 percent SE-30 on 80-100 mesh Varoport-30; (3) a 6 ft. x 1/8 in. column of 15 percent Dexsil on 80-100 mesh Varoport-30; and (4) a 12 ft. x 1/8 in. column of 15 percent Dexsil on 80-100 mesh Varoport-30. The fractions were programmed from 100 to 300° C at 6° C/min., and maintained at that temperature for the remainder of the run; the flow rate ≈25 cc/min.

Infrared Spectrometry of Undecanoic Acid and its Methyl Ester -

IR spectra were recorded on a Beckman IR-4 spectrophotometer, using CCl_4 solutions in NaCl cells. The solvent was compensated by using a variable path length cell in the reference beam. The undecanoic acid and the methyl undecanoate used were analytical standards from Poly Science Corporation.

Gas Chromatography of Fraction XIII-6-D-2 - Fraction XIII-6-D-2 was further fractionated by gas chromatography using packed 12' x 1/8" columns of 5% SE-30 on 80-100 mesh Chromosorb W and 5% GC SE-30 on 80-100 mesh Varoport-30. The oven temperature was programmed at 6° per minute from 100° to 300° and maintained isothermally at that temperature for the remainder of the run. For collection of esters, samples of 10 μℓ were injected.

Gas Chromatography and IR Spectrometry of Possible Sources of Dioctylphthalate -

The possibility of having introduced dioctylphthalate into our collected samples as a contaminant was investigated. Samples from possible sources of contamination were analyzed by IR spectrometry and by gas chromatography. The IR spectra were determined versus air with a Beckman IR-4 spectrophotometer in 0.4 mm fixed path length cells and using CCl_4 as a solvent unless otherwise specified. IR Spectrum No. 34 was run on AgCl discs, using benzene as a solvent. The GC spectra were run on a Varian GC 204-1B using 6' x 1/8" columns of 15% Dexsil on Varoport-30.

Silica Gel Chromatography of Fraction D - Fraction D, before esterification, was subjected to further separation by passage through silica gel in an attempt to further purify Fraction D. For silica gel chromatography, Grade 923, silica gel mesh 100-200 of Grace/Davison Company was used. To remove any contaminants present in the adsorbent, the silica gel column was rinsed with benzene, ether, and

mixtures of the two. Then a solution of 0.75 g of Fraction 6-D-6 in benzene was placed on a silica gel column (60 by 4.6 cm I.D.). Elution with benzene, ether, and methanol produced three fractions as shown in Figure 5. Fractions were reduced using a rotary flash evaporator. Total recovery was 99.6% of Fraction 6-D-6. The same procedure was followed for Fraction 6-D-5. The combined ether fractions from both columns were combined and then chromatographed on 93 by 2 cm I.D. silica gel columns. The various fractions obtained from this second silica gel chromatography are shown in Figure 5. Please note that Fractions 6-D-5 and 6-D-6 are the only ones that were passed twice through silica gel. The reason for this was that at the time that we were running the first silica gel chromatography, we were not fully aware of all the precautions that could be taken to prevent contamination. As can be seen from Figure 5, the ether fraction resulting from passing Fractions 6-D-5 and 6-D-6 twice through silica gel was labelled Fraction 6-D^{+}.

Each of the remaining Fractions D in benzene solutions were passed over a silica gel column only one time and eluted with benzene (Fraction D-Benzene), ether (Fraction D-Ether), and methanol (Fraction D-Methanol). The corresponding ether fractions were then methylated. A list of the weights of the fractions thus obtained is compiled in Table XXIV.

TABLE XXIV

SILICA GEL CHROMATOGRAPHY AND METHYLATION OF FRACTION D

Fraction No.	Weight, g
6-D_2	4.00
6-D_2-Benzene	0.43
6-D_2-Ether	2.70 (2.58)*
6-D_2-MeOH	0.68
Methyl Esters of 6-D_2-Ether	2.62
6-D_3	3.74
6-D_3-Benzene	0.24
6-D_3-Ether	2.78 (2.66)
6-D_3-MeOH	0.55
Methyl Esters of 6-D_3-Ether	2.72
6-D_4	3.97
6-D_4-Benzene	0.28
6-D_4-Ether	2.88 (2.77)
6-D_4-MeOH	0.66
Methyl Esters of 6-D_4-Ether	2.83

*The values in parentheses are the actual amount of Fraction 6-D_2-Ether that was methylated.

Infrared Spectrometry of Various Isolated Fractions -

A series of IR spectra (IR No. 35 through IR No. 44) were obtained of fractions taken at various stages of our separation scheme. This study was mainly conducted in an effort to detect the presence of acids or esters (phthalates, most probably) in the various fractions. Infrared spectra were obtained on 1-2% solutions in carbon tetrachloride (unless otherwise specified) using a Beckman Model IR-4 spectrophotometer in 0.4 mm NaCl cells. The solvent was compensated by using a variable path length cell in the reference beam. Some samples were examined using both CS_2 and CCl_4, the reason being that CS_2 is a better solvent to use for detecting aromatic bands. It is practically impossible to properly compensate for CCl_4 around 800 cm^{-1}.

Separation Scheme Applied to Fraction 6-D^+ (Figure 5) -

Fraction 6-D^+ was methylated and subjected to urea and thiourea chromatographies. The total recovery from the first urea chromatography on the methyl esters of Fraction 6-D^+ was 99.2%. The total recovery from the thiourea chromatography of Fraction VII-6-D^+ was 97.0%. Another step added to our complex separation scheme was to divide Fraction I-6-D^+ into compounds soluble in n-hexane (Fraction V-6-D^+) and those insoluble in n-hexane, but soluble in benzene (Fraction VI-6-D^+). This step was a means of narrowing the fractions before injecting them

into the gas chromatograph. Fraction VI-6-D^+ should contain the higher molecular weight methyl esters based on solubility data of methyl esters in polar and nonpolar solvents. Figure 5 shows the exact separation procedure followed in the case of Fraction 6-D^+.

Gas Chromatography of Fraction II-6-D^+ - Fraction II-6-D^+ was further fractionated by gas chromatography using 12' x 1/8" columns containing 15% Dexsil on 80-100 mesh Varoport-30.

Gas Chromatography of Fraction Urea Clathrate-6-D^+ - Final separation of Fraction Urea Clathrate-6-D^+ was carried out on gas chromatography columns of 5% SE-30 on 80-100 mesh Varoport-30 (10' x 1/8") programmed at 6°C/minute with a helium flow rate of 25 ml/minute. The oven temperature was programmed from 100° C to 300° C and maintained isothermally at that temperature for the remainder of the run. A total of six runs, using 10 μℓ charges, was made on the 1/8" O.D. column.

Gas Chromatography of Fraction XV-6-D^+ - Fraction XV-6-D^+ was subjected to gas chromatography on a 10' x 1/8" column of 5% SE-30 on Varoport-30, 80-100 mesh. The oven temperature was programmed at 6° per minute from 100° to 300° and maintained isothermally at that temperature for the remainder of the run. Seven consecutive runs were conducted in order to collect enough samples for mass spectral analysis.

Summary

This research involved the extraction of a high viscosity crude oil (3185 centistokes at 100° F) with sodium hydroxide-alcohol-water and separation of polar compounds by ion exchange and silica gel chromatography. The polar fraction, consisting mostly of acids and esters, was further separated by urea and thiourea chromatography and by gas chromatography. Identification of individual compounds was achieved mostly by mass spectrometry.

Our work constitutes the first detailed study of acids and esters present in a high viscosity crude oil. The polar fraction represented 1.6 wt. % of total crude oil with a number average molecular weight of 570.

This study has isolated cyclic acids from petroleum from C_{14}-C_{25}. The actual identification of these compounds involved their methyl esters because this simplified the gas chromatographic work. It is assumed in this work that all the methyl esters identified were originally present as carboxylic acids in petroleum. Reports in the literature of cyclic acids from petroleum have only been accomplished with up to C_{12} cyclic acids. The general formula for cyclic acids is $C_nH_{2n-2}O_2$ or $C_nH_{2n-1}COOCH_3$.

Three different normal fatty acids, C_{16}, C_{18} and C_{22}, were also identified. The formula for these acids is $C_nH_{2n}O_2$. One of the acids, stearic acid (n-C_{18}), was observed in five different fractions.

Phthalate esters are beyond doubt also present in these crude oils. Dioctylphthalate was observed in at least three different fractions. The other two phthalates, ethyl and butyl, have not been previously found in petroleum to my knowledge.

Also the mass spectra of two reference compounds, methyl cyclopentylpropanoate and methyl 2-cyclopentyl-n-hexanoate were recorded. A list of the acids and esters identified by mass spectrometry is included in Table XXV.

TABLE XXV

ANALYSIS OF ESTERS FROM HEAVY OILS BY MASS SPECTROMETRY

Fraction No.	MW	Formula	Identification	Compound As Present In Heavy Oil
73	222	$C_{12}O_4H_{14}$	Ethyl phthalate	Phthalate esters
79-6	278	$C_{16}O_4H_{22}$	Butyl phthalate	
9; 138; 173	390	$C_{24}O_4H_{38}$	Dioctyl phthalate	
75-4	270	$C_{15}H_{31}CO_2CH_3$	Methyl palmitate	Normal fatty acids
132; 135; 136; 139; 169	298	$C_{17}H_{35}CO_2CH_3$	Methyl stearate	
131	354	$C_{21}H_{43}CO_2CH_3$	Methyl behenate	
72-1	220	C_6H_{13}-Ø-CO_2CH_3	Aromatic ester	Aromatic acids
4	232	C_7H_{12}-Ø-CO_2CH_3	Aromatic ester with a double bond in the hydrocarbon chain.	

TABLE XXV (Cont'd.)

Fraction No.	MW	Formula	Identification	Compound As Present In Heavy Oil
75-1	240	$C_{13}H_{25}CO_2CH_3$	Methyl ester of cyclopentyl carboxylic acids.	Cyclopentyl carboxylic acids.
75-3	254	$C_{14}H_{27}CO_2CH_3$		
77-2	268	$C_{15}H_{29}CO_2CH_3$	" "	"
81-2; 136	324	$C_{19}H_{37}CO_2CH_3$	" "	"
83; 83-1; 84; 137	338	$C_{20}H_{39}CO_2CH_3$	" "	"
84-1; 138	352	$C_{21}H_{41}CO_2CH_3$	" "	"
139	366	$C_{22}H_{43}CO_2CH_3$	" "	"
140	380	$C_{23}H_{45}CO_2CH_3$	" "	"
85-4; 141	394	$C_{24}H_{47}CO_2CH_3$	" "	"

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