FRACTIONATION AND ANALYSIS OF MAYA CRUDE OIL

THESIS

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CHAPTER 1

INTRODUCTION

For the conversion of crude oils into more valuable fuels by pyrolysis and hydrocracking, more information about their chemical composition is needed. Crude oil is known to contain molecules with large aromatic and naphthenic ring systems [1]. As the number of rings increases, the number of possible arrangements of naphthenic and aromatic rings relative to each other increases at an exponential rate. There is also variation in the number and type of alkyl side chains. The presence of heteroatoms also complicates the mixture. Fractionation into compound classes prior to chromatographic and/or mass spectral analysis is essential. Chapter two covers some of the fractionation methods that have been used for separation of petroleum samples into compound classes.

For non-volatile components such as those found in crude oil and crude oil distillation residues, liquid chromatographic analysis must be used. Because of the low resolution of high performance liquid chromatography (HPLC) relative to gas chromatography (GC), separation of individual components is often very difficult. Even after extensive pre-fractionation, HPLC chromatograms often appear as broad envelopes with individual components unresolved. Powerful analytical tools such as liquid chromatography / mass spectrometry (LC/MS) are often able to give only statistical results [1]. If complete separation of a component has been accomplished, elucidation of

the exact structure of high molecular weight components using mass spectral data is often difficult if not impossible.

Organosulfur compounds in crude oil can affect storage stability, cause corrosion of processing equipment, poison catalysts used in catalytic cracking processes, and exacerbate environmental problems such as acid rain [2,3,4]. Polycyclic Aromatic Sulfur Heterocycles (PASHs) are also known to be toxic to many biological systems [4,5]. Because of these deleterious effects, there is a continuing interest in finding methods suitable for the fractionation and analysis of this type of compound. The PASH components are the most problematic class of sulfur containing compounds found in crude oil. Because they have very similar properties to the polycyclic aromatic hydrocarbons, the PASH compounds are particularly difficult to separate from non-sulfur aromatics.

The most common method for fractionation of petroleum samples is the Saturate–Aromatic–Resin-Asphaltene (SARA) method [6,7]. The first step involves extraction into a hydrocarbon solvent such as pentane. The asphaltenes (aggregates of extended polyaromatics, naphthenic acids, fatty acids, metalloporphyrins, ionic salts, and polyhydric phenols) [8] are pentane insoluble and precipitate out of solution. Next, the pentane soluble portion (maltenes) is separated into saturates (alkanes and cycoparaffins) [8], aromatics (mono-, di-, and polycyclic aromatic hydrocarbons with alkyl side chains) [8], and resins (aggregates with a multitude of building blocks such as polycyclic aromatic hydrocarbons, sulfoxides, amides, thiophenes, pyridines, quinolines, and carbazoles) [8] by elution from an alumina column using solvents of increasing elutropic strength.

Another method that has been used for fractionation of petroleum products is called the Acid–Base–Neutral (ABN) method [9]. This method divides the oil into six fractions based on polarity and acid-base characteristics: saturates and aromatics, low polarity nitrogen-sulfur-oxygen (NSO) containing components, medium polarity NSO containing components, high polarity NSO containing components, acids, and bases. Separation of the neutral fractions is accomplished by fractionation on silica gel. The acidic components are retained on a silica gel column that has been impregnated with KOH, and the basic components are retained on a silica column impregnated with HCl.

Two fractionation schemes have been proposed for the separation of sulfur-containing species from the non-sulfur hydrocarbons. One approach involves oxidation of the PASH components into the corresponding sulfones [10]. The sulfones are more polar and can be separated by adsorption chromatography. Another separation scheme for sulfur containing components uses ligand exchange chromatography [3,4,5]. This method uses silica gel columns impregnated with a metal salt such as $PdCl_2$ or $CuCl_2$. The sulfur atoms form charge transfer complexes with the metal ions. These charge transfer complexes are more polar and hence highly retained on the sorbent surface. After the hydrocarbon fraction has been eluted, more mobile phase or a stronger mobile phase can be used to elute the sulfur containing species.

Once the crude oil sample is separated into compound classes, methods for analysis are needed. NMR is useful for determination of structural information as well as the relative percent hydrogen distribution in the average molecule. IR can be used to calculate the average chain length for the saturated part of the molecule, and it is also helpful in determining which kinds of functional groups are present. Mass spectrometry

is effective in determination of the molecular masses of the components and can also provide some structural information. Brown and Ladner have developed a method for calculation of the average structural parameters in petroleum fractions using ^{1}H NMR data, elemental analysis data, and average molecular weight [11,12]. Careful analysis of the average structural parameters for each fraction allows the researcher to gain a better understanding of their complex chemical composition.

Sulfur-containing species can be identified by a variety of gas chromatography approaches so long as the molecule has sufficient volatility. As the molecular weight increases, the volatility diminishes until GC is no longer a feasible approach for analysis. In an effort to develop a LC/MS approach for the analysis of organosulfur compounds in crude oil, we have used the Finnegan LCQ ion trap mass spectrometer with both electrospray ionization (ESI) and atmospheric pressure chemical ionization (APCI) sources for the mass spectrometric (MS) analysis of organosulfur compounds. The addition of a suitable central atom that can form charge transfer complexes with analytes has been used to increase the sensitivity of ESI/MS for polycyclic aromatic hydrocarbons [13]. We have developed a new method for sulfur-specific ESI/MS detection of PASH components in hydrocarbon matrices. The addition of $PdCl_2$ dissolved in methanol as a sheath liquid results in a dramatic increase in sensitivity for PASH components which can be detected as the molecular ions [2].

Many petroleum compounds contain polycyclic aromatic systems within their carbon skeletons. Many polycyclic aromatic hydrocarbons (PAH) are toxic because of their mutagenic properties. Because of their high thermal stability, these systems can sometimes remain after combustion and be released into the atmosphere. For this reason,

there is a continuing interest in finding methods for the fractionation and analysis of PAH components. Supercritical Fluid Extraction (SFE) can be used as a method of extracting PAHs from solid matrices [14,15]. The solubilities of Maya crude oil, Maya crude oil distillation residue, Maya crude oil asphaltene fraction, and the model compound fluoranthene in supercritical carbon dioxide were tested using SFE.

Capillary Electrophoresis is an efficient method for the separation of cations and anions. Neutral compounds can also be separated by adding a surfactant to the electrolyte mixture [16]. The surfactant forms micelles which act as a pseudo stationary phase. Separation is based on the partitioning of the analytes between the solution and the micelle. This technique is called Micellar Electrokinetic Capillary Chromatography (MECC). Separation of a PAH mixture was accomplished using MECC, and preliminary work for the separation of crude oil fractions was also done.

CHAPTER 2

FRACTIONATION PROCEDURES

2.1 EXPERIMENTAL

2.1.1 Reagents and Chemicals

Methanol, tetrahydrofuran (THF), pentane, hexane, heptane, chloroform ($CHCl_3$), dichloromethane (CH_2Cl_2), acetone, toluene, diethyl ether, HCl, and NaCl were obtained from EM Science (Gibbstown, NJ); Maya crude oil, from Mobil Oil Corporation (Beaumont, TX); Alumina (100/120 mesh) from MicroTek; NaOH and p-ethyl phenol from MCB Manufacturing (Cincinnati, OH). KOH and H_2SO_4 were reagent grade and used as supplied. Formic acid and sodium sulfate were obtained from Baker (Phillipsburg, NJ); sodium carbonate and 2-Naphthol from Fisher (Fair Lawn, NJ); $PdCl_2$, $CDCl_3$, tetradecane, naphthalene, flouranthene, dibenzothiophene (DBT), diethyl amine and silica gel (70-270 mesh) from Aldrich (Milwaukee, WI); $CuCl_2$ from Mallinckrodt (St. Louis, MO); nonane and ethylbenzene from Eastman Kodak (Rochester, NY).

2.1.2 Viscosity Measurements and Elemental Analysis

Specific gravities of the light and middle distillates were determined by direct measurement of mass using an analytical balance and volume using a graduated cylinder.

The specific gravity of the residue was determined by direct measurement of mass and measurement of volume of water displaced in a graduated cylinder. American Petroleum Institute (API) gravities were calculated as [142.5 / (specific gravity)] – 131.5, and expressed in degrees. Samples for elemental analysis were sent to Desert Analytics, P.O. Box 41838, Tucson, Arizona 85717. Percent carbon, hydrogen, and nitrogen were determined simultaneously by combustion using a CHN analyzer. Percent sulfur was determined by conversion of sulfur into sulfur dioxide followed by analysis on a detector that is specific for carbon dioxide, water, nitrogen, and sulfur dioxide.

2.1.3 Instrumentation

2.1.3.1 Supercritical Fluid Extraction

Supercritical carbon dioxide extraction was done with a Supelco SFE-400 Extractor. Carbon dioxide is heated in a 400 mL stainless steel reservoir until the desired pressure is reached. The sample was loaded into a 10 mL extraction vessel which was then connected to the pressurized reservoir. A valve was then opened, letting the supercitical fluid into the extraction vessel. The supercritical fluid is allowed to mix with the crude oil allowing for full dissolution of soluble components. After the static extraction period (5 - 15 min), a valve was opened at the other end of the extraction vessel allowing carbon dioxide from the pressurized reservoir to flow through the extraction vessel. Dynamic extraction was continued until the pressurized reservoir was empty (15 - 20 minutes). The supercritical fluid extract was collected in a solvent reservoir. THF was used as the collection solvent.

2.1.3.2 Infared Spectroscopy

Infared spectra were recorded on a Perkin Elmer 1600 FTIR instrument. Samples were dissolved in a small amount of CH_2Cl_2. A small amount of the solution was then transferred to a KBr salt plate, and the CH_2Cl_2 was evaporated under a stream of nitrogen gas. Spectra were scanned from 500 – 4000 cm^{-1}. Sixteen scans were averaged for the background blank. Sixteen scans were averaged for each spectrum obtained.

2.1.3.3 Nuclear Magnetic Resonance Spectroscopy

^1H NMR experiments were done on a Varian instrument operating at 400 MHz. A Sun workstation with UNIX software was used for data acquisition. The proton spectra were run in $CDCl_3$ solvent. TMS was used as an internal calibration standard. Solutions for the proton spectra were prepared by dissolving 5 mg of sample in 1.5 mL of $CDCl_3$. The following experimental conditions were used: spectrum width, 6000 Hz; data points, 44,928; pulse width, 7.0 μs; and number of transients, 16.

2.1.4 Fractionation Procedures

2.1.4.1 Distillation

The Maya crude oil was distilled (see figure 2.1). A light distillate boiling up to 200°C was collected under atmospheric pressure. A middle distillate was obtained under reduced pressure (20 torr) keeping the temperature below 180°C. The residue was the remaining non-volatile fraction.

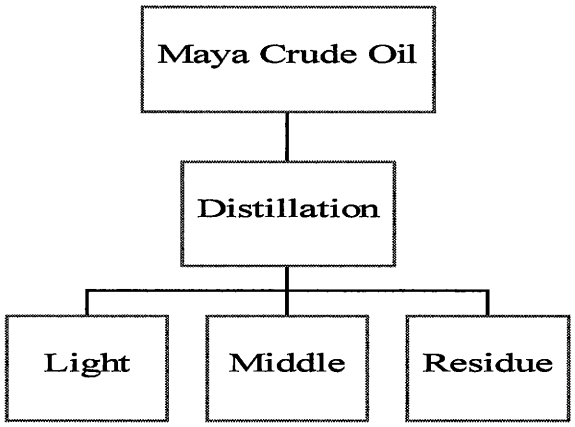

Figure 2.1 : Scheme for the Distillation of Maya Crude Oil

2.1.4.2 Supercritical Fluid Extraction

Supercritical fluid extraction (SFE) was performed on 0.073 g fluoranthene, 0.385 g Maya crude oil, 0.088 g Maya crude oil residue, and 0.055 g Maya crude oil asphaltene fraction. This procedure separates a petroleum sample into supercritical carbon dioxide soluble components and supercritical carbon dioxide insoluble components (figure 2.2). The run conditions for each extraction are listed in Table 2.5. Percent extracted was calculated after each extraction by washing the extraction vessel with CH_2Cl_2, evaporating the CH_2Cl_2, and weighing the amount that was insoluble in CO_2.

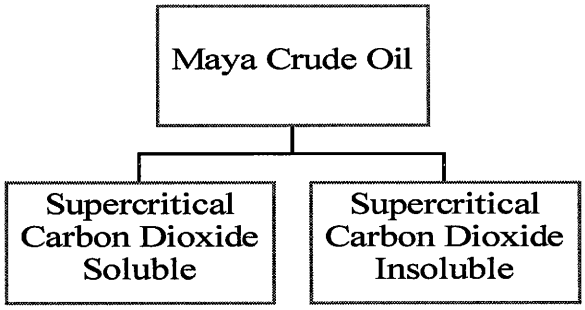

Figure 2.2 : Scheme for the SFE Fractionation of Maya Crude Oil

2.1.4.3 Saturate-Aromatic-Resin-Asphaltene Fractionation Method

The saturate-aromatic-resin-asphaltene (SARA) method (figure 2.3) first separates petroleum into pentane soluble maltenes and pentane insoluble asphaltenes. The maltenes can then be further fractionated by column chromatography on alumina. A Maya crude oil (CM) and a Maya crude oil distillation residue (CMh) were fractionated by extraction with pentane (40 mL / g) into maltenes and asphaltenes. The pentane soluble fractions were then separated into saturates, aromatics and resins by adsorption chromatography. Alumina was activated at 275°C for 72 hours. Alumina was then packed into a glass column (2.2 x 20 cm), and the column was pre-wet with 100 mL heptane. The sample (2.99 g) was dissolved in a small volume of heptane, and was loaded at the top of the column. The flow rate was set to two milliliters per minute. Saturates (CMSa or CMhSa) were washed from the column with 350 mL heptane. Aromatics (CMAr or CMhAr) were recovered with 430 mL heptane:toluene (1:1). Resins (CMResins or CMhResins) were recovered by eluting first with 430 mL toluene:methanol (1:1), and then with 215 mL methanol. Solvents were removed on a rotary evaporator.

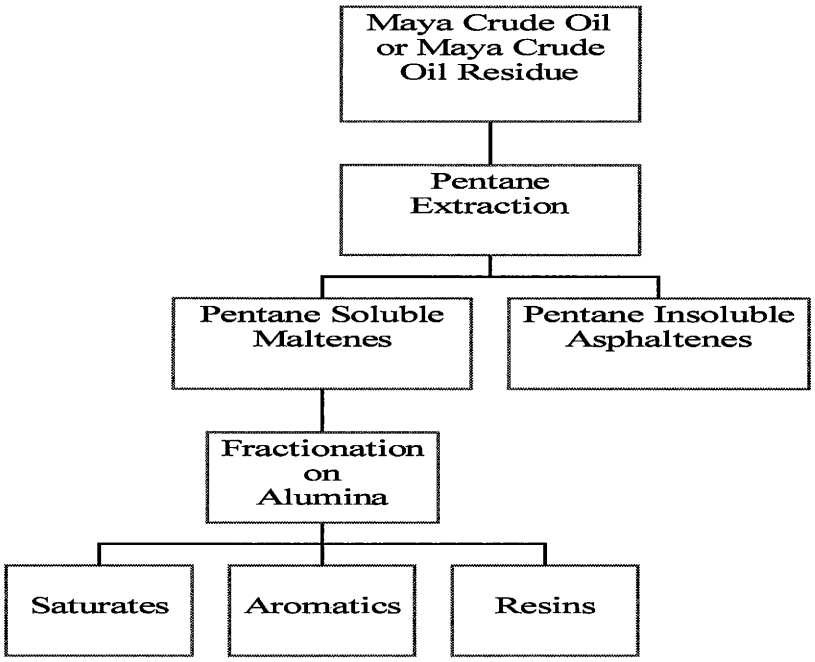

Figure 2.3: Scheme for the SARA Fractionation of Maya Crude Oil and Maya Crude Oil Residue

2.1.4.4 Solvent Extraction of Asphaltenes

Solvent extraction was tested as a means of further fractionating the asphaltene fraction from the Maya crude oil residue (see figure 2.4). Asphaltenes were subjected to Soxhlet extraction for four hours using 60 mL of solvent / gram of asphaltene. The asphaltenes were extracted first with methanol. The methanol soluble portion was designated as CMhAsMe. The methanol insoluble portion was extracted with acetone. The acetone soluble fraction was designated as CMhAsAcet. The remaining insoluble portion was found to be soluble in THF. The THF soluble fraction was designated as CMhAsTHF.

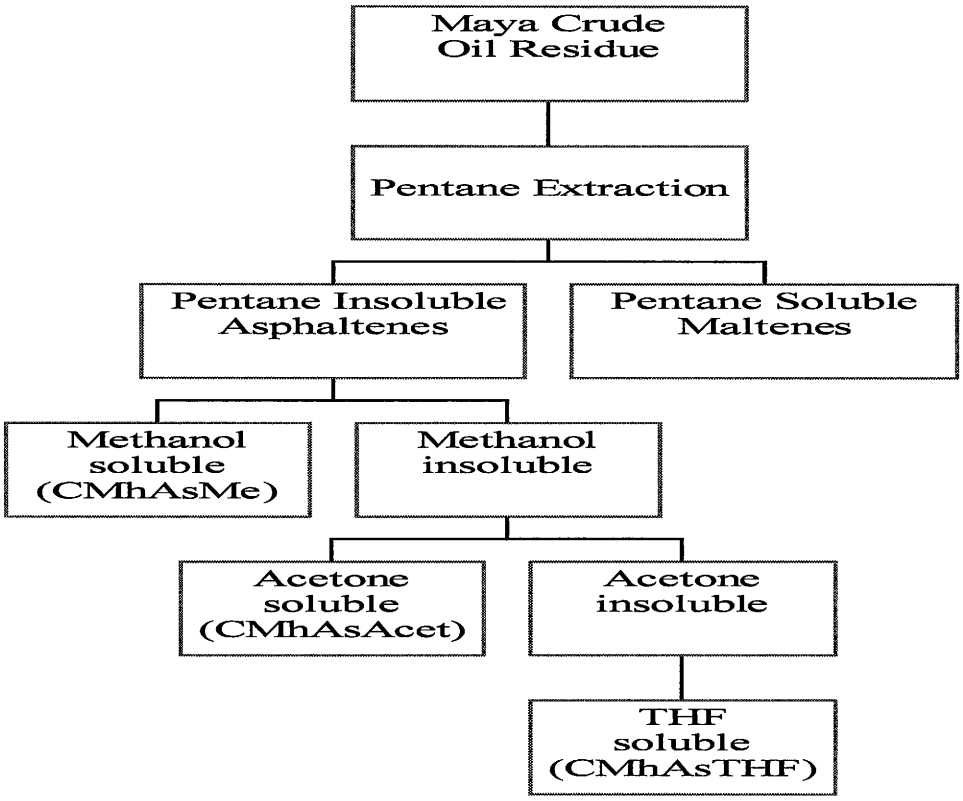

Figure 2.4 : Scheme for the Solvent Extraction of Asphaltenes

2.1.4.5 Acid-Base-Neutral Fractionation Method

Another method used for separation of crude oils is a hybrid of adsorption chromatography and acid / base extraction and is called the acid-base-neutral (ABN) method of separation (figure 2.5). This method utilizes a series of columns filled with three kinds of silica gel. Acidic silica was prepared by first bubbling gaseous hydrochloric acid gas (prepared by reaction of concentrated sulfuric acid with sodium chloride) into cold methanol. Silica was then allowed to sit in the acidic methanol solution for one hour, and then the solvent was removed by rotary evaporation. Acidic silica was stored in a dessicator below 0°C. Basic silica was prepared by dissolving 5%

KOH (mass KOH / mass silica) in methanol. Silica gel was then mixed with the basic methanol solution, and the solvent was evaporated by rotary evaporation. Basic silica was stored in a dessicator.

Preliminary experiments were done using model compounds to determine the cutoff points for saturates and aromatics, low polarity, and medium polarity fractions. The eluent used for the elution of the model compounds was CH_2Cl_2:methanol (99:1). 2,3-Benzanthracene was used to help predict the elution volume for aromatic components. The elution of 2,3-Benzanthracene was detected by the bright orange color of the eluent at the time of elution. 2,3-Benzanthracene eluted at 85 mL, and so it was decided to collect saturates and aromatics from 0 to 100 mL. 2-Naphthol was selected for prediction of the elution volume required for the medium polarity fraction. The elution of 2-Naphthol was detected by the faint yellow color of the eluent at the time of elution. 2-Naphthol eluted at 270 mL, and so it was decided to collect the medium polarity fraction from 200 to 400 mL. This means that the low polarity fraction was collected from 100 to 200 mL.

Mayan crude oil residue (0.9193 g) was dissolved in CH_2Cl_2:methanol (99:1) and loaded at the top of the first column. The initial eluent was CH_2Cl_2:methanol (99:1). Saturates and aromatics (CMhSa+Ar) were eluted through all four columns with 100 mL of this eluent. Low polarity nitrogen-sulfur-oxygen (NSO) containing components (CMhLP) were eluted from all four columns with an additional 100 mL of the eluent mixture. The medium polarity NSO fraction (CMhMP) was then eluted through all four columns with another 200 mL of the same eluent mixture. Column four was disconnected from the series, and the remaining medium polarity components were

washed from column four with CH_2Cl_2:methanol (90:10). Columns one, two, and three were then disconnected from each other for elution of the more polar fractions.

Column one was filled with activated silica and retained the highest polarity components. This high polarity NSO fraction (CMhHP#1) was removed from the silica by washing first with 50 mL of the azeotopic mixture CH_2Cl_2:acetone:methanol (47:23:30), and then with 50 mL methanol. The final step involved refluxing the silica with a large volume of azeotopic mixture to remove most of the remaining high polarity components (CMhHP#2). Some high polarity components were inevitably lost on column one as indicated by the darkened color of the silica even after this final step.

The second column uses acid impregnated silica to immobilize components that are basic. These basic components (CMhBases) were washed from column two with 100 mL of methanol. The bases were removed from their hydrochloride salts by passing the eluent through another column filled with base impregnated silica.

A third column filled with base impregnated silica selectively removes acidic components (CMhAcids). The acids were removed from column three using 150 mL CH_2Cl_2:formic acid (99:1). All of the fractions were stripped of solvent using a rotary evaporator, and the percent recovery for each was determined.

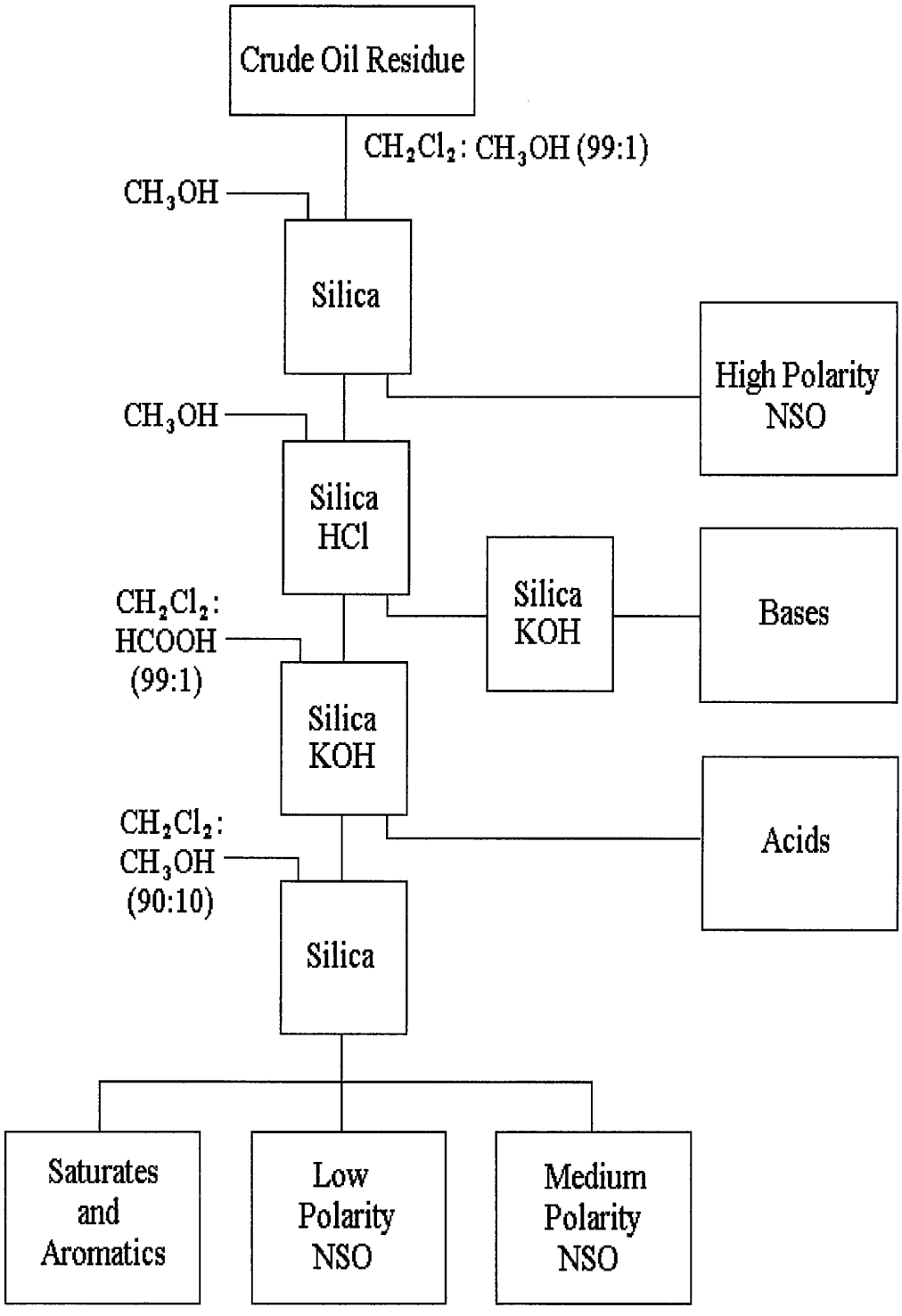

Figure 2.5: Scheme for the ABN Fractionation of Maya Crude Oil Residue

2.1.4.6 Ligand Exchange Chromatography

Sulfur-containing components were isolated by ligand exchange chromatography (LEC) using $PdCl_2$ or $CuCl_2$ impregnated silica gel. The initial eluent was chosen so that only non-sulfur-containing components are eluted. Sulfur-containing compounds form complexes with the metal ion and hence are highly absorbed onto the silica surface. A stronger eluent is then used to elute sulfur-containing components. Aliphatic sulfur compounds (Aliph-S) were separated from the aliphatic hydrocarbons (Aliph) using $CuCl_2$ / silica gel. Polycyclic Aromatic Sulfur Heterocycles (PASH) and other sulfur containing polycyclic aromatic compounds (SPAC) were separated from the polycyclic aromatic hydrocarbons (PAH) using $PdCl_2$ / silica gel. The fractionation scheme for the isolation of sulfur-containing components is illustrated in figure 2.6.

Alumina was dried at 200°C overnight. $CuCl_2$/silica was prepared by mixing 20 g of silica gel with 1g of $CuCl_2$ in distilled water. The mixture was dried to a wet powder with a rotary evaporator, and then dried at 200°C for 24 hours prior to use. $PdCl_2$/silica was prepared by mixing 20 g of silica gel with 1 g of $PdCl_2$ suspended in an aqueous solution. The mixture was dried in an oven at 95°C overnight, then held at 200°C for 24 hours prior to use.

Initial tests were performed to confirm the validity of the procedure for isolation of sulfur compounds. A mixture of 3 mg fluoranthene and 3 mg DBT were used for method validation. These components are both polycylic aromatic hydrocarbons, but DBT contains a sulfur atom. Fluoranthene should pass through quickly while DBT should be more highly retained. The fluoranthene/DBT mixture was dissolved in 5 mL

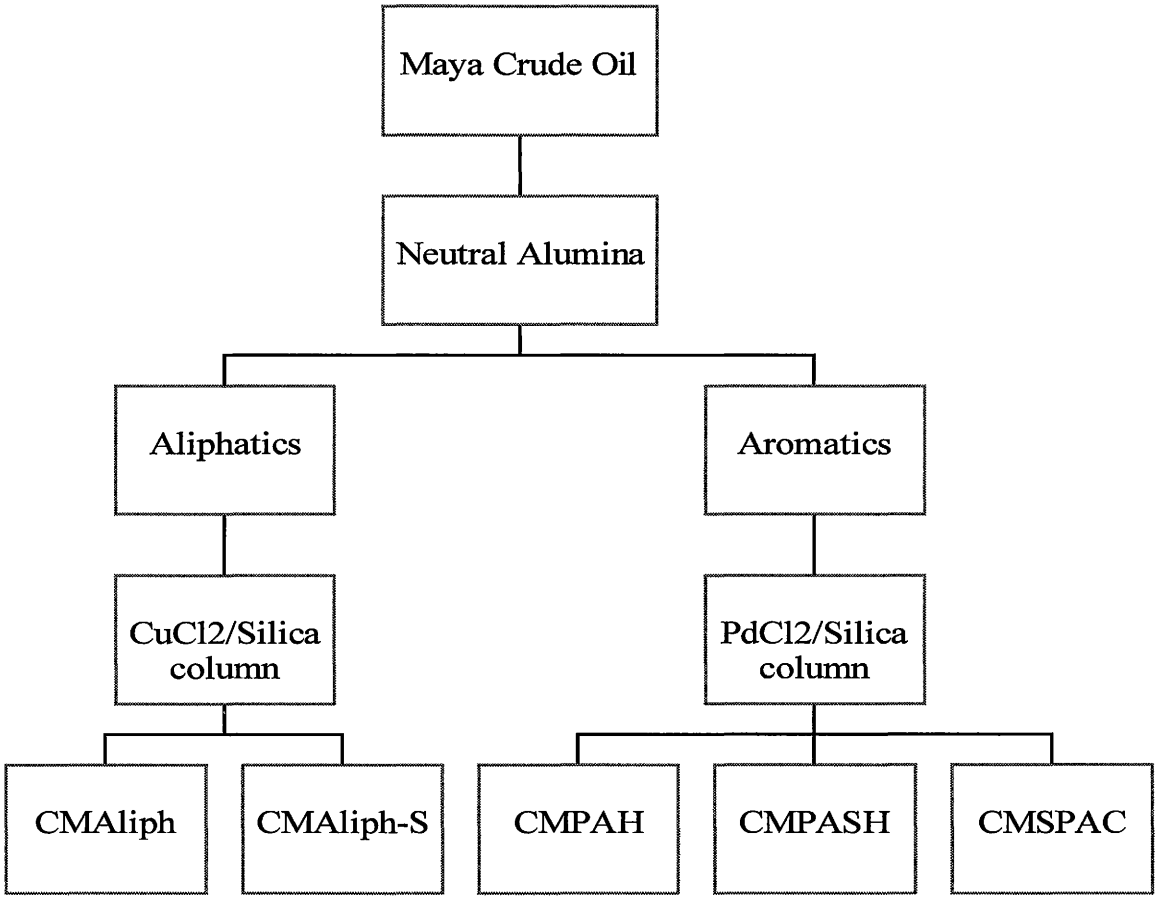

Figure 2.6: Scheme for the LEC Fractionation of Maya Crude Oil

CH_2Cl_2 and absorbed onto 0.5 g of the $PdCl_2$ / silica gel. The CH_2Cl_2 was evaporated under a gentle stream of nitrogen, and the mixture was packed on top of 5.0 g $PdCl_2$ / silica gel in a 10 x 125 mm column. 30 mL of $CHCl_3$:hexane (1:1) were used to elute the PAH fraction. A further 50 mL of the same eluent, $CHCl_3$:hexane (1:1), were used to elute the PASH fraction. 100 mL of $CHCl_3$:diethyl ether (9:1) were used to elute the SPAC fraction. The PASH and SPAC fractions were reduced in volume to approximately 1 mL by rotary evaporation, after which 100 μL of diethylamine was added to break up the Pd complexes. The PASH and SPAC fractions were further

cleaned by passing them through neutral alumina with 50 mL toluene. All of the fractions were evaporated to dryness and redissolved in 5 mL CH_2Cl_2. Reverse phase high performance liquid chromatography / electrospray ionization / mass spectrometry (HPLC/ESI/MS) using $PdCl_2$ sheath liquid (see section 4.1) was used to determine if the DBT was more highly retained than fluoranthene.

After the preliminary experiments with standard compounds, the Maya crude oil was fractionated. Approximately 0.2 g of the Maya crude oil was dissolved in 5 mL CH_2Cl_2. This mixture was absorbed onto 3 g of neutral alumina. The solvent was removed under a gentle stream of nitrogen gas. The alumina with the absorbed sample was then packed on top of 6 g neutral alumina in a 10 x 125 mm column. The sample was eluted first with 20 mL of hexane, which removes the aliphatics. The aromatics were then eluted with 50 mL of toluene. Solvents were removed from the aliphatics and the aromatics by rotary evaporation. The entire fractionation scheme for isolation of sulfur compounds is illustrated in Figure 2.6.

The aliphatics were dissolved in 5 mL CH_2Cl_2 and absorbed onto 0.5 g of the $CuCl_2$ / silica gel. The solvent was evaporated under a gentle stream of nitrogen gas, and the mixture was packed on top of 5 g of $CuCl_2$ / silica gel in a 10 x 125 mm column. 50 mL of hexane were used to elute the CMAliph fraction. 100 mL of $CHCl_3$:diethyl ether (9:1) were then used to elute the CMAliph-S fraction.

The aromatics were dissolved in 5 mL CH_2Cl_2 and absorbed onto 0.5 g of the $PdCl_2$ / silica gel. The solvent was evaporated under a gentle stream of nitrogen gas, and the mixture was packed on top of 5.0 g $PdCl_2$ / silica gel in a 10 x 125 mm column. 30 mL $CHCl_3$:hexane (1:1) were used to elute the CMPAH fraction. A further 50 mL of the

same eluent, $CHCl_3$:hexane (1:1), were used to elute the CMPASH fraction. 100 mL of $CHCl_3$:diethyl ether (9:1) were used to elute the CMSPAC fraction. The CMPASH and CMSPAC fractions were reduced in volume to approximately 1 mL by rotary evaporation, after which 100 μL of diethylamine was added to break up the Pd complexes. The CMPASH and CMSPAC fractions were further cleaned by passing them through neutral alumina with 50 mL toluene. All of the fractions were evaporated to dryness and re-dissolved in 5 mL CH_2Cl_2.

2.1.5 Calculation of Average Structural Parameters

Average structural parameters were calculated using the modified Brown and Ladner method [11,12]. These calculations take into account NMR spectral data, elemental analysis data, and the average molecular weight. Number average molecular weights were determined from the gel permeation chromatograms (figure 3.5) and the APCI mass spectra (figures 4.2 – 4.4). It was found that to reduce error in the calculations, it was necessary to average at least one thousand peaks from the mass spectrum. The NMR spectra were divided into four hydrogen types: H_A, H_α, H_β, H_γ. These types are described in Table 2.1.

Table 2.1: Assignments of Proton Bands in NMR Spectra

Symbol	Range (ppm, from TMS)	Assignment
H_A	6.0-9.0	Aromatic hydrogens
H_α	2.0-4.0	Hydrogens in saturated groups α to aromatic rings
H_β	1.0-2.0	Hydrogens of methylene and methine groups β or further to aromatic rings, and hydrogens in β methyl groups
$H\gamma$	0.5-1.0	Hydrogens in methyl groups γ or further removed from aromatic rings

Table 2.2 presents the designations of the symbols used in the Brown and Ladner calculations. The average structural parameters were calculated by the equations in Table 2.3.

Table 2.2: Designation of Symbols Used in Calculation of Average Structural Parameters

M	Number average molecular weight
C_T	Total number of carbon atoms in molecule
H_T	Total number of hydrogen atoms in molecule
C_A	Number of aromatic carbons in molecule
C_N	Number of naphthenic carbons in molecule
C_P	Number of paraffinic carbons in molecule
C_S	Number of saturated carbons in molecule
f_A	Fraction of aromatic carbon
f_N	Fraction of naphthenic carbon
f_P	Fraction of paraffinic carbon
R_T	Total number of rings in molecule
R_A	Number of aromatic rings in molecule
R_N	Number of naphthenic rings in molecule
H_{AU}/C_A	H/C atom ratio of hypothetical unsubstituted aromatic system
σ	Degree of substitution of the aromatic system
L	Average length of side chain

Table 2.3: Calculation of Average Structural Parameters

$$f_A = \frac{C/H - (H_\alpha + H_\beta + H_\gamma) / 2H_T}{C/H}$$

$$\sigma = \frac{H_\alpha / 2H_T}{H_A / H_T + H_\alpha / 2H_T}$$

$$\frac{H_{AU}}{C_A} = \frac{H_A/H_T + H_\alpha/2H_T}{C/H - (H_\alpha + H_\beta + H_\gamma) / 2H_T}$$

$$C_A = C_T f_A$$

$$R_A = (C_A - 2) / 4 \quad \text{(for cata condensed aromatic system)}$$

$$R_A = (C_A - 4) / 3 \quad \text{(for peri condensed aromatic system)}$$

$$R_T = C_T + 1 - H_T/2 - C_A/2$$

$$R_N = R_T - R_A$$

$$C_N = 4 R_N \quad \text{(for cata condensed six membered ring system)}$$

$$C_N = 3 R_N \quad \text{(for peri condensed six membered ring system)}$$

$$C_S = C_T - C_A \qquad C_P = C_S - C_N$$

$$f_N = C_N / C_T \qquad f_P = C_P / C_T$$

The average side chain length was calculated from IR data. It has been previously reported [6] that there is a linear relationship between the relative intensities of the 1460 and 1380 cm^{-1} bands (A_{1460} / A_{1380}) and the ratio of methylene to methyl groups (nCH_2/nCH_3) in the molecule. The relationship is as follows:

$$nCH_2/nCH_3 = 2.93\ A_{1460}/A_{1380} - 3.70$$

With the assumption that the saturated part of the molecule has twice as many hydrogen atoms as carbon atoms (H/C = 2.0), the number of methyl groups in the molecule can be obtained by the equation:

$$nCH_3 = C_S / (nCH_2/nCH_3 + 2)$$

The number of side chains of the average molecule is equal to nCH_3, and the average chain length (L) is defined as C_p/nCH_3.

2.1 RESULTS

2.2.1 Distillation

Table 2.4 shows the results from the distillation of the Maya crude oil:

Table 2.4: Results From the Distillation of Maya Crude Oil

Fraction	Boiling Point	Recovery (%)	Density (g/mL)	API Gravity (degrees)
Crude Oil	n.a.	100	0.92	23
Light Distillate	<200°C @760torr	38	0.80	47
Middle Distillate	<180°C @20 torr	22	0.92	23
Residue	>180°C @20 torr	40	1.0	9.6

2.2.2 Supercritical Fluid Extraction

Several samples were extracted to determine solubility in supercritical carbon dioxide. Extraction conditions as well as percent solubility for each component are listed in Table 2.5. It is clear that the high molecular weight species present in the residue and asphaltene fractions are quite insoluble in supercritical carbon dioxide. This would suggest that supercritical fluid extraction (SFE) using carbon dioxide could be used as an alternative to distillation for obtaining high molecular weight residues. Because the high temperatures used in distillations of crude oils can result in thermal decomposition of components, SFE residues should be more representative of the actual high molecular weight species that are present in the crude oil.

Table 2.5: SFE Results for Fluoranthene, Maya Crude Oil, and Fractions from Maya Crude Oil.

Sample	% Soluble	Pressure (psi)	Temperature (°C)	Extraction Time (min) (static + dynamic)
Fluoranthene	82	3000	60	5+15
Crude Oil	63	2000	80	5+15
Crude Oil Residue	8.2	2800	80	5+15
Asphaltenes	4.4	3000	70	15+20

2.2.3 Saturate-Aromatic-Resin-Asphaltene Fractionation Method

Saturate-aromatic-resin-asphaltene (SARA) fractionation was done on the crude oil and the crude oil residue. Results have been tabulated in Table 2.6. As expected, the

distillation residue contains significantly higher proportions of resins and asphaltenes. Resins and asphaltenes contain components that have high molecular weight and/or highly polar functional groups. Many of these components have large condensed aromatic ring systems and long aliphatic chains which contribute to their extremely low volatility.

Table 2.6: SARA Fractionation of Maya Crude Oil and Maya Crude Oil Residue

	% of Crude Oil	% of Crude Oil Residue
Saturates	19	14
Aromatics	36	15
Resins	15	33
Asphaltenes	24	38

The FT-IR spectrum for Maya crude oil is shown in appendix A.1, and the FT-IR spectra for the SARA fractions of a Maya crude oil are in appendix A.2. The spectra are useful in determining the validity of the fractionation method. The Maya crude oil shows C-H stretching at 2850 and 2925 cm^{-1} and C-H bending at 1380 and 1460 cm^{-1}. A four band pattern present at 720 – 900 cm^{-1} indicates the presence of substituted and/or condensed aromatic rings. The slight absorbance from 3000 to 3100 cm^{-1} is due to Ar-H stretching. The absorbance above 3100 cm^{-1} is due to O-H and N-H stretching. The increased absorbance in the 1200 to 1450 cm^{-1} region is due to C-H and N-H bending as well as C=S stretching. The broad absorbance band from 1450 to 1750 cm^{-1} is indicative of C=C and C=N stretching. The saturate fraction shows only C-H stretching and bending frequencies. The absence of absorbance at 1600 cm^{-1} confirms that no aromatics

are present. The aromatic fraction also shows C-H stretching and bending frequencies. Very little absorbance is seen above 3100 cm^{-1} indicating the absence of O-H and N-H groups. The increased absorbance in the 1200 to 1450 cm^{-1} region due to C-H bending and C=S stretching is noticeable. The C=C and C=N stretching band from 1450 to 1750 cm^{-1} is present as expected. The resin fraction shows many similarities to the aromatics; the major difference is the broad absorbance band from 3100 to 3700 cm^{-1} indicating the presence of O-H and N-H groups. The resins also show increased absorbance in the C=N stretching region. A medium band from 1000 to 1100 cm^{-1} that was not present in the aromatic fraction can be attributed to C-O and C-N stretching. The asphaltene fraction has all of the same absorbance bands that are present in the resin fraction with slight variations in intensity.

The ^1H NMR spectrum for the Maya crude oil is shown in appendix B.1, and the ^1H NMR spectra for the SARA fractions are shown in appendix B.2. Elemental analysis data (Table 2.7), APCI-MS data (Table 2.7), and ^1H NMR data (Table 2.8) were used to calculate the average structural parameters (Table 2.9) using the modified Brown and Ladner method. The results show that the average molecule contained in the crude oil (CM) and the aromatic fraction (CMAr) contains five aromatic rings. Although the calculations predict that the average molecule in the saturate (CMSa) fraction has one aromatic ring, this could be due to a high degree of branching which increases the C/H ratio. The resin fraction (CMResin) has an average molecule with almost seven aromatic rings. All of the fractions have an average of 3.3 – 4.2 naphthenic rings per molecule. The σ parameter is an indication of the degree of substitution of the aromatic system. For the saturate fraction, the σ parameter is equal to 1.0. This is because there are no

aromatic hydrogens present. The aromatic rings in the crude oil, aromatic fraction, and resin fraction are about 50% substituted ($\sigma \approx 0.5$). Average chain lengths vary from 2.0 to 4.3 carbon units in the order: resins < crude oil < aromatics < saturates. The parameter H_{AU}/C_A is the hydrogen to carbon ratio of the hypothetical unsubstituted aromatic system. This can also be thought of as an indication of the degree of condensation of the aromatic system. The results of this analysis indicate that the degree of condensation of the aromatic system increases in the order: saturates < aromatics < crude oil ≈ resins.

Table 2.7: Elemental Analysis Data, H/C Ratio and Average Molecular Weight (M) of Maya Crude Oil and SARA Fractions.

Fraction	%C	%H	%S	H/C	M
CM	83.0	10.8	3.71	1.55	1209[a]
CMSa	83.2[b]	12.5[b]	0.78[b]	1.79	995[c]
CMAr	82.7	10.5	5.07	1.51	1103[d]
CMResins	73.3	7.47	3.95	1.21	1017[a]

[a] From the gel permeation chromatograms (Figure 3.5)
[b] From the elemental analysis data of CMAliph and CMAliph-S fractions (Table 2.13)
[c] From the APCI-MS Spectra of CMAliph and CMAliph-S fractions (Figure 4.2).

Table 2.8: Percent Hydrogen Distribution in the ^{1}H NMR Spectra of Maya Crude Oil and SARA Fractions.

Fraction	H_{ar}	H_α	H_β	H_γ	H_T
CM	0.05	0.09	0.56	0.30	1.00
CMSa	0	0.15	0.68	0.18	1.01
CMAr	0.06	0.17	0.59	0.18	1.00
CMResin	0.12	0.18	0.55	0.15	1.00

Table 2.9: Brown and Ladner Structural Parameters of Maya Crude Oil and SARA Fractions.

Fraction	f_A	f_N	f_P	R_A	R_N	σ	L	H_{AU}/C_A
CM	0.26	0.14	0.60	5.0	3.8	0.47	2.9	0.56
CMSa	0.10	0.14	0.75	1.3	3.3	1.00	4.3	1.27
CMAr	0.29	0.14	0.57	5.0	3.5	0.59	3.7	0.76
CMResins	0.47	0.2	0.33	6.7	4.2	0.43	2.0	0.55

2.2.4 Solvent Extraction of Asphaltenes

The asphaltene fraction was extracted into solvents of various polarity to gain a better understanding of the nature of this fraction. Only 2.11% of the asphaltene fraction was found to be soluble in methanol. The methanol insoluble, acetone soluble fraction constituted 15.4% of the total asphaltenes. The remaining 82.5% was found to be soluble in tetrahydrofuran.

2.2.5 Acid-Base-Neutral Fractionation Method

The Maya Crude Oil Residue (CMh) was fractionated using the acid-base-neutral (ABN) fractionation scheme. The residue was found to contain saturates and aromatics (CMhSa+Ar, 23%), low polarity NSO (CMhLP, 57%), medium polarity NSO (CMhMP, 9%), high polarity NSO (CMhHP#1 and CMhHP#2, 2%), acids (CMhAcids, 6%), and bases (CMhBases, 8%). The total recovery was greater than 100% due to the elution of a small amount of silica gel. FT-IR spectra for the ABN fractions are shown in appendix

A.3. Assignments for the IR absorbances are not listed here because they are nearly identical to the assignments listed for the SARA fractions (section 2.2.3).

^{1}H NMR spectra for the ABN fractions are shown in appendix B.3. Elemental analysis data (Table 2.10), APCI-MS data (Table 2.10), and ^{1}H NMR data (Table 2.11) were used to calculate the average structural parameters (Table 2.12) using the modified Brown and Ladner method. The results show that the average molecule contained in the fractions analyzed contains four aromatic rings. The average molecule in the CMhSa+Ar fraction contains 4.2 naphthenic rings, while the average molecule in the CMhLP and CMhMP polarity fractions has only 3.2 – 3.4 naphthenic rings. The aromatic rings in the CMhSa+Ar and CMhLP fractions are slightly less than 50% substituted. In the CMhMP fraction the aromatic rings are 60% substituted.

Average chain lengths (L) for the fractions analyzed vary from 1.6 to 2.1 carbon units. The values for the parameter H_{AU}/C_A indicate that the degree of condensation of the aromatic system increases as the polarity of the fraction decreases. This would seem to contradict the results from the SARA fractionation method where the more polar fraction (CMResin) had a higher degree of condensation. When the SARA method of fractionation is used, the low polarity fractions have the lowest average molecular weight. If the ABN method is used, the low polarity fractions have the highest molecular weight. The data seems contradictory, however one must realize that the mobile phases used are very different. Perhaps the ABN method is much better at separating by polarity, while the SARA method fractionates according to the number of aromatic rings present. This reasoning would certainly help to explain the apparent contradiction in molecular weight and H_{AU}/C_A data.

Table 2.10: Elemental Analysis Data, H/C Ratio, and Average Molecular Weights (M) of ABN Fractions from Maya Crude Oil Residue.

Fraction	%C	%H	%N	%S	H/C	M[a]
CMhSa+Ar	82.19	10.79	0.24	2.85	1.56	1073
CMhLP	83.89	11.03	0.34	4.02	1.56	1000
CMhMP	78.93	9.77	0.94	4.18	1.47	877

[a] Calculated form the APCI-MS spectra

Table 2.11: Percent Hydrogen Distribution in the ^{1}H NMR Spectra of the ABN Fractions from Maya Crude Oil Residue.

Fraction	H_{ar}	H_α	H_β	H_γ	H_T
CMhSa+Ar	0.03	0.05	0.63	0.26	0.97
CMhLP	0.05	0.09	0.58	0.26	0.98
CMhMP	0.05	0.15	0.51	0.15	0.86

Table 2.12: Brown and Ladner Structural Parameters of the ABN Fractions from Maya Crude Oil Residue.

Fraction	f_A	f_N	f_P	R_A	R_N	σ	L	H_{AU}/C_A
CmhSa+Ar	0.24	0.17	0.59	3.9	4.2	0.46	1.9	0.37
CMhLP	0.26	0.14	0.61	4.0	3.2	0.47	2.1	0.59
CMhMP	0.31	0.18	0.52	3.9	3.4	0.60	1.6	0.70

There are several signals present in the ^{1}H NMR spectra of the ABN fractions that are not pertinent to the Brown and Ladner method. All of the fractions from the ABN method have a signal at 4.2 ppm which has been attributed to hydrogens bonded to

oxygen. Another signal that is present in all of the fractions is due to residual CH_2Cl_2 and is seen at 5.3 ppm. The CMhBase fraction has several signals in the 3 – 4 ppm region corresponding to protons bonded to nitrogen. The CMhHP#1, CMhHP#2, and CMhMP fractions all show signals in the 3 – 4 ppm region which can be attributed to methylene protons where the methylene group is bonded to oxygen.

2.2.6 Ligand Exchange Chromatography

The Maya crude oil was found to contain 19% aliphatic hydrocarbons (CMAliph), 11% aliphatic sulfur containing hydrocarbons (CMAliph-S), 20% polycyclic aromatic hydrocarbons (CMPAH), 3.5% polycyclic aromatic sulfur heterocycles (CMPASH), and 4.9% other sulfur containing polycyclic aromatic compounds (CMSPAC). These results show higher numbers for percent saturates and lower numbers for percent aromatics when compared to the SARA results (Table 2.6). This can be attributed to the differences in column size, sorbent mass, and eluent volumes. Results for the elemental analysis of all of the fractions obtained are summarized in Table 2.13. The sulfur content of the aliphatic fractions is <2%, while the aromatic fractions have higher sulfur content (4.25% – 8.12%). This is consistent with the fact that sulfur is usually found within heterocyclic structures rather than in aliphatic chains. Complete separation of the sulfur containing aromatics from their hydrocarbon counterparts was not achieved. Aromatic hydrocarbons with large condensed aromatic ring structures are highly retained on the silica surface and hence could possibly coelute with some of the sulfur containing aromatics. Furthermore, based on $PdCl_2$/amino propyl bonded solid phase extraction (SPE) of mixtures containing several PASH standard compounds [18] we have come to

realize that the low molecular weight PASH compounds have a lower affinity for the Pd^{2+} ion and hence will very probably appear in the PAH fraction.

Table 2.13: Elemental Analysis Data for LEC Fractions from Maya Crude Oil.

Fraction	%C	%H	%S
CMAliph	86.0	13.4	0.18
CMAliph-S	78.3	10.9	1.81
CMPAH	83.0	10.0	4.25
CMPASH	75.1	9.78	6.34
CMSPAC	75.2	9.52	8.12

^{1}H NMR spectra for the sulfur enriched fractions can be found in appendix B.4. All of the sulfur enriched fractions show a sharp band at 2.3 ppm corresponding to protons adjacent to sulfide linkages. All of the fractions have a sharp signal at 1.5 ppm which has been attributed to R-SH protons where R is alkyl. The SPAC fraction shows a band at 3.8 ppm due to the presence of Ar-SH protons. Brown and Ladner calculations of the average structural parameters were not done for these fractions because they should be very similar to the numbers obtained for the CMSat and CMAr fractions of the SARA method.

CHAPTER 3

CHROMATOGRAPHIC SEPARATIONS

3.1 EXPERIMENTAL

3.1.1 Reagents and Chemicals

Methanol, acetonitrile, THF, H_3PO_4, sodium borate, hexane, chloroform ($CHCl_3$), and acetone were obtained from EM Science (Gibbstown, NJ); Maya crude oil, from Mobil Oil Corporation (Beaumont, TX); the sixteen component mixture of polycyclic aromatic hydrocarbons (EPA 610 PAH) from Supelco (Bellefonte, PA); Chlorosulfonic acid from MCB Manufacturing (Cincinnati, OH); Brij-30 from Aldrich (Milwaukee, WI).

3.1.2 Preparation of Brij-S

Sulfonated Brij-30 (Brij-S) was prepared from Brij-30 by sulfonation. Brij-30 (9.33 g) was placed in a three neck round bottom flask. As the temperature was lowered to 15°C, chlorosulfonic acid (3.0 g) was added slowly over 15 minutes with constant stirring. Then the ice bath was removed, and the mixture was stirred for another five minutes. Sodium hydroxide (1.6 g) was dissolved in 30 g of ice slurry. The basic ice slurry was added to the reaction mixture, and stirring was continued for five minutes.

The pH was adjusted to 8.0 with 0.5 M H_3PO_4. The sodium Brij-30 sulfate was then used for preparation of the buffers used in micellar electrokinetic capillary chromatography.

3.1.3 Preparation of Standards

3.1.3.1 EPA 610 PAH mixture

For micellar electrokinetic capillary chromatography experiments (figure 3.5), the EPA 610 PAH stock solution as obtained from Supelco (Bellefonte, PA) was diluted to a concentration suitable for MECC. A 50 μL syringe was used to transfer 100 μL of the stock solution into a 10 mL vial, and 0.5 mL of the buffer solution was added using a 2 mL pipet.

For normal phase HPLC (figure 3.1a), the EPA 610 PAH stock solution as obtained from Supelco (Bellefonte, PA) was diluted to a concentration suitable for HPLC. A 50 μL syringe was used to transfer 20 μL of the stock solution into a 10 mL vial. The solvent was removed under a gentle stream of nitrogen, and 0.4 mL of hexane:CH_2Cl_2 (1:1) was added using a 2 mL pipet.

3.1.3.2 Gel Permeation Chromatography

Naphthalene (188 ppm), benzo [k] fluoranthene (150 ppm), rubrene (150 ppm), 5, 10, 15, 20 tetraphenyl 21H, 23H porphine (133 ppm), and 1320 molecular weight polystyrene (150 ppm) were prepared by weighing a small amount of each standard into a 10 mL vial and adding 5 mL of tetrahydrofuran using a 5 mL pipet.

3.1.4 Cleanup of Asphaltene Fractions Prior to HPLC Analysis

Because the fractionation procedures used in the isolation of the saturate, aromatic, and resin fractions require them to pass through a silica or alumina column, there is no need for solid phase extraction (SPE) with a silica cartridge (Supelco LC-Si, 3 mL) prior to normal phase HPLC analysis. In contrast, the asphaltene fraction contains many polar and/or high molecular weight species that are very highly retained on both normal and reverse phase columns. For this reason, it was necessary to sub-fractionate the asphaltenes using SPE prior to any attempt at HPLC analysis.

SPE was performed on the extracts from the asphaltene fraction using reverse phase SPE cartridges (Supelco ENVI-18, 3 mL). Methanol extract (7.2 mg) was evaporated to dryness under a gentle stream of nitrogen. One mL of methanol:water (4:6) was added and the mixture was placed in a sonicator for one minute to allow for dissolution of soluble components. The supernatant liquid was then loaded at the top of the SPE cartridge. Another 2 mL of methanol:water (4:6) was then passed through the cartridge to elute the fraction CMhAsMe40. This cycle was repeated collecting 2 mL of eluent at 60, 4 mL of eluent at 80, and 6 mL of eluent at 100% methanol to elute the remaining fractions (CMhAsMe60, 80, and 100).

Acetone and THF extracts were eluted using a THF:water step gradient system. Acetone extract (7.8 mg) or THF extract (17.9 mg) was evaporated to dryness under a gentle stream of nitrogen. One mL of THF:water (1:4) was added and the mixture was placed in a sonicator for one minute to allow for dissolution of soluble components. The supernatant liquid was then loaded at the top of the SPE cartridge. Another 4 mL of THF:water (1:4) was then passed through the cartridge to elute the fraction

CMhAsAcet20 or CMhAsTHF20. This cycle was repeated collecting 5 mL of eluent at 40, 60, 80, and 100% THF to elute the remaining fractions (CMhAsAcet40, 60, 80, 100 and CMhAsTHF40, 60, 80, 100).

Asphaltene THF extract was also extracted on a normal phase SPE cartridge (Supelco LC-Si, 3 mL). This was done using a hexane:$CHCl_3$ step gradient. THF extract (6.2 mg) was evaporated to dryness under a gentle stream of nitrogen. One mL of hexane:$CHCl_3$ (4:1) was added and the mixture was placed in a sonicator for one minute to allow for dissolution of soluble components. The supernatant liquid was then loaded at the top of the SPE cartridge. Another 4 mL of hexane:$CHCl_3$ (4:1) was then passed through the cartridge. This cycle was repeated collecting 5 mL of eluent at 40, 60, 80, and 100% chloroform.

3.1.5 Preparation of Mobile Phases for HPLC and GPC

HPLC grade solvents were filtered three times through a 0.45 μm nylon filter prior to use to remove particulates and dissolved gasses.

3.1.6 Instrumentation and Run Conditions for Chromatographic Separations

3.1.6.1 High Performance Liquid Chromatography

A Beckman 110B LC Pump connected to a Beckman 210A 20μL injector and a Beckman 160 single wavelength detector set at 254 nm were used for HPLC experiments. A Hitachi D-2500 Chromato-Integrator was used for collection of the data. Normal phase HPLC was done on an amino-propyl bonded phase column (IBM Amino, 2.1 x 150

mm, dp = 5 μm) with 100% hexane mobile phase at a flow rate of 0.5 mL/min. Reverse phase HPLC was done on a C-18 bonded phase column (Supelco LC-18, 2.1 x 150 mm, dp = 5 μm). The mobile phase for the CMhAsMe fractions was 100% methanol at a flow rate of 0.5 mL/min. The mobile phase for the CMhAsAcet60 and CMhAsTHF40 fractions was THF:water (60:40) at a flow rate of 0.3 mL/min.

3.1.6.2 Gel Permeation Chromatography

A Tracor 951 LC Pump connected to a Rheodyne 20 μL injector and a Tracor 970A variable wavelength detector set at 254 nm was used for GPC. A Varian 4400 integrator was used for collection of the data. The mobile phase for GPC experiments was 100% THF at a flow rate of 0.5 mL/min.

3.1.6.3 Micellar Electrokinetic Capillary Chromatography

Capillary Electrophoresis was done on a Waters Quanta 4000 CE System. The run voltage for all separations was 15 kV. Capillaries were 75 μm in diameter. Samples were introduced by hydrostatic injection for ten seconds. The electrolyte solution consisted of sodium borate (5-8 mM) and Brij-s (40 mM). The electrolyte solution was adjusted to pH = 9.0 prior to addition of organic modifier (either acetonitrile or THF, 30 – 40%).

3.2 Results

3.2.1 Cleanup of Asphaltene Fractions Prior to HPLC Analysis

The CMhAsMe fraction passed completely through the reverse phase SPE cartridge when the methanol:water gradient was used as the eluent. This suggests that methanol extracts might be amenable to HPLC analysis using a reverse phase column and methanol:water mobile phase.

The CMhAsAcet fraction passed completely through the reverse phase SPE cartridge when the THF:water gradient was used for elution. Elution was not complete until the gradient reached 100% THF. This indicates that acetone extracts are amenable to HPLC analysis using a reverse phase column and THF:water mobile phase.

The CMhAsTHF fraction did not pass completely through the reverse phase SPE cartridge even when 100% THF was used as the eluent. Components with large condensed aromatic ring structures and/or long alkyl chains are highly retained on reverse phase packings. This means that only part of the THF soluble fraction can be analyzed by reverse phase HPLC only after SPE.

The CMhAsTHF fraction did not pass completely through the normal phase SPE cartridge even when 100% $CHCl_3$ was used as the eluent. The components with highly polar funtional groups and/or large numbers of aromatic rings are highly retained on normal phase columns; therefore, only part of the THF soluble asphaltenes can be analyzed using normal phase HPLC only after SPE.

These results seem to suggest that while many of the components contained in the asphaltene fraction are amenable to analysis by HPLC, those components containing

large aromatic ring systems cannot be separated by chromatography using columns that are currently available. Gel Permeation Chromatography is the only technique that will work for these kinds of molecules.

3.2.2 Chromatographic Separations

3.2.2.1 High Performance Liquid Chromatography

The polycyclic aromatic hydrocarbon standard mixture (EPA 610 PAH) was separated on a bonded phase amino column with 100% hexane mobile phase (figure 3.1a). Because there are no polar groups present in the polycyclic aromatic hydrocarbons, separation of these components with hexane mobile phase is based solely on the number of aromatic rings. Compounds with more aromatic rings are more highly retained because they are more polarizable. Nine peaks were detected from the sixteen component (EPA 610 PAH) mixture. This was expected as many of the components are isomeric and cannot be separated by using an amino or silica column. Although the individual standard compounds were not analyzed, peak assignments were made based on our knowledge of adsorption chromatography.

For the crude oil residue fractions, the result was group separation with individual components unresolved (Figures 3.1, 3.2, and 3.3). Comparison of the chromatogram for the CMhAr fraction to the chromatogram for the EPA 610 PAH mixture indicates that the CMhAr faction contains mostly one to three ring aromatic systems. The calculation of Brown and Ladner average stuctural parameters for the CMAr fraction indicated that there were five aromatic rings in the average molecule. It would seem that the molecules

in the CMhAr fraction should contain more aromatic rings on average than the molecules in the CMAr fraction. Perhaps the large number of bulky alkyl side chains prevent the aromatic systems in the CMhAr molecules from making adequate contact with the sorbent surface resulting in artificially lowered ring numbers by normal phase HPLC; however, the unavailability of crude oil like standard compounds complicates the confirmation of this theory. Ring number analysis by normal phase HPLC cannot be applied directly to the CMhResin fraction because of the presence of heteroatom containing species which are more highly adsorbed than are hydrocarbons.

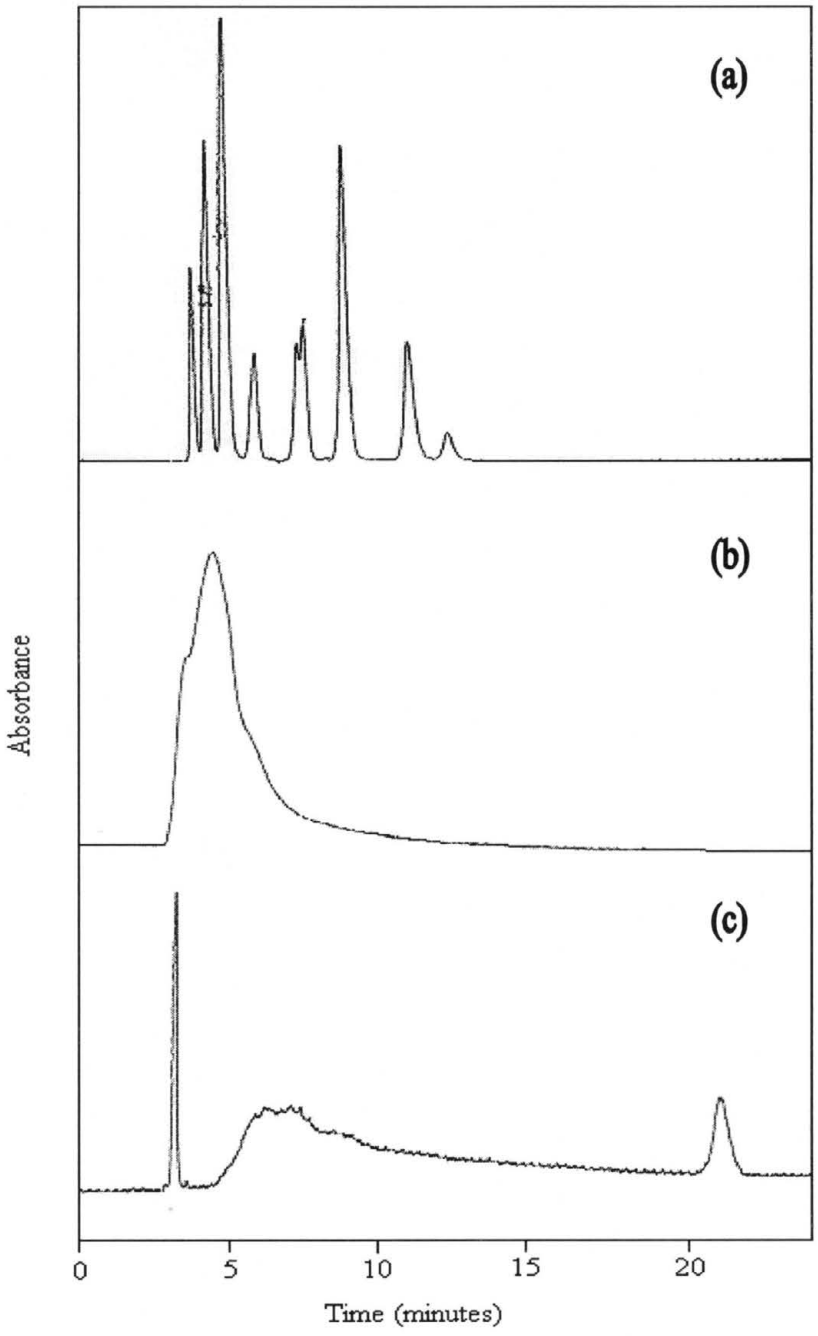

Figure 3.1: Normal phase HPLC Separation of a) EPA 610 PAH mixture, b) CMhAr, and c) CMhResins. Column, LiChrosorb Amino, 4.6 x 150 mm, dp = 5μm; Mobile Phase, 100% Hexane; Flow Rate, 0.5 mL/min.

Separation of the more polar fractions was done by reverse phase chromatography with an appropriate mobile phase. For the CMhAsMe fractions, 100% methanol mobile phase was used (figure 3.2). The CMhAsAcet and CMhAsTHF fractions were separated on a C-18 column with THF:water mobile phase (figure 3.3). Because some of the components in the THF extract of asphaltenes were found to be highly absorbing, SPE was done prior to any attempt at chromatography on an analytical column.

Because methanol is an excellent solvent for electrospray ionization / mass spectrometry (ESI/MS), the CMhAsMe fraction was chosen for further analysis using ESI/MS (figure 4.1). It is probable that the THF:water mobile phase used for the separation of the CMhAsAcet60 and CMhAsTHF40 fractions would be compatible with ESI/MS; however these experiments have not yet been performed.

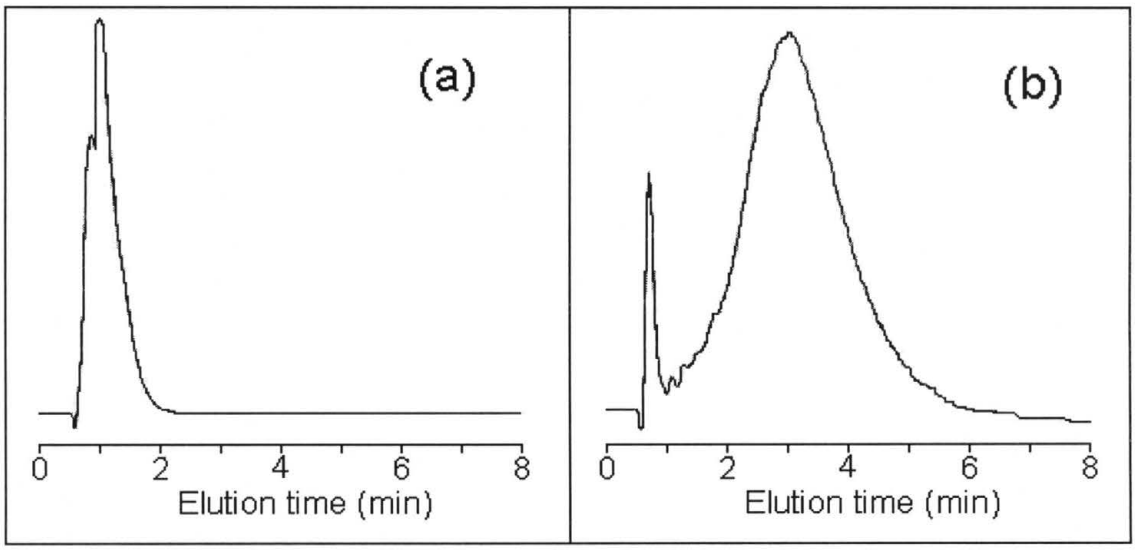

Figure 3.2: Reverse phase HPLC Separation of a) CMhAsMe80 and b) CMhAsMe100. Column, Supelco LC-18, 2.1 x 150 mm, dp = 5μm; Mobile Phase, 100% Methanol; Flow Rate, 0.5 mL/min.

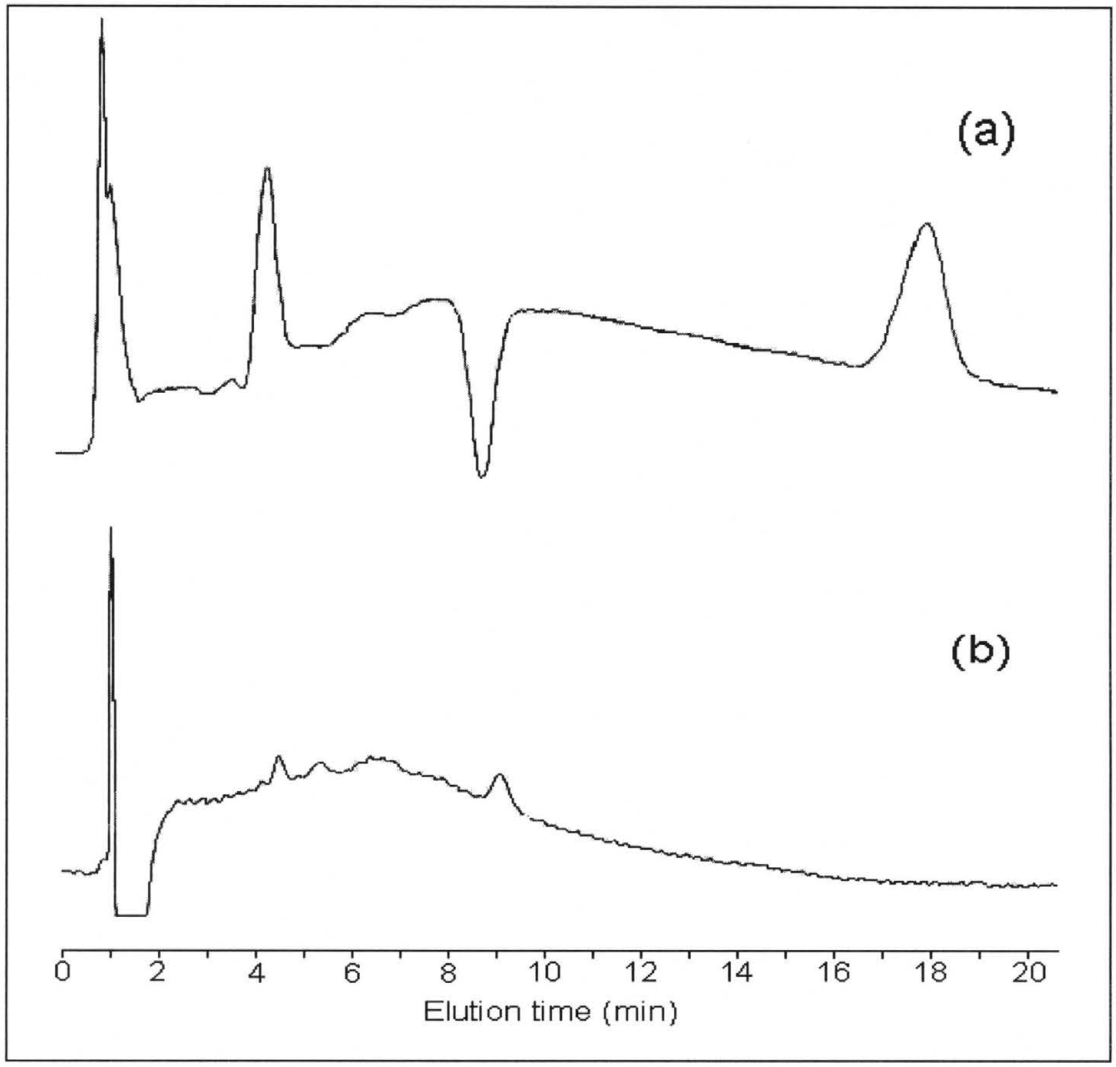

Figure 3.3: Reverse phase HPLC Separation of a) CMhAsAcet60 and b) CMhAsTHF40. Column, Supelco LC-18, 2.1 x 150 mm, dp = 5μm; Mobile Phase, THF:water (6:4); Flow Rate, 0.3 mL/min.

3.2.2.2 Gel Permeation Chromatography

GPC was performed on the standard components listed in section 3.1.3.2. The results were plotted as retention volume (mL) vs. log molecular weight (figure 3.4). GPC was performed on several fractions from the Maya crude oil residue in order to gain a better understanding of the molecular mass distribution of the residue and its subfractions

(figure 3.5). The calibration curve that was obtained for the standard compounds was used to convert the units for the x-axis of the GPC chromatograms from elution time (min) to units of molecular mass (amu). Number average molecular weights for some of the fractions (see Table 2.7) were calculated using the equation:

$$MW_{average} = \sum_{i} \frac{Area_{fraction}}{Area_{chromatogram}} * MW_{fraction}$$

where $MW_{average}$ = the average molecular weight of the entire crude oil fraction

$Area_{fraction}$ = the area of a specific elution interval

$Area_{chromatogram}$ = the area of the entire GPC chromatogram

$MW_{fraction}$ = the molecular weight at the midpoint of the elution fraction

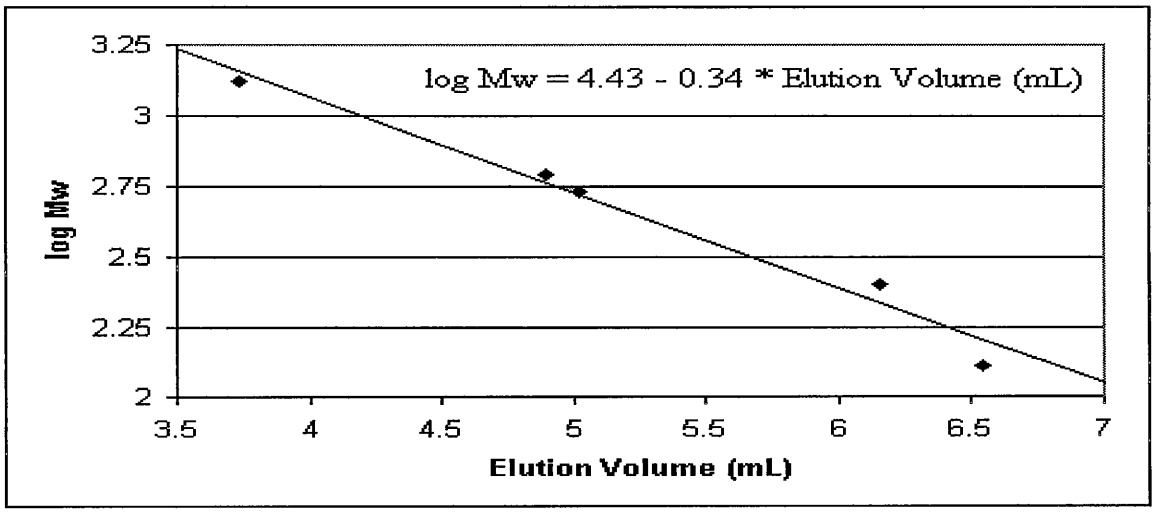

Figure 3.4: Calibration of GPC Column

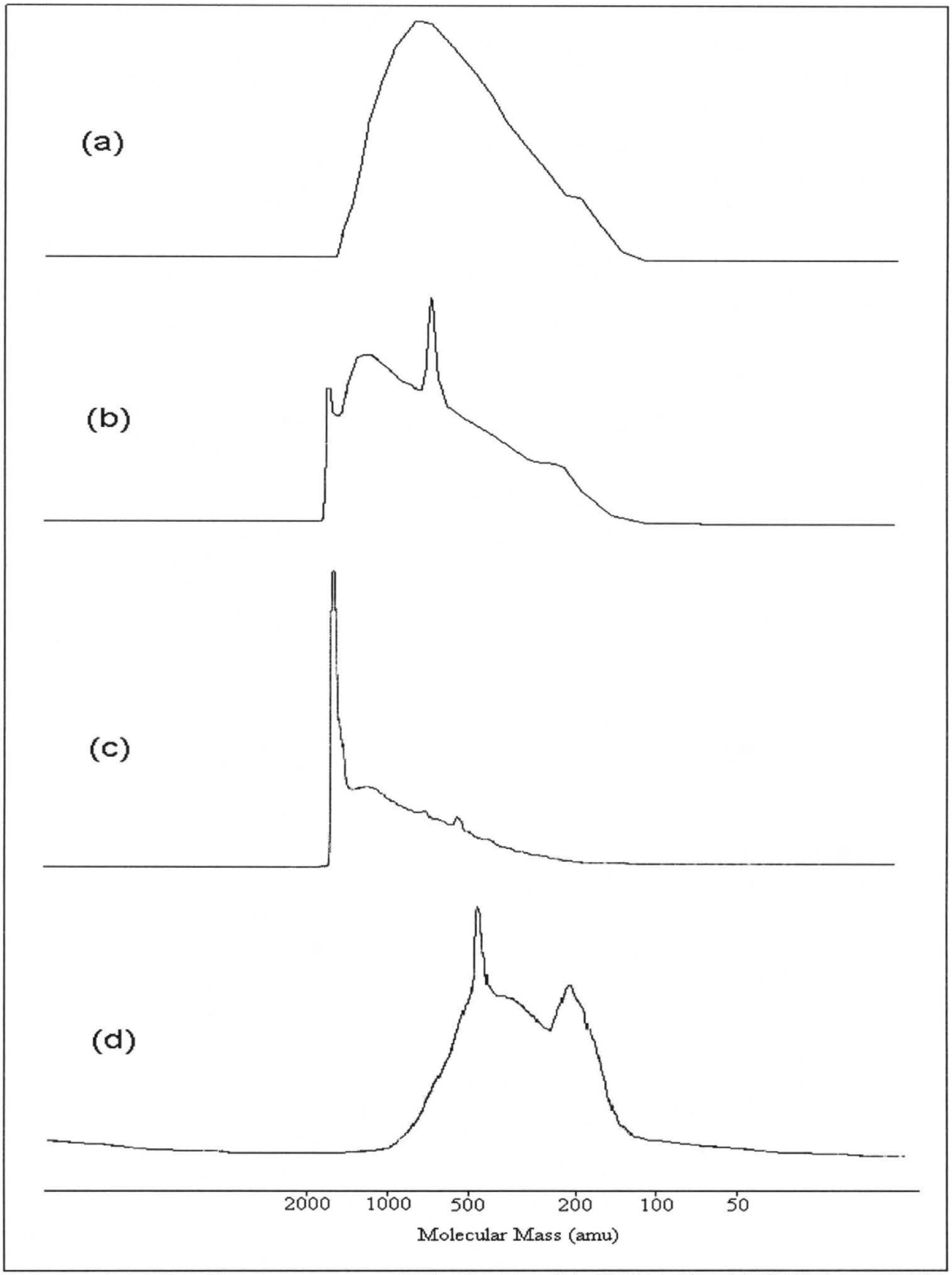

Figure 3.5: GPC Chromatogram of (a) CMhAr, (b) CMhResin, (c) CMhAs and (d) CMhAsMe. Column: Supelco Progel TSK G1000 HXL, 7.8 x 300 mm, dp = 6 μm; Mobile Phase, 100% THF; Flow Rate, 0.5 mL/min.

3.2.2.3 Micellar Electrokinetic Capillary Chromatography

The sixteen component EPA 610 PAH mixture was separated into fourteen peaks using micellar electrokinetic capillary chromatography (MECC) with acetonitrile:water electrolyte solution (figure 3.6). This separation was inferior to that which can be obtained using reverse phase HPLC [17]. MECC was also tested as a means of separating crude oils and their residues. It was found that the crude oil fractions were completely insoluble in the acetonitrile:water electrolyte solution used for electrophoresis of the standard mixture. An electrolyte solution with THF:water was more successful at dissolving some of the fractions, but not many experiments were performed. It is conceivable that MECC could be used for separation of some of the fractions. Further experimentation was not performed because HPLC is a better technique for separation of the crude oil fractions and standard compounds.

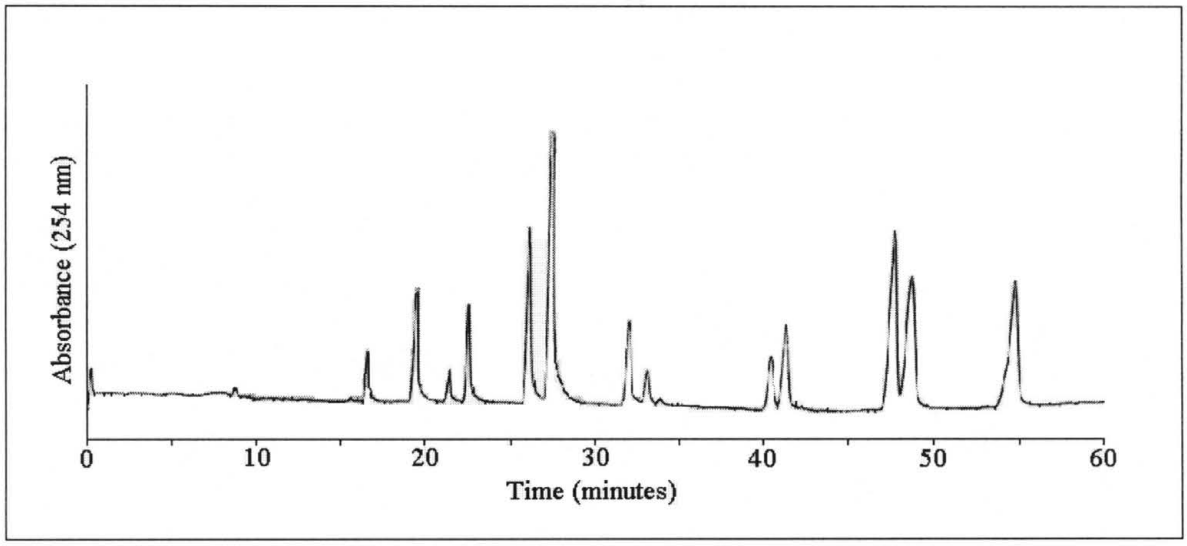

Figure 3.6: MECC of EPA 610 PAH Mixture. Electrolyte: 40mM Brij-S, 8mM sodium borate, 40% acetonitrile, 60% water, ph = 9.0; applied voltage, 15kV; current, 76μA; hydrostatic injection time, 20 sec; capillary, 53 cm x 75μm i.d.; detection, 254 nm.

CHAPTER 4

MASS SPECTROMETRY

4.1 EXPERIMENTAL

4.1.1 Reagents and Chemicals

Sources for all materials are listed: methanol, hexane, CH_2Cl_2 and $CHCl_3$ from EM Science (Gibbstown, NJ); $PdCl_2$ from Aldrich (Milwaukee, WI).

4.1.2 Preparation of Standards

A mixture containing benzothiophene (5.68 mM), dibenzothiophene (1.31 mM), 4,6-dimethyl-dibenzothiophene (1.06 mM), benzo-naphthothiophene (0.333 mM), and thianthrene (0.841 mM) was prepared by weighing a small amount of each component into a 50 mL volumetric flask. The flask was filled to the 50 mL mark, and placed in a sonicator for five minutes to allow for dissolution of all components.

A mixture containing fluoranthene (2.43 mM), acenaphthene (2.52 mM), phenanthrene (2.95 mM), and dibenzothiophene (3.23 mM) was prepared by weighing a small amount of each component into a 10 mL vial. A 10 mL pipet was used to add 8.75 mL of methanol to the mixture, and the mixture was placed in a sonicator for five minutes to allow for dissolution of all components.

4.1.3 Instrumentation

Mass spectra were obtained on a Finnigan LCQ equipped with atmospheric pressure chemical ionization (APCI) or electrospray ionization (ESI) source. A Thermo Separation Products vacuum membrane degasser and P4000 LC Pump comprised the solvent delivery system. A Thermo Separation Products AS3000 autosampler was used for injection of 20μL samples.

Ionization by APCI was accomplished by passing the sample through a heated vaporizer tube from which it emerges near the corona discharge needle as a nebulized spray. The needle is held at high voltage which creates a corona discharge current that forms reagent ions through a series of chemical reactions with solvent molecules and nitrogen sheath gas. These reagent ions react with sample molecules to form sample ions. APCI is used for samples with medium polarity that have some volatility.

ESI is used for samples that readily form ions in solution. The ESI needle sprays the sample solution into a fine mist of droplets that are charged at the surface. As the solvent evaporates, the electrical charge density at the surface of the droplets increases. From the very small, highly charged droplets, sample ions are ejected into the gas phase by electrostatic repulsion. For both APCI and ESI sources, nitrogen was used as the sheath and auxiliary gas. For detection of PASH species, 0.59mM $PdCl_2$ in methanol was added as a sheath liquid coaxially to the ES needle.

The ions formed in the atmospheric pressure region are pulled through a heated capillary by a decreasing pressure gradient. After passing through a tube lens and skimmer arrangement, the ions are transferred by the ion optics into an ion trap mass analyzer. A Gateway 2000 computer with Navigator 2.1 software was used for collection

and analysis of the data. The run conditions for the LC/MS analysis of all samples are reported with each Figure.

4.1.4 Bacterial Desulfurization Studies

Bacterial Desulfurization studies were performed in Dr. Linette Watkins' lab [19]. The bacterial strain *Rhodococcus strain sp.* IGTS8 was obtained from the American Type Culture Collection (ATCC 53968). Two different media were used for the growth of the *Rhodococcus strain sp.* IGTS8: a Difco nutrient media and a sulfur-free minimal media designated BSM2 by Li, et al [20].

Rhodococcus strain sp. IGTS8 was grown overnight to saturation in 5 mL tubes containing nutrient media. The cells were then concentrated by centrifugation and washed twice with BSM2 media, before suspension in 50 mL of BSM2 media. The sulfur source necessary for the growth was excluded in the negative control and was 200 uM DBT in the positive control. A 50 mg aliquot obtained from the aromatic fraction was dissolved in 5 mL of decane, filtered through a 0.2 μm filter (Corning, N. Y.) and then added to flasks inoculated with the bacteria. Cultures and sterile controls were incubated for 5 days at 30°C, with shaking at 220 rpm.

After 5 days, the pH of each culture was lowered to 2.0 by adding 3 M HCl prior to extraction with CH_2Cl_2. Each flask containing 50 mL of culture was extracted three times with 10 mL of CH_2Cl_2. The combined fractions were evaporated to remove CH_2Cl_2 and decane. The remaining desulfurized fraction was dissolved in a 1 mL mixture of CH_2Cl_2:methanol (30:70). This must be done by first adding 0.3 mL CH_2Cl_2 and then adding 0.7 mL methanol, otherwise the fraction will not dissolve. The fraction

was then passed through an SPE cartridge (Supelco ENVI-18, 3mL) with 10 mL of methanol:CH_2Cl_2 (70:30) before analysis with the LC/ESI/MS protocol (*vide infra*). The non-desulfurized aromatics were also subjected to the same SPE procedure prior to analysis by LC/ESI/MS.

4.2 RESULTS

4.2.1 HPLC/ESI/MS Results for the Methanol Extract from the Asphaltene Fraction of the Maya Crude Oil Residue.

Some of the components contained in the asphaltene fraction are easily ionizable and hence can be efficiently detected using LC-MS. Compounds such as metalloporphyrins, ionic salts, polyhydric phenols, and naphthenic acids are soluble in methanol and hence can easily be removed from the rest of the asphaltene fraction which consists of mostly high molecular weight extended polycyclic aromatic ring systems. Asphaltenes were extracted with methanol (60 mL / g, 4 hours). The methanol extract was then analyzed with HPLC/ESI/MS in the negative ion mode (see Figure 4.1).

Inspection of the mass spectrum for the CMhAsMe fraction shows the presence of many peaks that are separated by 14 amu. Differences of 14 are indicative of the presence of a homologous series. Careful inspection reveals at least two homologous series starting at m/z = 272.8 and the other starting at m/z = 294.9.

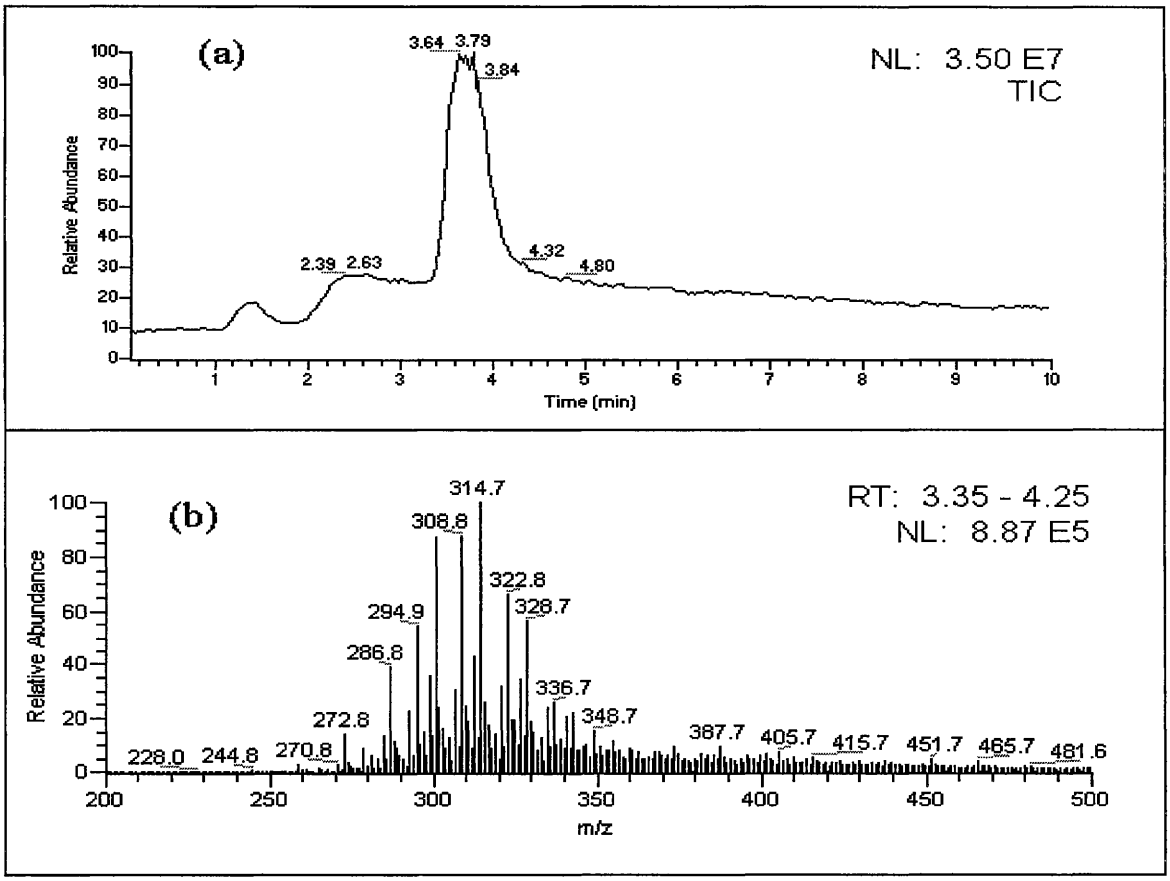

Figure 4.1: HPLC/ESI/MS of CMhAsMe Fraction: (a) Total Ion Chromatogram and (b) Mass Spectrum. Column, Supelco LC-18 (2.1 x 150 mm, dp = 5 μm); mobile phase, 100% methanol; flow rate, 0.2 mL/min; spray voltage, - 4.5 kV; capillary temperature, 200°C; sheath gas, N_2 at 60; auxillary gas, N_2 at 20.

4.2.2 HPLC/APCI/MS Results for Low Polarity Fractions

APCI/MS was used for the analysis of several fractions from a Maya crude oil (Figures 4.2 and 4.3) and a Maya crude oil residue (Figure 4.4). Number average molecular weights were calculated from the 1000 largest peaks in each spectrum. Using

less than 1000 peaks resulted in significant error in the calculation. The mass spectra obtained can be considered fingerprint chromatograms for the Maya crude oil residue; however, elucidation of molecular structure for individual components is practically impossible due to the extreme complexity of the mixtures. Furthermore, because of the very low sensitivity of APCI/MS for the PAH standard compounds, it is questionable if the ions being detected are actually saturate, aromatic, and low polarity compounds. The presence of metal-analyte complexes and/or heteroatomic species could be contributing significantly to the signals that are obtained.

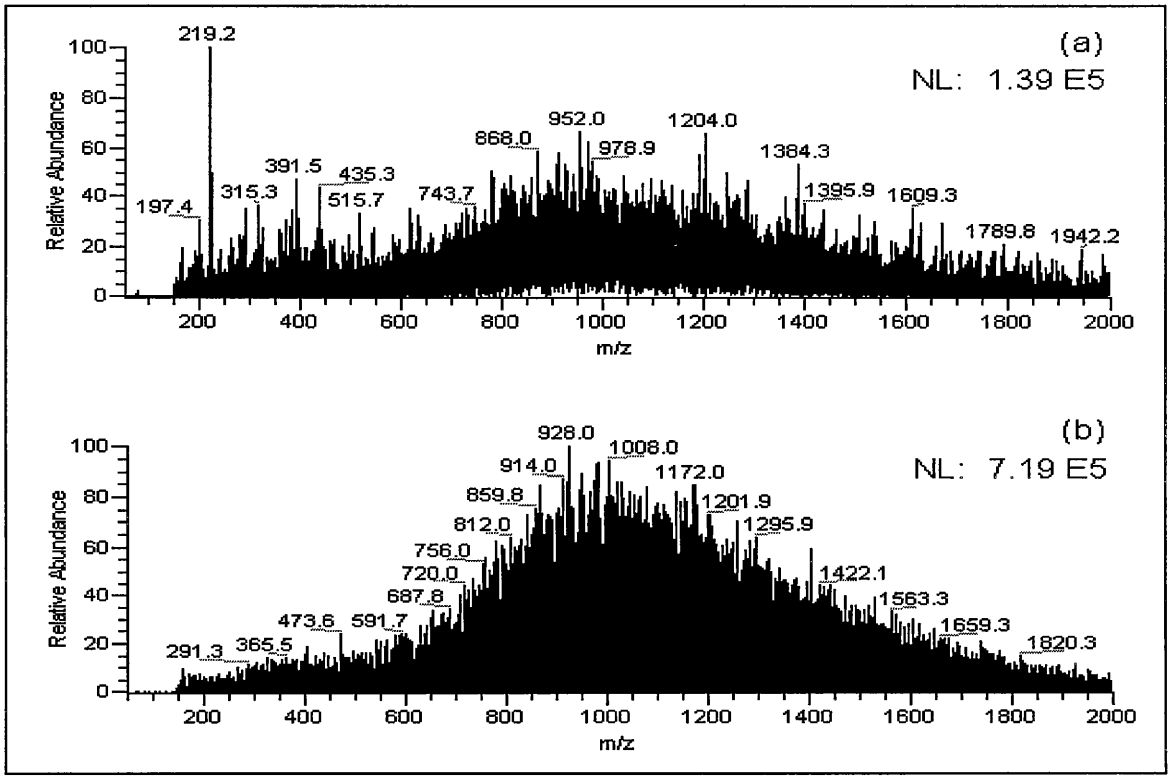

Figure 4.2: APCI-MS of (a) CMAliph and (b) CMAliph-S Fractions from Maya Crude Oil. Column, Supelco LC-NH_2 (3.0 x 250 mm, dp = 5 μm); Mobile Phase, Hexane:$CHCl_3$ (99:1); Flow Rate, 1.00 mL/min; Vaporizer Temperature, 450°C; Corona Discharge Current, 5 μA; Capillary Temperature, 150°C; sheath gas, N_2 at 80; auxiliary gas, N_2 at 20.

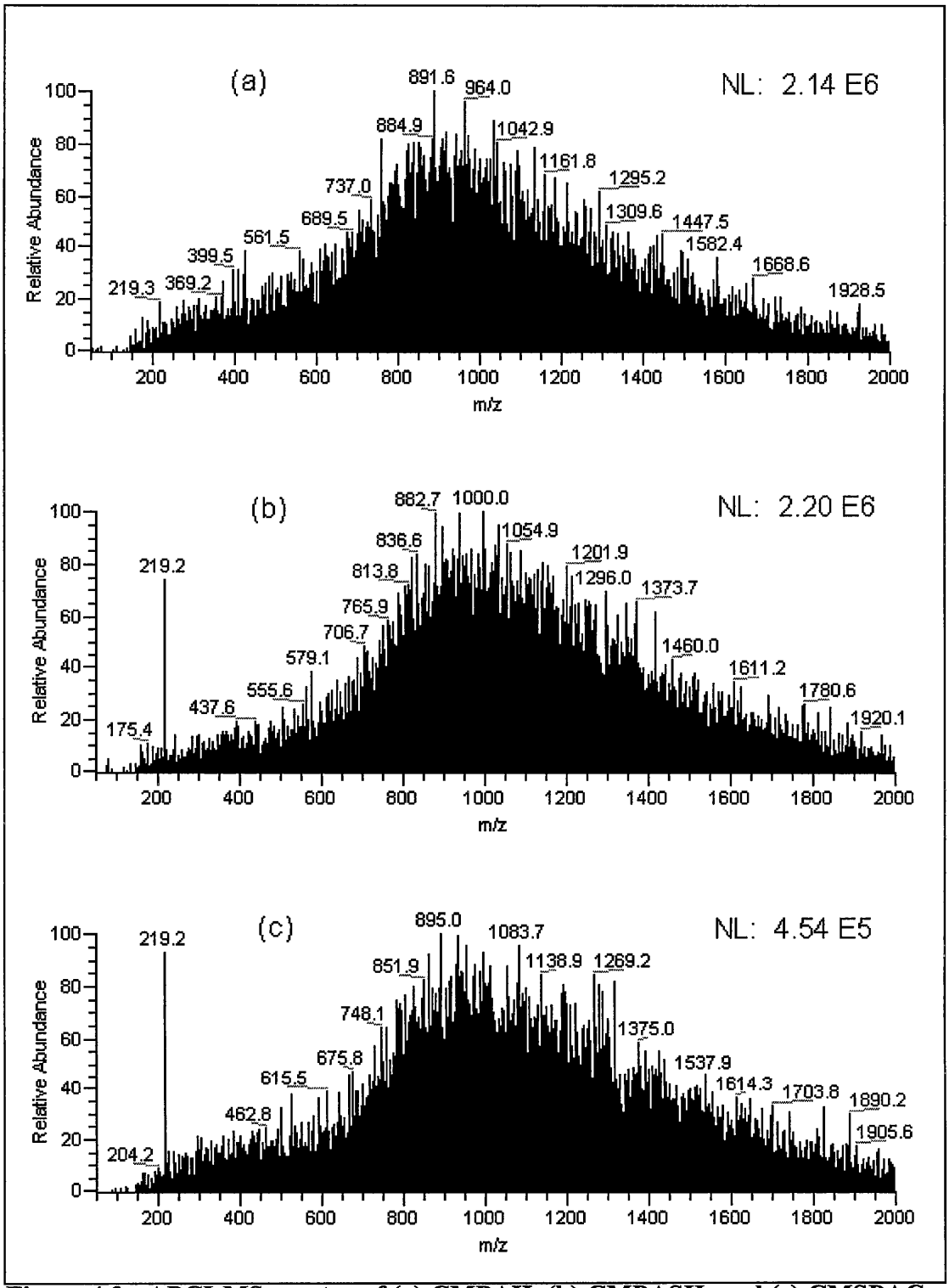

Figure 4.3: APCI-MS spectra of (a) CMPAH, (b) CMPASH, and (c) CMSPAC Fractions from Maya Crude Oil. Column, Supelco LC-NH_2 (3.0 x 250 mm, dp = 5 μm); Mobile Phase, Hexane:$CHCl_3$ (1:1); Flow Rate, 1.00 mL/min; Vaporizer Temperature, 450°C; Corona Discharge Current, 5 μA; Capillary Temperature, 150°C; sheath gas, N_2 at 80; auxiliary gas, N_2 at 20.

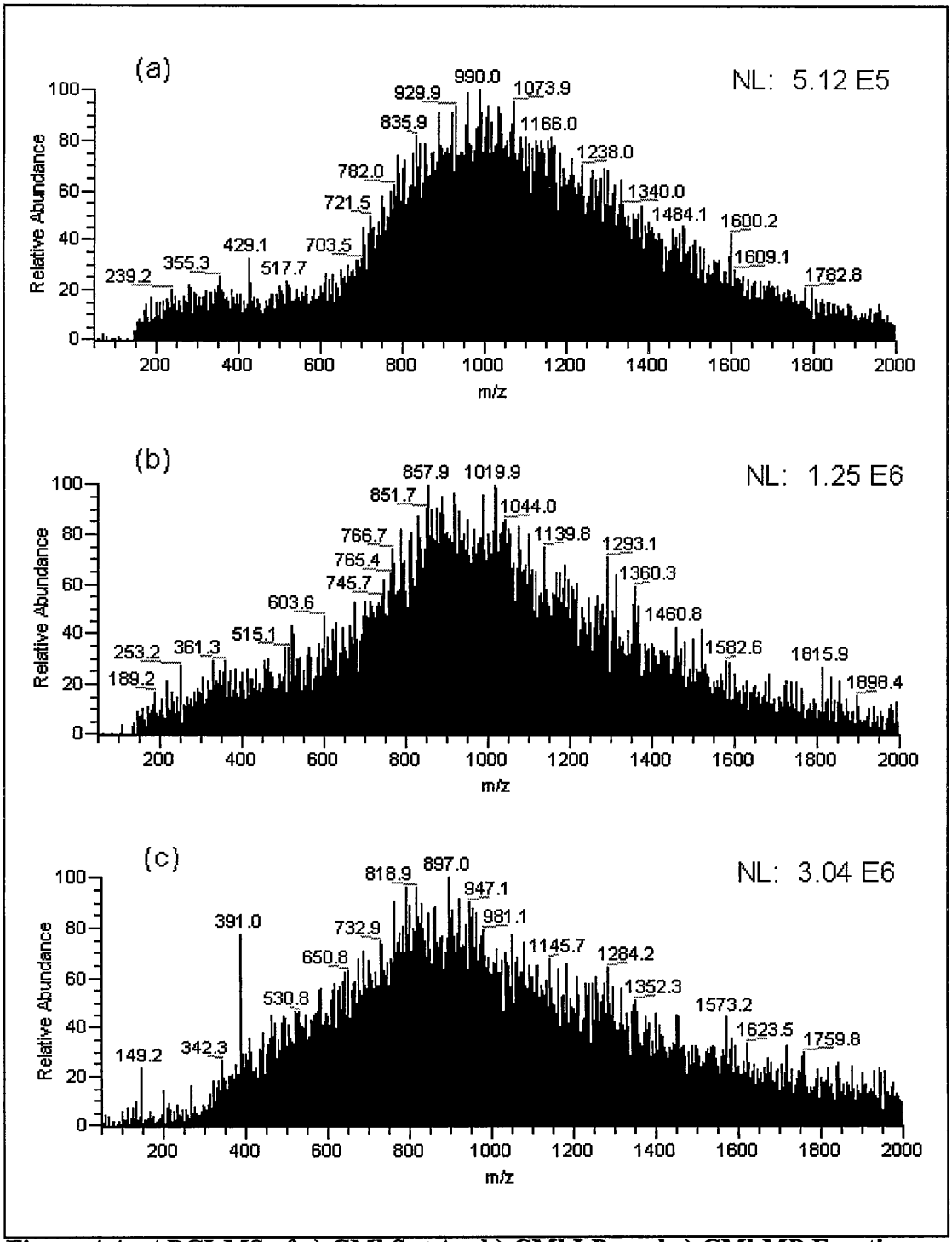

Figure 4.4: APCI-MS of a) CMhSa+Ar, b) CMhLP, and c) CMhMP Fractions From Maya Crude Oil Residue. Column, Supelco LC-NH_2 (3.0 x 250 mm, dp = 5 μm); mobile phase gradient, hexane:$CHCl_3$ (a & b = 0 – 10 % $CHCl_3$ in 20 minutes, c = 0 - 50 % $CHCl_3$ in 30 minutes); flow rate, 1.00 mL/min; vaporizer temperature, 450°C; corona discharge current, 5 μA; capillary temperature, 150°C; sheath gas, N_2 at 80; auxiliary gas, N_2 at 20.

4.2.3 MS Results for Organosulfur Compounds

Both ESI/MS and APCI/MS of dibenzothiophene (DBT) yield poor mass sensitivity even at a concentration of 1420 mg/L (Figure 4.5a). The peak for the protonated species is barely distinguishable from the noise. This is attributed to the low proton affinity of the DBT [2]. Oxidation of DBT into the corresponding sulfone can increase the sensitivity. The oxidation is carried out in CH_2Cl_2 using the oxidizing agent *m*-chloroperoxybenzoic acid. This converts the dibenzothiophene into the sulfone with 100% yield. Excess acid is removed by extraction with 10% sodium bicarbonate solution. The sulfone is easily protonated and can be detected at a concentration of 10 ppm with a linear dynamic range from 10 – 500 ppm. Dibenzothiophene sulfone has a sensitivity approximately 3700 times greater than that of the DBT (compare Figure 4.5b with Figure 4.5a).

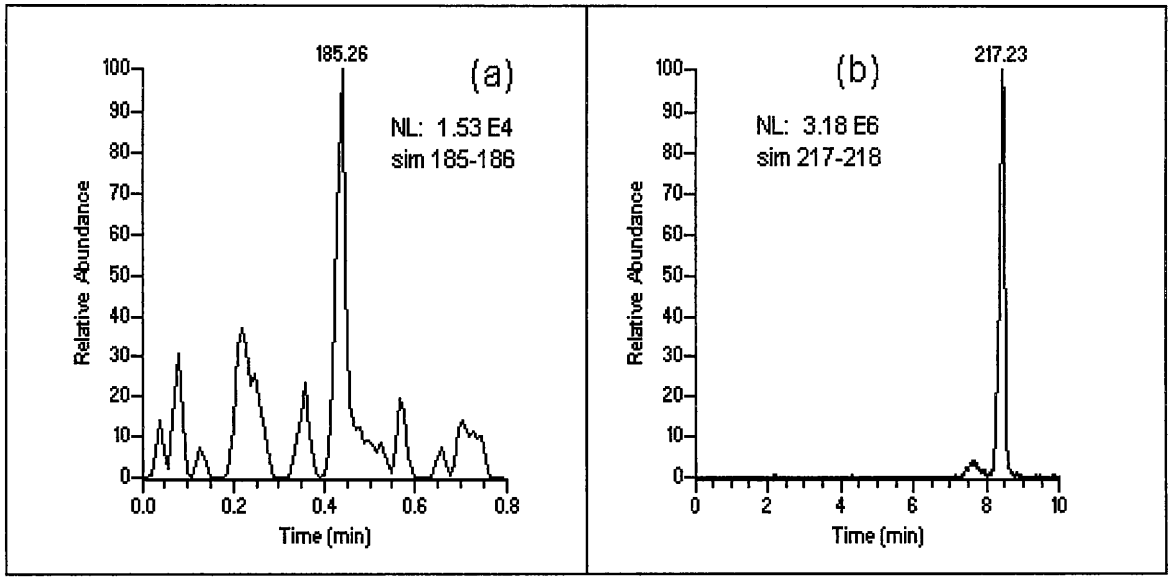

Figure 4.5: APCI-MS of a) DBT (1420ppm) and b) DBT sulfone (80 ppm). Column, Supelco LC-NH_2 (3.0 x 250 mm, dp = 5 μm); mobile phase, hexane:$CHCl_3$ (85:15); flow rate, 1.0 mL/min; vaporizer temperature, 450°C; corona discharge current, 5 μA; capillary temperature, 150°C; sheath gas, N_2 at 80; auxiliary gas, N_2 at 20.

PASHs are known to form charged complexes with Pd^{2+}. By adding $PdCl_2$ in methanolic solution after the chromatographic separation, the sensitivity for dibenzothiophene is increased (figure 4.6). This procedure uses a reverse phase chromatographic column with methanol mobile phase for introduction of the sample into an electrospray interface. Dibenzothiophene was detected as the molecular ion (m/z = 184). Several peaks at higher m/z values can also be attributed to dibenzothiophene-Pd complexes. Dibenzothiophene could be detected with a detection limit of 10 ppm. The sensitivity of ESI-MS for dibenzothiophene is 400 times greater when palladium is added as a sheath liquid.

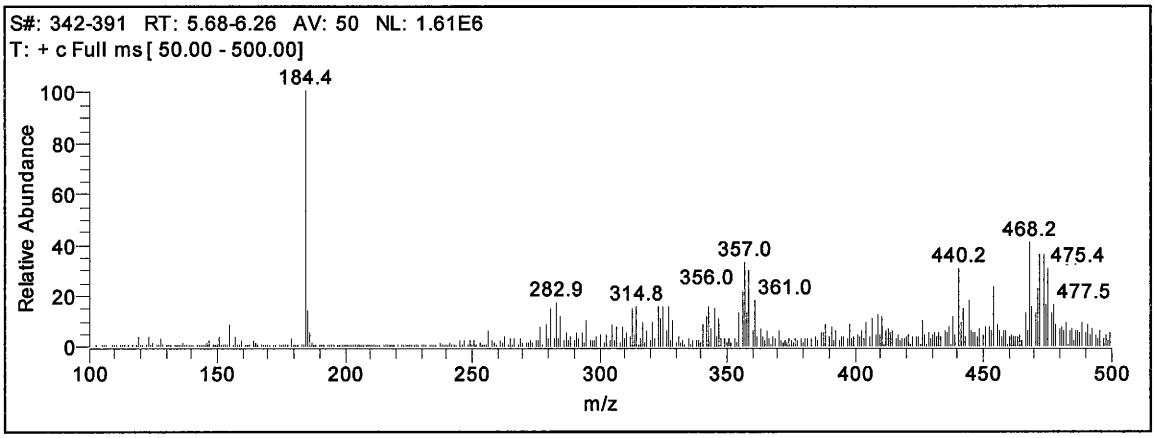

Figure 4.6: ESI-MS of DBT (708 ppm) with $PdCl_2$ Sheath Liquid. Column, Supelco LC-18 (2.1 x 150 mm, dp = 5 μm); Mobile Phase, 100% Methanol; Flow Rate, 0.1 mL/min; Spray voltage, 4.5 kV; Sheath Gas, Auxillary Gas, N_2; sheath liquid, $PdCl_2$ in methanol (0.56 mM) at 20 μL/min.

Experiments were conducted to determine optimal conditions for the reverse phase HPLC/ESI/MS detection of DBT using $PdCl_2$ sheath liquid (Table 4.1). The most important factors that affect sensitivity are the sheath gas flow rate, mobile phase flow rate, and the $PdCl_2$ sheath liquid flow rate. Typical sheath gas flow is 60 – 80 units for a mobile phase flow of 0.1 – 0.2 mL/min; however, at this setting there is too much

Table 4.1 Sheath Liquid Experiment Data on DBT using ESI-MS

Sheath Gas Flow	Mobile Phase Flow (mL/min)	$PdCl_2$ Sheath Flow (μL/min)	ES Voltage (kV)	Response ($x10^3$)
80	0.2	30	3.5	19500
80	0.2	30	1.0	12800
80	0.2	10	3.5	9050
40	0.2	20	4.5	1570
20	0.4	20	4.5	76
20	0.2	20	4.5	370
20	0.2	20	3.5	226
20	0.2	20	2.5	171
20	0.2	20	1.5	27
20	0.2	10	4.5	2
20	0.1	20	4.5	2650

DBT, 3.04 mM in methanol; $PdCl_2$, 0.56 mM in methanol; capillary temperature, 200°C; auxiliary gas, N_2 at 10; sheath gas, N_2.

background noise for efficient detection of the PASH components. Although the absolute response for DBT is much lower at a sheath gas flow of 20 units, the signal to noise ratio is greatly improved. Typically ESI gives better response with lower mobile phase flow rates because there is less solvent that needs to be evaporated. Adjustment of the $PdCl_2$ sheath liquid flow also seems to be critical. At a mobile phase flow of 0.2 mL/min, a sheath gas flow setting of 20, and a sheath liquid flow of 10 μL/min the response was too low. With these settings, the DBT:Pd mole ratio in the mobile phase entering the ES needle is 109:1. A much greater response is seen with a mobile phase flow of 0.1 mL/min and sheath liquid flow of 20 μL/min (DBT:Pd = 29.8:1). It was

determined that the optimal conditions are: mobile phase flow rate = 0.1mL/min and sheath liquid flow rate = 10 μL/min. These settings not only maximize the absolute response but also minimize the amount of solvent and $PdCl_2$ entering the mass spectrometer.

HPLC/ESI/MS using $PdCl_2$ sheath liquid was tested as a means of selectively detecting PASH components in mixtures containing both sulfur and non-sulfur polycylic aromatic species. Only dibenzothiophene was detected when the standard mixture containing fluoranthene (2.43 mM), acenaphthene (2.52 mM), phenanthrene (2.95 mM) and dibenzothiophene (3.23 mM) was analyzed. Conditions for the analysis are listed in Table 4.2. Another standard mixture containing benzothiophene (5.68 mM), dibenzothiophene (1.31 mM), 4,6-dimethyl-dibenzothiophene (1.06 mM), benzo-naphthothiophene (0.333 mM), and thianthrene (0.841 mM) was analyzed using the same experimental conditions. The response for PASH species appears to depend on the number of aromatic rings and the number of sulfur atoms in the molecule (Table 4.2). In general PASH compounds containing three or more aromatic rings were amenable to detection using this method.

Results from SPE studies [18] using $PdCl_2$ impregnated aminopropyl bonded SPE cartridges (Supelco LC-NH_2, 3 mL) seem to agree with the results of the ESI/MS experiments. The affinity of PASH components for the Pd^{2+} ion increases with the number of aromatic rings and the number of sulfur atoms.

Table 4.2 Comparison of ESI-MS Response and $PdCl_2$-SPE Retention for Polycyclic Aromatic Compounds

Compound	ESI[a]		SPE		
	Response[b]	Relative response	HPLC response before[c]	HPLC response after[c]	% Retained
Phenanthrene	0	0	57.9	57.9	0
Chrysene	- -	- -	28.7	28.6	0
Benzo(k)-Fluoranthene	- -	- -	20.2	18.2	10
Benzo(ghi)perylene	- -	- -	4.0	1.6	60
Benzothiophene[d]	0.02	0	29.0	21.1	27
Dibenzothiophene[d]	3.43	1.00	20.5	10.6	48
4,6-dimethyl dibenzothiophene[d]	3.30	0.96	- -	- -	- -
Benzo-Naphthothiophene[d]	90.4	26.4	- -	- -	- -
Thianthrene[d]	133.2	38.8	31.3	0	100

[a] Mobile phase, 100% methanol; flow rate, 0.1 mL/min; spray voltage, 4.5 kV; sheath gas, N_2 at 20; auxillary gas, N_2 at 10; sheath liquid, $PdCl_2$ in methanol (0.56 mM) at 10 µL/min. [b] area/concentration x 10^{-5} [c] area x 10^{-3}
[d] these are three and four ring polycyclic aromatic sulfur heterocycles

4.2.4 Bacterial Desulfurization Studies

HPLC/ESI/MS analysis of the aromatic fraction was conducted before and after bacterial biodesulfurization both in the absence (see Figure 4.6) and in the presence (Figure 4.7) of $PdCl_2$ sheath liquid. Without the $PdCl_2$ sheath liquid, the sulfur containing species can be expected to give zero response, and any response that is seen can be attributed to the presence of oxygen and nitrogen containing species. The response (see Figure 4.6) after biodesulfurization for the aromatic fraction was larger than the response before biodesulfurization in the absence of $PdCl_2$ sheath liquid. This can be attributed to the extraction of biological materials from the cells. The presence of these biological compounds does not significantly interfere with the measurement of percent sulfur reduction because most of the biological compounds elute before five minutes while most of the sulfur compounds elute after five minutes. With $PdCl_2$ sheath liquid, most of the response that is seen after five minutes is due to the presence of sulfur containing species.

Table 4.3 lists the area response for elution fractions before and after biodesulfurization for the aromatic fraction. After six minutes, there is a general decrease in area response for all of the elution intervals. Because the sulfur containing species are expected to elute in this range, this is evidence that the bacteria are indeed reducing the sulfur levels in the aromatic fraction. The total area for the 6 – 15 min fraction is reduced by 43%. The positive change in area response after bio-desulfurization for the 2.75 – 6 min range is attributed to the presence of extracted biological materials as well as polar biodesulfurization products.

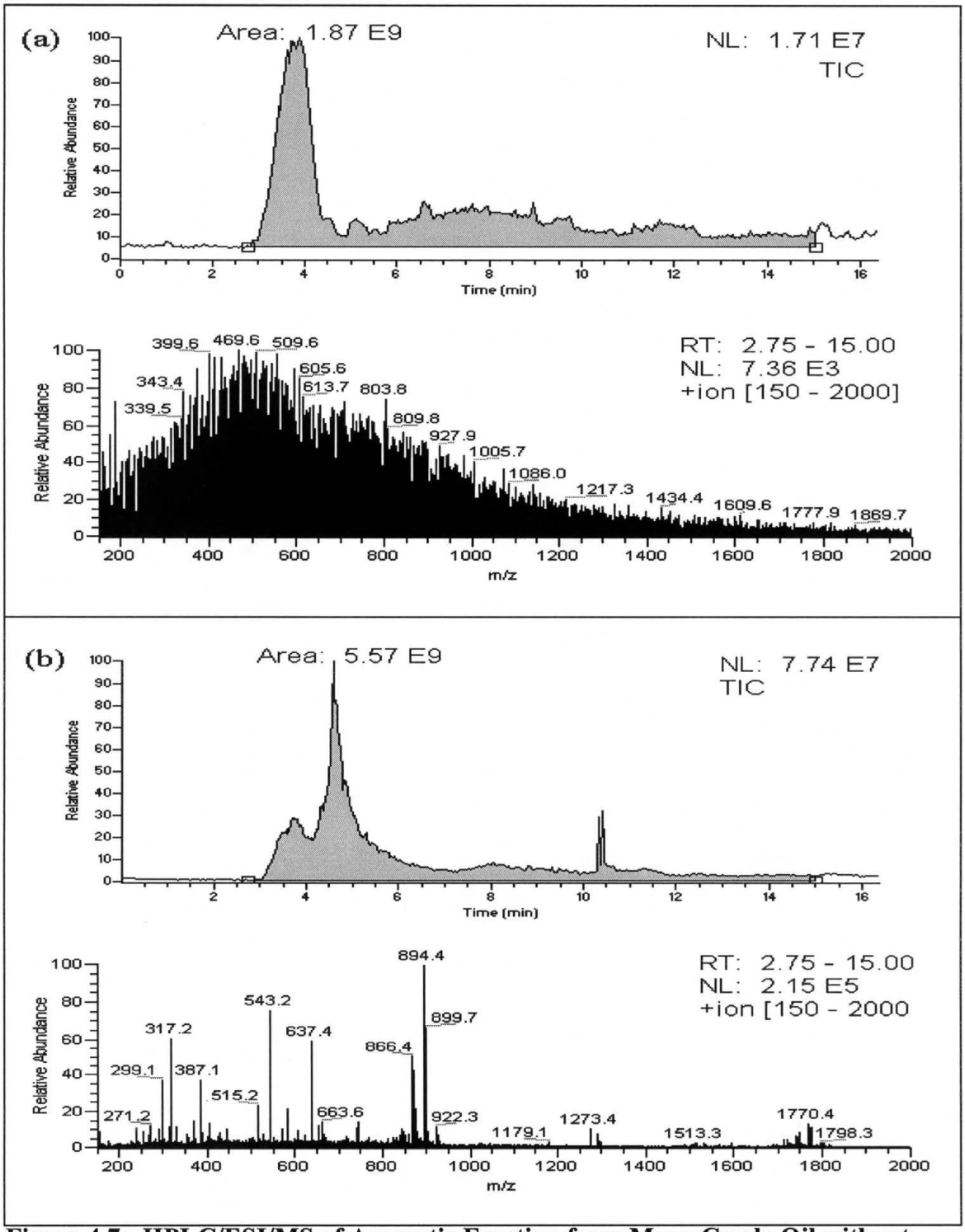

Figure 4.7: HPLC/ESI/MS of Aromatic Fraction from Maya Crude Oil without $PdCl_2$ Sheath Liquid (a) Before (9029 ppm) and (b) After (8308 ppm) Bio-desulfurization. Column, Supelco LC-18 (2.1 x 150 mm, dp = 5μm); mobile phase, methanol:CH_2Cl_2 (8:2); flow rate, 0.1 mL/min; spray voltage, 4.5 kV; sheath gas, N_2 at 80; auxillary gas, N_2 at 20.

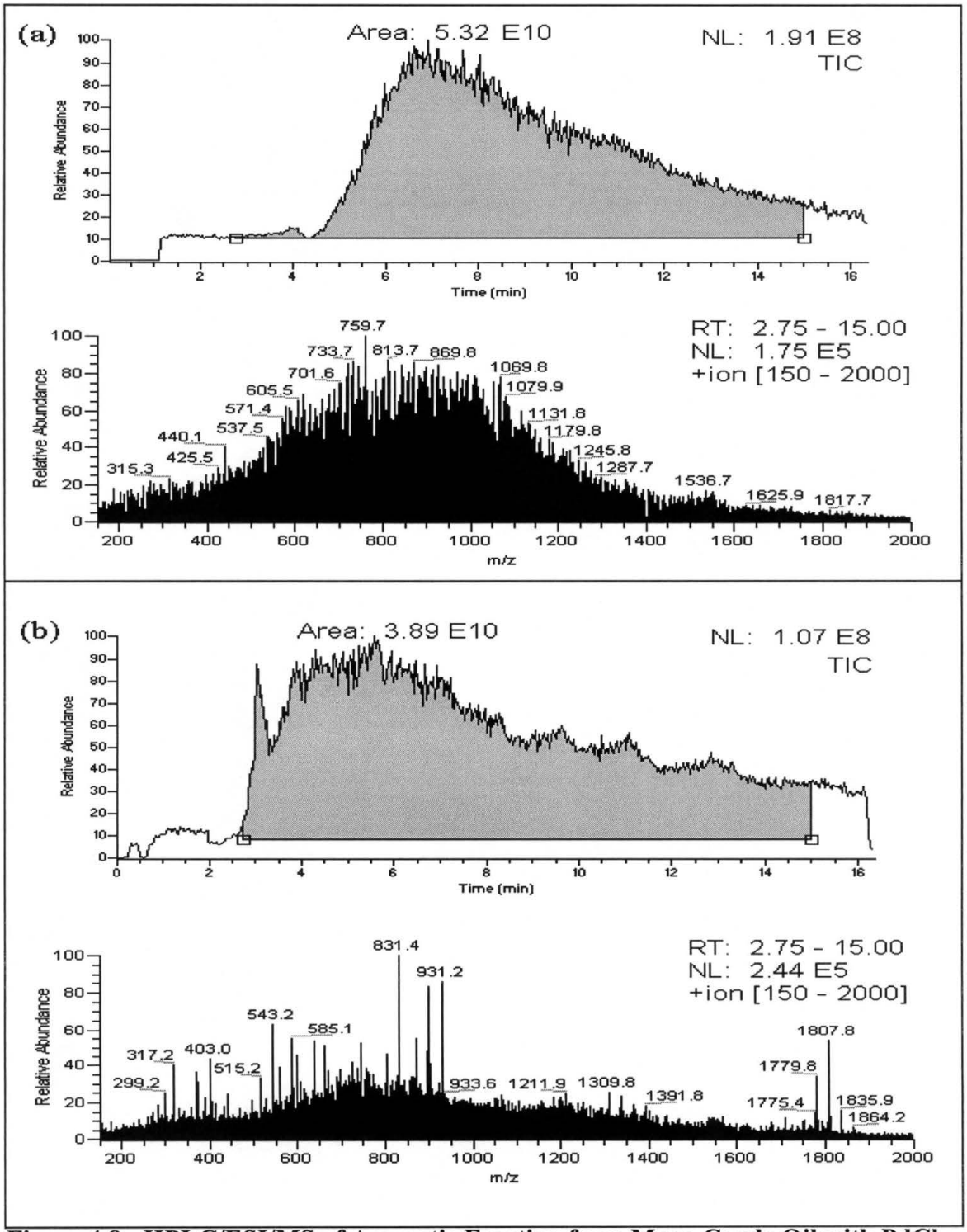

Figure 4.8: HPLC/ESI/MS of Aromatic Fraction from Maya Crude Oil with $PdCl_2$ sheath liquid (a) Before (9029 ppm) and (b) After (8308 ppm) Bio-desulfurization. Column, Supelco LC-18 (2.1 x 150 mm, dp = 5μm); mobile phase, methanol:CH_2Cl_2 (8:2); flow rate, 0.1 mL/min; spray voltage, 4.5 kV; sheath gas, N_2 at 80; auxillary gas, N_2 at 20; sheath liquid, $PdCl_2$ in methanol (0.56 mM) at 20 μL/min.

Table 4.3: ESI/MS Response of HPLC Elution Fractions from CMAr Before and After Bio-desulfurization.

Elution Time (min)	Area Response Before Bio-desulfurization[a]	Number Average Molar Mass[b]	Area Response After Bio-desulfurization[a]	% Change
2.75-5	10	– – –	109	+ 990
5-6	48	673	62	+ 29
6-7	97	794	56	- 42
7-8	94	872	46	- 51
8-10	146	928	71	- 51
10-12	106	955	59	- 44
12-15	87	909	69	- 21
Σ 6-15	531	889	301	- 43

[a] area x 10^{-4} [b] calculated from the largest 1000 masses

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APPENDIX A

FT-IR SPECTRA OF MAYA CRUDE OIL FRACTIONS

A.1: FT-IR of Maya Crude Oil

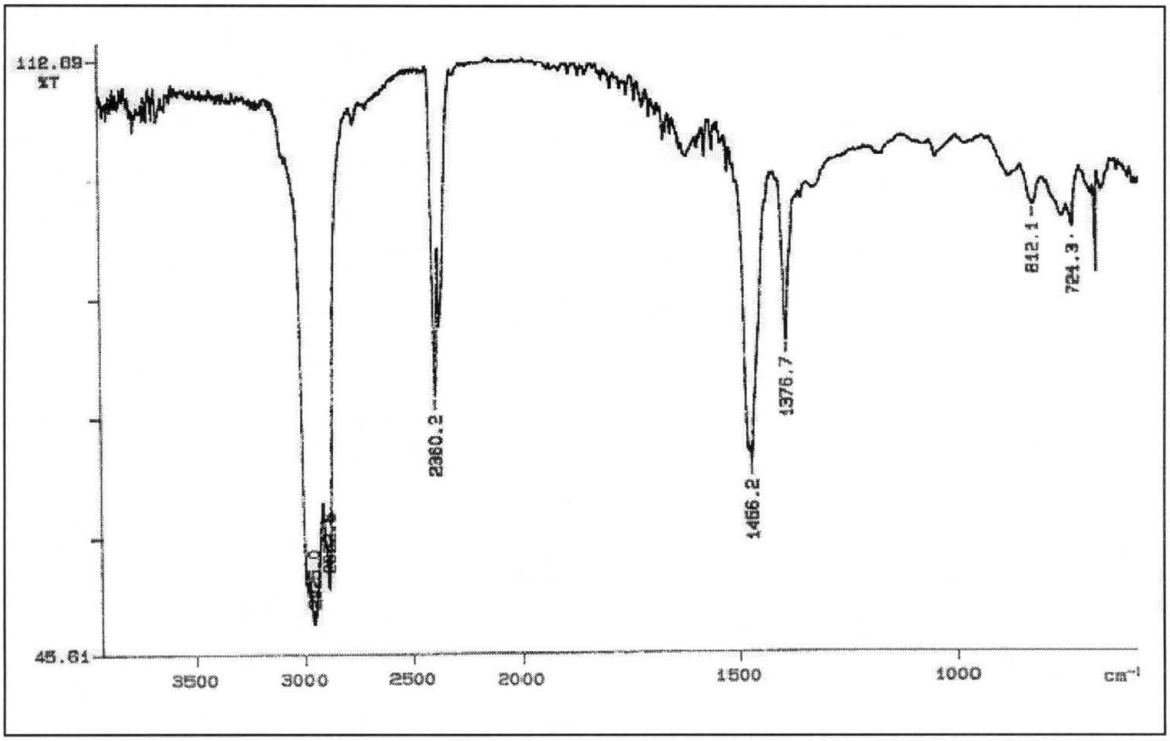

Figure A.1.1: FT-IR Spectrum of Maya Crude Oil

A.2: FT-IR of SARA Fractions from Maya Crude Oil

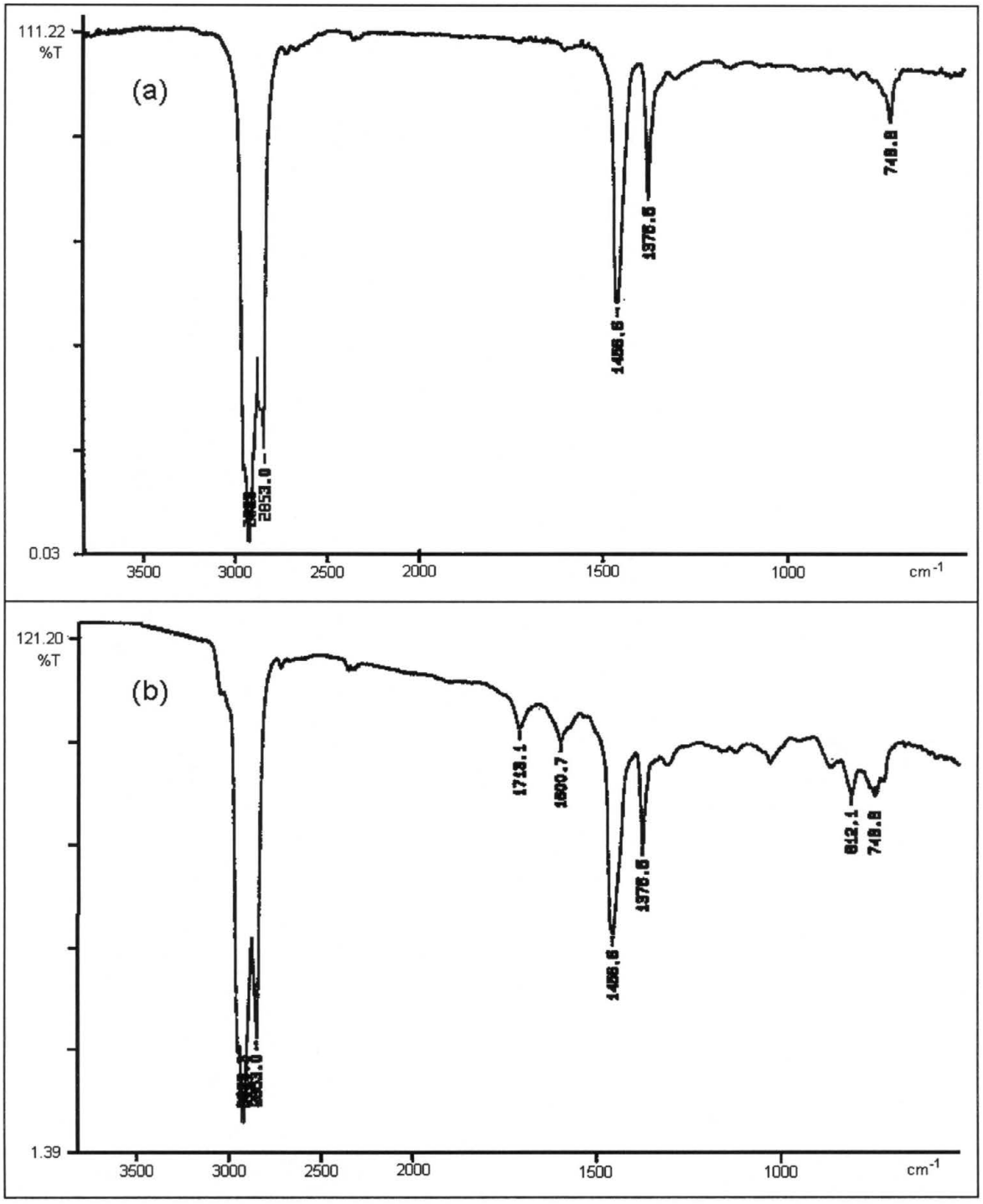

Figure A.2.1: FT-IR Spectra of (a) Saturate and (b) Aromatic Fractions from Maya Crude Oil

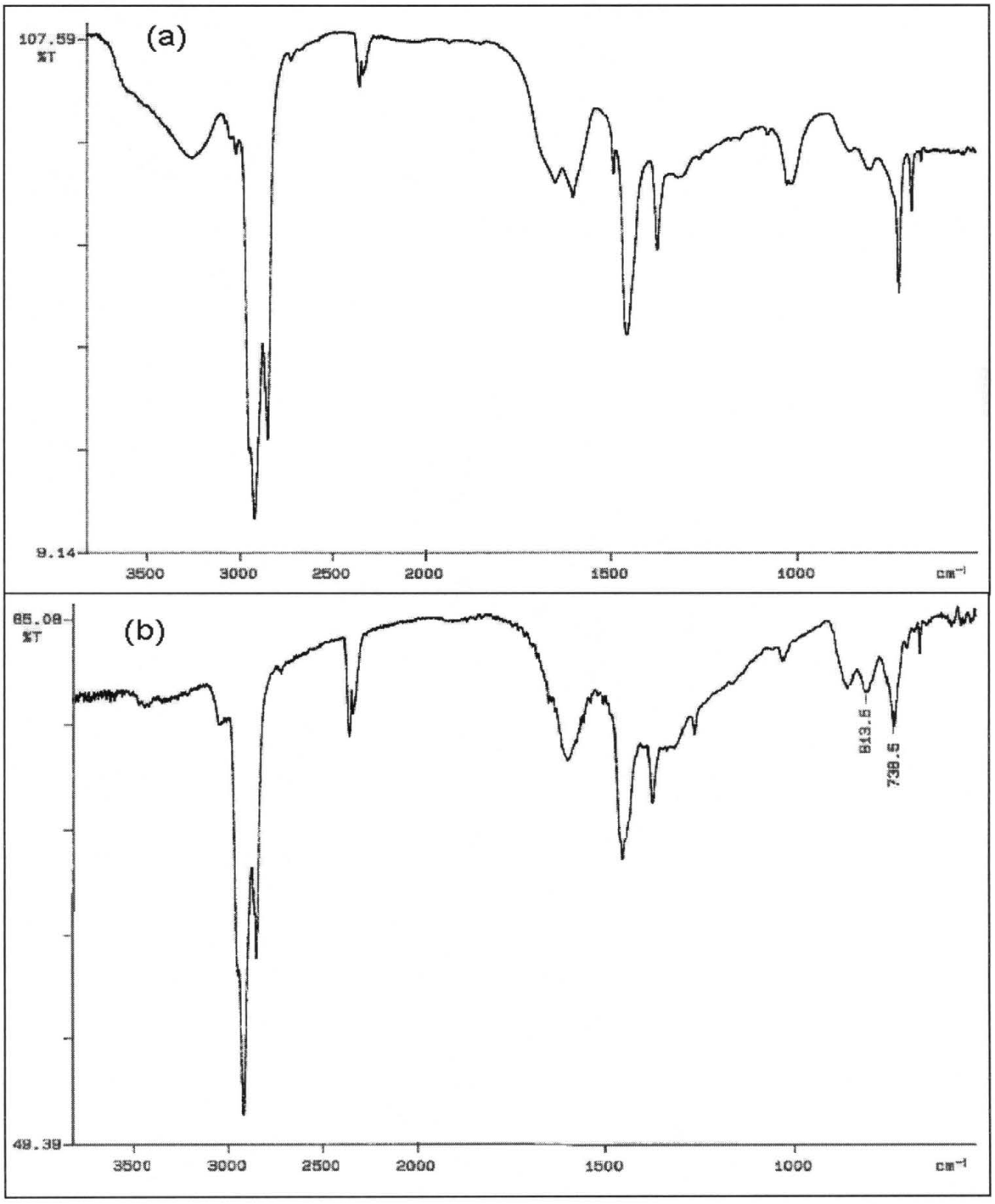

Figure A.2.2: FT-IR Spectra of (a) Resin and (b) Asphaltene Fractions from Maya Crude Oil

A.3: FT-IR of ABN Fractions from Maya Crude Oil Residue

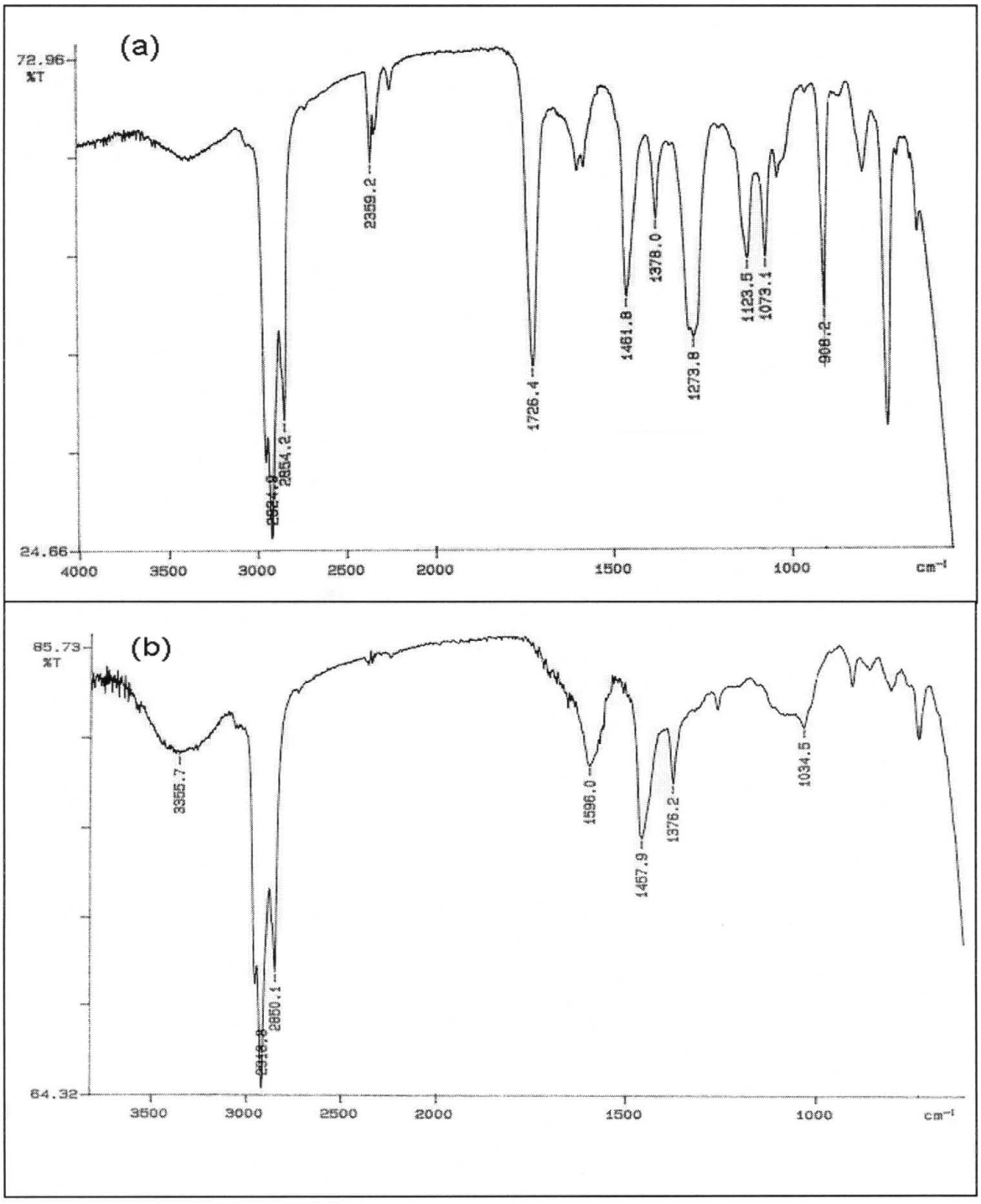

Figure A.3.1: FT-IR Spectra of (a) Acidic and (b) Basic Fractions from Maya Crude Oil Residue

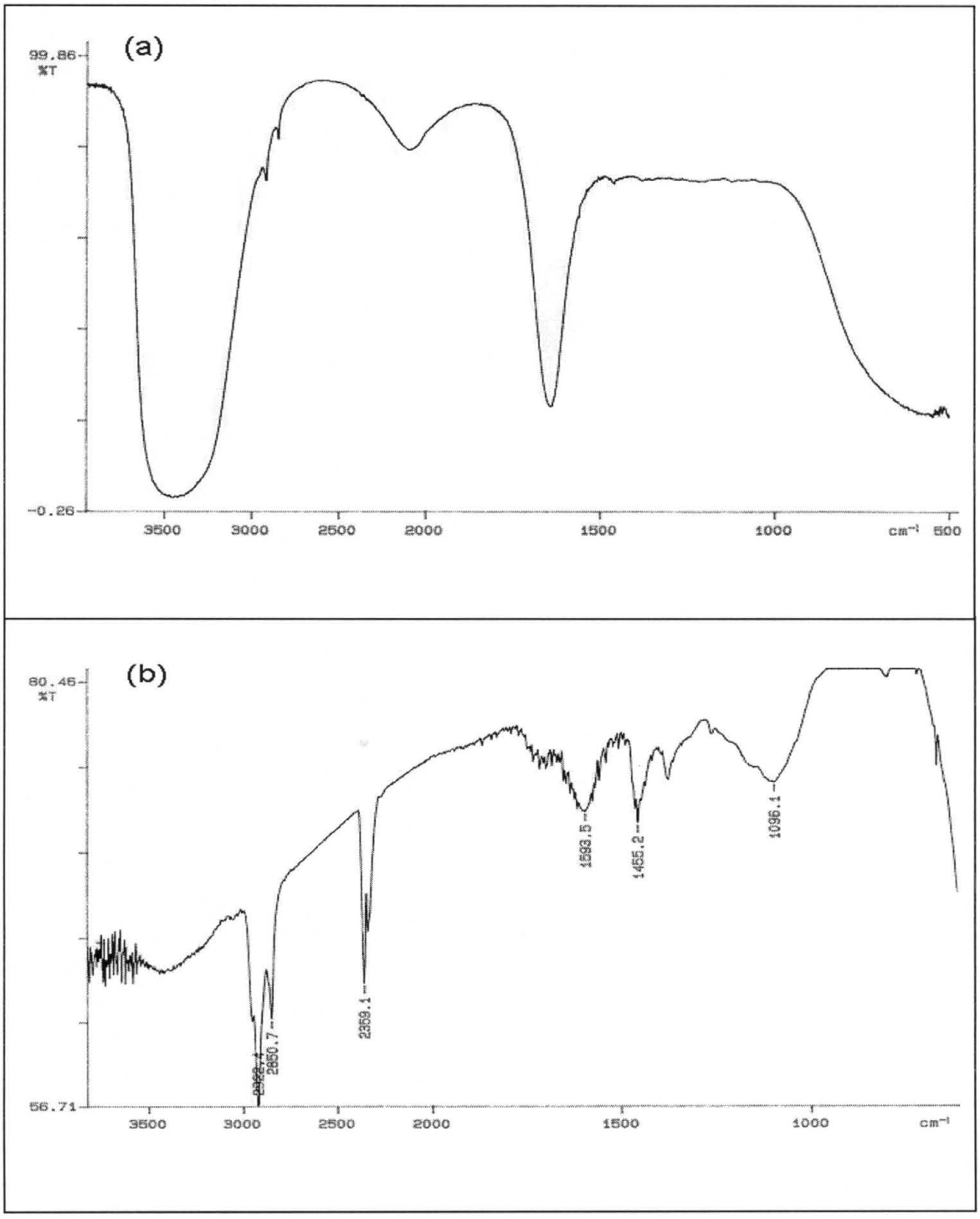

Figure A.3.2: FT-IR Spectra of (a) High Polarity #1 and (b) High Polarity #2 Fractions from Maya Crude Oil Residue

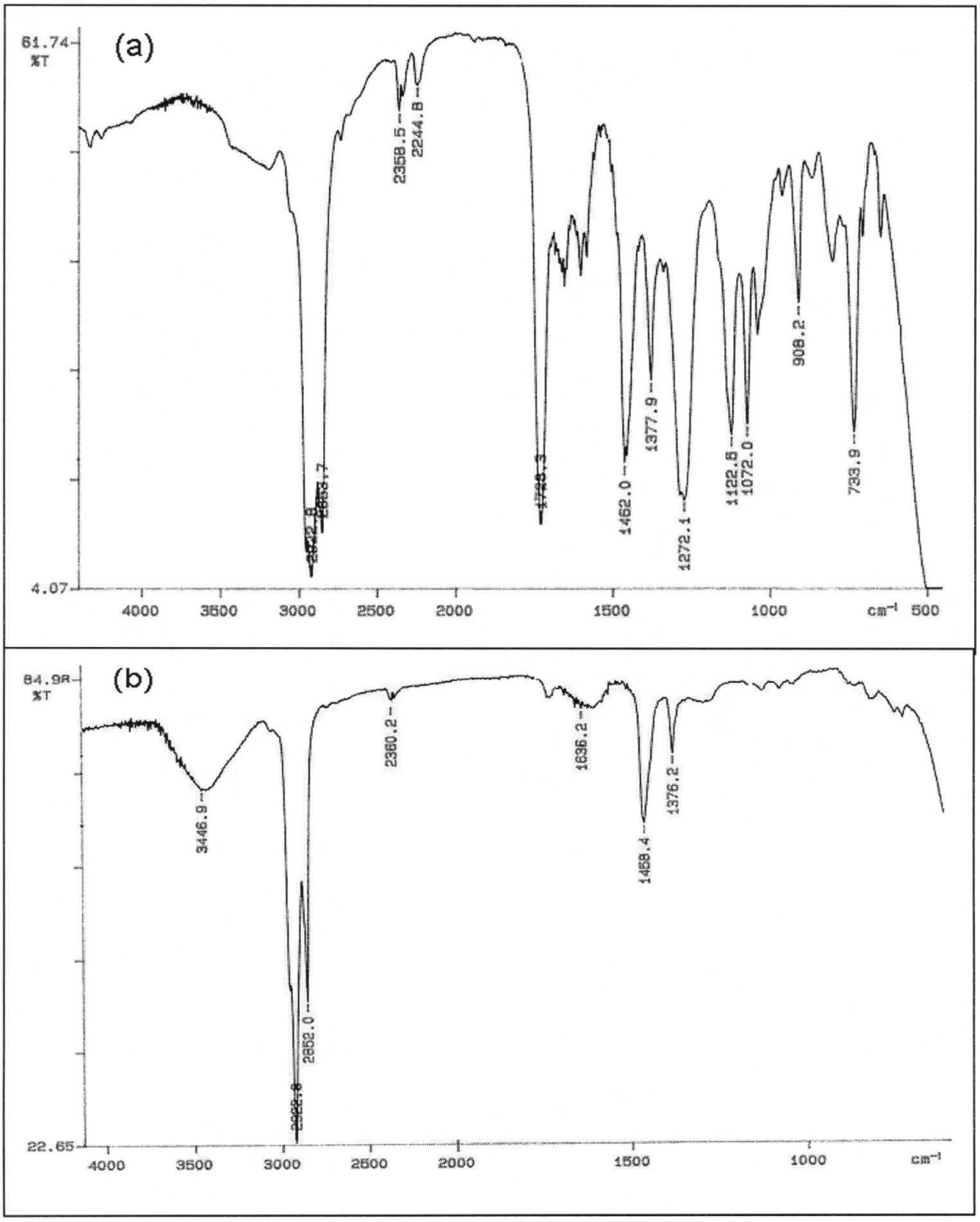

Figure A.3.3: FT-IR Spectra of (a) Medium Polarity and (b) Low Polarity Fractions from Maya Crude Oil Residue

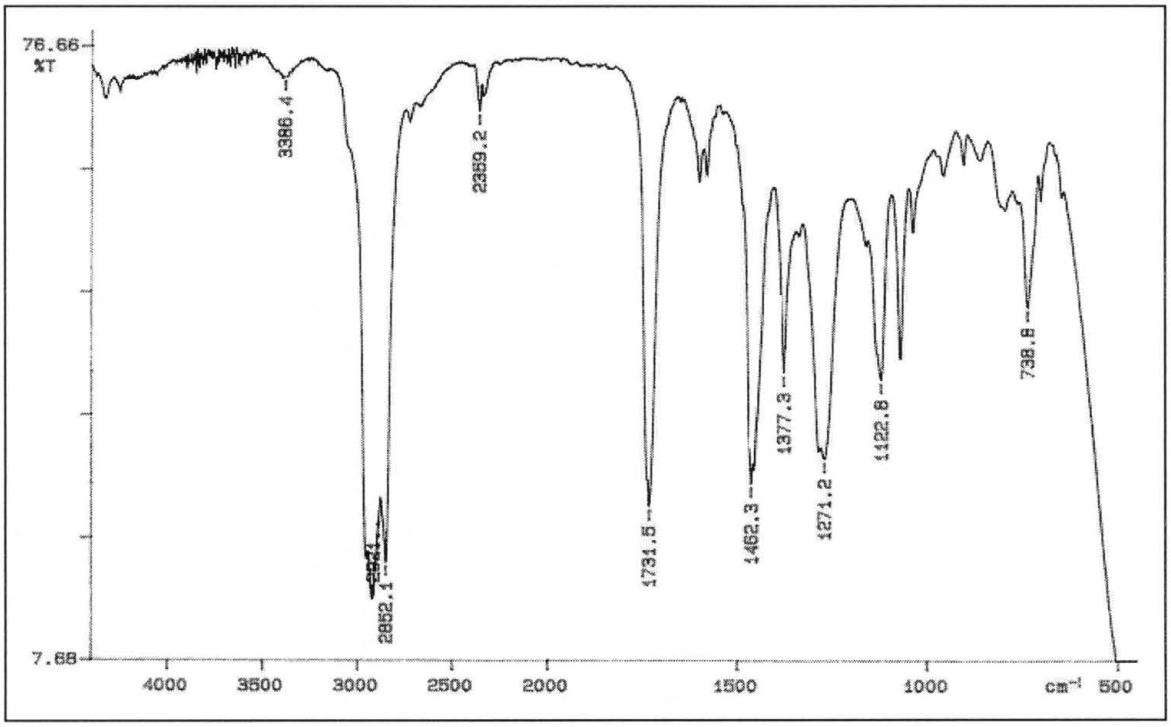

Figure A.3.4: FT-IR Spectrum of Saturate and Aromatic Fraction from Maya Crude Oil Residue

APPENDIX B

^{1}H NMR SPECTRA OF MAYA CRUDE OIL FRACTIONS

B.1: ^{1}H NMR of Maya Crude Oil

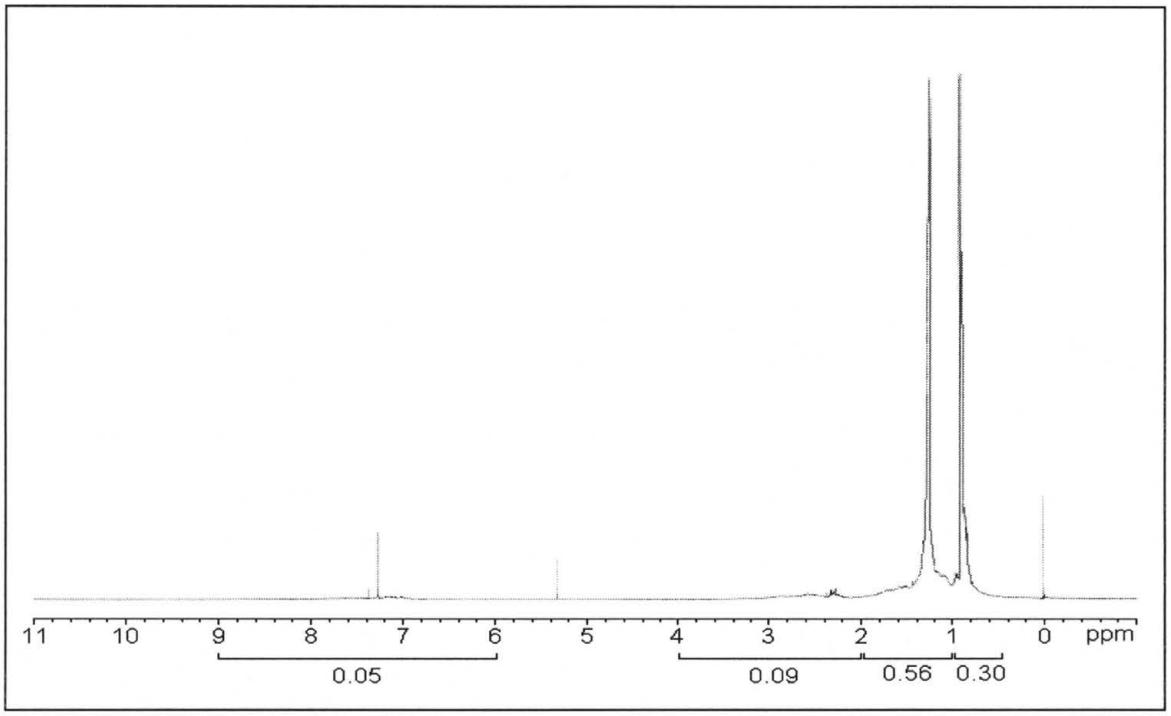

Figure B.1.1: ^{1}H NMR of Maya Crude Oil

B.2: ^{1}H NMR of SARA Fractions from Maya Crude Oil

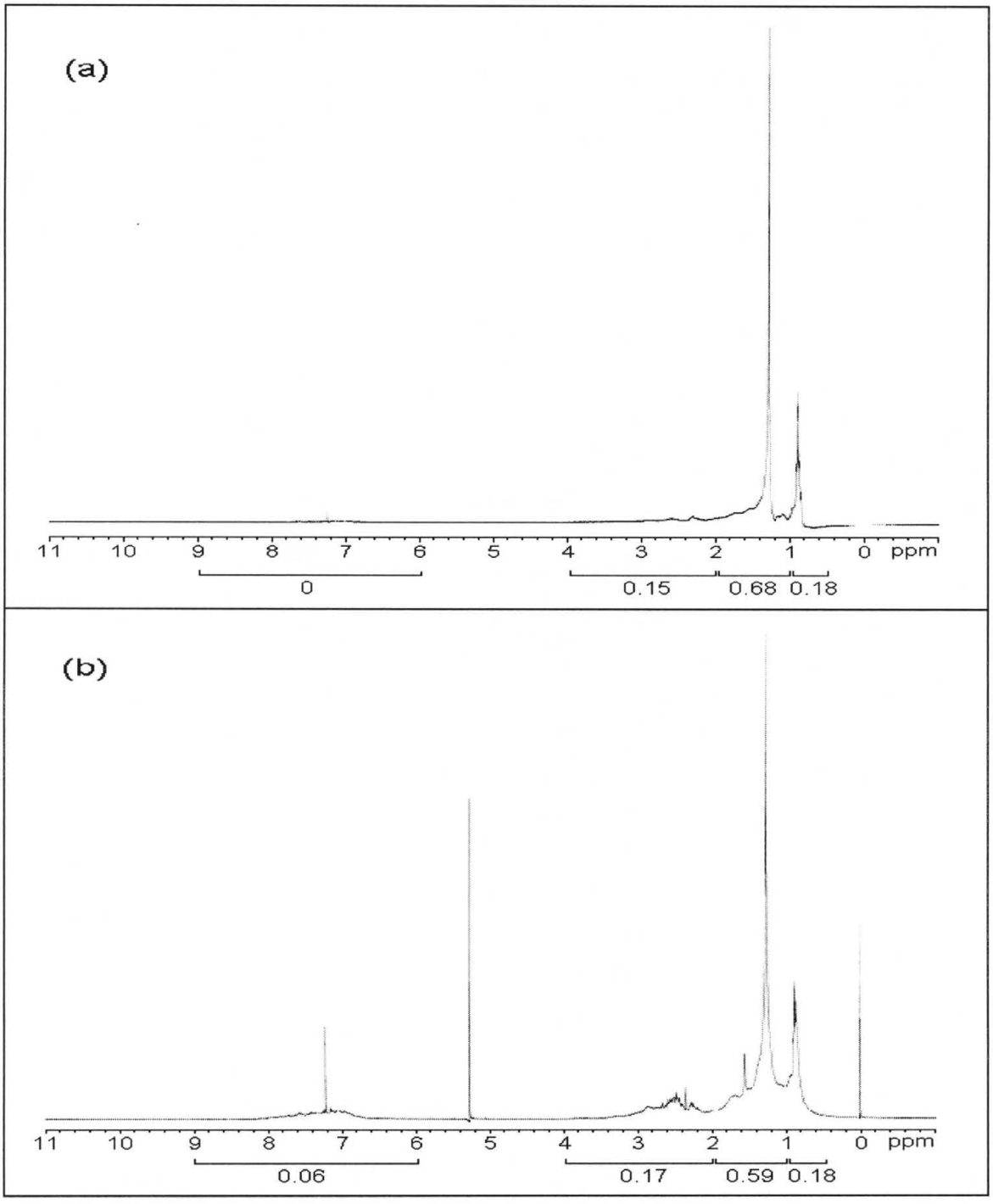

Figure B.2.1: ^{1}H NMR of (a) Saturate and (b) Aromatic Fraction from Maya Crude Oil

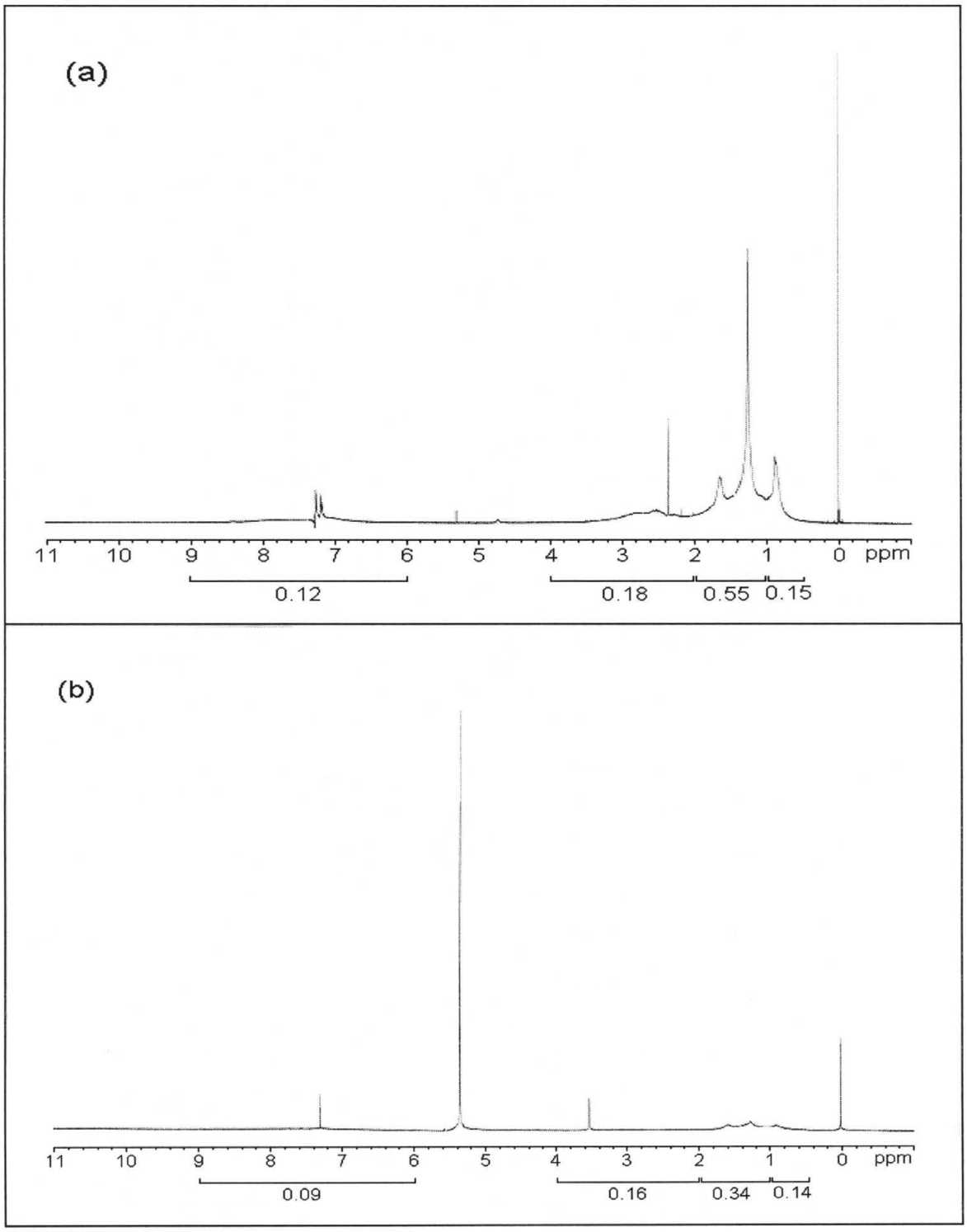

Figure B.2.2: ^{1}H NMR Spectra of (a) Resin and (b) Asphaltene Fraction from Maya Crude Oil

B.3: ^{1}H NMR of ABN Fractions from Maya Crude Oil Residue

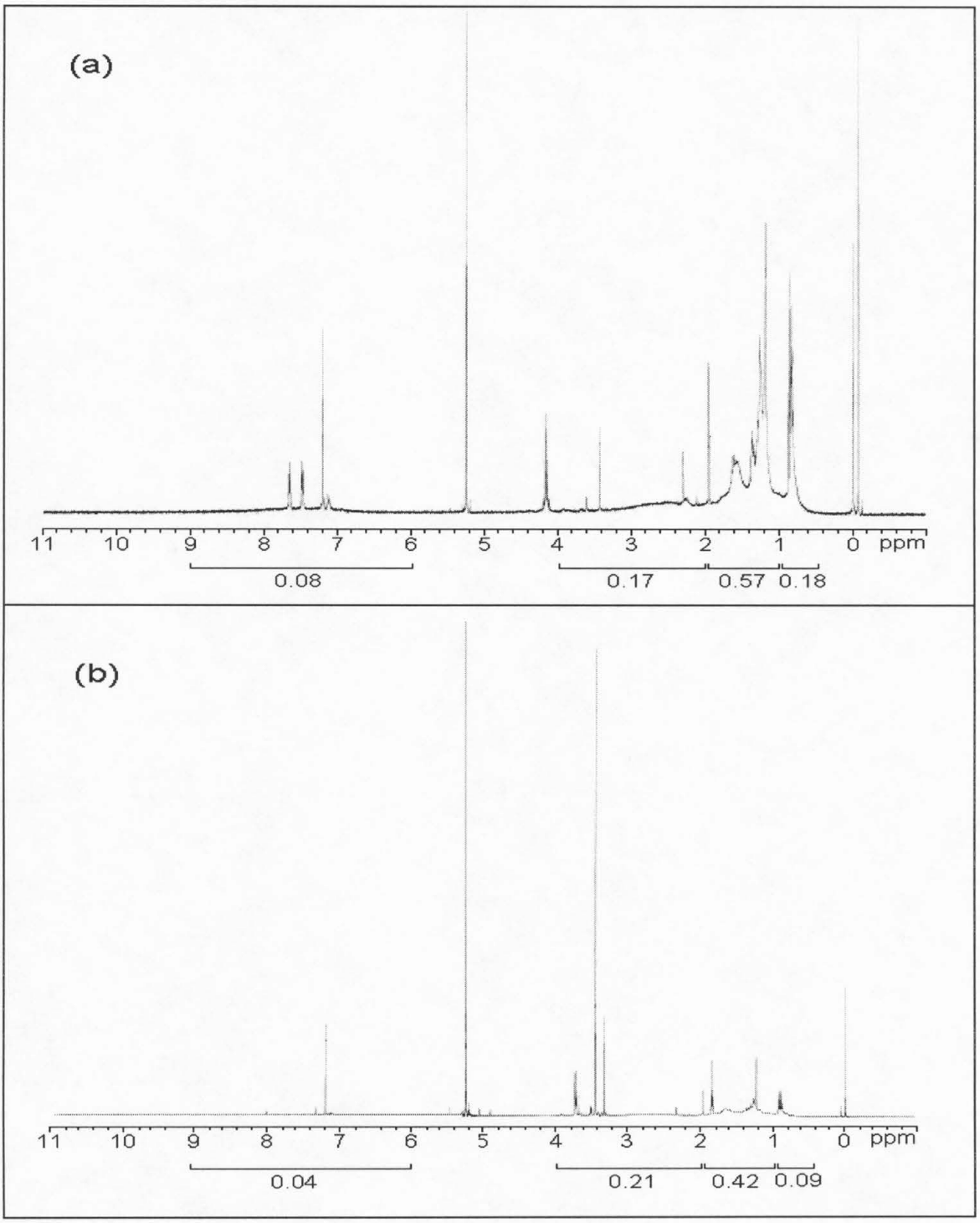

Figure B.3.1: ^{1}H NMR Spectra of (a) Acidic and (b) Basic Fractions of Maya Crude Oil Residue

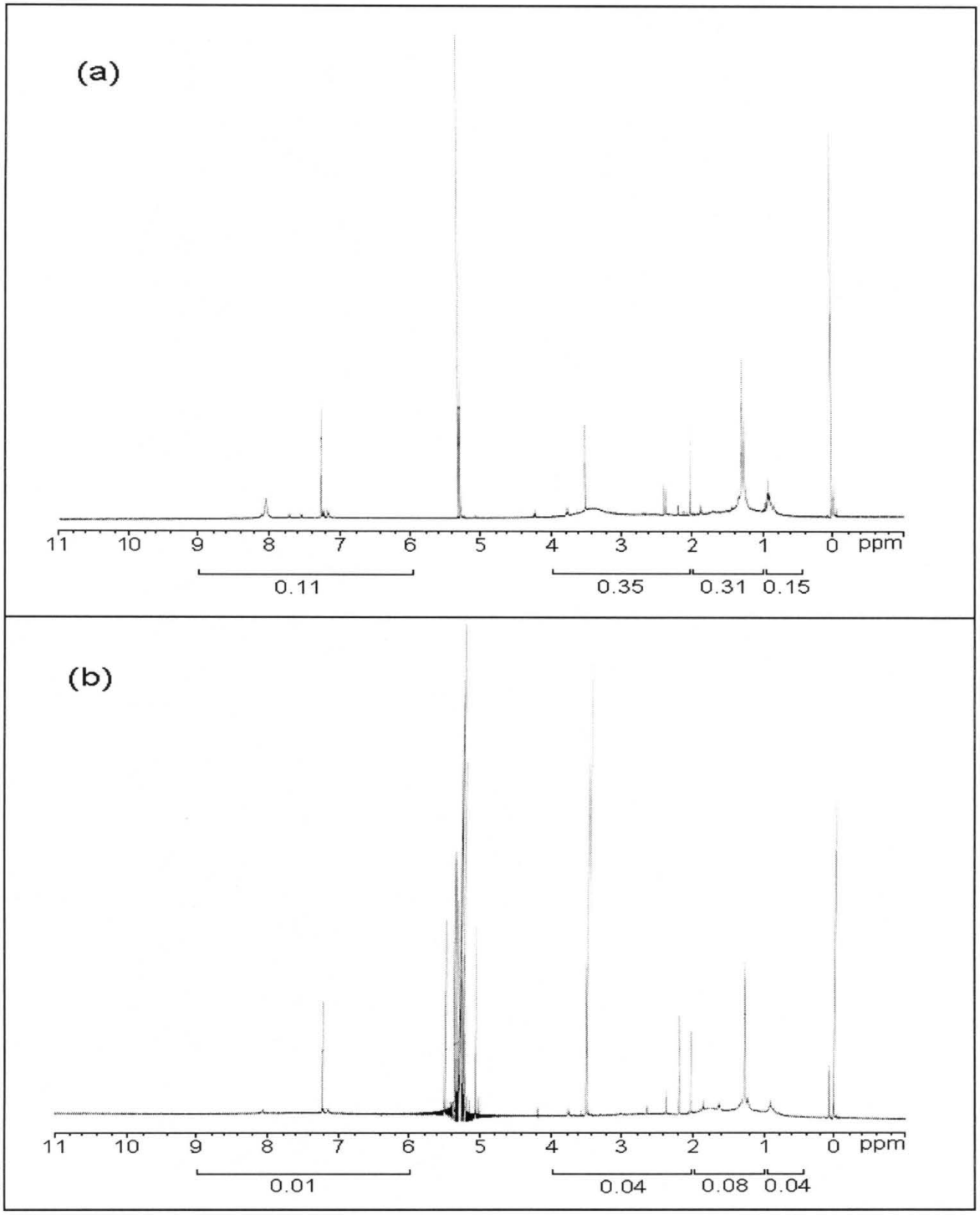

Figure B.3.2: ^{1}H NMR of (a) High Polarity #1 and (b) High Polarity #2 Fractions from Maya Crude Oil Residue

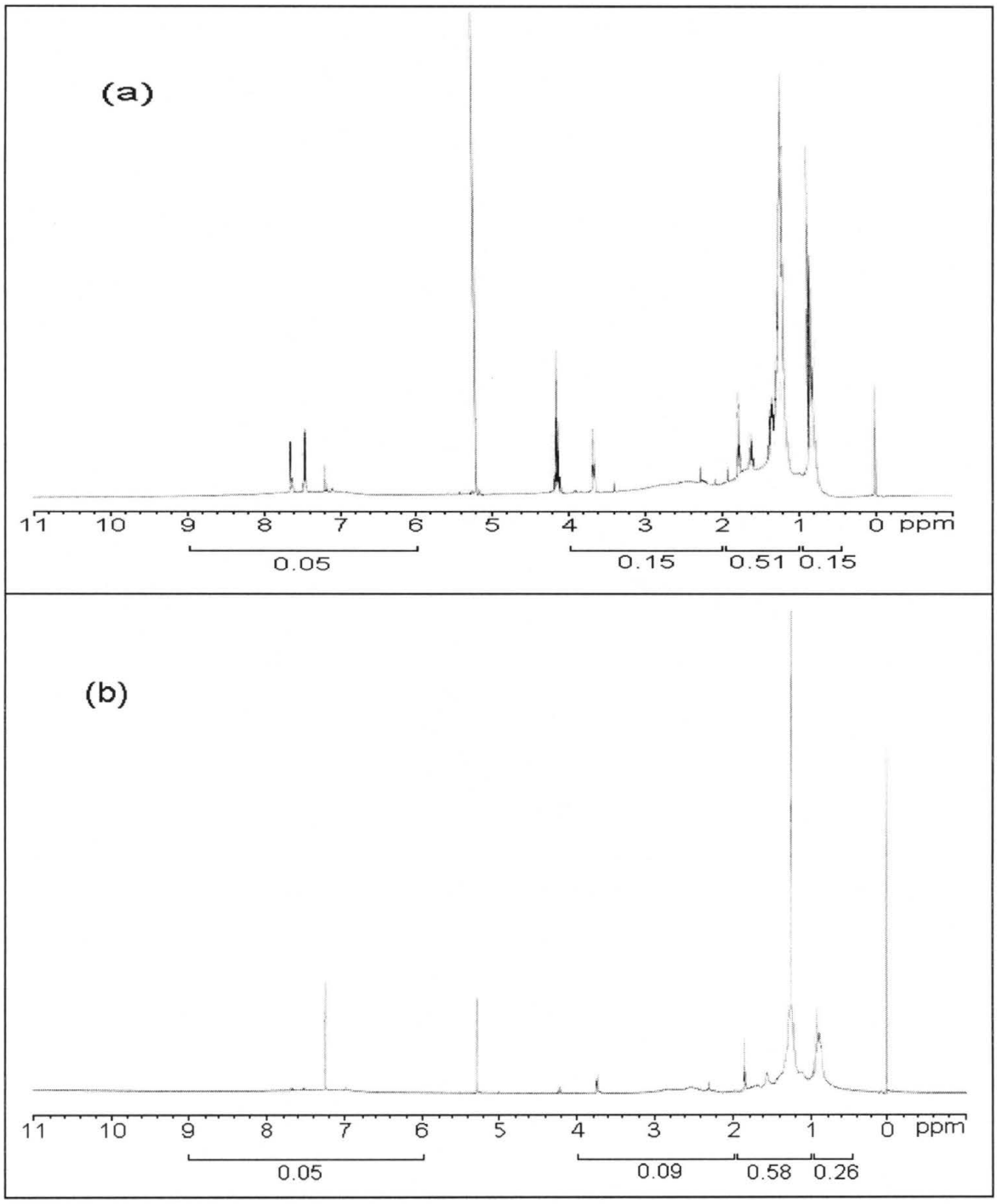

Figure B.3.3: ^{1}H NMR of (a) Medium Polarity and (b) Low Polarity Fractions from Maya Crude Oil Residue

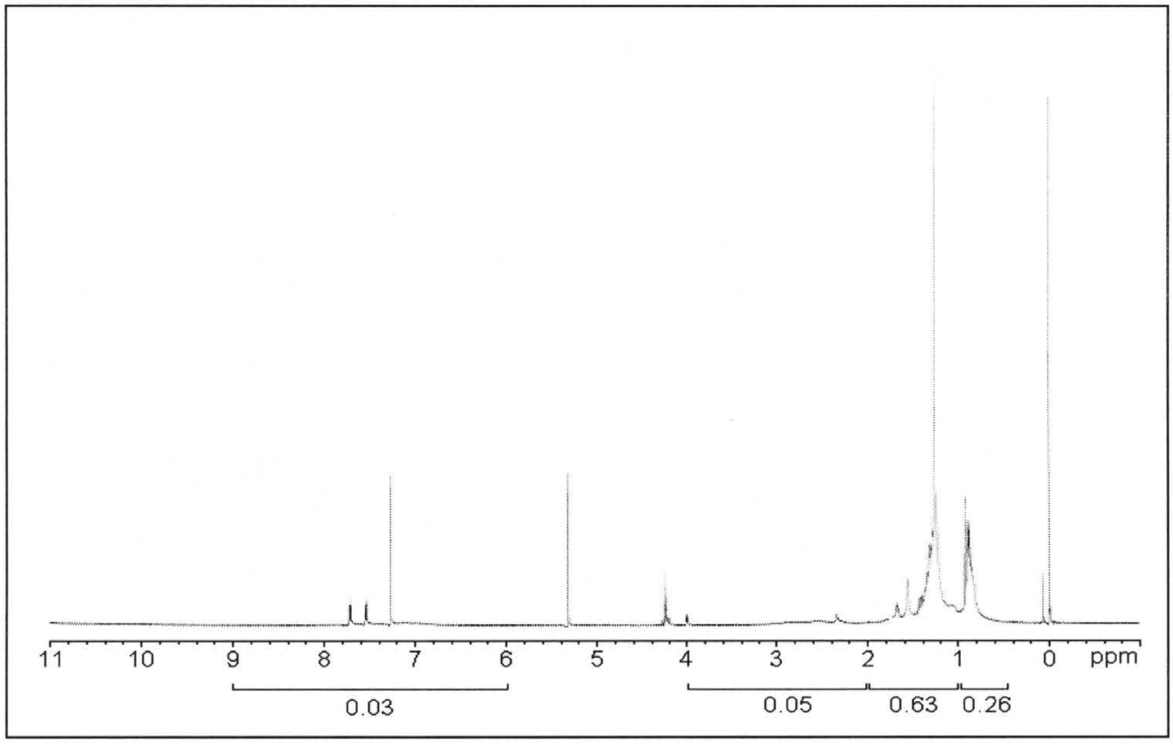

Figure B.3.4: ^{1}H NMR of Saturate and Aromatic Fraction from Maya Crude Oil Residue

B.4: ^{1}H NMR of LEC Fractions from Maya Crude Oil

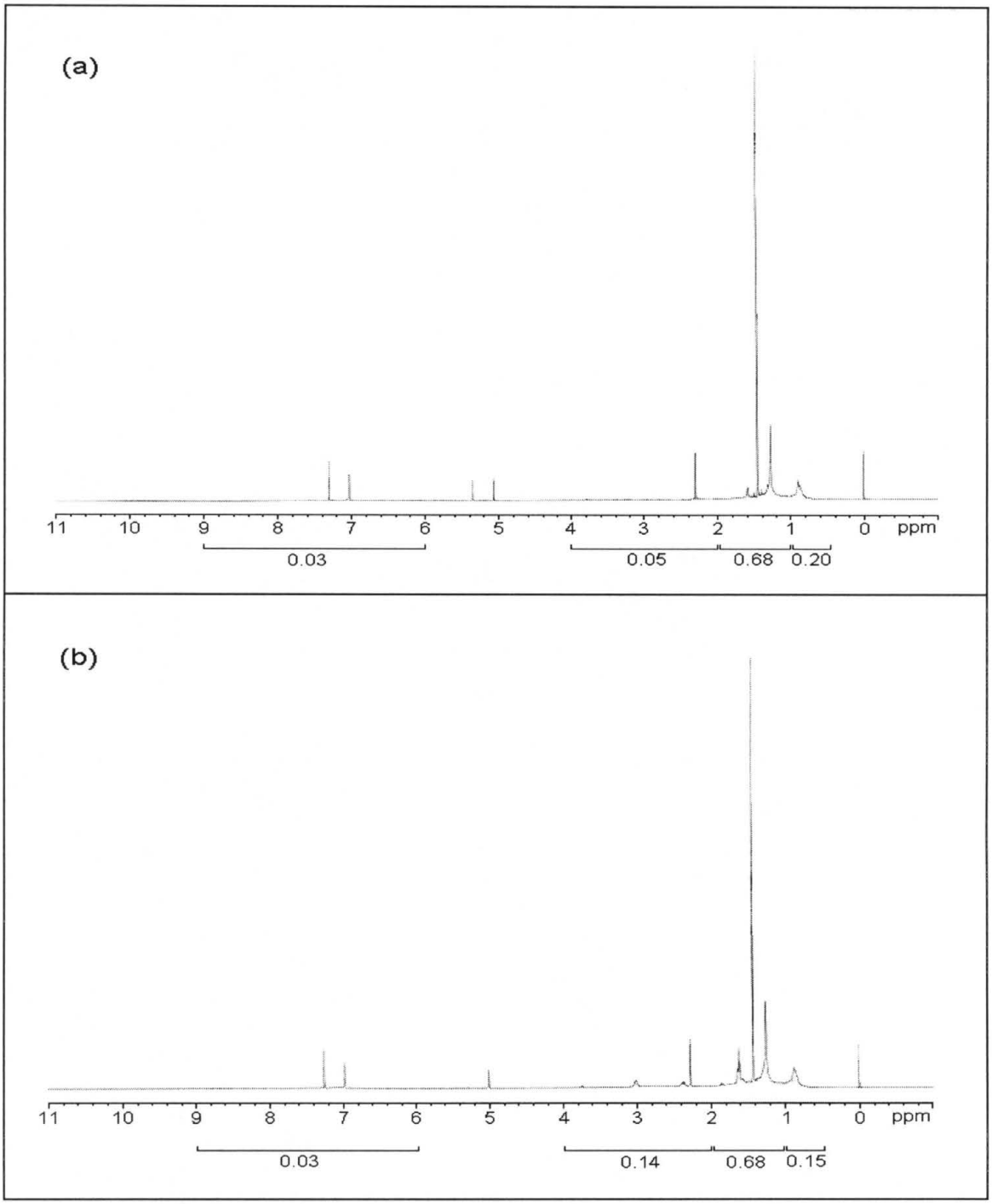

Figure B.4.1: ^{1}H NMR of (a) Aliphatic-S and (b) PASH Fractions from Maya Crude Oil

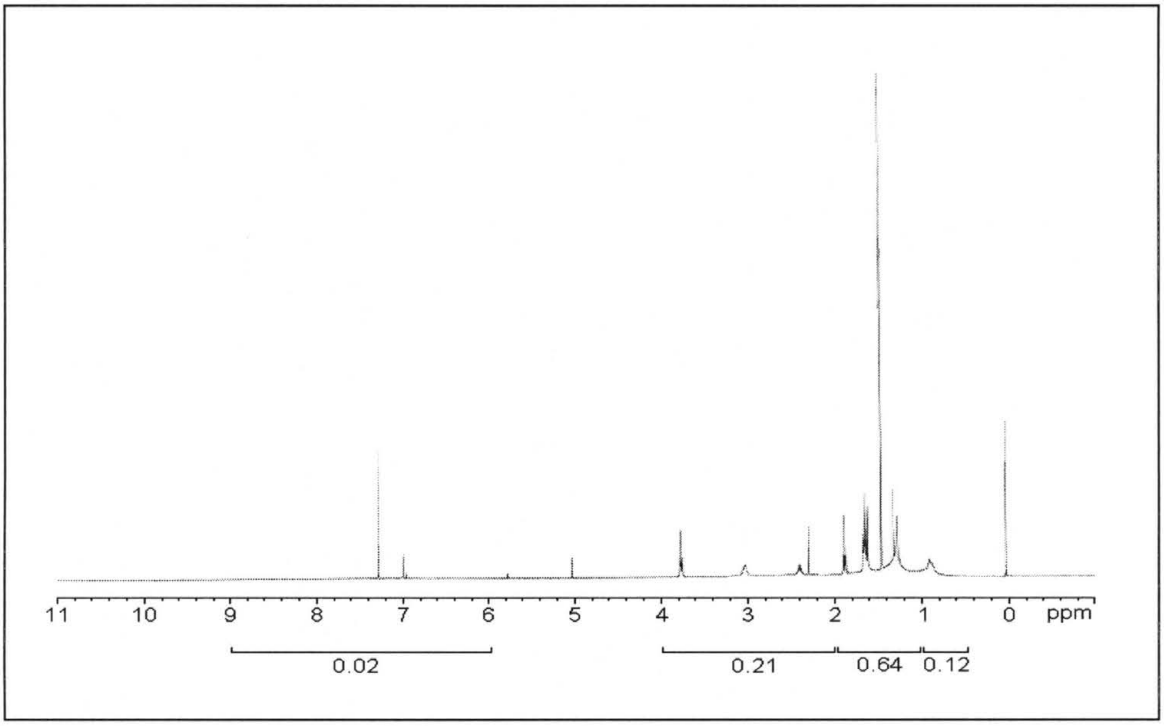

Figure B.4.2: ^{1}H NMR of SPAC fraction From Maya Crude Oil