

**PETROLEOMICS OF COLOMBIAN ASPHALTENES:
HIGH RESOLUTION MASS SPECTROMETRY VIEW OF ASPHALTENE
ADSORPTION, REACTIVITY, OCCLUSION AND AGGREGATION**

MARTHA LILIANA CHACÓN PATIÑO

**UNIVERSIDAD INDUSTRIAL DE SANTANDER
FACULTAD DE CIENCIAS
ESCUELA DE QUÍMICA
DOCTORADO EN QUÍMICA
BUCARAMANGA**

2016

**PETROLEOMICS OF COLOMBIAN ASPHALTENES:
HIGH RESOLUTION MASS SPECTROMETRY VIEW OF ASPHALTENE
ADSORPTION, REACTIVITY, OCCLUSION AND AGGREGATION**

Presented by:

MARTHA LILIANA CHACÓN PATIÑO

**Doctoral Dissertation As Requirement To Get The Degree of
Doctor in Chemistry**

Advisors:

Dr. Marianny Yajaira Combariza Montañez

Universidad Industrial de Santander

Dr. Andrea Gómez Escudero

Instituto Colombiano del Petróleo

Dr. Cristian Blanco Tirado

Universidad Industrial de Santander

UNIVERSIDAD INDUSTRIAL DE SANTANDER

FACULTAD DE CIENCIAS

ESCUELA DE QUÍMICA

DOCTORADO EN QUÍMICA

BUCARAMANGA

2016

ACKNOWLEDGMENTS

My gratitude goes to six people without whose help, support, and love, the dream of my PhD would had not been possible:

Doctor Marianny Yajaira Combariza and Cristian Blanco Tirado, for showing me the steps of the scientific method

Andrea Gomez Escudero, for giving me the opportunity of working at Ecopetrol

Martha Elena Patiño, my faithful mother

Jaime Alberto Huerfano, my godfather

Ivan Aguilera, from the beginning of my graduate school, because of his patience, love and support

CONTENTS

	Pag.
INTRODUCTION	17
1. FOURIER TRANSFORM ION CYCLOTRON RESONANCE MASS SPECTROMETRY AND PETROLEOMICS	21
1.1 FOURIER TRANSFORM ION CYCLOTRON MASS SPECTROMETRY	22
1.2 BUILDING THE ION CYCLOTRON CELL	23
1.3 THE NEED FOR HIGH RESOLVING POWER	27
1.4 MOLECULAR ASSIGNMENT AND DATA ANALYSIS	28
1.5 DATA ANALYSIS IN PETROLEOMICS	31
1.6 SAMPLE PREPARATION COUPLED TO FT-ICR MASS SPECTROMETRY ANALYSIS	36
1.7 COMBINING SAMPLE PREPARATION AND IONIZATION SOURCES FOR CHEMICAL SPECIATION	38
1.7.1 Electrospray ionization (ESI)	38
1.7.2 Atmospheric Pressure Photo-ionization (APPI)	40
1.8 IMPROVING COMPOSITIONAL SPACE ACCESSIBILITY AND CHEMICAL SPECIATION THROUGH SAMPLE FRACTIONATION	42
1.9 IMPROVEMENT OF ASPHALTENE MOLECULAR VIEW THROUGH FT-ICR MASS SPECTROMETRY	44
2. HIGH RESOLUTION MASS SPECTROMETRIC VIEW OF ASPHALTENE-SiO ₂ INTERACTIONS	48
2.1 ABSTRACT	48
2.2 INTRODUCTION	49
2.3 EXPERIMENTAL	52
2.3.1 Materials.	52
2.3.2 Asphaltene sample.	52

2.3.3 High-Performance Thin Layer Chromatography (HPTLC).	53
2.3.4 Extraction of asphaltene fractions from silica.	53
2.3.5 APPI FT-ICR mass spectrometry.	54
2.4 RESULTS AND DISCUSSION.....	55
2.5 CONCLUSIONS.....	72
 3. TRACING THE COMPOSITIONAL CHANGES OF ASPHALTENES AFTER HYDROCONVERSION AND THERMAL CRACKING PROCESSES BY HIGH- RESOLUTION MASS SPECTROMETRY”	74
3.1 ABSTRACT.....	74
3.2 INTRODUCTION	75
3.3 EXPERIMENTAL	79
3.3.1 Sample preparation.....	79
3.3.2 Elemental Analysis.....	80
3.3.3 Sample Preparation.	80
3.3.4 APPI (+) FT-ICR Mass Spectrometry.....	81
3.4 RESULTS AND DISCUSSION.....	82
3.5 CONCLUSIONS.....	107
 4. EXPLORING ASPHALTENE OCCLUSIONS THROUGH HIGH RESOLUTION MASS SPECTROMETRY.....	109
4.1 ABSTRACT.....	109
4.2 INTRODUCTION	110
4.3 EXPERIMENTAL SECTION	115
4.3.1 Materials.	115
4.3.2 Sample preparation.....	115
4.3.3 Column chromatography fractionation.	116
4.3.4 UV-Vis spectroscopy.....	116
4.3.5 APPI FT-ICR mass spectrometry.	116
4.4 RESULTS AND DISCUSSION.....	118

4.5 CONCLUSIONS.....	138
5. THE ROLE OF MOLECULAR COMPOSITION IN ASPHALTENE AGGREGATION	140
5.1 ABSTRACT	140
5.2 INTRODUCTION	141
5.3 EXPERIMENTAL SECTION	145
5.3.1 Materials.	145
5.3.2 Asphaltene samples.....	145
5.3.3 Asphaltene purification.....	146
5.3.4 Asphaltene fractionation by High-Performance Thin Layer Chromatography and asphaltene recovery.....	146
5.3.5 Aggregation tests.	147
5.4 RESULTS AND DISCUSSION.....	147
5.5 CONCLUSIONS.....	168
BIBLIOGRAPHY	169

LIST OF FIGURES

	Pag.
Figure 1.1. a) Ion motion in a homogeneous magnetic field and b) ion movement parallel to the magnetic field.	23
Figure 1.2. a) Trapping motion as a result of applying voltages trough the trapping plates; b) magnetron motion.	25
Figure 1.3. a) Ion motion and the b) ion cyclotron resonance cell.....	26
Figure 1.4. The sequence of events in an FT-ICR MS experiment.	27
Figure 1.5. Comparison between mass spectra of Colombian heavy crude oil: Blue corresponds to FT-ICR MS, and Red corresponds to Time of flight MS.	28
Figure 1.6. (+) APPI FT-ICR mass spectrum for a heavy Colombian crude oil. ...	29
Figure 1.7. Kendrick mass diagram for the class S1 in a Colombian heavy oil. ...	30
Figure 1.8. DBE versus carbon number plots for the class N1 in a Colombian heavy oil and its asphaltenes.	33
Figure 1.9. Planar limits as a result of molecular structure.	34
Figure 1.10. Van Krevelen diagrams for N ₁ compositions in a Colombian heavy oil and its asphaltenes.	35
Figure 1.11. Van Krevelen diagrams for the classes O ₁ , O ₂ , and O ₃ in a Colombian heavy oil.....	35
Figure 1.12. DBE versus carbon number plots for the class S ₁ of the ring HPLC-2 fractions from a Colombian heavy vacuum gas oil (427–482 °C).	37
Figure 1.13. Scheme of the electrospray ionization source (Fenn et al. 1989).	40
Figure 1.14. Scheme of the atmospheric pressure photoionization source.....	41
Figure 1.15. Ionization routes of N-compounds in positive and negative ESI and APPI.....	43
Figure 1.16. DBE versus carbon number plots for class S ₁ in a whole Colombian VGO, and its non-reactive and reactive sulfur fractions.	44

Figure 1.17.	Left: Van Krevelen diagrams for O ₁ species in a Colombian heavy oil and its asphaltenes; right: asphaltene core structures from CID experiments.	46
Figure 2.1.	Graphical abstract for High Resolution Mass Spectrometric View of Asphaltene–SiO ₂ Interactions	48
Figure 2.2.	(a) HPTLC SiO ₂ plate showing three characteristic bands after asphaltene fractionation with n-C ₆ , Tol, and CH ₂ Cl ₂ /MeOH; (b) whole and fractioned asphaltenes desorbed from the SiO ₂ ; (c) compound class distribution; (d) molecular weight distributions for W, WD, NE, DM, and T asphaltene fractions.	57
Figure 2.3.	Asphaltene recovery profiles from silica.....	60
Figure 2.4.	Van Krevelen diagrams for N ₁ , N ₂ , and N ₃ classes in W and WD samples.	62
Figure 2.5.	Van Krevelen diagrams for classes S ₁ , S ₂ , and HC in W and WD. ...	64
Figure 2.6.	Van Krevelen diagrams for N _n O ₀ S _s and O ₀ S _s classes in the fraction T.	65
Figure 2.7.	Van Krevelen diagrams for O ₀ and N _n O ₀ classes in the fraction NE. ...	68
Figure 2.8.	Van Krevelen diagrams for N ₃ O ₁ , N ₃ O ₂ , O ₁ S ₁ , and S ₁ in the DM fraction.....	69
Figure 2.9.	Van Krevelen diagrams for common classes to W, WD, NE, DE, and T samples.	70
Figure 2.10.	Model compounds for a) non-eluted, b) DCM/MeOH eluted, and c) Toluene eluted asphaltenes.....	72
Figure 3.1.	Graphical abstract for “Tracing the Compositional Changes of Asphaltenes after Hydroconversion and Thermal Cracking Processes by “High-Resolution Mass Spectrometry”	74
Figure 3.2.	Sample evolution and mass balances for a Colombian asphaltene sample exposed to hydroconversion and thermal cracking upgrading process.	83

Figure 3.3.	Compound class distributions and MWDs for raw asphaltenes and residual asphaltenes (Asp1, Asp2) and maltenes (M1, M2) after upgrading.....	86
Figure 3.4.	DBE versus carbon number plots for HC, S ₁ , O ₁ , N ₁ , and N ₂ compound classes in raw asphaltenes and residual asphaltenes (Asp1 and Asp2) and maltenes (M1 and M2) after upgrading.	90
Figure 3.5.	DBE distribution for the radical N ₁ class in maltenes (M1 and M2) after hydroconversion and thermal cracking processes.	91
Figure 3.6.	DBE distribution for the S ₁ class in maltenes (M1 and M2) after hydroconversion and thermal cracking processes.	94
Figure 3.7.	a). Planar aromatic limits in DBE versus carbon number plots; b). Planar limit slopes as result of molecular structure.	97
Figure 3.8.	H/C versus carbon number plots for the HC class in raw asphaltenes, and residual asphaltenes (Asp1 and Asp2) and maltenes (M1 and M2) after upgrading.....	100
Figure 3.9.	Van Krevelen diagrams for classes HC, S ₁ , O ₁ , N ₁ , N ₂ in raw asphaltenes, and residual asphaltenes (Asp1 and Asp2) and maltenes (M1 and M2) after upgrading.	103
Figure 3.10.	Hypothetical reaction pathways of model raw asphaltenes under hydroconversion or thermal cracking conditions: an archipelago-type structure producing maltenic products.	105
Figure 3.11.	Hypothetical reaction pathways of model raw asphaltenes under hydroconversion or thermal cracking conditions for an island-type structure with dibenzothiophene substituent, producing molecular compositions present in residual asphaltenes and produced maltenes.	106
Figure 4.1.	Graphical abstract for Exploring asphaltene occlusions through High-Resolution Mass Spectrometry.	109
Figure 4.2.	Extraction protocol for occluded compounds in <i>n</i> -C ₇ asphaltenes including weight percentages of the extracted fractions.	117

Figure 4.3.	Compound class distributions and molecular weight distributions for fractions E ₅ , E ₁₀ , E ₁₅ and E ₂₀ , measured by (+) APPI FT-ICR mass spectrometry.	120
Figure 4.4.	Isoabundance-contoured plots of DBE <i>versus</i> carbon numbers for classes HC, S ₁ , O ₁ and N ₁ in fractions E ₅ , E ₁₀ , E ₁₅ and E ₂₀	122
Figure 4.5.	From top to bottom, Van Krevelen diagrams for S-, N-, and O-compounds in fractions E ₅ , E ₁₀ , E ₁₅ , and E ₂₀	126
Figure 4.6.	Column chromatography of sample E ₂₀ including weight percentages of the eluted subfractions.	129
Figure 4.7.	UV-Vis spectra of subfractions F ₁₋₆	130
Figure 4.8.	Compound class distributions and molecular weight distributions for samples F ₁₋₆	132
Figure 4.9.	Contoured plots of DBE <i>versus</i> carbon number for subfractions F ₁₋₆ , resulted from the column chromatography separation carried out on the fraction E ₂₀	133
Figure 5.1	Graphical abstract for <i>The Role of Molecular Composition in Asphaltene Aggregation</i>	140
Figure 5.2.	High-performance thin-layer chromatography fractionation of asphaltenes from three different Colombian crude oils, with their respective retention factors (R _f) and the weight percentages.	149
Figure 5.3.	a) Turbiscan principle and backscattering profiles for b) a stable crude oil solution, c) O/W emulsion going under coalescence, d) W/O emulsion from extra heavy crude oil upon creaming, e) W/O emulsion from medium crude oil going through precipitation, and f) asphaltene flocculation in HepTol 70:30 v/v.	151
Figure 5.4.	Backscattering profiles for whole #1, #2, and #3 asphaltenes, WD samples, and the respective HPTLC subfractions.	154
Figure 5.5.	Molecular composition in terms of van Krevelen diagrams of the HPTLC fractions of the three asphaltene samples.	156

Figure 5.6.	Aggregation profiles for whole asphaltenes, WD samples and the HPTLC subfractions in terms of the Turbiscan Stability Index.	159
Figure 5.7.	Examples of the DLA and RLA mechanisms for asphaltene aggregation. We include the fitting curves for the TSI curves for the whole and WD samples from asphaltenes #1.	161
Figure 5.8.	Effect of SARA aromatics and resins on the stability of whole asphaltene dispersion.	164
Figure 5.9.	Molecular composition of aromatics, resins and asphaltenes from crude oils #1, #2, and #3.	165
Figure 5.10.	Compound class distribution in terms of heteroatom groups for aromatics, resins and asphaltenes from crude oils #1, #2, and #3.	166
Figure 5.11.	Wiehe solubility model for petroleum.	168

LIST OF TABLES

	Pag.
Table 3.1. APPI source conditions for raw asphaltenes, residual asphaltenes and produced maltenes.	81
Table 3.2. Elemental Composition of Raw Asphaltenes and Their Hydroconversion and Thermal Cracking Products*	84
Table 3.3. Planar aromatic limit slopes for raw asphaltenes, residual asphaltenes and produced maltenes.	98
Table 4.1. C#/DBE ratios for classes HC, S ₁ , O ₁ , and N ₁ in fractions E ₅ –E ₂₀	123
Table 4.2. Planar limit slopes for compound classes HC, S ₁ , O ₁ , and N ₁ in fractions E ₅ , E ₁₀ , E ₁₅ , and E ₂₀	125

ABSTRACT

TITLE: Petroleomics of Colombian Asphaltenes: High Resolution Mass Spectrometry View of Asphaltene Adsorption, Reactivity, Occlusion and Aggregation*

AUTHORS: Martha Liliana Chacón Patiño**

KEYWORDS: Asphaltenes, mass spectrometry, aggregation

DESCRIPTION

Asphaltenes are considered the least valuable components of crude oil because they are responsible for serious problems during production, transporting, and upgrading. Changes in petroleum composition or physical conditions like pressure or temperature dropping, can produce asphaltene deposits, causing formation damage, pipeline plugging, and catalysts poisoning. In this sense, understanding the role of molecular composition in asphaltene behavior is fundamental to improve production and processing of petroleum.

Three Colombian asphaltene samples (ASTM D6560-12) were purified via four cycles of successive maceration–Soxhlet extraction with *n*-heptane to extract occluded hydrocarbons. The purified asphaltene sample was then fractionated according to the degree of adsorption on SiO₂ particles, by elution with *n*-hexane, toluene, and dichloromethane-methanol. Molecular characterization of the samples was performed on a FT-ICR mass spectrometer (15 T) using an APPI source, and the aggregation kinetics was followed using Turbiscan Lab.

Fractionation of purified asphaltenes yielded three individual fractions, with different adsorption behaviors on the SiO₂ surface. We observed strong correlations between asphaltene adsorption on SiO₂ and molecular features such as aromaticity, heteroatom/C ratios, and degree of alkylation. We have experimental evidence suggesting that a synergy between aromaticity, steric hindrance, and heteroatom content are crucial factors in asphaltene adsorption on mineral surfaces. We also found a direct relationship between adsorption degree on the SiO₂ surface and the fractions aggregation behavior in solution phase. These results suggest that π -stacking is not the only driving force in asphaltene aggregation; we found that polar interactions such as hydrogen bonding, and acid-base associations play important roles in aggregation.

* Trabajo de grado

** FACULTAD DE CIENCIAS, ESCUELA DE QUÍMICA, Director: Dr. Marianny Yajaira Combariza Montañez, Codirectores: Dr. Andrea Gómez Escudero, Dr. Cristian Blanco Tirado.

RESUMEN

TITULO: Petroleomics of Colombian Asphaltenes: High Resolution Mass Spectrometry View of Asphaltene Adsorption, Reactivity, Occlusion and Aggregation*

AUTORES: Martha Liliana Chacón Patiño**

PALABRAS CLAVE: Asfaltenos, Espectrometría de Masas, Agregación

DESCRIPCION

Los asfaltenos son considerados los componentes de menor valor agregado del petróleo, debido a que son los responsables directos de una serie de problemas operacionales a lo largo de toda la cadena de producción del crudo. Cambios mínimos en la composición química de la matriz petroquímica, así como cambios en condiciones de presión y temperatura, promueven la agregación de asfaltenos, lo cual resulta en la formación de depósitos, daño a la formación, obstrucción de sistemas de transporte y envenenamiento de catalizadores. En este sentido, la comprensión de la naturaleza molecular de los asfaltenos es fundamental para mejorar la producción y el procesamiento de petróleo pesado.

Tres muestras de asfaltenos (ASTM D6560-12) fueron purificados por medio de cuatro ciclos de maceración-extracción Soxhlet con heptano, con el objetivo de extraer los compuestos malténicos ocluidos. Las muestras purificadas fueron fraccionadas de acuerdo al grado de adsorción en partículas de sílica, por medio de una elución secuencial con hexano, tolueno y diclorometano-metanol. La caracterización molecular de las muestras de asfaltenos fue realizada en un espectrómetro de masas de resonancia ciclotrónica de iones por transformada de Fourier, acoplado con fotoionización a presión atmosférica. Los estudios de agregación se realizaron con la herramienta Turbiscan Lab.

El fraccionamiento de las muestras de asfaltenos produjo tres fracciones distintivas, caracterizadas por su grado de adsorción sobre las partículas de sílica. Se observaron fuertes correlaciones entre el nivel de adsorción en SiO_2 y características moleculares como aromaticidad, impedimento estérico, contenido de N y O. En este documento se presenta evidencia que sugiere que las mismas características moleculares, y por consiguiente interacciones intermoleculares, que gobiernan los procesos de adsorción de asfaltenos en minerales, juegan papeles importantes en procesos de agregación en solución. Estos resultados sugieren sinergia de interacciones polares e interacciones tipo π , en la formación de agregados de asfaltenos.

* Trabajo de grado

** FACULTAD DE CIENCIAS, ESCUELA DE QUÍMICA, Director: Dr. Marianny Yajaira Combariza Montañez, Codirectores: Dr. Andrea Gómez Escudero, Dr. Cristian Blanco Tirado.

INTRODUCTION

In a world with rapid population growth and insatiable hunger for energy, it is imperative to develop methodologies for the rational and efficient use of the resources we still have. Although in 2015, *G7—leaders* claimed the era of fossil fuels has a nearby ending, we still lack a definitive and consolidated solution that meets the global demand for energy. Moreover, the main concern is that light, sweet and easy-to-process crude oils are depleted, and now the world depends mostly on non-conventional fossil resources, presenting challenging issues in all steps of the petroleum industry chain due to the high concentration of asphaltenes.

Asphaltenes are possibly the most analyzed and yet the least comprehended compounds of the crude oil matrix. Everything about asphaltenes seems to be ambiguous, unpredictable and intricate. Moreover, asphaltenes are considered the least valuable component of the crude oil: they are responsible for most of the problems during oil production, transporting, and upgrading because they present a strong tendency to precipitate. Change of conditions, such as diluent addition and pressure drop, can generate asphaltene deposition, causing wellbore or pipeline plugging, the production of stable water-in-oil emulsions, and catalysts poisoning in refinery processes. In this sense, for improving exploration, production and processing of petroleum, understanding asphaltene molecular structure and its aggregation is crucial.

Nowadays, asphaltene description remains as a shadowy story. For instance, a fundamental chemical property of any substance is the molecular weight, but in the case of asphaltenes, it remained controversial for a long time. Distinguishing between monomeric and aggregated asphaltenes is challenging, and thus, the debate on macromolecule or small molecule size nature of asphaltene continued for decades. Recently, the picture of asphaltenes like polymeric species having weights

up to 10^5 g/mol is fading away, and a description of asphaltenes such an ultra-complex and polydisperse mixture with molecular weight distributions along ~200-1500 g/mol is emerging.

Despite that, the debate continues regarding the fundamental molecular architecture of asphaltenes and the driving molecular interaction for the strong aggregation. Two popular competing asphaltene molecular motifs differ in their distribution of the fused aromatic rings and the aliphatic moieties. In the island model, asphaltenes consist of a single polycondensed aromatic core of at least 8-10 fused rings with peripheral long alkyl side-chains. On the other hand, the archipelago motif depicts asphaltenes as smaller islands interconnected by aliphatic bridges. Moreover, an old-school of thought suggests that the island model supports the strong asphaltene aggregation considering the π - π stacking as the leading molecular association.

However, lately reports led by Gray, Strausz, and Rodgers suggest that asphaltenes consist of an extremely complex mixture where archipelago-type structures and island motifs coexist. Moreover, the big island model by itself does not reconcile the known chemistry of petroleum and its molecular structure. In this sense, asphaltene properties such as strong adsorption on mineral surfaces, its suitability for producing distillable hydrocarbons upon thermal processes, the porous nature of the asphaltic macro-aggregates and occlusion of hydrocarbons and biomarkers inside asphaltenes structures are experimental evidence fitting into the archipelago model, in which supramolecular assemblies induce strong aggregation.

As well as the asphaltene content in heavy crude oils continues rising, is critical to understand the molecular basis of asphaltene behavior because, in the light of such knowledge, solution alternatives will be developed. Along these lines, in this dissertation, Fourier transform ion cyclotron resonance mass spectrometry is used to study the molecular basis of the most paramount asphaltene properties: adsorption, reactivity, occlusion, and aggregation.

Chapter 1 introduces the readers into high-resolution mass spectrometry and Petroleomics. It is given a comprehensive review on data analysis, interpretation and the use of sample preparation strategies to improve understanding of crude oil composition. Through Chapters 2–5, it is combined sample fractionation methods with high-resolution mass spectrometric analysis to understand the molecular basis of asphaltene properties. Thus, Chapter 2 provides a detailed description of the chemical features essential for asphaltene adsorption on mineral surfaces, specifically silica. At this regard, it is addressed the role of aromaticity, steric hindrance, accessibility of polar functionalities and molecular size in asphaltene adsorption processes.

Chapter 3 focuses on the molecular composition of the products from upgrading processes applied on asphaltenes. Sample fractionation of the products and deep graphical analysis of the high-resolution mass spectrometry data were useful to establish trends between compound class reactivity and the suitability to produce valuable maltenic compounds. Importantly, the analysis of the products, the residual asphaltenes, and the original feedstock hinted the structure of asphaltenes' building blocks. Along this lines, structural reconstruction of the parent asphaltic compounds, such as archipelago or island architectures, were elucidated.

Chapter 4 presents a novel methodology to extract occluded compounds inside asphaltenes aggregates. Coupling of fractionation methods and high-resolution mass spectrometry for the analysis of the trapped fractions allowed concluding that asphaltenes occlude a wide diversity of hydrocarbons: from long-alkyl-chain saturates to highly condensed aromatic cores with polar functionalities. The occlusion of such as compounds is supported by macroporous structures, which according to theoretical considerations, can only occur if asphaltenes exhibit archipelago motifs.

Finally, Chapter 5 presents correlations between molecular composition and asphaltene aggregation. The aggregation kinetics of four asphaltene samples from different geological origins were followed by the Turbiscan Technology. Further fractionation of the asphaltene samples by size exclusion chromatography coupled to selective solvent extraction, and the analysis by high-resolution mass spectrometry, applying infrared multiphoton dissociations, allowed to study the effect of heteroatom content, aromaticity, alkyl-substitution, and structural motif in the degree of asphaltene precipitation. Remarkably, the experimental evidence suggests that π -stacking is not the driving force in asphaltene aggregation. Instead, the synergy between heteroatom content, high content of alkyl side-chains, and the existence of archipelago motifs, seems to be responsible for the strong deposition of asphaltenes.

1. FOURIER TRANSFORM ION CYCLOTRON RESONANCE MASS SPECTROMETRY AND PETROLEOMICS

Along the history of humankind, societies have always taken from nature the energy needed for the development of life. The discovery of fire was the biggest step toward civilization. The fire allowed man's survival, driving the evolution through processes such as cooking of food, handling of metals to make tools, and art development (Milton, 1999). As human civilization advanced, other energy sources became necessary, and it was in the late 19th century when the first controlled oil production became true in North America (Masters, Root, and Attanasi n.d.). Since then, oil has been the cornerstone of our civilization. It is not possible to visualize a modern society without the so-called “black gold” because, besides being the primary origin of energy in the transportation industry, it provides the molecular building blocks for the production of polymers, synthetic fibers, and elastomers. With time, the world will consume the oil supply, and a new source of energy will replace fossil fuels. However, technological developments are still years away from providing a consolidated and definitive solution to supply the global demand for energy (Owen, Inderwildi, and King 2010). Nuclear, solar and wind power may be suitable candidates, but these sources only contribute with a fraction of the energy requirement. Nowadays, crude oil provides near the 85% of the total energy demand. An important fraction of this fossil fuel supply has been provided by light and “sweet” petroleum, it means low sulfur concentration and viscosity. However, because of depletion of light and sweet crude oils, the production of hard-processing heavy oils has increased (Corilo et al. 2010; Herzog, Lipman, and Kammen 2001).

Heavy petroleum is undoubtedly one of the most complex mixtures in nature. Commonly referred as super-complex, heavy oils may have more than 100,000 different molecular compositions, which varies as a function of geological origin, thermal maturity, and biodegradation activity (Lababidi et al. 2013). Moreover, for

years, petroleum engineers have used macroscopic measurements to determine its physicochemical properties which in turn determine its quality. However, parameters such as density, viscosity, asphaltene content, and elemental analysis for some cases are not enough to understand the heavy petroleum behavior (Alvarez et al. 2011). In this sense, a comprehensive characterization of heavy petroleum is mandatory (Purcell et al. 2010). Molecular characterization of this kind of matrices offers great challenges. Firstly, most of the species that compose heavy oils have boiling point above 510 °C, which limits its detection and quantitation by gas chromatography; secondly its tremendous structural chemical diversity forces the use of NMR and other techniques towards average structural parameters (Orrego-Ruiz et al. 2015). Fortunately, with the development of soft ionization techniques, Fourier transform ion cyclotron resonance mass spectrometry (FT-ICR MS) has become the quintessential technique to understand complex samples, providing the most accurate picture of the petroleum composition. Along these lines, *Petroleomics* is aimed to predict petroleum behavior from molecular composition (Rodgers, Schaub, and Marshall 2005).

1.1 FOURIER TRANSFORM ION CYCLOTRON MASS SPECTROMETRY

Fourier transform ion cyclotron resonance mass spectrometry is a remarkable example of combining three different science phenomena to give one analytical technique: mass spectrometry, ion cyclotron motion, and Fourier transforms (Jackson et al. 1998). The scope of mass spectrometry (MS) is to generate ions from an analyte by any appropriate ionization technique, to separate these ions by their mass-to-charge ratio (m/z) and to detect them according to their respective m/z and abundance. The analyte may be ionized by either impacting energetic electrons, reactive ions or photons, high electric fields or by thermal effect. Ion separation may be performed by static or dynamic electric or magnetic fields, or simply by the time of flight of the charged particles in a free-field tube (Gross 2011). On the other hand,

Fourier transform provides a way to change the waveform from a time-domain to a frequency-domain (Jackson et al. 1998).

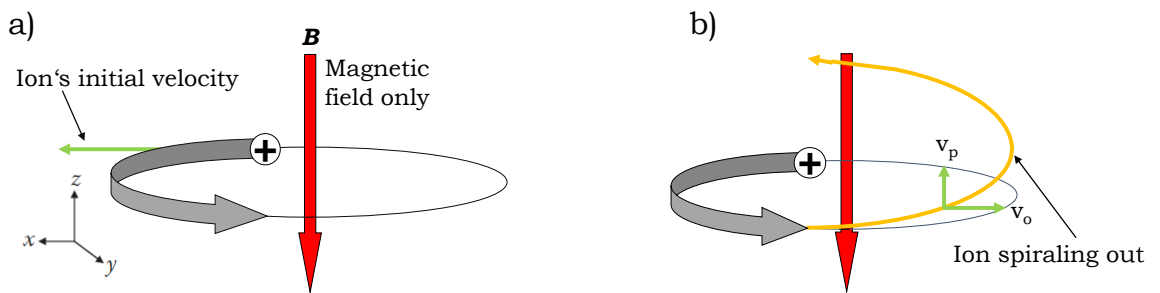
1.2 BUILDING THE ION CYCLOTRON CELL

When an ion enters into a space with a homogeneous magnetic field B , it experiences the Lorentz force, F_L as it is expressed by equation 1.1

$$F_L = mass \times acceleration = m \frac{dv}{dt} = qV \times B \text{ (Equation 1.1)}$$

Where m , q and V are mass, charge and velocity of the ion, respectively. The Lorentz force is perpendicular to the direction of the ion's velocity and the magnetic field (Jackson et al. 1998). As a result, in the absence of collisions, the ion's trajectory is bent into a circle of radius r . Therefore, the ion acquires radial velocity in the xy plane of the figure 1.1a.

Figure 0.1. a) Ion motion in a homogeneous magnetic field and b) ion movement parallel to the magnetic field.



From classical mechanics, the angular acceleration of the ion as a function of the velocity in the plane xy , is given by the equation 1.2; and the angular frequency of the ion (ω) is given by the equation 1.3.

$$a = \frac{V_{xy}^2}{r} \quad \omega = \frac{V_{xy}}{r} \quad (\text{Equations 1.2 and 1.3})$$

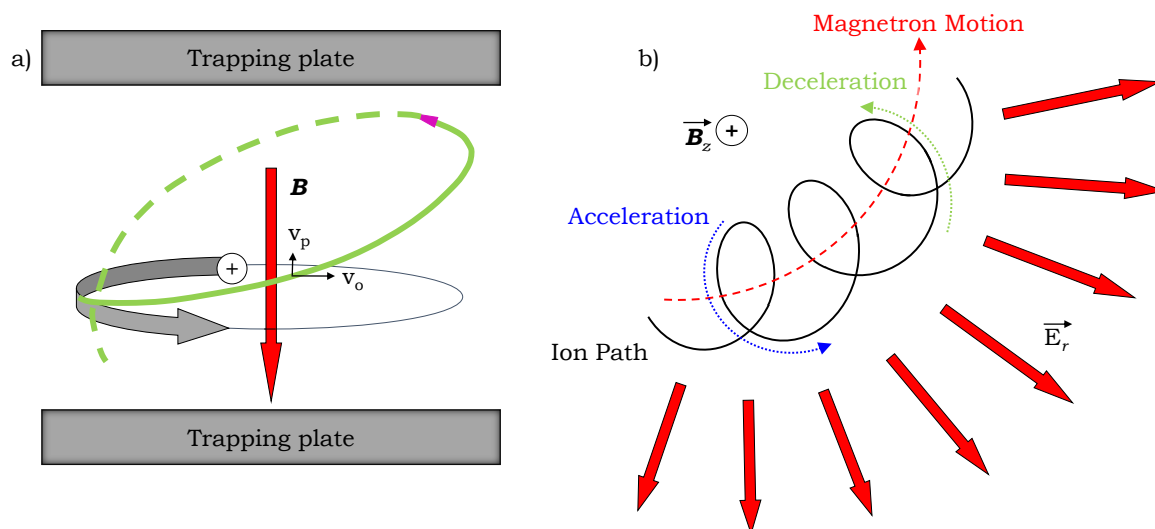
Basic rearranging of equations 1.1–1.3 results in the cyclotron equation, suggesting that the frequency of the cyclotron motion is inversely proportional to its m/z ratio. Equation 1.4 shows that by measuring the cyclotron frequency, m/z may be calculated (Jackson et al. 1998).

$$\omega = \frac{qB}{m} \quad (\text{Equation 1.4, cyclotron equation})$$

However, the ions have free movement in the parallel direction to the magnetic field, experiencing a spiral trajectory (figure 1.1b). So, it is necessary to put two electrodes at both ends of the ion cyclotron resonance cell (figure 1.2a) in order to trap the ions inside the cell. Thus it is produced a three-dimensional electric field, which produces two additional types of motion back and forth parallel to the direction of the magnetic field. As a consequence of the “trapping” potential applied on the ending-electrodes, the ions suffer a *trapping oscillation, which in turn generates a trapping potential* (figure 1.2a). Additionally, because of the trapping potential also has a radial component that produces an outward electrical force on the ion, the center of the cyclotron motion will describe a circular path perpendicular to the magnetic field called *magnetron motion* (figure 1.2b) (Cooper 2010).

In FT-ICR MS experiments, the goal is to measure the cyclotron frequency. This objective cannot be achieved by simply trapping the ions in the ICR cell. Two barriers must be overcome. First, ions with the same m/z are not necessarily in phase with each other, and second, the cyclotron motion needs to be at a radius sufficiently large to allow detection. For example, the cyclotronic ratio of a singly charged ion with $m/z < 1000$, in a 9.4 T magnetic field, is < 0.1 mm, and the average diameter of the ICR cell is between 8–10 cm (Senko et al. 1996).

Figure 0.2. a) Trapping motion as a result of applying voltages through the trapping plates; b) magnetron motion.



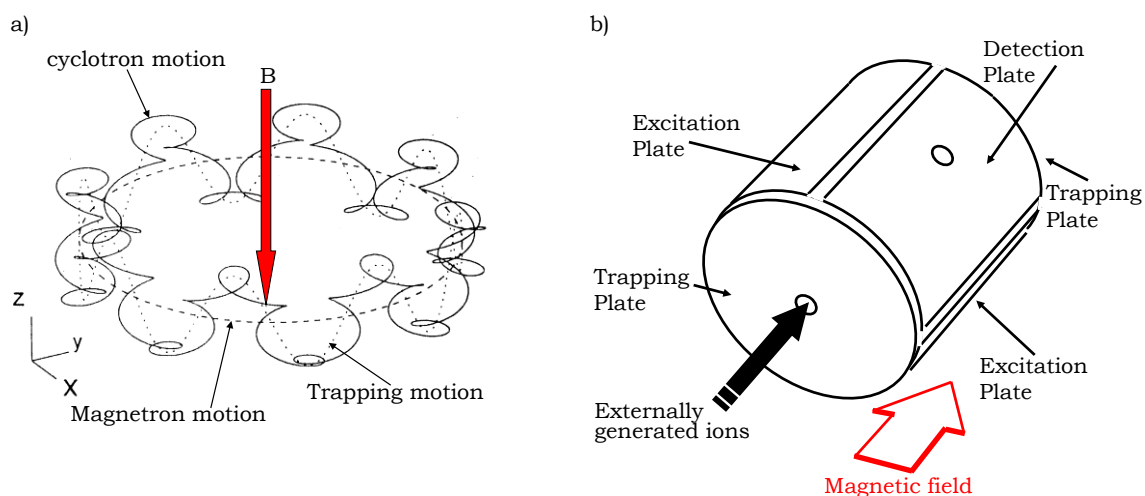
So, for the purpose of detecting the ions trapped in the ICR cell, it is needed to make the ions travel together in a single ion packet at a radius near to that of the ICR cell. This is possible by applying an oscillating electric field having the ions cyclotron frequencies. So, in a typical FT-ICR experiment, a radio-frequency potential containing frequencies that span the desired m/z -range is applied to two of the ICR cell electrodes. Ions orbiting at these frequencies absorb energy; increasing their kinetic energy and hence, their cyclotron radius. The cyclotron radius after excitation is independent of m/z ; all ions of a given m/z -range are excited to the same radius without any mass discrimination effects (Kilgour et al. 2012).

After the excitation, the second pair of electrodes is used to detect the image current induced by the clouds of ions. As a result, FT-ICR MS signal is a time-domain signal that contains the superimposed signals from each of the ion packets. Fourier Transform is applied to convert the time-domain signal into a frequency spectrum, and finally, a frequency to m/z calibration allows obtaining a mass spectrum. Figure 1.3 presents an illustrative picture of the ion motion inside the ion cyclotron resonance

cell, and a common setting of the ICR cell (Comisarow and Marshall 1974; Jackson et al. 1998)

In summary, a typical sequence of an FT-ICR MS experiment consists of successive steps of ion accumulation in the ICR cell, trapping the ion motion along the magnetic field, excitation of the ion cyclotron motion, and signal detection by measuring the oscillating image induced on the detection plates. Figure 1.4 presents the successive events in the FT-ICR MS measurements (Guan and Marshall 1995).

Figure 0.3. a) Ion motion and the b) ion cyclotron resonance cell.

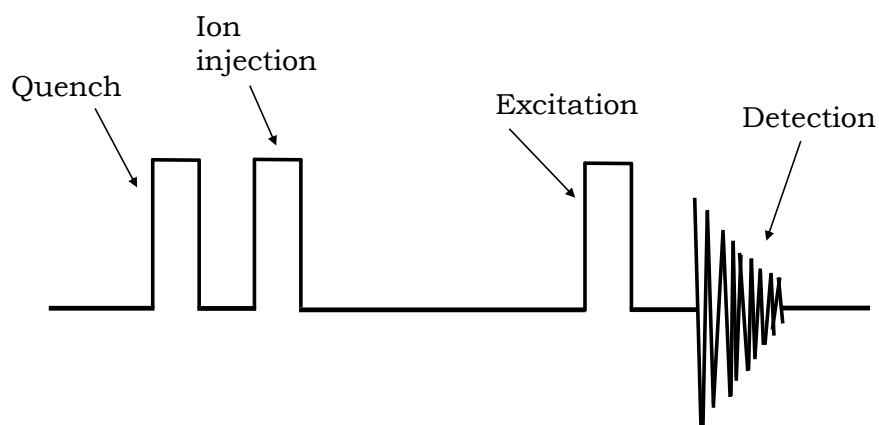


A remarkable feature of FT-ICR mass spectrometry is that the cyclotron frequency is independent of the ion kinetic energy and position, which makes the ICR cell intrinsically a high-resolution technique and unique when compared with other types of mass analyzers (Comisarow and Marshall 1976).

1.3 THE NEED FOR HIGH RESOLVING POWER

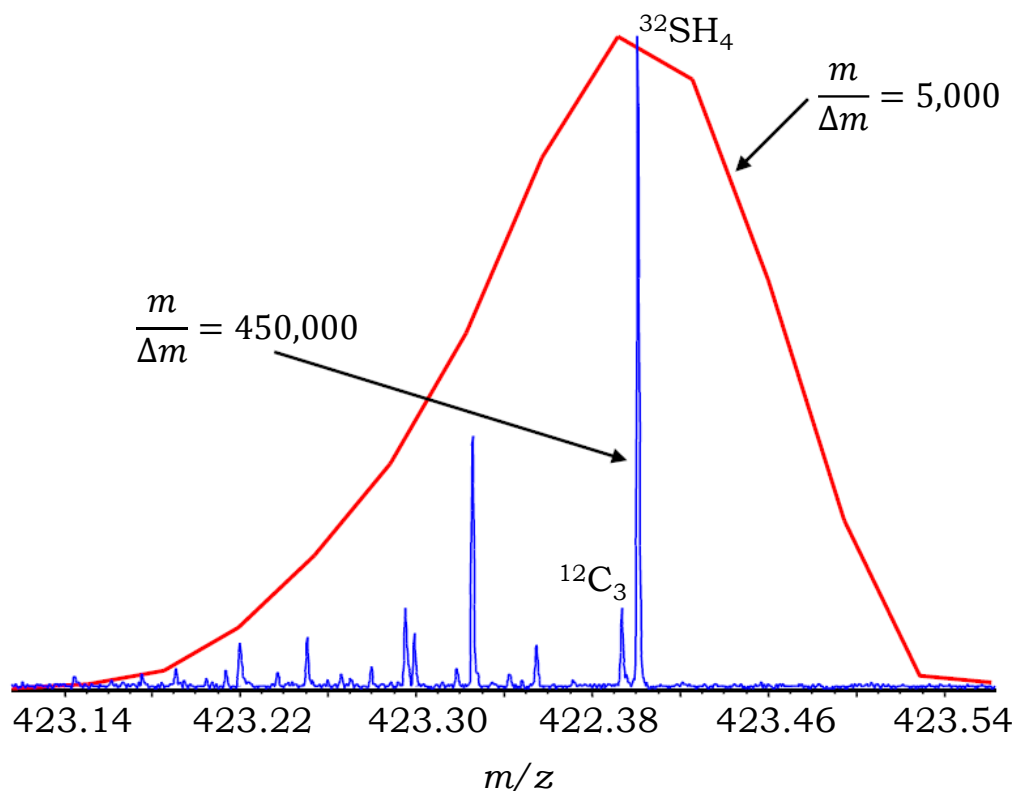
Petroleum is one of the most complex organic mixtures and its characterization has been a difficult task for analytical chemists. Crude oil characterization has usually been limited to average properties such as light scattering, UV-visible and infrared spectroscopy, nuclear magnetic resonance and X-ray diffraction. However, application of mass spectrometry in petroleum characterization has remarkably increased the molecular knowledge about this super-complex mixture.

Figure 0.4. The sequence of events in an FT-ICR MS experiment.



Only the high resolving power and the high mass accuracy offered by FT-ICR mass spectrometry allow assigning molecular formulas for thousands of compounds present in a crude oil sample. This can help to discern between isobaric species present in Crudes. For example, molecular species having the formulas $C_{31}H_{50}$ and $C_{28}H_{54}S_1$ differ in $^{12}C_3$ and $^{32}SH_4$ groups, having the same nominal mass, but differing exactly in 3.4 mDa. Importantly, the separation and the assignment of the doublets $^{12}C_3/^{32}SH_4$ have always been difficult and problematic by low-resolution LC-MS and GC-MS; this is an overcome issue by ultra-high resolution mass spectrometry as it can be seen in Figure 1.5.

Figure 0.5. Comparison between mass spectra of Colombian heavy crude oil: Blue corresponds to FT-ICR MS, and Red corresponds to Time of flight MS.



1.4 MOLECULAR ASSIGNMENT AND DATA ANALYSIS

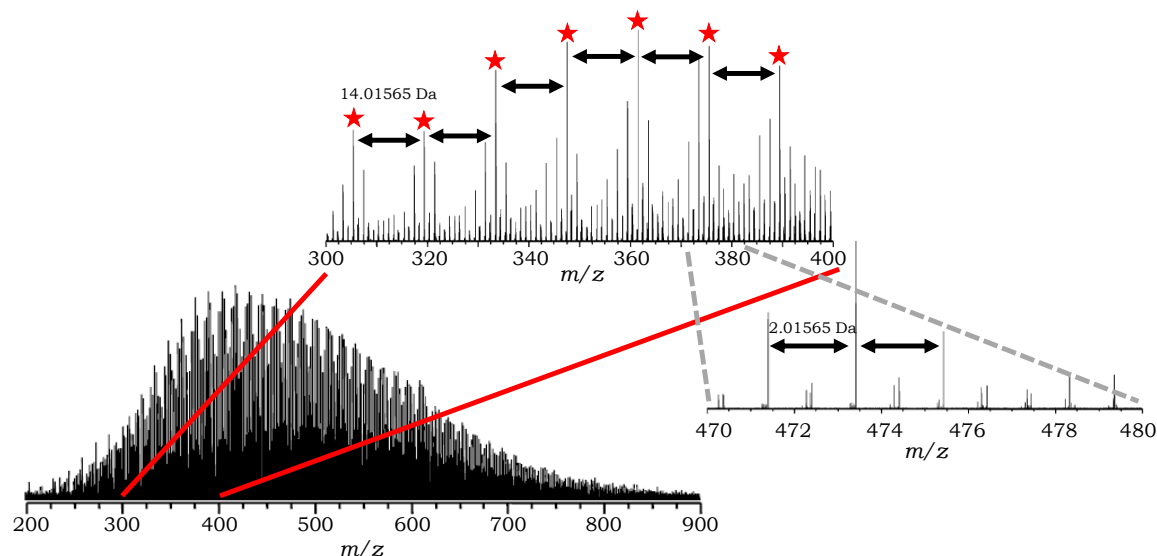
Data from FT-ICR mass spectrometry applied to crude oils is extremely complex. There are reports where are reported more than 80,000 resolved peaks in a single sample (McKenna, Purcell, et al. 2010). It means that the analysis of high-resolution mass spectrometry data cannot be carried out by visual inspection of each peak. Instead, mass spectrometrists take advantage of several features of the mass spectra. Close examination of one FT-ICR MS spectrum shows up repeated spacing patterns along the whole m/z range. Figure 1.6 presents a zooming of an FT-ICR mass spectrum of a South American heavy oil, where a series of 14 Da spacing is readily identified. Each consecutive 14.01565 Da spacing corresponds to an

additional methylene unit (CH₂) in the molecular structure, it means a homologous series with the same number of rings and double bonds (double bond equivalent – DBE), and the same number and type of heteroatoms (compound class), but a different content of CH₂ units are normally observed in a FTMS spectrum. Double bond equivalent is calculated through the equation 1.5, where C, H, and N are the number of carbon, hydrogen, and nitrogen atoms, respectively (Lobodin, Marshall, and Hsu 2012).

$$DBE = C - \frac{H}{2} + \frac{N}{2} + 1 \text{ (Equation 1.5)}$$

Additionally, it is possible to see another series with an exact mass spacing of 2.01565 (the mass of two ¹H atoms), which corresponds to species of the same class and carbon number, but different DBE (Islam et al. 2012).

Figure 0.6. (+) APPI FT-ICR mass spectrum for a heavy Colombian crude oil.



The Kendrick mass scale uses the 14.01565 Da spacing to classify the detected ions in homologous series. Conversion from exact IUPAC mass to exact Kendrick mass

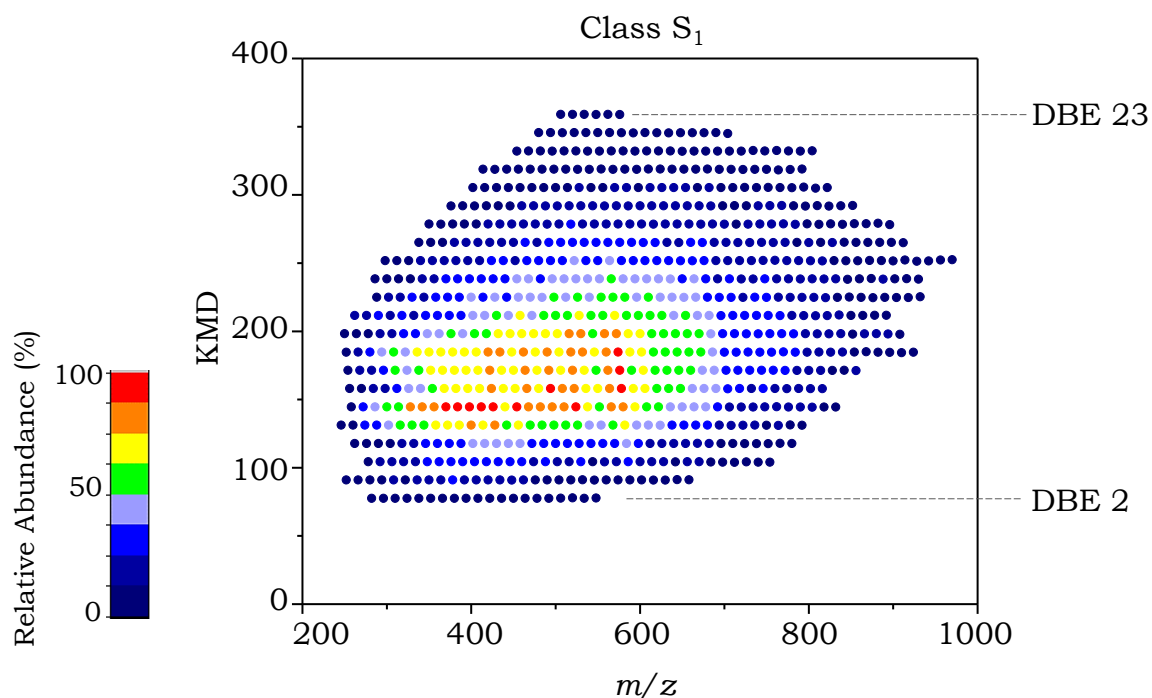
involves rescaling of the measured mass to the IUPAC mass of a CH₂ unit, as equation 1.6 indicates it (Kendrick 1963).

$$\text{Kendrick mass} = \text{IUPAC mass} \times \frac{14}{14.01565} \quad (\text{Equation 1.6})$$

Therefore, the Kendrick mass of the CH₂ unit —14.01565 becomes exactly 14. In consequence, members of the same class and DBE present identical Kendrick mass defect (KMD), as equation 1.7 indicates: (Kendrick 1963)

$$\text{KMD} = (\text{Nominal mass} - \text{Kendrick mass}) \times 1000 \quad (\text{Equation 1.7})$$

Figure 0.7. Kendrick mass diagram for the class S₁ in a Colombian heavy oil.



It is necessary to bear in mind that even at ppb mass accuracy, assignment of elemental formulas above m/z 400 is challenging because the number of structural rearrangements increases exponentially. In this sense, the utility of the Kendrick

sorting is that once few members of a compound class at low m/z (<400) have been identified, extension along the horizontal row of data with identical KMD lets on confident elemental composition assignment (Hughey et al. 2001). Figure 1.7 displays the Kendrick plot for the S_1 class in a Colombian heavy oil, analyzed by (+) APPI FT-ICR MS. In this Kendrick plot, the relative abundance is normalized within the S_1 family, and each point corresponds to a molecular composition. Additionally, higher Kendrick mass defect is the result of higher unsaturation.

1.5 DATA ANALYSIS IN PETROLEOMICS

Commercial software specialized in FT-ICR MS data analysis, such as Composer (Sierra Analytics) and PetroOrg (Future Fuels Institute–National High Magnetic Field Laboratory) perform molecular assignment as follows:

- First step: conversion of the mass scale from IUPAC to Kendrick mass scale, according to the equation 1.5; and calculation of the Kendrick mass defect using the equation 1.6.
- Classification of the peaks in groups by their Kendrick mass defect.
- Computation of the elemental composition of the lowest m/z peak of each group. It follows three criteria: sufficiently S/N ratio (>5), high mass accuracy and isotopic pattern.
- Expansion of the assignment to the rest of the series, by adding the repeat unit CH_2 to the base chemical formula.
- Determination of chemical class, carbon number, and double bond equivalents (DBE).

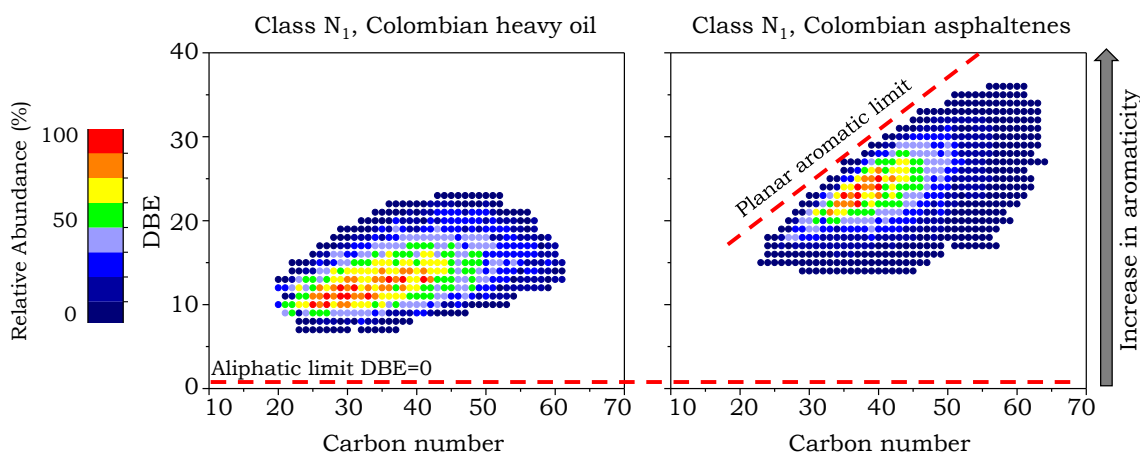
Usually, matching tolerance in the assignment process is ≈ 1 ppm. However, compounds classes with high heteroatom content, such as O_8 class (tetra-carboxylic

naphthenic acids, so-called ARNⁱ acids – naphthenate deposits precursors) and N₄O₁V₁ class (vanadyl porphyrins), need assignment error below 0.5 ppm to get unambiguous chemical assignment (Hughley et al. 2001; Hughey, Rodgers, and Marshall 2002).

Thousands of molecular formulas found in a crude oil are usually sorted in countered plots of DBE *versus* carbon number and van Krevelen diagrams (Hsu, Hendrickson, et al. 2011). These representations are useful to make a graphical differentiation between samples of different chemical nature, different thermal maturity, and petroleum from different geological origin (McKenna, Blakney, et al. 2010). Figure 1.8 presents the countered plots of DBE *versus* carbon number for all the molecules having one nitrogen atom (class N₁), in a Colombian heavy crude oil and its asphaltene fraction. Each point in the diagram represents a molecular composition, featured by its DBE and carbon number. It is clear that aromaticity increases from the bottom to the top, and there are upper and lower limits for the DBE. On the other hand, it is the aromatic planar limit, defined as the line generated by connecting maximum double-bond equivalence values at given carbon numbers. The features of this line, the slope and the y-intercept, are useful to predict and understand the molecular structure of molecules in crude oil. At this regard, Marshall (Lobodin et al. 2012) proposed the “90% rule”; that is, no hydrocarbon molecule DBE can exceed the 90% of the number of carbons in its molecular formula. In this sense, the planar limit slope of petroleum compound classes cannot exceed 0.900 (Hsu, Lobodin, et al. 2011).

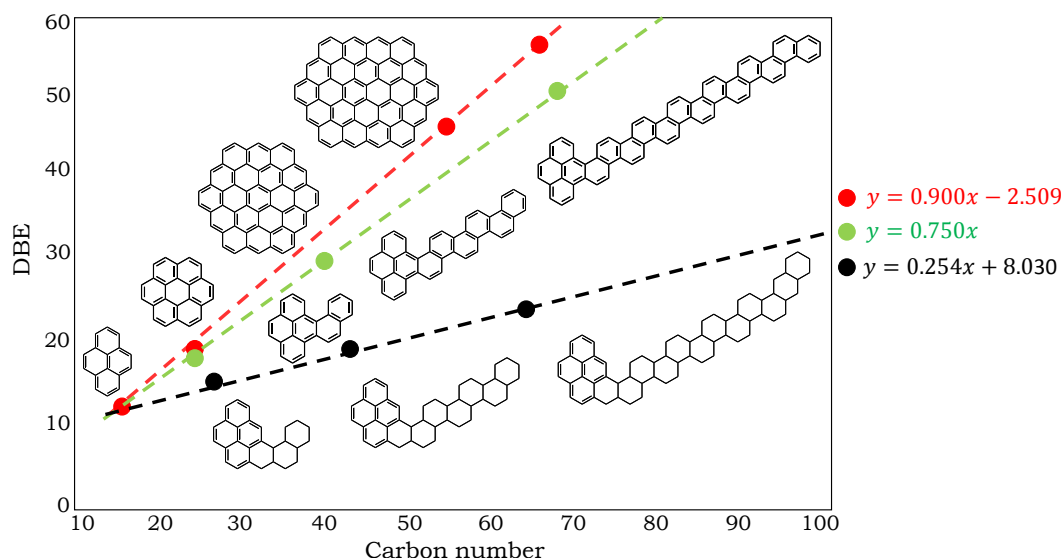
ⁱ ARN is derived from Norwegian and means “eagle”.

Figure 0.8. DBE versus carbon number plots for the class N₁ in a Colombian heavy oil and its asphaltenes.



In another contribution, Cho (Cho, Kim, and Kim 2011) reported the usefulness of planar limit slopes in structural elucidation of the SARA fractions from a heavy crude oil. There is a direct correlation between the aromatic structural condensation and the planar limit slope (Figure 1.9). Just to clarify these concepts, let us consider a hypothetical contoured plot of the class HC, where the molecular formula with the lowest DBE and carbon number, located at the planar limit, is C₁₆H₁₀, likely pyrene. As we move towards higher carbon numbers, pericondensed architecture is the unique molecular motif that meets the DBE increase. This type of structures, resulting from the non-linear addition of benzene rings to the starting structure, originates petroleomes with planar limit slopes around 0.900. On the other hand, if the aromatic rings are linearly added, the result is a catacondensed structure, and the planar limit slope is around 0.750. Finally, the addition of saturated rings to the starting pyrene originates planar limits slopes around 0.250. Cho and coworkers found that saturates, aromatics, resins, and asphaltenes fractions have planar limit slopes of 0.28, 0.73, 0.69, and 0.90, respectively (Cho et al. 2011).

Figure 0.9. Planar limits as a result of molecular structure.



Another strategy to analyze FT-ICR MS data from petroleum samples consists of the van Krevelen diagrams. These diagrams, made by plotting H/C versus O/C (or N/C or S/C) atomic ratios, are extremely useful to make a graphical differentiation between petrochemical samples (Kim, Kramer, and Hatcher 2003). As examples, van Krevelen plots have been used to see chemical differences among thin layer chromatography subfractions of asphaltenes, crude oils samples from different geological origin and vacuum gas oils (VGO) from liquefaction processes (Martha L. Chacón-Patiño et al. 2015; Wu, Rodgers, and Marshall 2004).

Figure 1.10 shows the van Krevelen diagrams for the class N_1 from the same samples previously shown in figure 1.8. Molecular assignments corresponding to compounds in different alkylation series appear along lines intersecting at $H/C=2$. It is easy to see why: if the elemental composition, $C_cH_hN_1$, loses n CH_2 units, its composition becomes $C_{c-n}H_{h-2n}N_1$. The new H/C (y-value) and N/C (x-value) are represented by the equations 1.8 and 1.9 (Wu et al. 2004).

$$y = \frac{h-2n}{c-n} \quad \text{and} \quad x = \frac{1}{c-n} \quad (\text{Equation 1.8 and 1.9, respectively})$$

Elimination of n gives:

$$y = 2 + x(h - 2c) \text{ (Equation 1.10)}$$

Figure 0.10. Van Krevelen diagrams for N₁ compositions in a Colombian heavy oil and its asphaltenes.

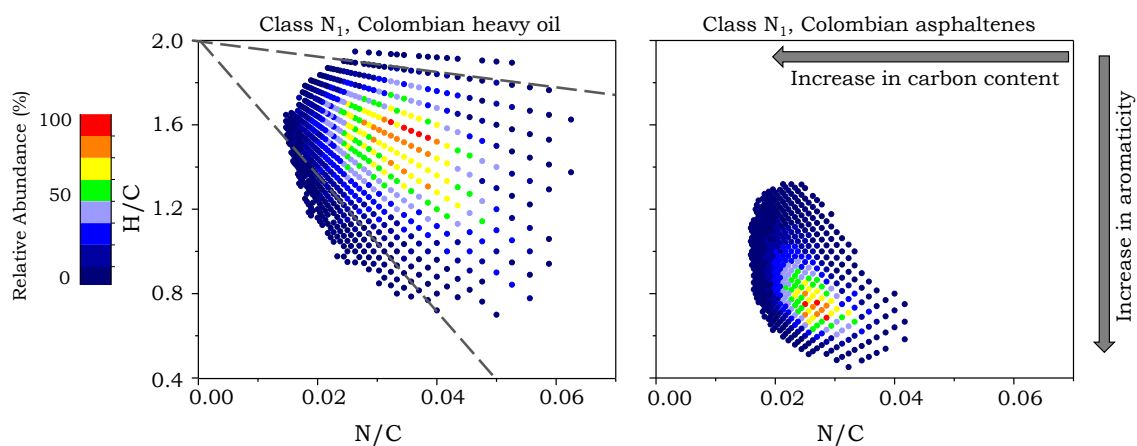
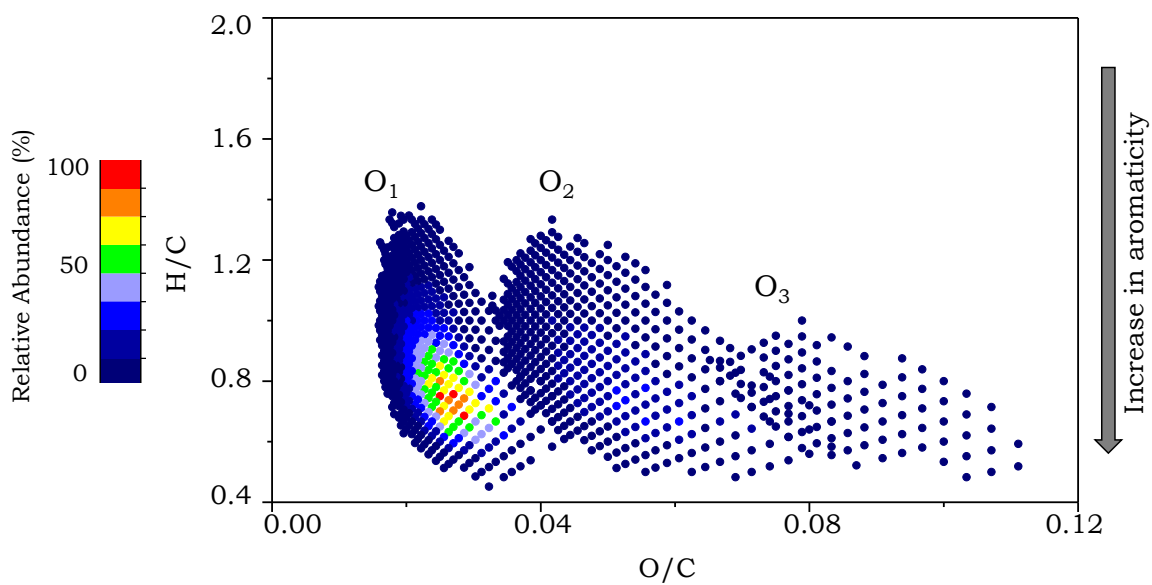


Figure 0.11. Van Krevelen diagrams for the classes O₁, O₂, and O₃ in a Colombian heavy oil.



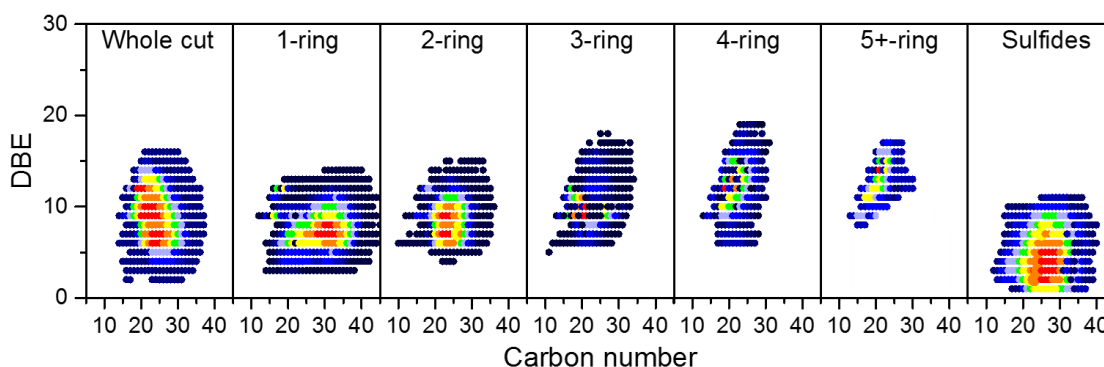
In van Krevelen diagrams, alkylation increases from right to left, and aromaticity from top to bottom. The utility of these diagrams arises from their ability to separate molecular compositions with different heteroatom content in the same plot. For example, Figure 1.11 reveals how O-containing asphaltenes are separated graphically, because of their different oxygen content. Outstanding, high heteroatom content is supported by high aromaticity, in this sense, O₃ class presents the lowest H/C ratios along the three O-classes (Altgelt and Boduszynski 1992; Boduszynski 1987, 1988)

1.6 SAMPLE PREPARATION COUPLED TO FT-ICR MASS SPECTROMETRY ANALYSIS

FT-ICR mass spectrometry allows observation of thousands of compositions in crude oil samples. At the birthday of Petroleomics, Rodgers and Marshall believed that resolution was the only requirement for comprehensive petroleum characterization.³⁵ However, today it is widely known that sample fractionation techniques must be applied to achieve a complete characterization of petrochemical samples, and moreover, to reach molecular speciation (Martha L. Chacón-Patiño et al. 2015).

At this regard, Cho and co-workers have reported the analysis of the SARA fractions from a heavy Arabian crude oil (Cho et al. 2012). They demonstrated that the analysis of the whole crude oil only allowed observation of ~17,600 different molecular compositions; however, the sum of different molecular compositions found in the four SARA fractions was ~33,800. Two effects may cause observation of more compounds by fractionation: concentration of low abundance compounds present in the whole sample or elimination of high ionization efficiency compounds masking low ionization efficiency ones (G. C. Klein et al. 2006).

Figure 0.12. DBE versus carbon number plots for the class S₁ of the ring HPLC-2 fractions from a Colombian heavy vacuum gas oil (427–482 °C).



In another report, Rodgers (Podgorski et al. 2013) used a separation methodology based on high-performance liquid chromatography, called HPLC-2. This was used to fractionate a vacuum gas oils in six fractions: mono-aromatics, di-aromatics, tri-aromatics, tetra-aromatics, penta-aromatics and polars, and observed more chemical species by FT-ICR MS, when compared with the analysis of the original sample as a whole (Podgorski et al. 2013; Robbins 1998). Figure 1.12 shows the DBE *versus* carbon number plots for the class S₁ in the (1-5+) ring fractions for a Colombian heavy vacuum gas oil. Interestingly, higher DBE fractions are shifted towards lower carbon numbers. The reason is because it is a distillation cut defined by an upper and boiling temperature, the progression of the composition from less aromatic to more aromatic molecules, within the boiling cut should meet the continuum model of Boduszynski. Thus, the highest carbon number species must exhibit the lowest DBE, and the lowest carbon number species must be the most aromatic and most polar species.

To achieve molecular speciation, comprehensive characterization of crude oil is carried out by combining fractionation techniques and different ionization methods. First of all, let us introduce a short review of the most used ionization techniques in petroleum characterization.

1.7 COMBINING SAMPLE PREPARATION AND IONIZATION SOURCES FOR CHEMICAL SPECIATION

1.7.1 Electrospray ionization (ESI). The story of the development of mass spectrometry is the story of crude oil characterization. In the 1950's, it was achieved the first MS analysis of petroleum, carried out by electron impact ionization (EI) at 70 eV (Meter, Bailey, and Brodie 1951). This ionization technique produces extensive fragmentation, increasing the complexity of the analysis, and because of the analyte must be in gas phase for the ionization process, the analysis is only focused on volatile species. However, along 1950's-1970's, EI-MS provided valuable information on the relative concentration and the molecular weight distribution of the hydrocarbon types present in waxes. Then, in 1968, it was the first report on the application of a fragment-free mass spectrometry in the analysis of waxes from heavy petroleum fractions: field desorption ionization was used to find the relative concentrations and the mass range for compound types such as *n*-paraffins, iso-paraffins, cycle-paraffins, and alkyl-benzenes (Mead 1968).

While people around the world were working on new ionization strategies for non-volatile compounds, mass spectrometry advanced in mass analyzers and detectors. Along these works, Marshall and Comisarow worked in the development of the mass analyzer with the highest resolution and the highest mass accuracy: the ion cyclotron resonance cell back in the 70's (Comisarow and Marshall 1974; Comisarow and Marshall 1976). However, the ICR cell remained anonymous in complex-mixture characterization until the development of electrospray ionization mass spectrometry by John Fenn, in the late 1980's. However, it was just until 2000 when Marshall and co-workers coupled electrospray ionization with FT-ICR mass spectrometry to resolve and identify 3000 nitrogen-containing aromatic compounds in a whole crude oil sample (Hughley et al. 2001, 2002).

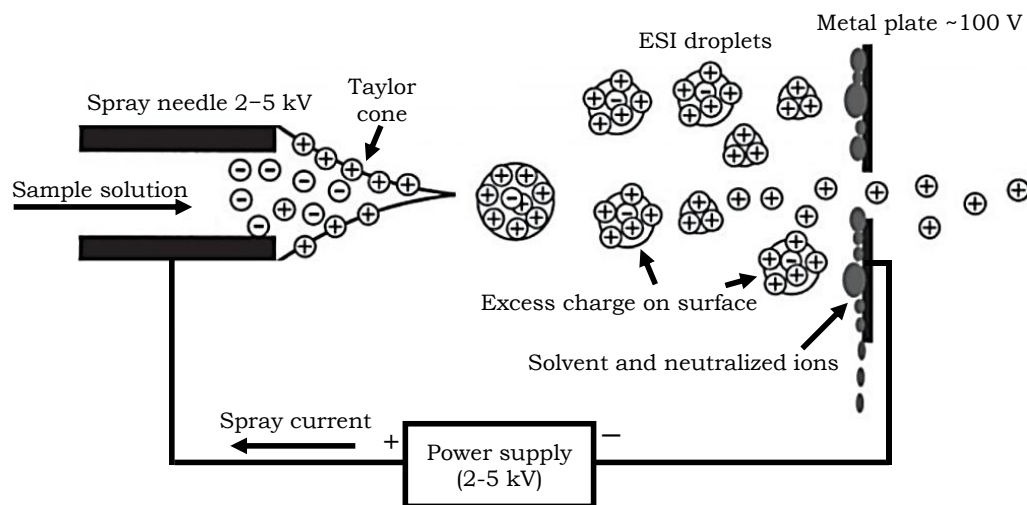
Electrospray ionization is a remarkable creation, for which John Fenn won the Nobel Prize in 2002. Described in colloquial terms by John Fenn, his invention consisted in “creating wings for molecular elephants”. Electrospray makes possible the intact ionization and volatilization of heavy and labile molecules, such as proteins, polymers, and heavy hydrocarbons. A solution of the sample passes through a capillary under a high electric potential ~ 5000 V. At the capillary exit, the solution is dispersed in a spray made up of small charged droplets, which undergo evaporation by two possible processes: electric field desorption or solvent evaporation. Thus, the charged molecules are released into the gas phase. Figure 1.13 presents a configuration of the electrospray source (Fenn et al. 1989).

Electrospray ionization is an useful technique for the analysis and speciation of polar compounds from petroleum such as naphthenic acids and neutral nitrogen in negative mode, and basic nitrogen in positive mode (Shi et al. 2010). In electrospray, ionization proceeds in the polar moieties of the chemical species, via proton transfer mechanisms. Thus, the analyte must be soluble in polar and protic solvents, such as water, methanol, and/or acetonitrile. In the analysis of petroleum, the sample is usually dissolved in mixtures of toluene and methanol, and doped with 1% of formic acid or 1% of ammonium hydroxide, for enhancing the ionization in positive and negative mode, respectively (Qian et al. 2004).

Besides oxygen and nitrogen compounds, one of the concerns in the petroleum industry is the accurate characterization of sulfur compounds. In the beginning, it was thought that electrospray ionization was not versatile for the analysis of sulfur-containing hydrocarbons (Liu, Shi, et al. 2010). However, recently Rodgers and co-workers have reported the application of ESI in the analysis of compound classes such as SO, SO₂, SO₃, SO₄, S₂O, and S₂O₂, by optimization of the ionization by the formation of lithium adducts $[M+Li]^+$ (V. V. Lobodin et al. 2014). Along similar lines, silver cationization also is used to access S_s and S_sO_o compounds in electrospray ionization, by the production of silver adducts $[M+Ag]^+$ (V. V Lobodin et

al. 2014). In this sense, Li^+ and Ag^+ cationization opens a pathway for detection and characterization of S_5O_6 compounds, which usually concentrate at the interface in oil/water emulsions, avoiding tedious separation and time-consuming derivatization (V. V Lobodin et al. 2014).

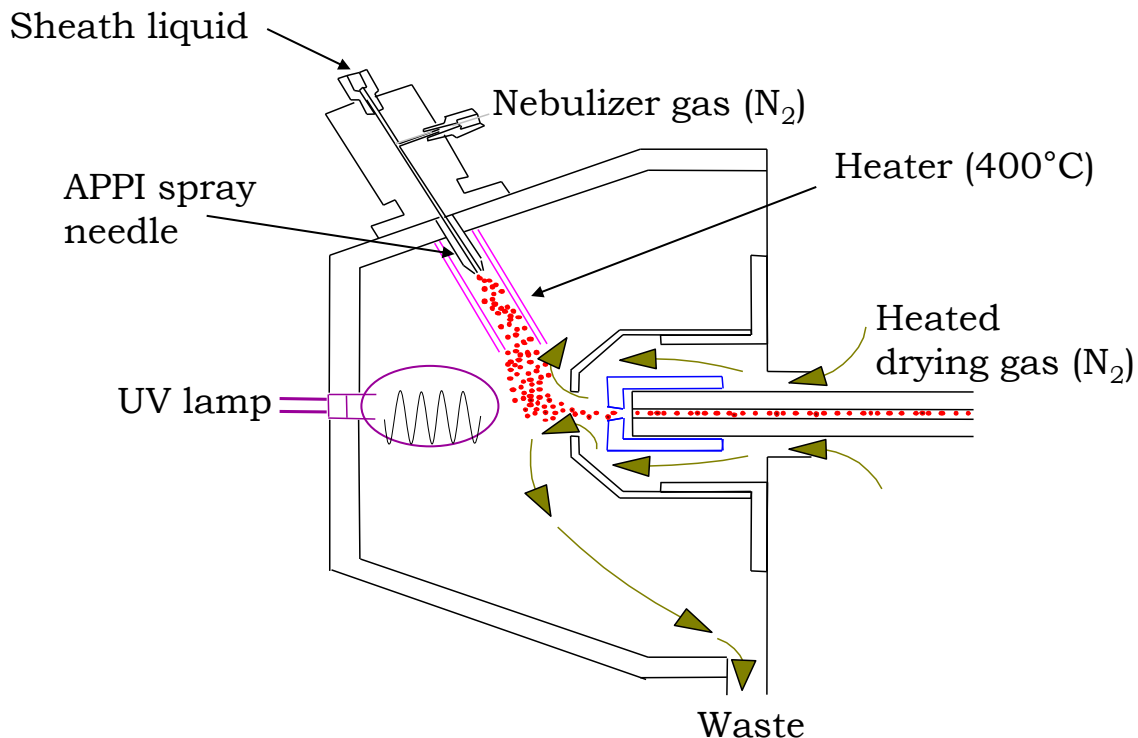
Figure 0.13. Scheme of the electrospray ionization source (Fenn et al. 1989).



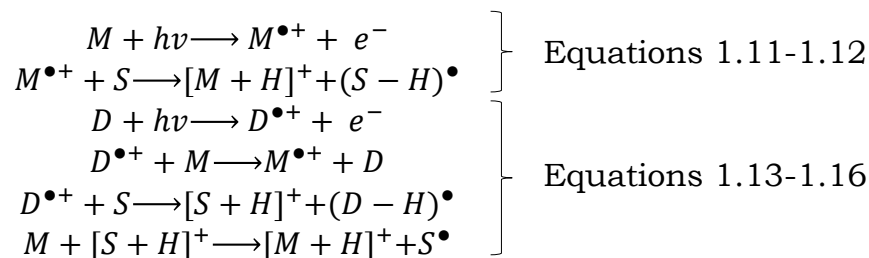
1.7.2 Atmospheric Pressure Photo-ionization (APPI). On the other hand, atmospheric pressure photoionization was developed in the early 2000's to analyze low volatile and non-polar compounds; and it has become the preferred technique for the characterization of non-polar petroleum fractions (Kauppila et al. 2002). As its main feature, APPI ionizes both low polarity and polar petroleum compounds. In the APPI source (Figure 1.14), the sample solution is vaporized by a hot nebulizer. After spraying, the analyte interacts with photons emitted from a discharge lamp (Krypton lamp ~ 10.6 eV). These photons induce a series of gas-phase reactions that may produce both radical $[\text{M}]^{\bullet+}$ and adduct ions $[\text{M}+\text{H}]^+$. Most of the aromatics molecules from crudes have ionization energies between 7 and 10 eV. On the other hand, air components (N_2 and O_2) and most of the common solvents (methanol, water, acetonitrile, and heptane) have higher ionization potentials than petroleum

compounds, promoting useful gas-phase reactions. Figure 1.14 shows the scheme of the APPI source.

Figure 0.14. Scheme of the atmospheric pressure photoionization source.



Nevertheless, the direct ionization of the analyte is characterized by a poor efficiency, due to the solvent property to absorb photons producing photoexcitation without ionization (Robb, Covey, and Bruins 2000). This reduces the number of photons available for the direct ionization of the sample, resulting in low the ionization efficiency. To solve this problem, it is used a dopant at relatively high concentrations that commonly are toluene or acetone (Jeremiah M Purcell et al. 2007). Thus, the ionization process is initiated by the photoionization of the dopant and then spreading toward the analytes. Equations 1.10-1.15 show the ionization mechanisms in free-APPI (equations 1.11 and 1.12) and dopant-APPI (equations 1.13 to 1.16) —where M, S, and D correspond to analyte, solvent, and dopant molecules, respectively.

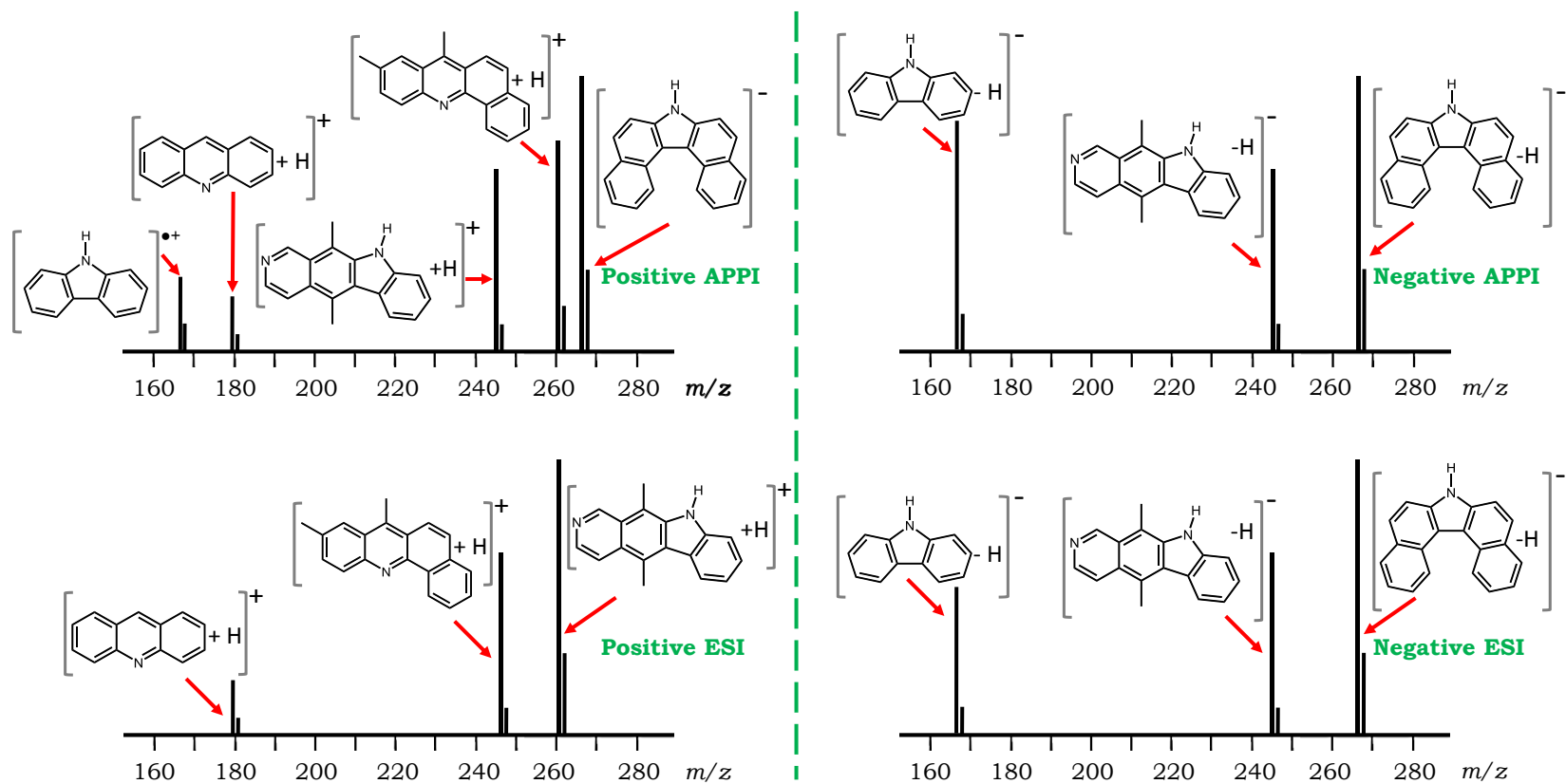


One of the first works reported in the characterization of polar compounds in crudes was done by Purcell and co-workers, who reported the speciation of N-compounds by ESI and APPI FT-ICR mass spectrometry (Jeremiah M. Purcell et al. 2007). They showed that when N atom is present in the pyridine form, it ionizes as protonated cation in positive ESI and positive APPI. Otherwise, when N is neutral –carbazole functional group, it ionizes as a radical cation in positive APPI and a deprotonated anion in negative ESI. Figure 1.15 presents several model compounds and their ionization paths in ESI and APPI. Remarkably, negative electrospray has been used to unveil the molecular composition of the fractions of naphthenic acids, by combining sample preparation techniques, FT-ICR MS, and suitable ionization source.

1.8 IMPROVING COMPOSITIONAL SPACE ACCESSIBILITY AND CHEMICAL SPECIATION THROUGH SAMPLE FRACTIONATION

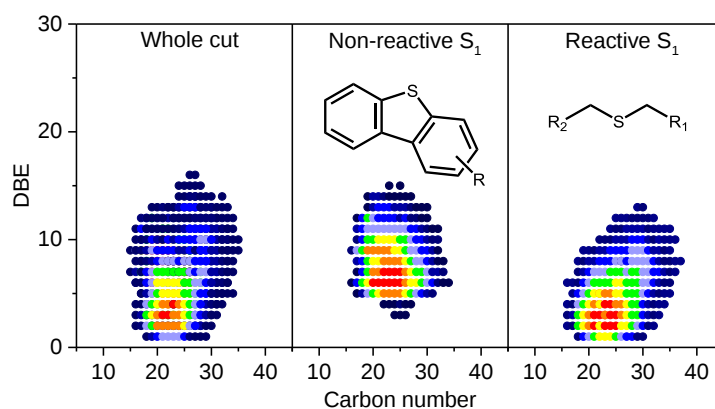
Sample preparation can be achieved by the isolation of the naphthenic acids through solid phase extraction. Sample is placed in contact with amino-silica (Si–R–NH₂) packed in cartridges and then it is eluted with *solvents of different polarities in order* to separate saturates, aromatics, and acid compounds. Even it is possible to achieve selective elution of naphthenic acids based on their molecular size, as Rowland (Rowland et al. 2014) reported.

Figure 0.15. Ionization routes of N-compounds in positive and negative ESI and APPI.



On the other hand, APPI has been used to access the molecular composition of sulfur compounds in petroleum after sulfur speciation (Schaub et al. 2005). Lobodin (Lobodin et al. 2015) reported the use of propyl sulfonic acid silica (PRS-Si) for selective separation of nonreactive and reactive sulfur. Separation was carried out in two stages; the first one consists in the modification of the stationary phase with silver and the second stage consists in sequential elution of sulfur fractions by the use of eluents with different strength. For instance, Figure 1.16 presents the APPI FT-ICR MS characterization of the sulfur fractions of vacuum gas oil from a Colombian heavy oil. This kind of procedure opens the possibilities of a better characterization of sulfur compounds either buy or sell crudes to refining.

Figure 0.16. DBE versus carbon number plots for class S_1 in a whole Colombian VGO, and its non-reactive and reactive sulfur fractions.



1.9 IMPROVEMENT OF ASPHALTENE MOLECULAR VIEW THROUGH FT-ICR MASS SPECTROMETRY

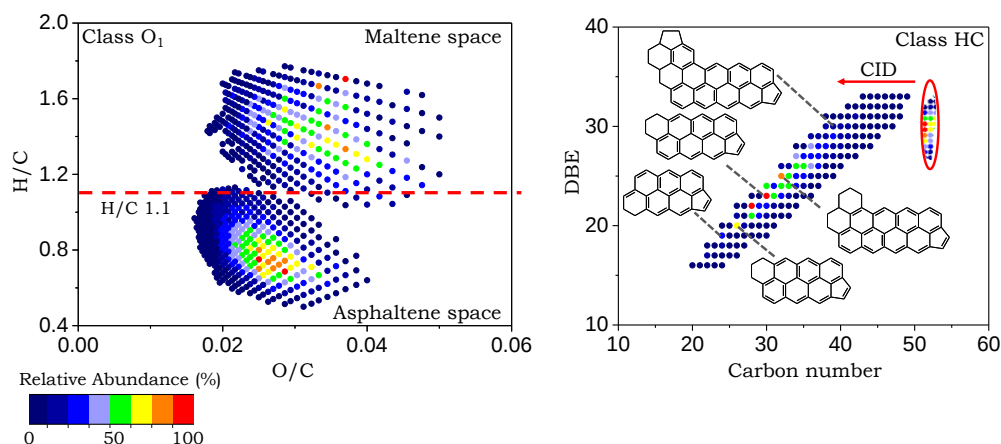
At the present time, the definition of asphaltenes is based on the solubility behavior of petroleum compounds in several solvents (Sheu and Storm 2013). Historically, there have been efforts to create a molecular definition of asphaltenes based on structure and function, beyond the operational description wholly founded on the

principle of solvent fractionation (Snape and Bartle 1985; Speight 2004; Speight et al. 1982). Early works began in 1950's, with the development of standard methodologies to precipitate asphaltenes from crude oils and bitumen (Anon 1979, Anon 2002, Anon n.d.; Knotnerus 1967). On the other hand, current works are focused on unraveling asphaltenes' molecular architecture, aiming to predict reactivity of heavy oils in upgrading processes (Purcell et al. 2010), adsorption of asphaltenic material on mineral surfaces (Adams 2014), and asphaltene deposition (Rogel, Moir, and Witt 2015).

Asphaltenes, operationally defined as the fraction of crude oil insoluble in *n*-alkanes such as *n*-pentane and *n*-heptane but soluble in aromatic solvents such as benzene and toluene, negatively impact production, transportation and refining of crude oils, as direct consequence of their particular molecular attributes (Akbarzadeh et al. 2007; Buenrostro-gonzalez et al. 2001). Asphaltenes are an ultra-complex sample (McKenna, Marshall, and Rodgers 2013), having a wide diversity of functional groups that give rise to non-covalent associations through acid-base interactions between carboxylic acids and pyridine groups, hydrogen bonding, formation of coordination complexes because of vanadium and nickel content, generation of hydrophobic pockets driven by van der Waals interactions between alkyl and naphthenic moieties, and π - π stacking between parallel aromatic cores. Those associations, although individually weak, together bring strongly associated structures that exist even at low concentrations ~ 50 $\mu\text{g/mL}$ (McKenna et al. 2013) and relative high temperatures ~ 70 $^{\circ}\text{C}$ (Agrawala and Yarranton 2001). And it is this strong aggregation, the direct cause of the historical controversies and limitations in accurate and complete characterization of asphaltenes: the evolution on the view of asphaltenes' molecular weights from the range of 10^4 – 10^5 amu, observed by gel permeation chromatography and vapor pressure osmometry (Acevedo et al. 1985) to weights between 200–1700 amu, detected by molecular diffusion measurements and two-step laser mass spectrometry (Groenzin and Mullins 2000; Pomerantz et al. 2008).

In order to improve the molecular definition of asphaltenes, a set of sophisticated analytical techniques that provide the most accurate molecular information is required. Infrared spectroscopy (IR), X-ray diffraction (XRD), nuclear magnetic resonance (NMR), and fluorescence depolarization (FD) have shed some light on the molecular structure of petroleum asphaltenes. Along these lines, some molecular features such as functional groups content by IR (Benkhedda et al. 1992), degree of condensation and substitution of aromatic rings by NMR (Calemma et al. 1995; Trejo, Centeno, and Ancheyta 2004), and number of aromatic cores in an asphaltene cluster by XRD have been determined (Shirokoff, Siddiqui, and Ali 1997; Yen, Erdman, and Pollack 1961). Although these techniques are responsible for the contemporary molecular picture of asphaltenes, they provide only average information (Akbarzadeh et al. 2007).

Figure 0.17. Left: Van Krevelen diagrams for O_1 species in a Colombian heavy oil and its asphaltenes; right: asphaltene core structures from CID experiments.



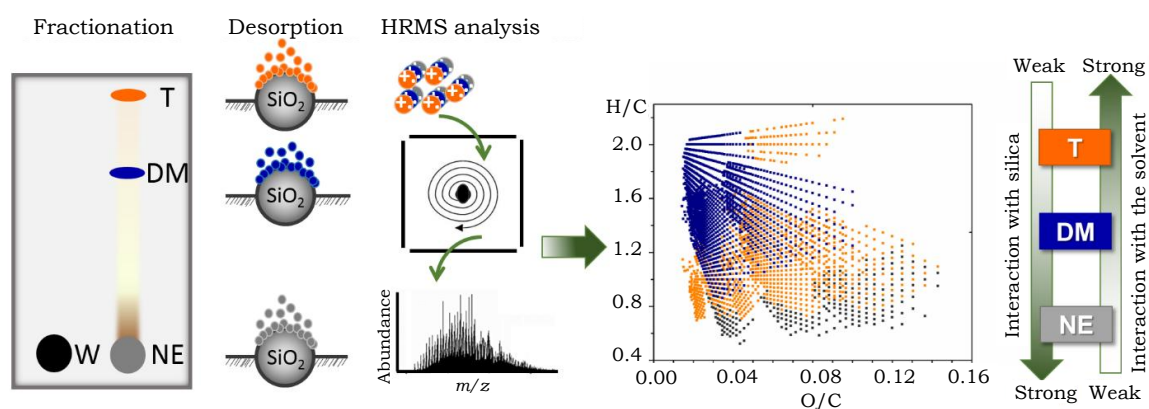
On the other hand, as stated before, since the advent of soft ionization techniques such as electrospray ionization (ESI) and atmospheric pressure photoionization (APPI), the application of Fourier transform ion cyclotron resonance mass spectrometry (FT-ICR MS) has developed to become the quintessential technique

for analysis of super-complex mixtures such as heavy oils, vacuum residues and asphaltenes (Marshall and Rodgers 2004; McKenna, Blakney, et al. 2010; Podgorski et al. 2013; Rodgers and Marshall 2007; Rodgers et al. 2005). By taking advantage of the high resolution and mass accuracy from FT-ICR mass spectrometry, in 2013, Rodgers *et al.* described asphaltenes as the chemical species with DBE values between 20 and 50, and carbon numbers from 30 to 60, with a well-defined aromatic limit, not exceeding a planar limit slope of 0.900 (McKenna et al. 2013). In other words, asphaltenes have H/C ratios below 1.1, being an extension of the maltene compositional space to higher degrees of aromaticity instead of higher carbon numbers, with a maximum structural condensation meant to be pericondensed. Additionally, infrared multiphoton dissociation (IRMPD) was employed to understand the molecular structure, and it allowed to deduce that island and archipelago motifs may co-exist in *n*-alkane insoluble materials. In others words, FT-ICR MS analysis enabled to conclude what Mullins, Gray, and Yen *et al* (Gray et al. 2011; G. Klein, Kim, Rodgers, A. G. Marshall, et al. 2006; Pomerantz et al. 2008, 2009) concluded separately: asphaltenes are not as big as they were believed. However, the controversy around the true molecular architecture remains (Ramírez and Morales 2013; Sabbah et al. 2011). Figure 1.17 displays the petroleomes of the class O₁, in maltenes and asphaltenes samples from a Colombian heavy oil. There is a clear gap between the asphaltene and the maltene compositional space, corresponding to H/C=1.1. Also, Figure 1.17 displays asphaltene core structures found by collision induce dissociations coupled to FT-ICR MS. The isolated compositions (red circled) are fragmented by collisions with N₂. The detected fragments, located in the planar limit region, correspond to molecular formulas proposed as asphaltenes' core structures from theoretical calculations. The detection of fragments with lower DBE values than the precursors hints a parent asphaltene made up by archipelago motifs.

2. HIGH RESOLUTION MASS SPECTROMETRIC VIEW OF ASPHALTENE-SiO₂ INTERACTIONS

**Redrafted from Martha L. Chacón-Patiño, Cristian Blanco-Tirado, Jorge A. Orrego-Ruiz, Andrea Gómez-Escudero, and Marianny Y. Combariza; Energy Fuels, 2015, 29 (3), pp 1323–1331.*

Figure 0.1. Graphical abstract for High Resolution Mass Spectrometric View of Asphaltene-SiO₂ Interactions



2.1 ABSTRACT

The study of asphaltene adsorption on mineral surfaces is fundamental for understanding wettability changes in rocks and fluid behavior in reservoirs. In this chapter, it is reported an analytical approach to investigate the molecular features responsible for asphaltene-silica interactions. It was used high performance thin layer chromatography silica plates and an elutropic series of solvents to fractionate asphaltenes according to their particular affinity with the mobile and stationary phases. There were observed three characteristic asphaltene fractions (with R_fs of 0, 0.69, and 0.90), which were in turn desorbed and analyzed by positive

atmospheric pressure photoionization FT-ICR mass spectrometry. In general, polar non-eluted compounds, highly retained by the silica surface with $R_f=0$, exhibit molecular compositions with N_1 , N_nO_0 and O_0 classes and the lowest H/C ratios compared to the other subfractions. Polar CH_2Cl_2 : MeOH-eluted compounds with $R_f=0.69$, have predominantly HC, N_1 , N_3 , N_1O_1 , N_3O_1 , N_3O_2 , O_1S_1 , O_1S_2 , and S_1 compound classes. Finally, toluene-eluted compounds with $R_f=0.90$ exhibit mostly $N_nO_0S_1$ and O_0S_1 compound classes. It was possible to establish trends between compound retention characteristics and molecular features such as aromaticity, heteroatom/C ratios, and degree of alkylation. Additionally, we show that fractionation of asphaltene samples on silica surfaces expose many species not observed when analyzing the whole asphaltene mixture by APPI-FT-ICR-MS.

2.2 INTRODUCTION

Asphaltene stability is perhaps one of the most challenging issues in the oil industry, since it affects dramatically all the steps in the production chain due to adsorption, aggregation and deposit formation in reservoir rocks, transport pipeline walls, refinery equipment and catalysts surfaces (Syunyaev et al. 2009). Since heavy, extra-heavy and unconventional crude oils, with high asphaltene concentrations, have become appealing to the petroleum industry, analytical efforts using the state of the art high-resolution mass spectrometric methodologies have been used to ascertain the molecular characterization of these complex mixtures. Detailed knowledge of molecular compositions could be translated into efficient production, transport and processing schemes (Mullins et al. 2012; Wiehe and Kennedy 2000).

Asphaltenes are defined by their solubility as the crude oil fraction insoluble in *n*-heptane and soluble in aromatic solvents such as benzene, toluene, and pyridine (Strausz, Mojelsky, and Lown 1992). As a complex mixture, they are commonly described regarding their molecular weight distributions (MWD), recently reported in

the range of 500-1200 g/mol with an average of ~750 g/mol (Pomerantz et al. 2009), however these measurements still pose an analytical challenge. At this regard, Otto P. Strausz *et al.* (Kaminski et al. 2000) using gel permeation chromatography, vapor pressure osmometry, and mass spectrometry concluded that structural polydispersity and aggregation are the main difficulties in the determination of asphaltene MWD. The authors reported MWD in the range of m/z ~200 to m/z ~900 with maxima between $200 < m/z < 350$ for two Mayan asphaltene fractions. In agreement, Pomerantz *et al.* (Pomerantz et al. 2008) observed asphaltene MWD between $200 < m/z < 1200$ using two-step laser desorption/laser ionization mass spectrometry, a technique where asphaltene aggregation is minimized.

Also, the complexity of asphaltenes is reflected by the wide variety of structural motifs found in many asphaltic samples. This diversity includes pericondensed aromatic rings with heteroatoms such as N, O, and S, present in the form of thiophene, sulfide, sulfoxide, hydroxyl, carbonyl, carboxyl, pyrrolic, pyridinic and quinolinic functional units, among others (Gray et al. 2011). These compounds also contain metals such as V, Ni, and Fe bound to porphyrins in the form of metal complexes (Dechaine and Gray 2010). Due to their structural characteristics asphaltenes tend to self-aggregate, even at low concentrations (Tanaka et al. 2004). Importantly, intermolecular forces such as acid-base interactions, hydrogen bonding, metal coordination, π - π stacking and association of non-polar moieties in hydrophobic pockets are linked to asphaltene aggregation. Although individually weak, together they become strong enough to promote aggregation and subsequent precipitation (Gray et al. 2011). These intermolecular forces are also important in asphaltene adsorption on surfaces: the cause of most problems in formation damage and fluid behavior in reservoirs.

Many comprehensive reviews concerning asphaltene adsorption properties on surfaces and its aggregation behavior are available in the literature (Adams 2014). Regarding identification of specific functionalities in asphaltene molecules

interacting selectively with inorganic surfaces, Clementz *et al.* (Clementz 1976) found that basic nitrogen is mainly concentrated in asphaltene adsorbed layers on montmorillonite, while its concentration decreased in the non-adsorbed asphaltene fraction. Also, infrared spectroscopy has been useful for determining the functionalities strongly adsorbed on mineral surfaces. Plancher *et al.* (Plancher, Dorrence, and Petersen 1977) reported that oxygen and nitrogen asphaltene compounds are the predominantly adsorbed species on mineral surfaces. Another report by Dudasova *et al.* (Dudášová *et al.* 2008) showed that trends in adsorption of asphaltenes are strongly influenced by the nature of the surface and not by the source of the asphaltenes. They found a correlation between the amount of nitrogen in asphaltenes and their adsorption density on the solid surface. Recently, Murray R. Gray *et al.* (Zahabi, Gray, and Dabros 2012) reported the characterization of adsorbed asphaltenes on Kaolinite by X-ray photoelectron spectroscopy (XPS) and time-of-flight secondary ion mass spectrometry (SIMS). XPS deep profiling analysis of asphaltenes adsorbed on Kaolinite indicated selective adsorption of oxygenated asphaltenes. However, XPS and SIMS data did not provide evidence for selective adsorption of N or S compounds (Wang *et al.* 2013).

Despite numerous scientific efforts focused on understanding asphaltene adsorption properties, many unsolved questions remain related mostly to the molecular composition of specific compound classes interacting with inorganic surfaces. In this contribution, we use normal phase HPTLC and APPI-FT-ICR-MS to assess the molecular signature of compounds classes in an asphaltene sample interacting with different affinities with a polar inorganic surface such as silica. Asphaltenes extracted from an extra-heavy Colombian crude oil (API=8.2) were placed in contact with silica microparticles and subjected to sequential elution with an elutropic series of solvents. The strength of the interaction between the particular asphaltene compound classes and the silica surface is related to the retention factor (R_f) of the compounds within the HPTLC plate. Three asphaltene subfractions were identified after elution, and they were sequentially extracted from the silica stationary phase

using THF spiked with N-methyl-pyrrolidone. Asphaltene fraction extracts were directly analyzed by APPI-FT-ICR-MS to establish molecular features such as molecular weight distributions (MWD), compound classes, H/C, O/C, N/C, and S/C ratios. The correlation of these molecular characteristics with solvent elution strength and SiO₂ surface properties allowed us to establish the molecular composition of compound families interacting, at different levels, with silica. Additionally, FT-ICR MS analysis of the HPTLC eluted asphaltene fractions exhibit signals for compounds not detected when analyzing directly the whole asphaltene sample. We believe this is a useful approach to study selective interactions between active inorganic surfaces and complex mixtures such as whole crude oils, bitumen, humic/fulvic acids and protein mixtures among others.

2.3 EXPERIMENTAL

2.3.1 Materials. In this study, it was used analytical grade (99.5%) toluene (Tol), tetrahydrofuran (THF), dichloromethane (DCM), *n*-hexane (*n*-C₆), methanol (MeOH) and *n*-heptane (*n*-C₇), which were purchased from Merck (Darmstadt, Germany). *N*-methyl-2-pyrrolidone (NMP) was acquired from Sigma-Aldrich (St Louis, MO). HPTLC plates of silica 60 sorbent (particle size of 4-8 µm with aluminum backing and pore size of 60 Å) were purchased from Merck Millipore (Darmstadt, Germany).

2.3.2 Asphaltene sample. Asphaltene samples from a Colombian extra-heavy crude oil were used throughout this study. The sample was obtained according to the ASTM D6560-12 method. In short, 1 g of crude oil was mixed with 40 mL of *n*-C₇ and heated for 40 minutes under reflux. The mixture was stored in the dark overnight, and the precipitated raw asphaltenes were collected by filtration. Asphaltene cleaning was performed using a Soxhlet apparatus with *n*-C₇ as recycling solvent during 150 hours. Purified solid asphaltenes were redissolved in hot toluene. Finally, toluene was eliminated by roto-evaporation leaving the purified

and reprecipitated solid asphaltene fraction. The weight percentage content of asphaltene in the crude oil sample was $15.5 \pm 0.2\%$. This procedure was repeated several times to accumulate enough samples for analysis. For HPTLC fractionation, a stock solution of asphaltene in toluene (5 mg/mL) was always used.

2.3.3 High-Performance Thin Layer Chromatography (HPTLC). 2 μ L of a purified asphaltene stock solution (5000 ppm) were directly applied on pre-activated HPTLC plates using a micropipette. Fractionation was performed by sequential elution of the asphaltene sample using an eluotropic series of solvents consisting of *n*-C₆ (running time 40 minutes), Tol (running time 7 minutes) and DCM/MeOH 9/1 v/v (running time 5 minutes). The selected eluotropic series is routinely used in a standard TLC method for analysis of saturates, alkyl-aromatics, and polars in petrochemical samples (Vela et al. 1995). The elution process was carried out several times to collect enough sample for mass spectrometry analysis.

2.3.4 Extraction of asphaltene fractions from silica. To establish appropriate conditions for asphaltene recovery from the silica stationary phase, we considered the effect of different solvents and the extraction time. Additionally, to determine extraction recovery factors (R%) from silica, the stationary phase of a non-eluted HPTLC plate loaded with a known amount of purified asphaltenes (5000 ppm) was removed from the aluminum plate and combined with Tol/NMP and THF/NMP mixtures (2 mL for 200 mg of stationary phase) with different NPM content (0, 5, 10 and 15%). This approach is a modification of the one previously reported by T. Pernyeszi *et al.* (Pernyeszi and Dékány 2001). The resulting mixture (silica-asphaltene-solvent/NMP) was subjected to sonication (110 W, 40 kHz, Branson, Danbury, CT) for 1 hour, collected and centrifuged for 20 minutes at 4000 rpm (SiGMA 4-16KS, Germany). The supernatant was collected and the recovered asphaltenes were quantified by UV-Vis spectroscopy using a Genesys10S spectrometer (Thermo Scientific, Billerica, MA) with a built-in calibration curve (linear dynamic range: 15 to 50 mg/L) for asphaltenes. The calibration curve was

constructed using molar absorptivity values calculated for purified asphaltene samples at $\lambda=532$ nm. On the other hand, separation of the asphaltene fractions from the eluted plates of silica was achieved by dissolving the organic phase adsorbed on the silica with THF/NMP 90/10 V/V in an ultrasound bath for 60 minutes. Three samples were collected, labeled NE, DM and T from the HPTLC silica plates. Samples were centrifuged for 20 minutes at 4000 rpm, the supernatants collected and the mixture roto-evaporated. Finally, samples were redissolved in toluene to a final concentration of 0.8 mg/mL.

2.3.5 APPI FT-ICR mass spectrometry. Mass spectrometric analysis was performed on a FT-ICR mass spectrometer (Solarix 15 T, Bruker Daltonics, Billerica, MA) equipped with an APPI source, fitted with a Krypton photoionization lamp (10.6 eV). FT-ICR mass spectra of whole asphaltene and its HPTLC subfractions were acquired in positive mode. Samples were injected at a flow rate of 600 μ L/h. The APPI source was operated under the following conditions: nebulizer gas pressure of 2.0 bar; capillary voltage and end plate offset of 2 kV and 500 V, respectively; vaporizer and dry temperature of 280 and 200 °C, respectively; the dry gas flow was set at 4.0 L/min. Ions were accumulated in a hexapolar collision cell for 50 ms and transported to the ICR cell through a multipole ion guide system with a time of flight of 0.8 ms. Each spectrum was recorded by the accumulation of 200 scans of time-domain transient signals in 4 mega-point time-domain data sets. The front and back trapping voltages in the ion cyclotron cell were set at +0.70 V and +0.50 V, respectively. The ICR cell was calibrated using a NaTFA solution (m/z from 200 to 1200). All mass spectra were internally recalibrated using a series of homologous alkylated compounds. A resolving power, at m/z 400, of 520,000 and a mass accuracy threshold <1.0 ppm provided unambiguous molecular formula assignments for singly charged molecular ions with relative abundance above 0.1%. Mass spectra were processed using the Composer software version 1.0.6 (Sierra Analytics, Modesto, CA, USA).

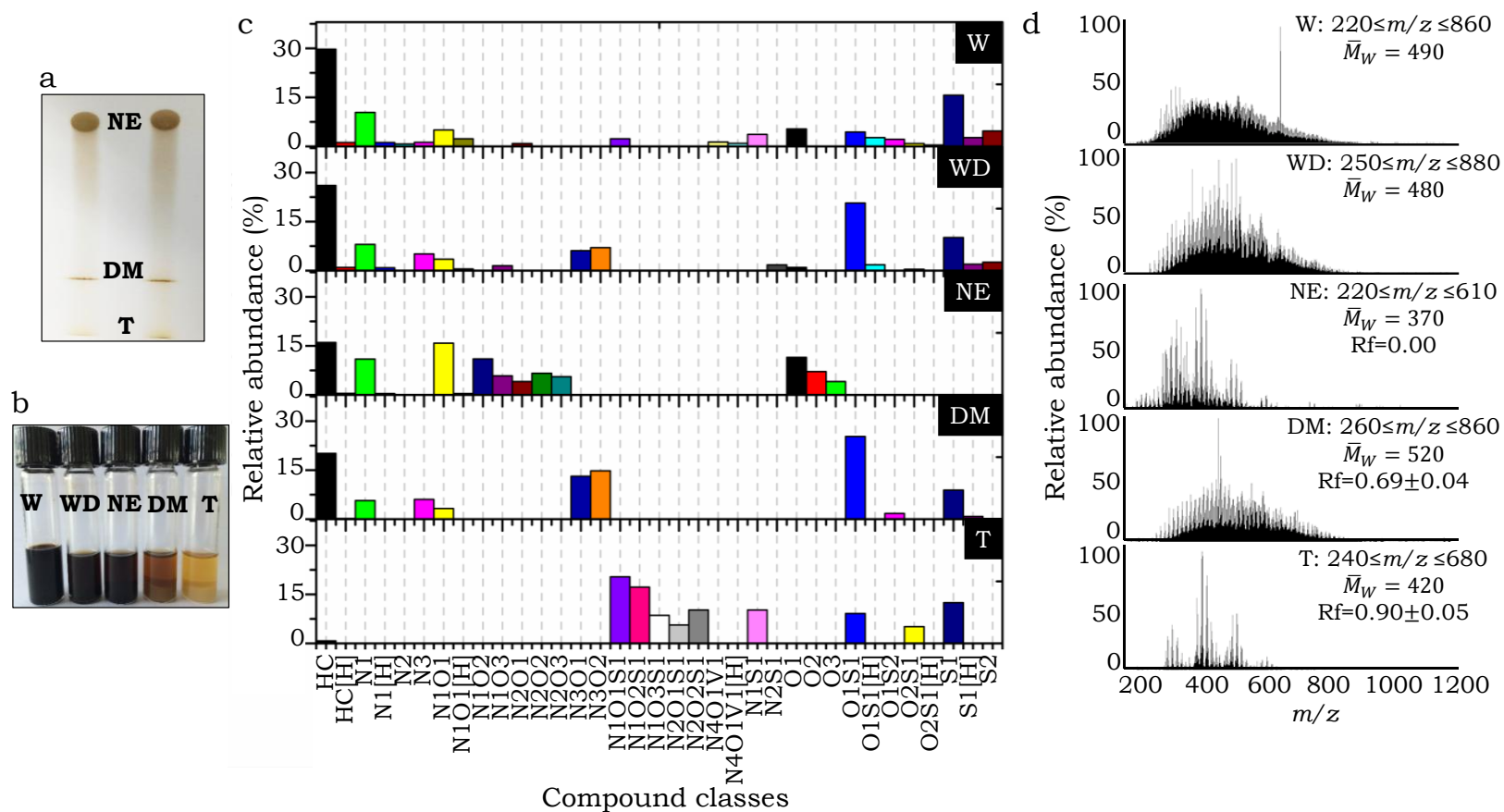
2.4 RESULTS AND DISCUSSION

Fractionation of complex mixtures such as crude oils can provide a comprehensive molecular characterization by allowing access to compounds not detectable when the original sample is analyzed as a whole, particularly in analytical approaches concerning high-resolution mass spectrometry (HRMS). We believe that separation of asphaltenes according to their degree of interaction with a common mineral surface (such as SiO_2) could potentially provide a tool to establish molecular features particular to compounds prone to causing problems due to adsorption issues. In this sense, purified asphaltene samples were fractionated by HPTLC following the procedure described in the *Experimental Section*. Figure 2.2a shows a silica HPTLC plate after eluting the whole asphaltene sample (W) using the following eluotropic series: *n*-C₆, Tol, and 9/1 (v/v) CH₂Cl₂/MeOH. Elution with *n*-C₆ caused no fractionation of the asphaltene sample on the HPTLC plate, as expected considering the solvent's hydrophobic nature and the polar character of both asphaltene and SiO_2 . The second elution with toluene effectively fractionated the sample and dragged, almost with the solvent front, a band located on the far right side of the plate labeled T. The third elution solvent (9/1 (v/v) CH₂Cl₂/MeOH) produced a second band labeled DM. Finally, a band of non-eluted compounds corresponding to the fraction that did not migrate with any of the solvents in the eluotropic series was labeled NE.

Interactions between the analyte molecules and the stationary and the mobile phases drive the asphaltene fractionation on the HPTLC silica plates. Regarding the stationary phase, the surface of silica microparticles consists of various kinds of silanols and siloxanes. Siloxanes are usually regarded as hydrophobic, with no bearing on polar solute retention according to some authors, while silanols are considered active adsorption sites. The high polarity surface of silica is due mostly to the presence of silanol groups, in single, geminal, or vicinal forms and additional hydrated silanols in the case of hydrated silica (Pernyeszi and Dékány 2001; Shen,

Zettlemoyer, and Klier 1980). These active sites can interact strongly with molecules containing basic nitrogen, carbonyl carbons, fused aromatic rings, oxygen functionalities -carboxylic acids, ketones, and metal complexes, such as vanadyl porphyrins (Parida et al. 2006). For the mobile phase, we used an eluotropic series with a solvation capability gradient appropriate for interactions with medium to high polarity samples, such as asphaltenes. The polarity indexes (ϵ_0) for *n*-C₆, Tol, and 9/1 DCM/MeOH are respectively 0, 0.29, and 0.47. We calculated the retention factor (R_f) for each of the fractions in the plate, as the ratio between the distance traveled by a band on the TLC surface and the distance traveled by the solvent front. NE, DM, and T fractions have R_f values of 0, 0.69±0.04, and 0.90±0.05, respectively. As R_f's reflect on the polarity of the eluted fraction, and ϵ_0 on the solvation capability of the solvent, we could argue that a lack of separation with *n*-C₆ is in agreement with the solubility definition of asphaltenes and it indicates that all asphaltene molecules have a relatively high polar character and are also retained by the polar SiO₂. Along the same lines, molecules interacting weakly with the polar SiO₂ support and strongly with the aromatic solvent are present in the T fraction, while mobile polar molecules with high capability of interact preferably with DCM/MeOH mixture through hydrogen bonds are found in the DM band. Finally, highly polar molecules, perhaps amines and oxygenated asphaltenes, interacting strongly with the polar silica support and weakly with the eluotropic series of solvents, might be present in the NE fraction.

Figure 0.2. (a) HPTLC SiO₂ plate showing three characteristic bands after asphaltene fractionation with n-C₆, Tol, and CH₂Cl₂/MeOH; (b) whole and fractionated asphaltenes desorbed from the SiO₂; (c) compound class distribution; (d) molecular weight distributions for W, WD, NE, DM, and T asphaltene fractions.



Using the same eluotropic series of solvents for the analysis of a deasphalted heavy oil by TLC-FID, Vela *et al.* (Vela *et al.* 1995) observed bands corresponding to saturates (eluted with *n*-C₆), alkyl aromatics (eluted with toluene), and polar compounds (eluted with DCM/MeOH), while the non-eluted fraction was assigned to asphaltenes. In another report, Rodgers (Smith *et al.* 2014) performed direct FT-ICR MS analysis of petroleum fractions separated by thin layer chromatography. They employed a mixture of *n*-C₇/isopropanol to separate compounds by polarity, which produced the farthest migration of the least polar molecules —saturates, followed by polar molecules such as aromatics and resins, and ending with the most polar molecules —asphaltenes. In this study, FT-ICR MS analysis allowed the establishment of characteristic molecular features for each fraction.

While the nature of the solid support is fundamental for selective adsorption of organic compounds, the silica pore diameter and particle size (4–8 µm and 60 Å, respectively) have a direct effect on separation performance in terms of resolution and analysis time. Small particle size and pore diameters translate into better resolution and shorter analysis time. In general, HPTLC plates offer improved separation when compared with conventional TLC. On the other hand, it is important to take into account the so-called super-complexity of petrochemical samples, which combined with the continuous petroleum composition, makes chromatographic separation a tough task. In this sense, the work was focused on optimizing mobile phase and asphaltene concentration. The eluotropic series used was selected because it is a standard for crude oil SARA analysis using TLC (G. C. Klein *et al.* 2006; Vela *et al.* 1995). The rationale for this selection lies in the fact that considering the continuous nature of crude oil composition, and its fractions, asphaltenes will also have a continuous composition and will exhibit a tendency similar to that of the mixture they were extracted from. On the other hand, the concentration was varied so it was possible to minimize band tailing and collect significant amounts of sample for mass spectrometric analysis. Under these conditions, asphaltene multilayer formation and nanoaggregation could be an issue (Abu Tarboush and Husein 2012).

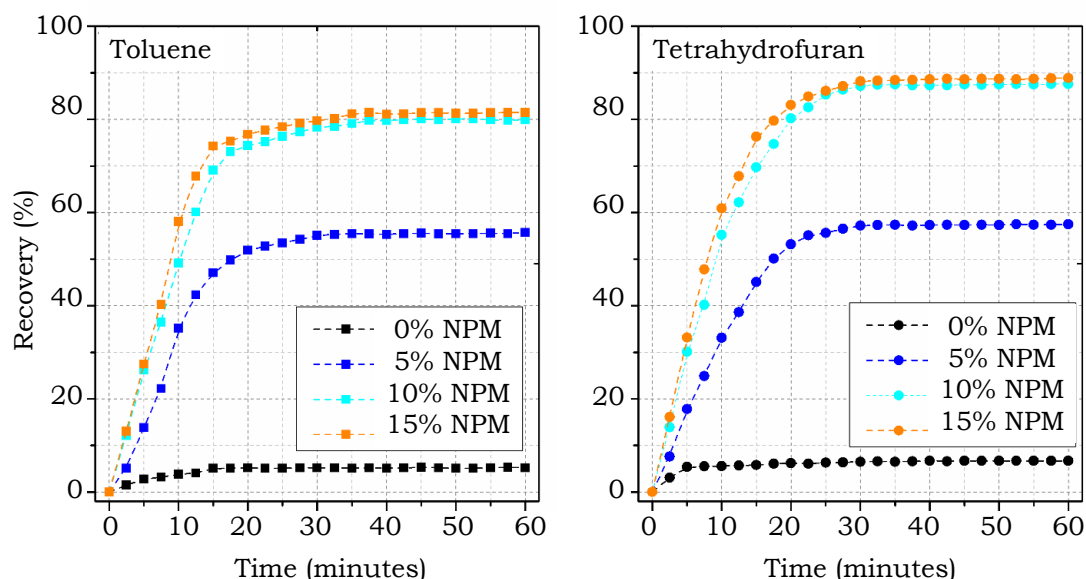
However, it is important to emphasize that the amount of the sample used for the HPTLC fractionation (10 µg) does not exceed the mass loading capacity of the HPTLC plates (14.14 µg). Along these lines, there are enough active sites on the stationary phase surface for interactions with the sample, preventing sample overloading and multilayer formation.

Once asphaltenes are fractionated on the HPTLC plate, fraction recovery from the silica is of fundamental importance for subsequent molecular characterization. At a first and simple approximation, it was considered confining fraction retrieval exclusively to the NE, DM, and T bands. Band tails were not considered in the extraction process, and future efforts will be focused on the molecular characterization of asphaltenes located in band tails to extend the accessible asphaltene molecular compositional space. Due to strong interactions between asphaltenes and silica, many research groups report the need for polar (and extremely toxic) solvents, such as dimethylsulfoxide (DMSO) or pyridine, for efficient desorption.^{21,43–45} However, Pernyeszi *et al.* (Pernyeszi and Dékány 2001), testing asphaltene adsorption and desorption processes on various silica sorbents, proposed the use of *N*-methylpyrrolidone as toluene additive to effectively desorb asphaltenes from silica.

Along these lines, we tested Tol and THF, spiked with 5, 10 and 15% NMP, as desorption solvents in an attempt to recover whole asphaltene samples from the silica. We followed the recovery efficiency evolution for a whole asphaltene sample, by UV-Vis, when using different solvent/NMP mixtures (Figure 2.3). In general, the THF/NMP system was more efficient for asphaltene recovery from the SiO₂. Also, neither pure toluene nor THF is useful as recovering solvent, since only 7% (approx.) of the sample can be extracted in this way. On the other hand, the amount of extracted asphaltenes with 10 and 15% of NMP is almost the same for both solvents, still with the THF system being the most efficient. Now, considering the high boiling point of NMP (204 °C), which could be difficult to remove from the sample by roto-

evaporation and interfere with the HRMS analysis, it was decided to use the minimal amount of NMP for efficient asphaltene recovery. In the case of the THF/NMP system, it was selected 10% of NMP to achieve $90\pm2\%$ of asphaltene desorption from the silica. Figure 2.2b shows the asphaltene fractions (NE, DM, and T) and the whole asphaltene (WD) recovered from the silica using THF+10% NMP as solvent.

Figure 0.3. Asphaltene recovery profiles from silica.



After optimizing fraction recovery efficiency from the SiO_2 stationary phase and extracting TLC bands accordingly, we performed positive APPI FT-ICR MS analysis to establish the molecular composition of the whole and fractioned asphaltenes. Most of the ion signals observed in the mass spectra of the asphaltene sample and its fractions correspond to radical cations. We observed protonated ions for classes such as HC, N_1 , $\text{N}_4\text{O}_1\text{V}_1$, N_1O_1 , O_2S_1 , O_1S_1 , and S_1 in low abundance ($<2.8\%$). Figure 1 displays compound classes observed for whole and desorbed asphaltenes (W, WD) and the NE, DM and T fractions recovered from the silica, together with their MWDs. Observed MWD ranges are in agreement with previous literature reports, ranging from m/z 200 to m/z 1800 (Altgelt and Boduszynski 1992; Badre et al. 2006). McKenna *et al.* (McKenna et al. 2013) found, by electrospray ionization coupled to

time-of-flight mass spectrometry, that MWDs of asphaltene monomers should range within $350 < m/z < 800$, while MWDs of asphaltene dimers are located around m/z 2000 and MWDs of multimers extend up to 22 kDa.

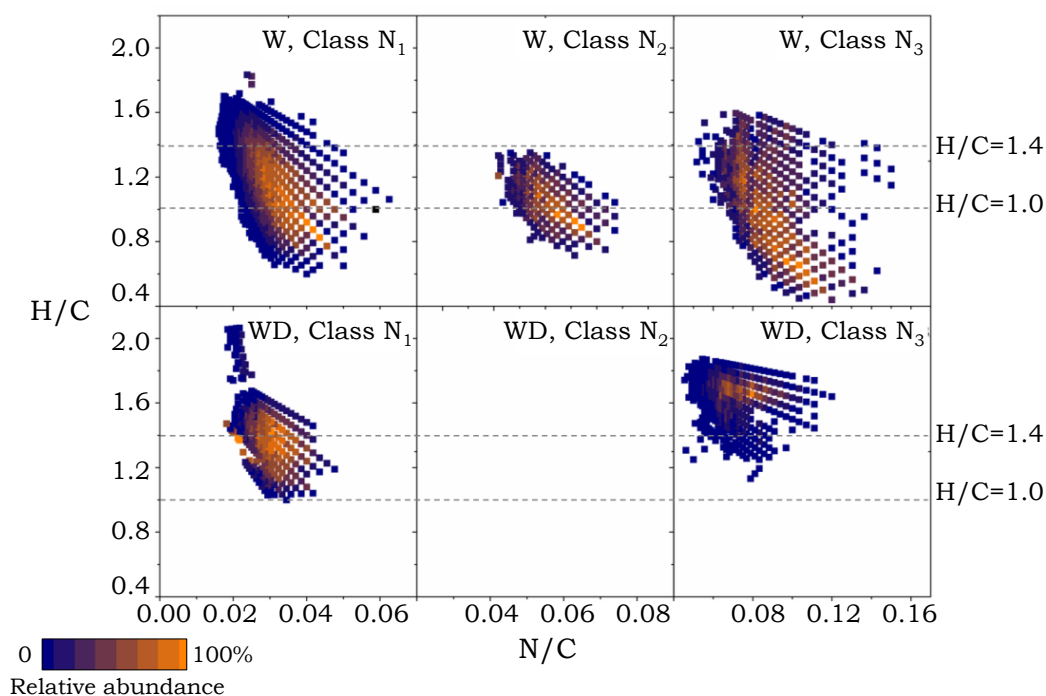
Though MWD hints at differences among the samples, variations in compound class distributions, as the asphaltene sample is fractioned or simply desorbed as a whole from the silica, effectively expose selective interactions between the sample with the SiO₂ and the solvents. Our initial approach was to compare the whole asphaltene sample before and after interacting with the SiO₂, without any chromatographic separation. The APPI analysis of whole asphaltenes results in the observation of 15 different compound classes detected as radical cations (sample W, Figure 2.2) while only 13 compound classes are detected in the SiO₂-desorbed asphaltenes (sample WD, Figure 2.2). Comparing W and WD samples composition, one can easily see which types of molecules are selectively bound to the SiO₂ surface after desorption with the THF-NMP mixture. Compound classes N₂, N₂O₁, and N₄O₁V₁, among others, are actively retained by SiO₂ and are not observed in sample WD.

Strong interactions between N- and O-containing compounds and SiO₂ are in agreement with many reports and reviews in the literature (Ford et al. 1981; Marczewski and Szymula 2002). Silanol groups, in the SiO₂ surface, are recognized as active adsorption sites where hydrogen bonding and ionic interactions can occur with heteroatoms present in asphaltene molecules. It is believed that N-containing molecules adsorb on acidic isolated, or isolated silanols, while solutes with O-containing groups interact preferentially with vicinal, or hydrogen bonded silanols (Li et al. 1994). Also, π - π interactions can occur with siloxane groups once aromatic or hydrophobic molecules with double bonds are close enough to the SiO₂ surface favoring short range interactions such van der Waals intermolecular forces.

A report by Murrell *et al.* (Grenoble, Long, and Murrell 1984) demonstrated the efficient adsorption of basic asphaltenes using mineral sorbents. Along the same

lines, the observed selective retention of vanadyl porphyrins ($\text{N}_4\text{V}_1\text{O}_1$) by SiO_2 was expected. Interactions between SiO_2 and $\text{N}_4\text{V}_1\text{O}_1$ are synergistic and caused by the interaction of siloxane oxygens with the porphyrin π system, hydrogen bonding between the oxo-vanadyl group and silanols, and complexation between siloxane oxygens and vanadium. Takagi *et al.* (Takagi *et al.* 2002) have used these effects for the efficient adsorption of cationic porphyrins on clay, and Osaheni *et al.* (Fyvie *et al.* 2012) reported the use of silica for the removal of vanadium impurities from partially deasphalted crude oil samples.

Figure 0.4. Van Krevelen diagrams for N_1 , N_2 , and N_3 classes in W and WD samples.

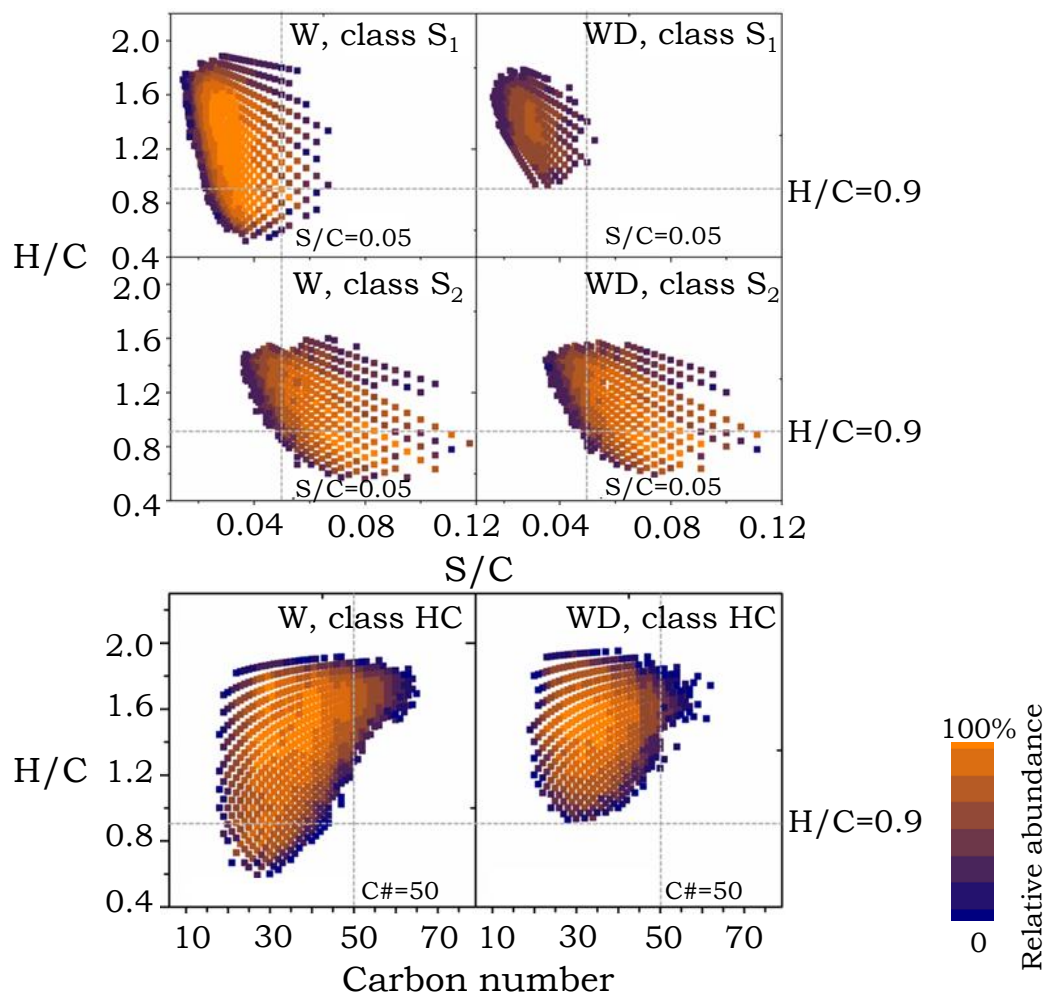


Thus, for a closer compositional examination of Figure 2.2, we used van Krevelen diagrams. As described in Chapter 1, these diagrams result from plotting H/C vs. O/C (or N/C or S/C) atomic ratios, being extremely useful to interpret FT-ICR MS data (Wu *et al.* 2004). In van Krevelen diagrams, homologous series of compounds correspond to diagonal lines intersecting in $\text{H/C}=2$, with alkylation increasing from right to left along the diagonal. Interestingly, compounds not retained by silica, and

common to both W and WD samples, are the HC, N₁, N₃, O₁S₁, S₁, and S₂ classes, among others. No retention of N₁ and N₃ classes is in contradiction with the expected high adsorption of N-containing asphaltenes by mineral surfaces. Figure 2.4, representing the molecular composition of N₁, N₂, and N₃ asphaltenes in W and WD samples, shows compound family N₂ in W strongly retained by the SiO₂ surface and exhibiting an extremely aromatic distribution with the absence of compounds with H/C ratios above 1.4. On the other hand, for N₁ and N₃ compound classes, only structures with H/C ratios above 1.0 are observed in WD. This observation indicates that more aliphatic compounds were extracted from the SiO₂ surface, with the most aromatic fraction of these families remaining adsorbed on the SiO₂, in a fashion similar to that of N₂. It is important to mention that N-containing compounds in W, located below H/C = 1.0, have few methylene units (up to 13 -CH₂ units) while compounds located above H/C = 1.0 have up to 40 methylene units.

On the other hand, no retention of S₁ and S₂ classes is not surprising as sulfur-containing species, due to their high polarizability, are believed to interact weakly with highly polar (less polarizable) species such as silanol groups. For example, Frolov *et al.* (Li *et al.* 1994) found no correlation between sulfur content in asphaltenes and its adsorption on mineral surfaces. Figure 2.5 shows the van Krevelen plots for S₁ and S₂ classes and the H/C vs. carbon number plots for the HC class present both in W and WD samples. Although apparently, from Figure 2.2c, most S₁ and S₂ compounds were recovered from the silica, the absence of signals at the bottom left corner of the van Krevelen diagram for the S₁ class in the WD sample indicates retention of highly aromatic (H/C<0.9) S-containing species with 20–50 carbon atoms (0.02<S/C<0.05). Most of the S-containing compounds retained by SiO₂ correspond to the beginning of the homologous series in the van Krevelen diagram. These compounds have fewer alkyl groups (a maximum of 10 -CH₂- units) than the ones higher up in the same line of the diagram, in the upper left corner, containing up to 40 -CH₂- units, which are not retained.

Figure 0.5. Van Krevelen diagrams for classes S_1 , S_2 , and HC in W and WD.

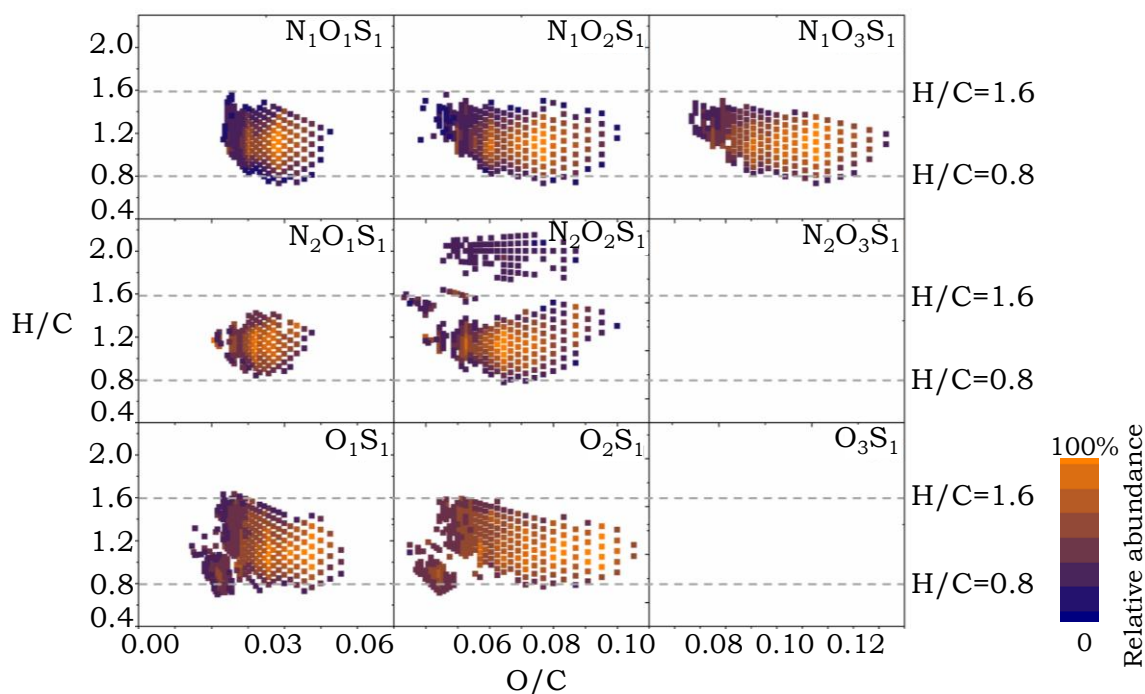


These observations indicate that both low H/C ratios and few methylene units in the structure of the S_1 , N_1 , N_2 and N_3 classes play important roles in compound retention by SiO_2 . Comparison of HC compound class for samples W and WD provides further evidence to support this conclusion. Compositional diagrams for the HC class in Figure 2.5 show aromatic asphaltenes, having 20–45 carbon atoms and $\text{H/C} < 0.9$, missing from the bottom left area of the WD graph, which means selective retention of highly aromatic and sizable compounds on the SiO_2 surface. This observation agrees with previous reports by Acevedo *et al.* (Acevedo *et al.* 1995) concerning strong interactions of highly aromatic asphaltenes with inorganic substrates. In

addition, adsorption tests on kaolinite have shown selective adsorption of highly aromatic asphaltenes with short alkyl chains.(Wang et al. 2013). Also, competitive adsorption of PAHs on mineral surfaces demonstrated a stronger adsorption of a five aromatic ring structure (1,2,3,4-dibenzanthracene) when compared with a two-aromatic ring molecule (naphthalene) (Parida et al. 2006). On the other hand, the composition of the S₂ class does not change significantly from sample W to WD. Examination of the van Krevelen diagrams for this class shows an absence of signals in the bottom left corner for W and WD samples. As discussed earlier, apparently SiO₂ retains mostly molecular species with low H/C and few methylene units, as seen in compound classes N₁, N₃, S₁, and HC. We hypothesize that absence of highly aromatic species in the S₂ compound class translates into no retention by SiO₂.

After sample elution on the TLC plate, HRMS analysis of the fractions (T, DM, and NE) resulted in the observation of 12 compound classes already identified in the whole asphaltene sample (HC, N₁, N₃, N₁O₁, N₂O₁, N₁O₁S₁, N₁S₁, O₁, O₁S₁, O₁S₂, O₂S₁, and S₁) and of 12 new families (N₁O₂, N₁O₃, N₂O₂, N₂O₃, N₃O₁, N₃O₂, N₁O₂S₁, N₁O₃S₁, N₂O₁S₁, N₂O₂S₁, O₂, and O₃) not detected by direct APPI analysis of W. Soft ionization techniques, such as APPI and ESI, are routinely used for the detailed characterization of crude oils, asphaltenes, and bitumen by FT-ICR MS (Islam et al. 2012). Although resolving powers >500,000 allow observation of thousands of molecules in petrochemical samples, resolution alone is not enough for complete sample characterization. For instance, Cho *et al.* (Cho et al. 2012) performing HRMS analysis of the SARA fractions from crude oil, found several compound classes not observed in the whole crude oil sample. This fact suggests fractionation as a tool for a complete molecular characterization of complex mixtures.

Figure 0.6. Van Krevelen diagrams for N_nO_oS_s and O_oS_s classes in the fraction T.



The observation of twelve new compound classes implies either access to low ionization efficiency or low concentration compounds in the asphaltene sample upon fractionation. Separation may cause two effects: concentration of low abundance compounds present in the original sample or elimination of high ionization efficiency compounds masking low ionization efficiency ones (Cho et al. 2011). Starting with the toluene eluted fraction (T), we observe in Figure 2.6 a wide variety of $N_nO_oS_1$ compound classes, four of which, out of the ten observed, are absent in the whole asphaltene sample. Sample DM, the fraction eluted with $CH_2Cl_2/MeOH$, exhibits seven compound families common to W and two unique compound classes N_3O_o ($o=2,3$). For the non-eluted fraction (NE), we observed eleven compound families, six of which are not present in the W sample; this fraction is mostly made up of N_nO_o and O_o compound classes.

Close examination of the T fraction, with a R_f of 0.90 signaling weak sample interaction with silica and good affinity for the eluting solvent (toluene), shows the presence of HC, $N_1O_1S_1$, $N_1O_2S_1$, $N_1O_3S_1$, $N_2O_2S_1$, $N_2O_1S_1$, N_1S_1 , O_1S_1 , O_2S_1 , and

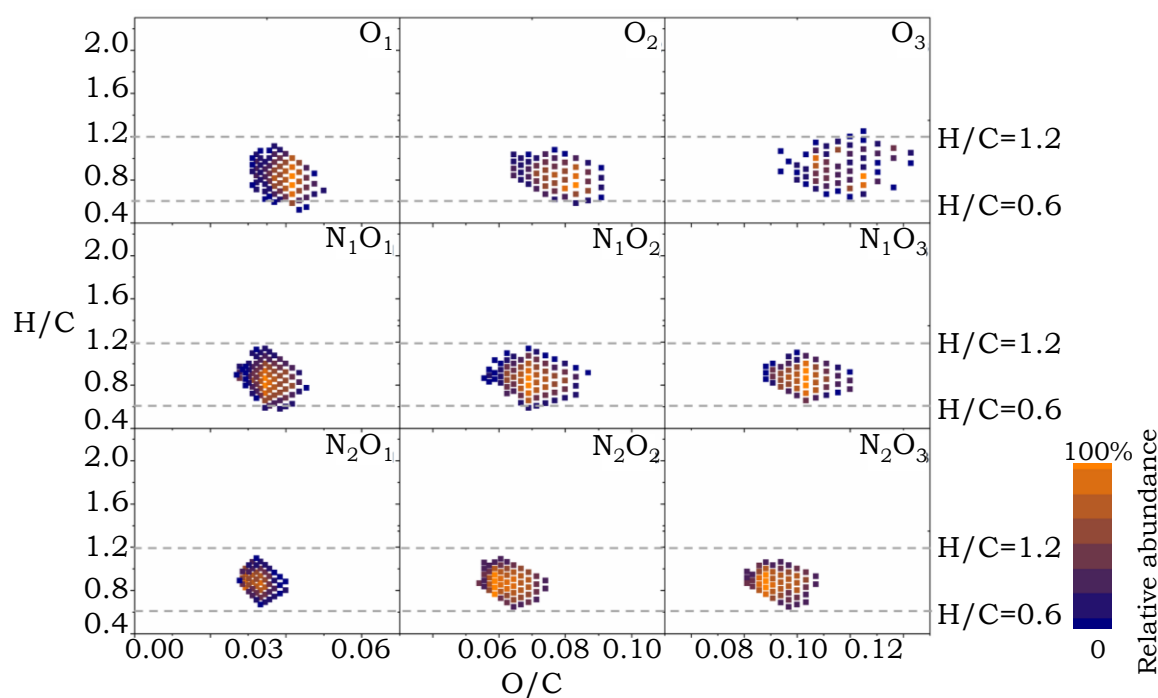
S₁ classes. The low affinity of sulfur functional groups toward silica surfaces could be explained from two points of view. First, being more polarizable than nitrogen or oxygen, sulfur interaction with the silica (through hydrogen bonds) is weaker. Second, sulfur may be present in functional groups having significant steric effects (i.e., thioether) which could make the adsorption processes difficult. However, it is puzzling that the high abundance of O and N atoms in the T fraction appears not to have any bearing on the interactions with the SiO₂ surface, as they apparently do in the case of the NE fraction.

Comparison of the van Krevelen diagrams for the N_nO_oS_s and O_oS_s compound classes in the T fraction, with the N_nO_o and O_o classes in the NE fraction (Figures 2.6 and 2.7) sheds some light on this issue. Variations in the H/C ratio show that the T fraction is more saturated than the NE fraction. As an example N_nO_oS_s compounds in the fraction T exhibit H/C ratios between 0.8 and 1.6, indicating low aromaticity (Figure 2.6), while the analogous series of N_nO_o families, in the NE fraction, are more aromatic having H/C ratios between 0.6 and 1.2 (Figure 2.6). Additionally, from Figures 2.6 and 2.7 it is possible to draw information about the number of methylene units in the O_o, N_nO_o and N_nO_oS₁ compound classes in the fractions NE and T. Considering the beginning of each diagonal line in the van Krevelen diagram (bottom right) as the most aromatic compound of the series, each point in the diagonal indicates the addition of one methylene unit to the core structure. Hence, the number of points in the diagonal gives us an approximate value of compound alkylation. Clearly, compounds in the T fraction have homologous series with up to 25 methylene units, while compounds in the NE fraction have a maximum of 11 methylene units in their structures. An increase in alkylation, and hence in steric hindrance, seems to be responsible for the lack of interactions between compounds in the T fraction and the SiO₂ support, despite its high N and O content and the presence of aromatic cores. To the extent of our knowledge, this is the first observation regarding the effect of steric hindrance on the interaction between real asphaltene samples and inorganic surfaces. Reports by Vela *et al.* (Vela *et al.* 1995)

regarding the TLC-FID analysis of deasphalted heavy oil refer to the toluene-eluted fraction as the alkyl-aromatic portion of a complex hydrocarbon mixture. In fact, our observations indicate that the T fraction is made up of alkyl-aromatic structures not exclusively containing H and C but also containing N, O, and S heteroatoms. To this point, we can hypothesize that asphaltene molecules with heteroatoms and aromatic cores containing few alkyl substituents (low steric hindrances) are more likely to be strongly retained by the SiO₂ support. This adsorption process takes place through various kinds of intermolecular forces. On the other hand, species with a higher number of alkyl units in their structures (large steric hindrances), appear to have more affinity for the eluting solvent. Molecules in the NE fraction fulfill the former description while molecules in the T fraction match the later. In this regard, Larter *et al.* (Li et al. 1992) reported adsorption of alkylcarbazoles on silica as strongly dependent on steric hindrance around the pyrrolic nitrogen. Also, Adams (Adams 2014) pointed out that difficulty in proposing universal correlations between asphaltene composition (elemental and acid–base data) and adsorption behavior is due to the lack of information concerning the surrounding environment of asphaltene active sites (i.e., steric hindrance).

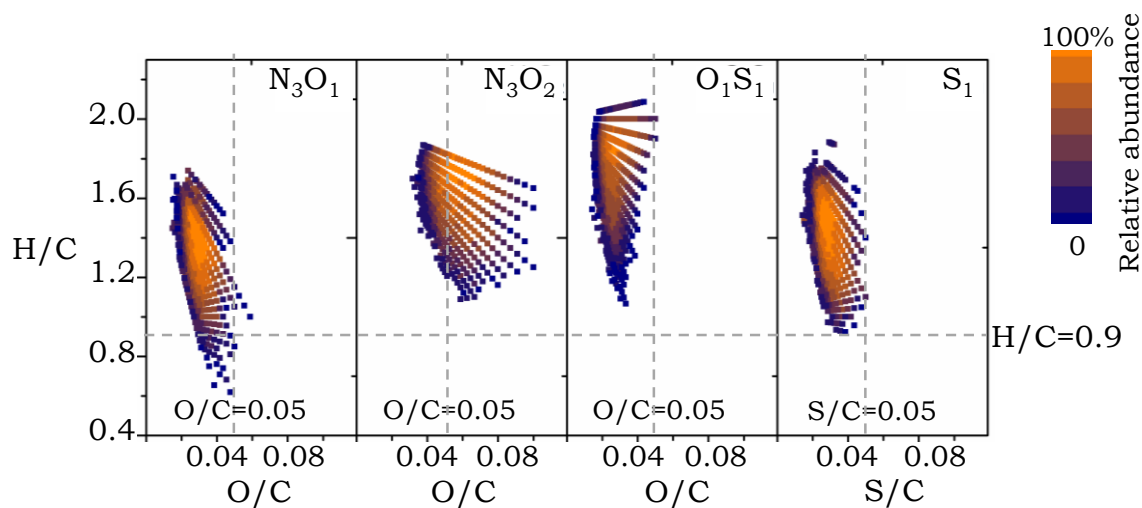
On the other hand, N_nO₀S₁ compounds in the T fraction are not detected by direct analysis of the whole asphaltene sample, and only fractionation gives a way to detect them, with high abundances, in these samples. It is found only one work by Rodgers *et al.* (Purcell et al. 2010) reporting O₁S₁, O₂S₁, N₁O₁S₁, N₁O₂S₁, and N₂O₁S₁ compound classes in asphaltenes extracted from an Iraqi vacuum residue, with low relative abundance <5%. In our case, the relative abundances of these compounds range from 5 to 20%. Finally, although the O₁S₁ compound class is present in all fractions —except the NE, in the fraction T the molecular characteristics of the O₁S₁ family are similar to those of the other compound classes eluted in this fraction (lower left Figure 2.6).

Figure 0.7. Van Krevelen diagrams for O₀ and N_nO₀ classes in the fraction NE.



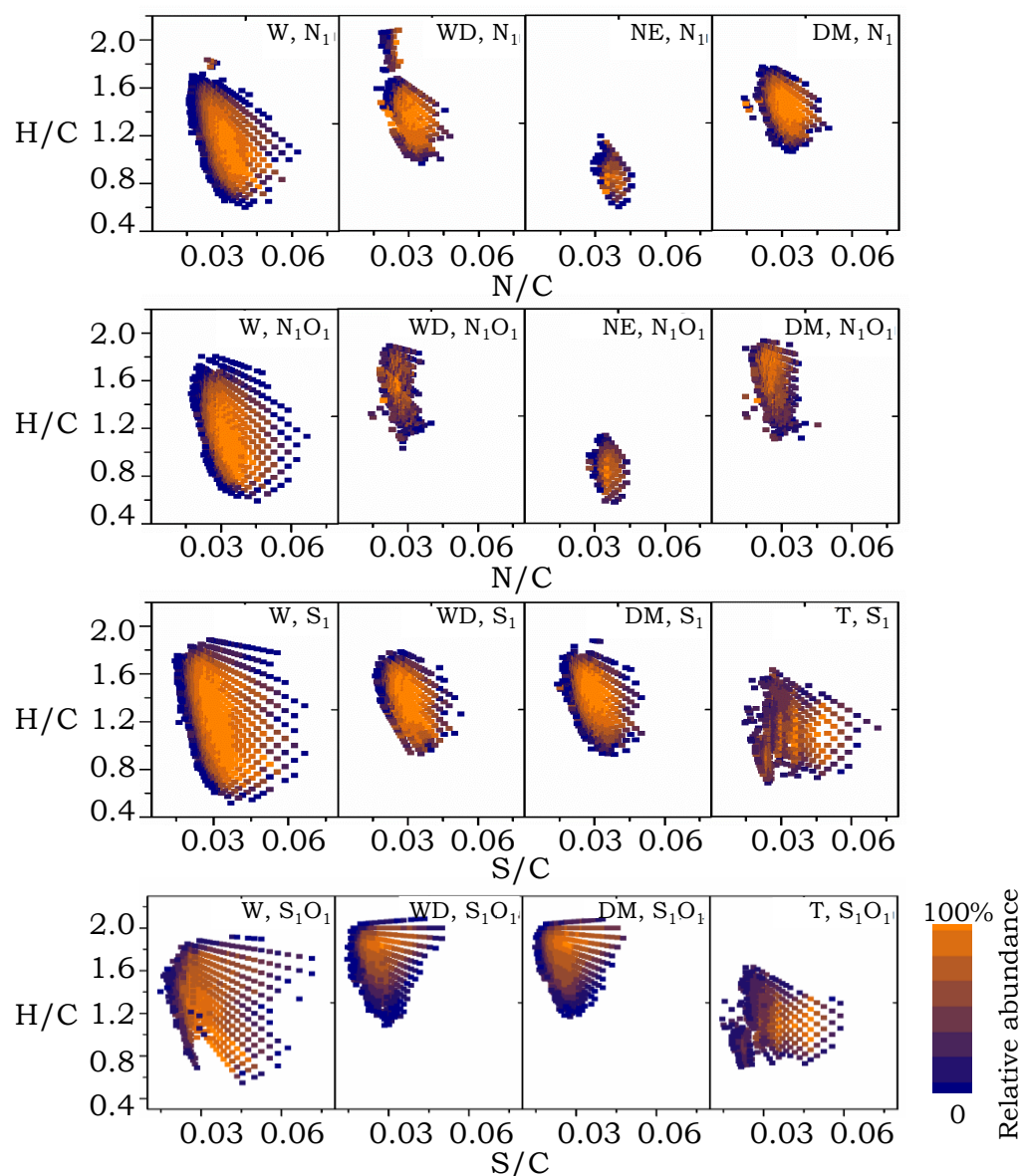
The non-eluted asphaltene fraction (NE), corresponding to compounds with high affinity to SiO_2 , has a molecular composition of HC, N_1 , N_1O_1 , N_1O_2 , N_1O_3 , N_2O_1 , N_2O_2 , N_2O_3 , O_1 , O_2 , and O_3 classes. Figure 2.7 shows van Krevelen diagrams for O_0 , N_1O_0 , and N_2O_0 compound classes found in the NE fraction where, in contrast to the T fraction, H/C ratios are relatively low ($0.6 < \text{H/C} < 1.2$) indicating lower content of methylene units (up to 11 CH_2 units) and higher insaturation, and therefore minimal steric hindrance. As discussed before, less alkylation might translate into more efficient interactions between nitrogen, oxygen, and aromatic moieties with the SiO_2 surface through hydrogen bonding and π - π interactions, respectively. Along the same lines, the class HC in the fraction NE is composed of highly aromatic compounds with H/C ratios between 0.4 and 1.0 and low methylene content (up to 11 CH_2 units). These HC molecules are strongly retained by the SiO_2 surface due perhaps to non-classic hydrogen bonds between aromatic moieties and silanols and van der Waals interactions with siloxanes.

Figure 0.8. Van Krevelen diagrams for N_3O_1 , N_3O_2 , O_1S_1 , and S_1 in the DM fraction.



For the DM fraction, with $R_f=0.69$, we observed nine compound classes: HC, N_1 , N_3 , N_1O_1 , N_3O_1 , N_3O_2 , O_1S_1 , O_1S_2 , and S_1 . Several features of the compounds found in the DM fraction (Figure 2.8) are evident when contrasted with the T and NE fractions. Molecules in the DM fraction cluster in the upper left corner of the van Krevelen diagrams indicating the lowest aromaticity, highest number of methylene units (up to 35), and highest carbon content of all of the asphaltene fractions. At first glance, this observation seems to be contrary to our initial conclusion regarding a large number of alkyl groups and steric effects as being responsible for the weak interaction between the T fraction and the SiO_2 support. However, in the case of the DM fraction we have to consider the nature and polarity of the elution solvent. The mixture $CH_2Cl_2/MeOH$ has the highest polarity of the eluotropic series, besides being the only protic solvent used for asphaltene elution. A protic solvent, capable of interacting with the silica surface through hydrogen bonding, can form a layer on the SiO_2 and displace polar components in the asphaltene interacting with the silica exclusively through hydrogen bonding (Badjic and Kostic 2000).

Figure 0.9. Van Krevelen diagrams for common classes to W, WD, NE, DE, and T samples.

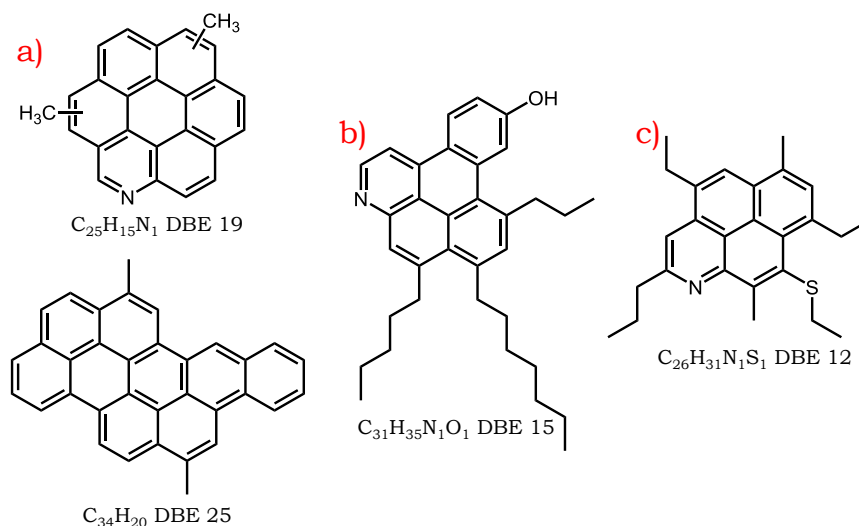


In this sense, we could argue that polar classes such as N_3O_1 , N_3O_2 , and O_1S_1 must have a high tendency to interact through hydrogen bonds, which is possible only if the active sites are accessible. Considering that S atoms may be present in the form of thioether groups with steric hindrance, N- and O-containing functionalities must be accessible to form hydrogen bonds with the silica. Additionally, we believe that the aromatic moieties of the molecules composing the DM fraction are also exposed so that they can interact with silanol groups through weak hydrogen bonds. Many authors have reported non-classic hydrogen bonding between aromatic moieties

and silica surfaces (Badjić and Kostić 2001). Figures 2.9 show the molecular composition of common classes in W, WD, NE, DM, and T, and supports the previous observations.

Finally, with an illustrative purpose, Figure 2.10 displays proposed molecular structures as representative of each fraction. Besides reflecting all the observations described above, these structures are in agreement with the elemental composition of each fraction and meet some reports on proposed structures from theoretical calculations and ms/ms analysis (Ruiz-Morales 2002).

Figure 0.10. Model compounds for a) non-eluted, b) DCM/MeOH eluted, and c) Toluene eluted asphaltenes.



2.5 CONCLUSIONS

Along this chapter, it was demonstrated that combination of normal-phase high-performance thin layer chromatography with FT-ICR mass spectrometry is a powerful tool to understand the molecular basis of asphaltene adsorption on inorganic surfaces. In agreement with previous reports, the presence of specific functional groups (N, O, and metal complexes) confers asphaltic compounds high affinity for the SiO_2 surface. However, a proper description of asphaltene–mineral

surface interactions requires additional consideration of molecular features such as H/C ratio (or aromaticity), the number of methylene units in the structure (or steric hindrance), and functional group accessibility. Van Krevelen diagrams clearly show that strongly retained (non-eluted) compounds have low H/C ratios (<0.9) and few alkyl units (<11 $-\text{CH}_2$ units) in their structure; these characteristics allow for a variety of cumulative short and long range bonding interactions between asphaltenes and silica. These molecules belong to compound classes such as N_1 , N_1O_1 , N_1O_2 , N_1O_3 , N_2O_1 , N_2O_2 , N_2O_3 , O_1 , O_2 , and O_3 .

In contrast, weakly retained (toluene-eluted) compounds exhibit H/C ratios between 0.8 and 1.6, and a relatively high number of alkyl units in their structure (with homologous series having up to 25 methylene units). Heteroatoms interaction with the silica surface in this fraction seems to be hindered by sterics, and bonding with silica is probably limited to weak π – π interactions easily disrupted by the aromatic nature of the solvent. These molecules belong to compound classes such as $\text{N}_1\text{O}_1\text{S}_1$, $\text{N}_1\text{O}_2\text{S}_1$, $\text{N}_1\text{O}_3\text{S}_1$, $\text{N}_2\text{O}_1\text{S}_1$, $\text{N}_2\text{O}_2\text{S}_1$, O_1S_1 , and O_2S_1 .

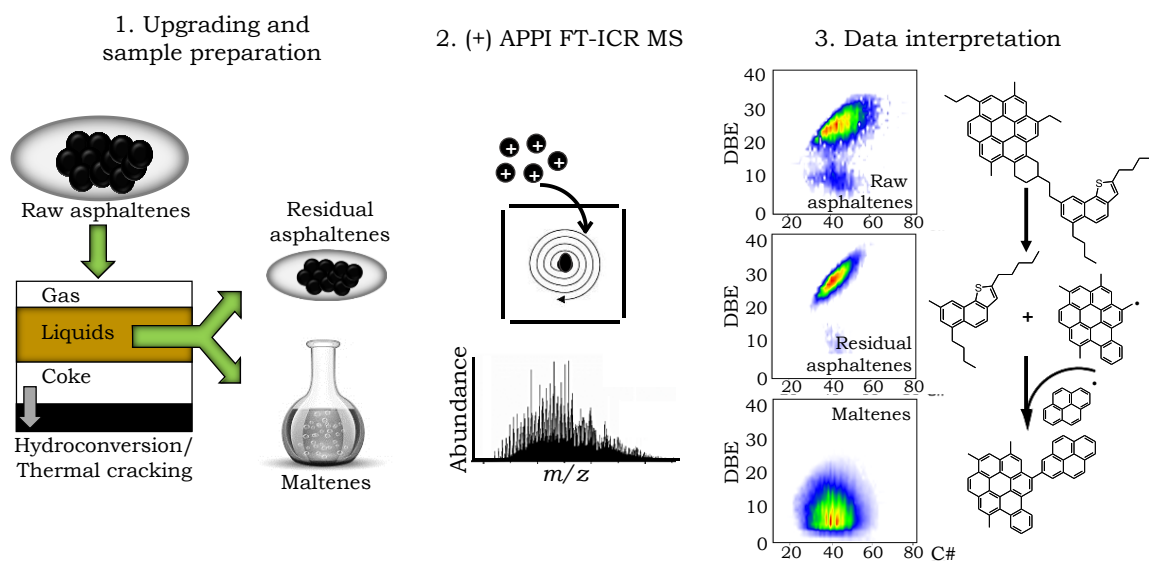
Finally, although the polar $\text{CH}_2\text{Cl}_2/\text{MeOH}$ -eluted fraction, which is composed of compound classes HC , N_1 , N_3 , N_1O_1 , N_3O_1 , N_3O_2 , O_1S_1 , O_1S_2 , and S_1 , exhibits the highest H/C ratios, and abundant alkylation (up to 35 methylene units). Its interactions with silica do not seem to be hindered by sterics. We hypothesize that compounds in this fraction interact mostly through hydrogen bonding between readily accessible functional groups and the silica surface and that these interactions can be disrupted to some extent by the protic nature of the solvent mixture used as eluent.

Finally, it is important to point out that deep understanding of asphaltene adsorption and aggregation processes require a deeper knowledge of molecular structure; this information can be effectively provided by selective sample fractionation combined with high-resolution mass spectrometry as demonstrated in this contribution.

3. TRACING THE COMPOSITIONAL CHANGES OF ASPHALTENES AFTER HYDROCONVERSION AND THERMAL CRACKING PROCESSES BY HIGH-RESOLUTION MASS SPECTROMETRY”

**Redrafted from Martha L. Chacón-Patiño, Cristian Blanco-Tirado, Jorge A. Orrego-Ruiz, Andrea Gómez-Escudero, and Marianny Y. Combariza; Energy Fuels 2015, 29, 6330–6341*

Figure 0.1. Graphical abstract for “Tracing the Compositional Changes of Asphaltenes after Hydroconversion and Thermal Cracking Processes by “High-Resolution Mass Spectrometry”



3.1 ABSTRACT

With heavy crude oil refining on the rise, upgrading strategies are fundamental to yield high-value products. Hydroconversion and thermal cracking are well-established and widely used upgrading processes for heavy oils' distillation cuts and

residues. Recognizing molecular changes in these fractions after upgrading, particularly of asphaltenic compounds, is fundamental to understand and optimize the processes. In this work, it is presented the tracking of the compositional changes in the asphaltene fraction of a Colombian heavy oil, after hydroconversion and thermal cracking, using high-resolution mass spectrometry. The liquid products from the upgrading processes were fractionated into maltenes and residual asphaltenes, and it was established that between 33% and 38% of the original feedstock is transformed into maltenic products. Contoured plots of double bond equivalents versus carbon number and van Krevelen diagrams show maltenic fractions exhibiting lower aromaticity, smaller molecular size, fewer heteroatomic species, and higher content of alkyl side chains than the starting asphaltenic material. Residual asphaltenes, on the other hand, consist of compounds with lower H/C ratios and reduced content of alkyl groups than the feedstock. Also, structural information about the feedstock, such as archipelago or island structures, can be derived from the plots. This information is useful to establish trends between compound class reactivity and the suitability to produce valuable maltenic compounds through upgrading technologies.

3.2 INTRODUCTION

Nowadays, with light crude oil reserves around the globe dramatically depleted, the petrochemical industry's attention is rapidly shifting to nonconventional hydrocarbons, such as heavy and extra-heavy oils and bitumens (Carbognani and Orea 1999; Corilo, Rowland, and Rodgers 2016). Refining strategies for these highly complex hydrocarbon mixtures involve a traditional atmospheric or vacuum distillation, followed by the upgrading of the residues by catalytic and non-catalytic processes (Gray, Khorasheh, and Wanke 1992). Hydroconversion, which is an example of the former, and thermal cracking, which is a representative of the latter, are well-known as upgrading alternatives widely used for synthetic petroleum

production (Jansen et al. 2014). Hydrogenation, deoxidization, denitrogenation, and desulfurization of polynuclear aromatics and heteroaromatic compounds occur simultaneously in hydroconversion processes while heavy metals such as nickel and vanadium are removed (Wiehe 2008). In thermal cracking processes, primary reactions involve thermolysis of aromatic-alkyl bonds, to produce light hydrocarbons, whereas secondary reactions, such as dehydrogenation of naphthenic moieties and condensation of aromatic units, produce coke precursors (Hurff and Klein 1983).

Macroscopic properties such as API grade, SARA composition, and elemental analysis are not enough to predict ideal operational conditions in upgrading processes, particularly for nonconventional hydrocarbons (Purcell et al. 2010). For this reason, tracking compositional changes in heavy oils before and after the upgrading processes is of fundamental importance to design tailored refining strategies aimed to increase yields of valuable products. However, not every fraction of the heavy oil has an “unpredictable” behavior upon upgrading. Among the saturated, aromatic, resin, and asphaltene fractions present in a crude oil, the latter has become increasingly abundant and is considered the most problematic during upstream and downstream stages (Purcell et al. 2010).

Operationally defined by solubility, asphaltenes are the crude oil fraction insoluble in *n*-heptane and soluble in aromatic solvents such as toluene or benzene (Schwager, Lee, and Yen 1977). These complex mixtures have molecular structures consisting mostly of pericondensed aromatic rings with heteroatoms (N, O, and S) present in the form of functional groups such as thiophene, sulfide, sulfoxide, hydroxyl, carbonyl, carboxyl, pyrrole, and pyridine, among others (Gray et al. 2011; Mullins et al. 2012). The asphaltenic fraction of a crude is also associated with high concentrations of heavy metals such as V and Ni, which are commonly found in the form of metal complexes as petroporphyrins (Ali et al. 1993a; Dunn et al. 2003). These molecular features promote the self-aggregation of asphaltenes, which is a

behavior restricting comprehensive structural characterization of these molecules. Recently, a supramolecular model for asphaltene aggregation has been proposed by Gray *et al.* (Gray *et al.* 2011), in which intermolecular forces such as hydrogen bonding, acid–base interactions, metal coordination, and π – π stacking play a role in producing highly stable asphaltene aggregates. Generally speaking, asphaltene aggregation causes damage in crude oil reservoirs, pipeline deposits in transport systems, and challenging situations in the refinery (Juyal *et al.* 2005). Also, because of their high heavy metal and sulfur contents, and their great potential to produce coke, asphaltenes are known as the “bad guys” in upgrading processes, particularly of catalytic and thermal nature.

Several analytical approaches, involving nuclear magnetic resonance (NMR), X-ray diffraction (XRD), and infrared spectroscopy (IR), have been used to follow the molecular modification of asphaltenes upon upgrading. For example, Ancheyta *et al.* (Trejo, Ancheyta, and Rana 2009), who used NMR to study molecular changes in Maya asphaltenes after hydrotreatment, reported the production of lighter asphaltenes with reduced sulfur content and increased aromaticity. Along the same lines, Merdrignac *et al.* (Merdrignac, A. Quoineaude, and Gauthier 2006) found that structural evolution during hydroconversion is marked by a decrease in asphaltene size and an increase in aromaticity, as a result of molecular dealkylation. On the other hand, Liu *et al.* (Liu *et al.* 2013) by combining XRD, NMR, and IR, determined that hydrocracking causes a decrease in aromatic and alkyl carbon content in Venezuelan asphaltenes while the amount of naphthenic carbon increases. Although these approaches provide useful chemical information, a detailed compositional picture of asphaltene evolution is still not available.

Recently, atmospheric pressure photoionization coupled with high-resolution Fourier transform ion cyclotron mass spectrometry (APPI FT-ICR-MS) has emerged as a robust and reliable technique for molecular characterization of complex mixtures such as asphaltenes and vacuum residues (Robb *et al.* 2000; Tachon *et al.* 2011).

In this regard, McKenna *et al.* (McKenna *et al.* 2013), using APPI FT-ICR-MS to study asphaltene's molecular composition were able to resolve more than 30,000 ion signals in a single sample, defining the asphaltene compositional space. Unlike spectroscopic techniques, which reflect average properties such as aromaticity or alkyl side-chain content, high-resolution mass spectrometry provides a detailed view of each accessible compound in a complex sample. Only the high resolving power and the mass accuracy offered by FT-ICR MS allow the assignment of elemental compositions to detected ions in mixtures having up to 50,000 different chemical species.³³ To the best of our knowledge, there is only one report dealing with the use of ultrahigh-resolution mass spectrometry for the thorough analysis of asphaltene's compositional changes after upgrading. Purcell *et al.* (Purcell *et al.* 2010), used APPI FT-ICR MS to follow the transformation of Iraqi asphaltenes after hydroconversion. The authors found not only that the upgrading process increases asphaltene aromaticity, as it has been reported by using other analytical techniques, but also that compound classes of the S_x type are transformed into S_{x-1} classes in the products.

Use of high-resolution mass spectrometry to track upgrading processes is beneficial from many points of view since it could be used to tune the upgrading process and observe the effects of operational conditions on the molecular features of the products. Besides, in the case of heavy fractions, the technique could also provide some insights into the molecular architecture of complex compounds such as asphaltenes. On this subject, Gray *et al.* (Rueda-Velázquez *et al.* 2013), through analysis of the distillable fraction from the hydrocracking products using gas chromatography field ionization time-of-flight high-resolution mass spectrometry (GC-FD-TOF HR-MS), suggested the presence of basic building blocks, such as 1–4 ring naphthenes, 1–3 ring aromatics, mono- and dibenzothiophenes, among others, in asphaltenes from different geological origins.

In this contribution, the asphaltene fraction from a heavy Colombian crude oil was subjected to hydroconversion and thermal cracking processes and was analyzed by APPI FT-ICR mass spectrometry to track compositional changes in the products after the upgrading. Liquid products from the thermal process and hydroconversion were fractionated into asphaltenes and maltenes, following the procedure described by the ASTM Standard D6560-12, and were further analyzed by APPI FTICR MS. It was found that the asphaltene feedstock undergoes quite similar chemical transformations with hydroconversion and thermal cracking. For example, the maltenic fraction originated from both processes exhibits lower molecular-weight distributions, higher H/C ratios, a high degree of alkyl substitution, and lower heteroatom content than the starting material. The residual asphaltenes also exhibit lower molecular-weight distributions and lower H/C ratios and degree of alkyl substitution than the original asphaltenes. Also, a retrospective analysis of the upgrading products, particularly the remaining asphaltenes, gave us some insights into the molecular architecture of the parent asphaltenes. In this context, we believe that a detailed description of asphaltene's transformation could guide efficient upgrading strategies that will eventually impact the economic value of heavy crude oils positively.

3.3 EXPERIMENTAL

3.3.1 Sample preparation. Colombian asphaltenes were extracted from a heavy crude (API=12), following the methodology described by ASTM Standard D6560-12. Briefly, 1 g of crude oil was mixed with 40 mL of *n*-C₇ and heated for 40 min under reflux. The mixture was stored in darkness overnight, and the precipitated raw asphaltenes were collected by filtration. Asphaltene cleaning was performed recycling *n*-C₇, in a Soxhlet apparatus, until the solvent was clear. Finally, the clean asphaltenes were recovered by dissolution of the remaining solid in hot toluene, which was subsequently rotoevaporated to produce a clean sample. The raw

asphaltene sample was subjected to hydroconversion and thermal cracking processes in a batch microreactor that was equipped with a fluidized sand bath, under the following conditions: for the thermal cracking process, 4 g of asphaltenes were heated for 20 min at 430 °C under a nitrogen atmosphere at a pressure of 0.69 MPa. For the hydroconversion process, 4 g of the asphaltene sample were mixed with 100 ppm of molybdenum naphthenate and reacted for one hour at 430 °C and 13.7 MPa under a hydrogen atmosphere. After the reactions, the liquid products were separated and kept for mass spectrometric analysis.

3.3.2 Elemental Analysis. Elemental analysis (CHNS) of the raw asphaltenes and the liquid products from hydroconversion and thermal cracking processes were carried out in a FLASH 2000 analyzer (Thermo Scientific, Billerica, MA, USA). The samples (2–3 mg) underwent catalytically combustion at 1200 °C.

3.3.3 Sample Preparation. The liquid products from the hydroconversion and the thermal cracking processes were also subjected to the ASTM Standard D6560-12 method for obtaining the maltenic and the residual asphaltenic fractions. In this fashion, five samples were obtained for mass spectrometric analysis: raw asphaltenes, residual asphaltenes and maltenes from hydroconversion (Asp1 and M1, respectively), and residual asphaltenes and maltenes from thermal cracking (Asp2 and M2, respectively). Figure 1 shows sample evolution and mass balances for each upgrading process. For FT-ICR MS analysis of the raw asphaltenes and their upgrading products, the samples were diluted in toluene to a concentration below 0.30 mg/mL.

3.3.4 APPI (+) FT-ICR Mass Spectrometry

Table 0.1. APPI source conditions for raw asphaltenes, residual asphaltenes and produced maltenes.

Parameter	Raw asphaltenes	Samples Asp1 and Asp2	Samples M1 and M2
Concentration (mg/mL)	0.20	0.20	0.10
Flow (μL/hrs.)	300	300	200
Capillary voltage (V)	-2200	-2200	-2200
End plate offset (V)	-800	-800	-800
Vaporization temperature (°C)	350	350	220
Transfer capillary temperature (°C)	250	250	210
Nebulizer gas pressure (Bar)	2.2	2.2	1.0
Drying flow (L/min)	3.0	3.0	2.5
Ion accumulation time (ms)	300	400	30

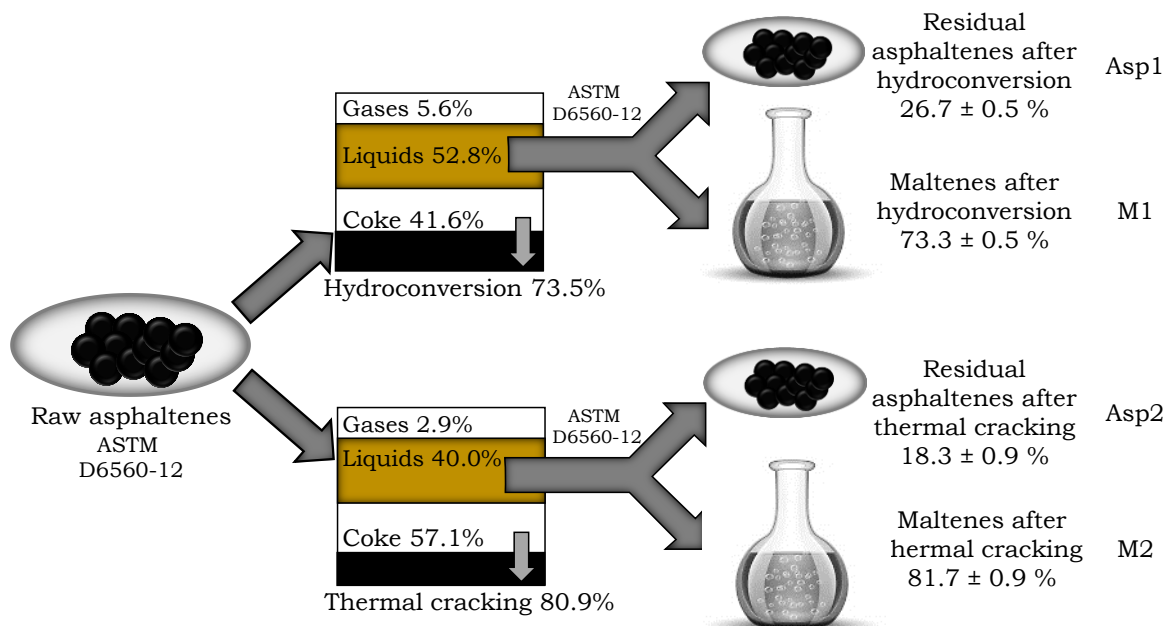
Mass spectrometric analysis was performed on a FT-ICR mass spectrometer (SolariX 15 T, Bruker Daltonics, Billerica, MA) equipped with an APPI source, operated in positive (+) mode, fitted with a Krypton photoionization lamp (10.6 eV). Each spectrum was recorded by the accumulation of 100 scans of time-domain transient signals in four mega-point time-domain data sets. The front and back trapping voltages in the ion cyclotron cell were set at +0.70 V and +0.50 V, respectively. The ICR cell was calibrated using a NaTFA solution (m/z from 200 to 1200). All mass spectra were internally recalibrated using a homologous series of alkylated compounds. A resolving power higher than 550,000 at m/z 400.0000 for all mass spectra, and a mass accuracy below 1.0 ppm provided unambiguous molecular formula assignments for singly charged ions, either as radical cations $M^{\bullet+}$ or protonated molecules $[M+H]^+$, with a relative abundance above 0.5%. Mass spectra were processed using the Composer software 1.0.6 (Sierra Analytics, Modesto, CA, USA). Detailed APPI source conditions for each sample and other instrumental parameters are provided in Table 3.1.

3.4 RESULTS AND DISCUSSION

Hydroconversion and thermal cracking are well-studied and widely used as upgrading technologies. Also, because of their high potential to produce valuable high-quality products from heavy fractions, they are also attractive for the molecular modification of asphaltenes (Ancheyta et al. 2003). Figure 3.2 includes mass balances for the upgrading processes, as well as catalytic hydroconversion and thermal conversion percentages. At first glance, it seems that the asphaltene feedstock behaves similarly in both processes, particularly regarding liquid and coke yields. However, fractionation of the liquids according to the procedure described by ASTM Standard D6560-12, results in different amounts of residual asphaltenes and produced maltenes. In the case of hydroconversion, 73.3% of the liquids are maltenes, according to the n -C₇ solubility definition, while after thermal cracking maltenes correspond to 81.7% of the liquid products (Figure 1). Thus, the net yield of maltenes in hydroconversion and thermal cracking were ~38% and ~33%, respectively. Our results indicate that both processes have the potential to produce valuable products from Colombian asphaltenes, with hydroconversion yielding more useful products. On this subject, Savage *et al.* (Savage, Klein, and Kukes 1985) have reported maltene yields between 20% and 35% from hydrocracking of North American asphaltenes, and found that the percentage of residual asphaltenes increases from 30% to 45% as the temperature increases from 350 °C to 450 °C. In this study, the liquid products from thermal cracking and hydroconversion, contain ~18% and ~27% of n -C₇ of insoluble material or residual asphaltenes, respectively.

In this work, it was found 48.4% and 51.9% of coke yield for hydroconversion and thermal cracking respectively, which is in agreement with the literature. For example, Karimi *et al.*³⁶ observed a conversion of ~50% of an asphaltene feedstock to coke under thermal cracking conditions. In another work, Gray *et al.* (Gray et al. 1992) reported minimal coke production (~10%) from hydrocracking asphaltenes when using a hydrogen-donor solvent and an iron-based catalyst.

Figure 0.2. Sample evolution and mass balances for a Colombian asphaltene sample exposed to hydroconversion and thermal cracking upgrading process.



Elemental composition of raw asphaltenes and the liquid products from each upgrading process are shown in Table 3.2. Conversion is characterized by an increase in H/C ratios, with thermal cracking yielding more saturated liquid products than hydroconversion. Also, both upgrading processes generate liquid products with reduced sulfur content, when compared with the raw asphaltenes. Similarly, Purcell *et al.* (Purcell *et al.* 2010) reported a decrease in sulfur content in the residual asphaltenes after hydroconversion and Liu *et al.* (Liu *et al.* 2013) observed an increase in H/C ratios and a reduction in sulfur and nitrogen contents for hydrocracked Venezuelan asphaltenes. On the other hand, the elemental analysis of the unfractionated liquid products revealed nonsignificant changes in nitrogen content. It is well-known that yields of coke, gasses, residual asphaltenes and maltenes in upgrading processes are a direct consequence of the feedstock composition and the reaction environment (Savage, Klein, and Kukes 1988).

Although many analytical approaches have been used to trace feedstock fate and upgrading products composition, the complexity of these samples prevents in-depth compositional analysis by traditional spectroscopic methods.

Table 0.2. Elemental Composition of Raw Asphaltenes and Their Hydroconversion and Thermal Cracking Products*

Sample	Raw asphaltenes	Hydroconversion products	liquid	Thermal cracking products	liquid
C (w %)	82.96	82.50		82.42	
H (w %)	7.10	7.98		8.61	
N (w %)	1.03	0.93		0.83	
S (w %)	7.45	6.84		6.08	
H/C	1.02	1.16		1.25	

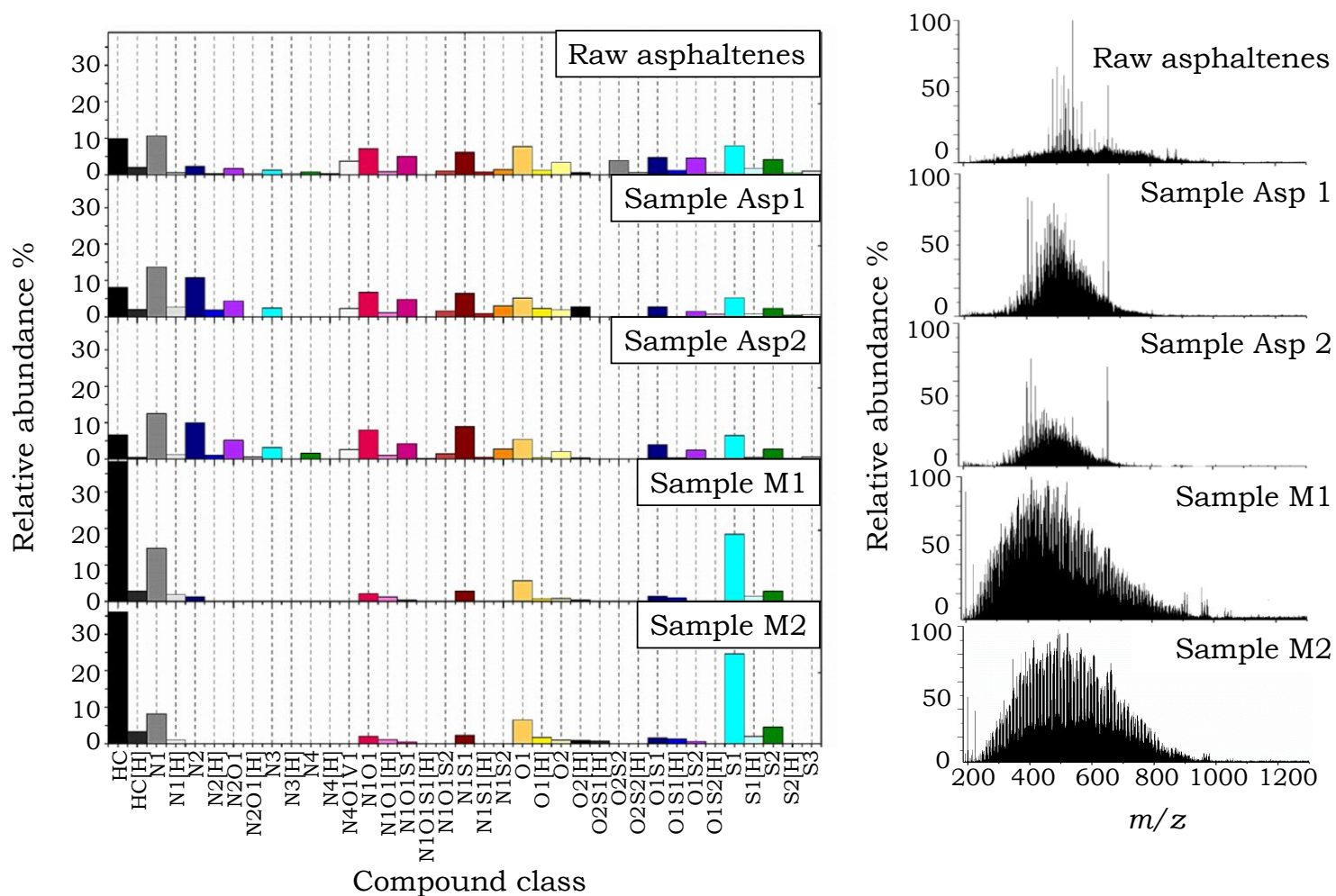
*The absolute errors of the elemental analysis measurements are between 0.02 and 0.04 wt %, and the standard errors of the mean are between 0.02 and 0.09.

Raw and residual asphaltenes (Asp1 and Asp2) and the produced maltene fractions (M1 and M2), from hydroconversion and thermal cracking processes, were analyzed by positive APPI FT-ICR mass spectrometry under optimized conditions. Ion signals in the mass spectra were analyzed with the software Composer, using exact masses and the Kendrick mass defect criteria for elemental composition assignment. Assignment errors fluctuate between 277 and 642 ppb. RMS values below 700 ppb provide unequivocal elemental composition assignment for practically all compound classes (McKenna, Blakney, et al. 2010). However, particular attention should be paid to elemental composition assignments of compound classes with high heteroatom content. For example, if there is more than one elemental composition for a signal, with a mass tolerance below 1 ppm, the assignment can be confirmed by looking at the isotopic fine structure (Stenson, Marshall, and Cooper 2003). Regarding the number of molecular assignments, one can conclude that sample polydispersity decreased after upgrading, because of the low number of elemental compositions found in the maltene fraction, when compared with the starting material. Similarly, Purcell *et al.* (Purcell et al. 2010) reported a decreased number of elemental assignments in Iraqi asphaltenes after hydroconversion.

Figure 3.3 shows the compound class distribution and the mass spectra for the raw asphaltene and the samples Asp1, Asp2, M1, and M2. The molecular weight distributions (MWDs) of maltenes suggest profound transformations of the asphaltene feedstock through hydroconversion and thermal cracking processes. While the feedstock exhibits a MWD along $220 < m/z < 980$, centered around m/z 600, compounds in M1 and M2 samples are shifted to lower masses with MWDs between $200 < m/z < 920$, centered around m/z 420 and m/z 490 for maltenes after hydroconversion and thermal cracking, respectively. Residual asphaltenes also have different mass spectral profiles than the original asphaltenes. Asp1 and Asp2 MWDs are $300 < m/z < 800$, and both are centered on m/z 500.

Sorting molecular assignments in compound classes provides a different vantage point to observe the effect of the upgrading processes on the asphaltene feedstock. Figure 3.3 displays the compound class distribution of the samples, presented as the relative abundance of each class, detected as either, or both, radical cations and protonated molecules.

Figure 0.3. Compound class distributions and MWDs for raw asphaltene and residual asphaltene (Asp1, Asp2) and maltene (M1, M2) after upgrading.



The raw asphaltenes exhibit 21 compound classes: HC, N₁, N₂, N₂O₁, N₃, N₄, N₄O₁V₁, N₁O₁, N₁O₁S₁, N₁O₁S₂, N₁S₁, N₁S₂, O₁, O₂, O₂S₁, O₂S₂, O₁S₁, O₁S₂, S₁, S₂, and S₃. The same trend in compound class distribution and relative abundances is present in the residual asphaltenes (Asp1 and Asp2), except for the classes O₂S₁ and O₂S₂, which are only detected in the raw sample. In contrast, maltenes' composition is very different to that of the raw asphaltenes. After hydroconversion, only 9 compound classes are detected in maltenes: HC, N₁, N₂, N₁O₁, N₁S₁, O₁, O₁S₁, S₁, and S₂. Moreover, maltenes from thermal cracking exhibit the following compound classes: HC, N₁, N₁O₁, N₁S₁, O₁, O₂, O₁S₁, S₁, and S₂.

Interestingly, the class HC, or hydrocarbons with no heteroatoms, is the most abundant (~35%–40%) in maltene fractions M1 and M2, although this compound class has very low abundance in the starting asphaltenic material (~10%). These results indicate the suitability of hydroconversion and thermal cracking processes to yield products enriched with useful hydrocarbons (of the HC kind), from Colombian asphaltenes.

Regarding the molecular composition of produced maltenes after upgrading, the absence of compound classes with high heteroatom content, such as N₂O₁, N₃, N₄, N₁O₁S₂, N₁S₂, O₂S₂, O₁S₂, and S₃, and the prevalence of the classes HC, N₁, N₂, N₁O₁, N₁O₁S₁, N₁S₁, O₁, O₂, O₁S₁, S₁, and S₂ with less heteroatoms could be indicative of nitrogen and sulfur loss from the raw asphaltenes. Similarly, an increase in the relative abundance of the classes HC and S in the maltenic products suggests that heteroatomic classes undergo breakup reactions where some N moieties and S moieties are lost, giving rise to simpler hydrocarbons. Supporting these observations, Purcell *et al.* (Purcell *et al.* 2010) used FT-ICR MS to follow the compositional changes in Iraqi asphaltenes during deep hydroconversion and reported a consecutive loss of S atoms from classes such as S₃ and N₁S₃ to produce S₂ and N₁S₂ classes, respectively. In another work, Karimi *et al.*, (Karimi *et al.* 2011) who performed hydrocracking of heavy residues from Alberta bitumen, concluded

that the conversion of asphaltenes occurs mostly through cracking of long aliphatic chains from the aromatic cores, with the loss of heteroatoms such as sulfur from sulfide-like functionalities.

Regarding sulfur-containing compounds, it is also important to consider that, under thermal cracking and hydroconversion conditions, alkyl sulfides would more likely turn into H₂S and paraffins, than thiophene moieties (Chakhmakhchev and Suzuki 1995; Lu et al. 2014). Consequently, it could be feasible to assume that sulfur-containing families in produced maltenes or residual asphaltenes are most likely present in the form of thiophenic functionalities. In this regard, Hauser *et al.* (Hauser et al. 2014) reported that sulfides in saturates or as side chains in aromatics, resins, and asphaltenes, are more likely to undergo thermal cracking reactions than thiophene moieties. The authors studied vacuum residues reactivity under thermal cracking conditions and suggested that non-thiophenic sulfur is removed from the feedstock and transformed to H₂S, which is detected in the produced gasses in high concentrations (14 wt %). On the other hand, refractory sulfur present in thiophenic functionalities is resistant to conversion and remains concentrated in the residual pitch. Supporting these observations, Jimenez *et al.* (Jiménez Mateos and Fierro 1996) using X-ray photoelectron spectroscopy, studied coke samples from the carbonization reaction of a Maya vacuum residue and found thiophenic sulfur to be the most abundant functionality.

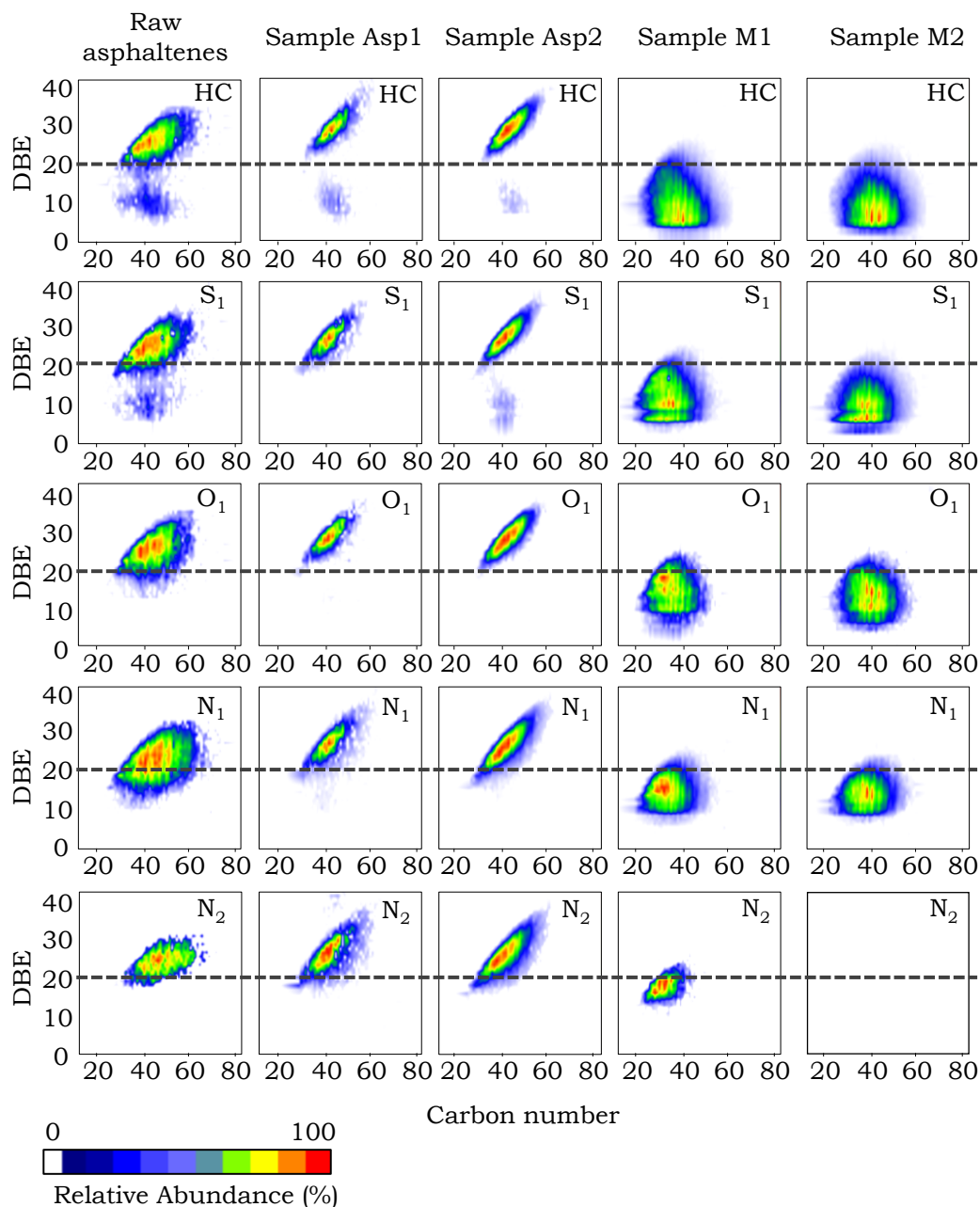
Histograms of compound class distribution in Figure 3.3, although useful as a general way to compare sample composition, do not contain detailed molecular information. Thus data analysis, by precise methods such as isoabundance-contoured plots of double bond equivalents (DBEs) versus carbon number, is useful to follow asphaltenes molecular changes after upgrading (Purcell et al. 2007). Figure 3.4 shows the isoabundance contoured plots of DBE versus carbon number for the compound classes HC, S₁, O₁, N₁, and N₂ for the raw asphaltenes and the residual asphaltenes (Asp1 and Asp2) and maltenes (M1 and M2) obtained after

hydroconversion and thermal cracking. The plots are normalized to the highest signal abundance for each class. Raw asphaltenes are highly aromatic compounds with DBE values ranging from 18 to 33, and carbon numbers between 30 and 65. On the other hand, contoured plots of DBE versus carbon number for samples M1 and M2, the maltenes product of hydroconversion and thermal cracking processes, exhibit significant differences when compared to the original and residual asphaltenes. Maltenes in Figure 3.4 have low DBE values (<22) and carbon numbers between 20 and 60. This distribution is quite similar to that reported by McKenna and co-workers for vacuum distillation cuts (371–510 °C and 510–538 °C) of a Middle Eastern heavy crude oil (McKenna, Blakney, et al. 2010; Podgorski et al. 2013).

Generally speaking, the composition of the residual asphaltenic and maltenic fractions after upgrading reflects the chemical complexity of the starting material. After upgrading, DBE values for residual asphaltenes range from 22 to 37, with carbon numbers from 32 to 56, in contrast with the values for the original sample—DBE from 18 to 33 and carbon numbers from 30 to 65. There are no significant differences between the contoured plots for residual asphaltenes from hydroconversion and thermal cracking processes. The slight increase in DBE in residual asphaltenes after upgrading, with little reduction in carbon number, can be associated with dehydrogenation of peripheral naphthenic rings in the asphaltene core structure, as has been previously reported. For example, Ancheyta *et al.* (Ancheyta, Trejo, and Rana 2010) reported that hydrotreated asphaltenes increase their aromaticity factors, in comparison with the feedstock. Also, similar trends have also been observed by Rodgers *et al* (Purcell et al. 2010).

On the other hand, regarding changes in the molecular structure during upgrading, Savage *et al.* (Savage et al. 1985; Savage and Klein 1988) reported that, below 400 °C, in thermal cracking processes, cleavage of alkyl side chains occurs mainly without altering the asphaltene's core.

Figure 0.4. DBE versus carbon number plots for HC, S₁, O₁, N₁, and N₂ compound classes in raw asphaltenes and residual asphaltenes (Asp1 and Asp2) and maltenes (M1 and M2) after upgrading.

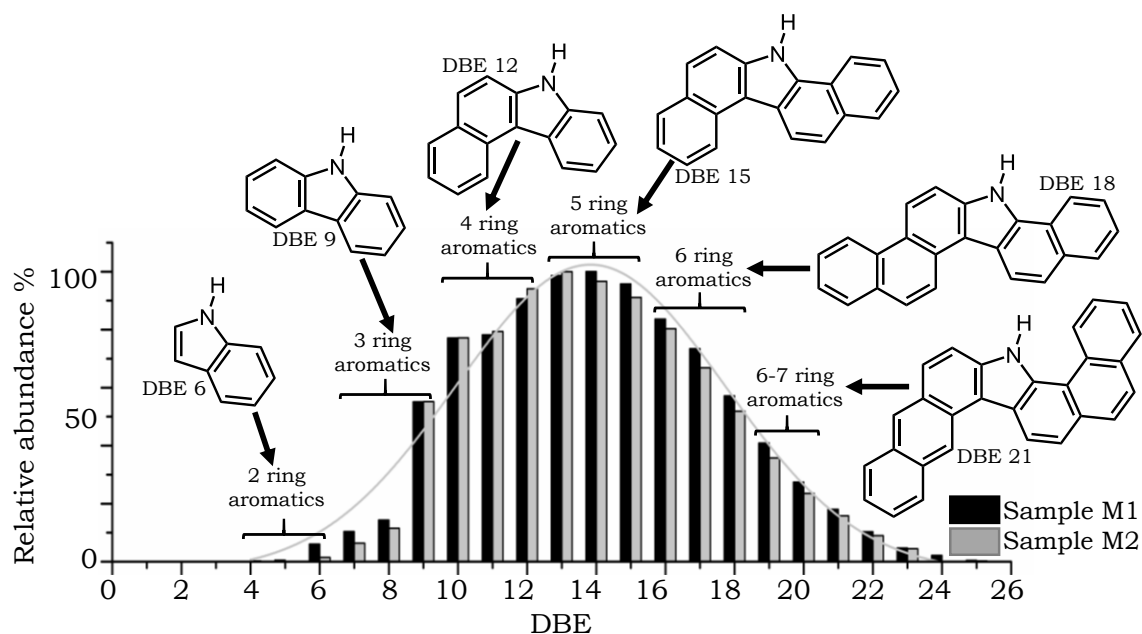


Hence, under the thermal and hydroconversion conditions in the experiments of this work (430 °C), it is possible to hypothesize that conversion of asphaltene's structure

begins with a loss of alkyl side chains with a further modification of the aromatic core architecture. According to Figure 3.4, the original asphaltene sample consists of molecules with aromatic cores containing up to 30 carbon atoms in their alkyl side chains, while residual asphaltenes consist in mostly condensed units with few alkyl groups, where the most abundant homologous series only have up to 18 methylene units in their structures. Plots of DBE *versus* carbon number provide a way to calculate an approximate number of carbon atoms in the alkyl side chains of asphaltenes. For instance, let us consider the molecular species located at the planar limit in the DBE *versus* carbon number plots, which correspond to the beginning of each homologous series. Each point in these homologous series (moving horizontally from left to right) indicates the addition of one carbon atom to the alkyl side chains, attached to the core structure. In other words, raw asphaltenes consist of aromatic cores having at least 30 carbon atoms in the alkyl side chains.

The observation of narrow distributions along the homologous series in the residual asphaltenes in Figure 3.4 suggests a massive loss of alkyl chains from the original asphaltene sample. Along the same lines, Chiaberge *et al.* (Chiaberge *et al.* 2009), using NMR and FTIR, concluded that thermal cracking of asphaltenes proceeds through reactions based on radical fragmentation, producing dealkylation and peripheral naphthenic unit aromatization. In another report, Kawai *et al.* (Kawai and Kumata 1998) presented a mechanism for thermal cracking of heavy oils where asphaltene dealkylation is a consequence of free radicals attacking the asphaltene structure; in this fashion, dealkylation reactions produce asphaltenes with smaller sizes. An alternative reaction pathway involves radical recombination to produce highly condensed asphaltenes, with few alkyl side chains, commonly associated with coke production.

Figure 0.5. DBE distribution for the radical N₁ class in maltenes (M1 and M2) after hydroconversion and thermal cracking processes.



Until this point, it is possible to conclude that residual asphaltenes must consist of highly condensed and very stable “cores,” left after the cleavage of alkyl chains from the original asphaltenes, which can survive the upgrading process. This idea coincides with several reports in the literature depicting asphaltenes as thermally stable polyaromatic molecules with alkyl side chains or peripheral naphthenes. Both alkyl and naphthenic groups in these structures are cracked in thermal or catalytic upgrading while the remaining aromatic cores may undergo further reactions to produce coke (Alvarez et al. 2011). Along these lines, the produced maltenes, essentially the *n*-C₇ soluble fraction of the cracked products must also contain information about the structure of the raw asphaltenes. This train of thought was the basis for a report by Gray *et al.* (Rueda-Velásquez et al. 2013) where analysis by GC-FD-TOF MS of the distillable fraction from hydrocracked asphaltenes showed that basic units in asphaltenes could consist of 1-, 2-, 3-, 4-ring naphthenes, 1-, 2-, 3-ring aromatics with thiophene functionalities, and 4+-ring aromatic units.

In our case, the DBE *versus* carbon number plots for maltenes in Figure 3.4 indicate a significant diversity of building blocks in the starting material. For example, the HC class, which is the most abundant in maltenes, has compounds with DBE values

between 4 and 12 that might be indicative of structures consisting of 1–4-ring aromatics with up to 20 carbon atoms in their alkyl side chains (G. C. Klein et al. 2006).

Regarding the class N₁ in samples the M1 and M2, molecular species with DBE values of 14–15 and carbon numbers between 30 and 36 exhibit the highest abundances. According to Purcell *et al.* (Jeremiah M. Purcell et al. 2007) nitrogen-containing aromatics under APPI conditions can produce radical cations if the N is contained within a pyrrolic moiety, and protonated molecules if the N is pyridinic. We observe a high abundance of radical cations in M1 and M2 samples, which indicate the presence of pyrrolic units. Supporting this assumption, Purcell *et al.* (Jeremiah M. Purcell et al. 2007) reported a high abundance of pyrrolic functionalities with DBE values of 14–15 in South American crude oils analyzed by positive APPI FT-ICR MS and suggested the presence of dibenzocarbazoles in the samples. Along the same lines, the N₁ class in residual asphaltenes is detected as radical cations in high abundance. It is important to note that nitrogen moieties in the residual asphaltic material must be embedded in large and highly condensed aromatic cores with few alkyl side chains. Based on reports, Figure 3.5, which presents the DBE distribution for the class N₁ in samples M1 and M2, also displays several model compositions for N-structures, likely in upgrading products.

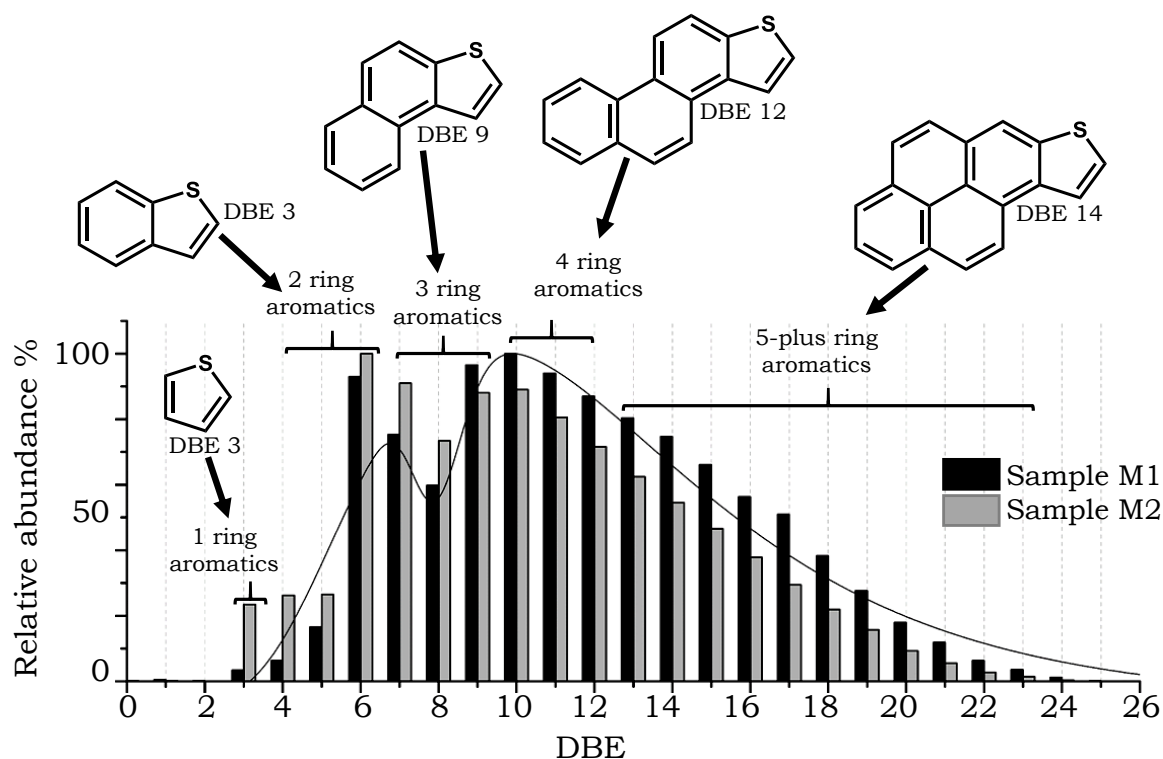
In the case of oxygen, the petroleome of class O₁ in raw asphaltenes spans from DBE=18 to DBE=33, increasing slightly in the residual asphaltenes —from DBE=22 to DBE=35; and carbon numbers in the raw asphaltenes go from 30 to 65, decreasing slightly in the residuals —carbon numbers go from 32 to 55. Mullins (Mullins et al. 2012) has suggested that the oxygen content of asphaltenes is low, in comparison to sulfur, and that O₁ may be present in polar groups such as phenols and furans. In the residual asphaltenes from both upgrading processes, the O₁ class presents a molecular composition with DBE values slightly increased and reduced number of methylene units, when compared with the starting material. On the other

hand, the low aromaticity of the O₁ class in produced maltenes contrasts with the high aromaticity of the asphaltene feedstock. In maltenes, DBE values for the O₁ class range from 8 to 22 with alkylation up to 22 methylene units, suggesting a broad variety of fundamental units in the starting material.

The N₂ class in residual asphaltenes presents a slight increase in DBE values and lower content of methylene units than the raw asphaltenes. Nitrogen in residual asphaltenes must be present as pyrrolic or pyridinic moieties within the aromatic core structure. On the other hand, N₂ class in produced maltenes after hydroconversion, exhibits a narrow molecular distribution with DBE values between 12 and 22 and a methylene content of up to 18 units.

One of the main concerns in upgrading schemes is related to sulfur content in the products, which not only affects the fuel price but also has an adverse impact on the atmosphere, where it is released as SO_x compounds after combustion. The S₁ compound class in maltenes from hydroconversion and thermal cracking displays a bimodal distribution, as seen in Figure 3.4. The plot of the DBE distribution for this compound class in Figure 3.6 clearly shows two series. The first homologous series has a maximum at DBE=6, which extends along carbon numbers 20 to 54 while the second series presents a maximum at DBE=10 and spans from carbon numbers 18 to 56. Regarding sulfur molecules in products from hydrocracked asphaltenes, Gray *et al.* (Rueda-Velásquez et al. 2013), using gas chromatography coupled to FI-TOF-MS, found that cracked sulfur-containing products consist mostly of 2-ring and 3-ring aromatics —benzothiophenes, naphthenobenzothiophenes and dibenzothiophenes, among other products such as sulfides. Based on reports, in Figure 3.6 it is included a set of S₁ aromatic model molecules as an illustration.

Figure 0.6. DBE distribution for the S₁ class in maltenes (M1 and M2) after hydroconversion and thermal cracking processes.



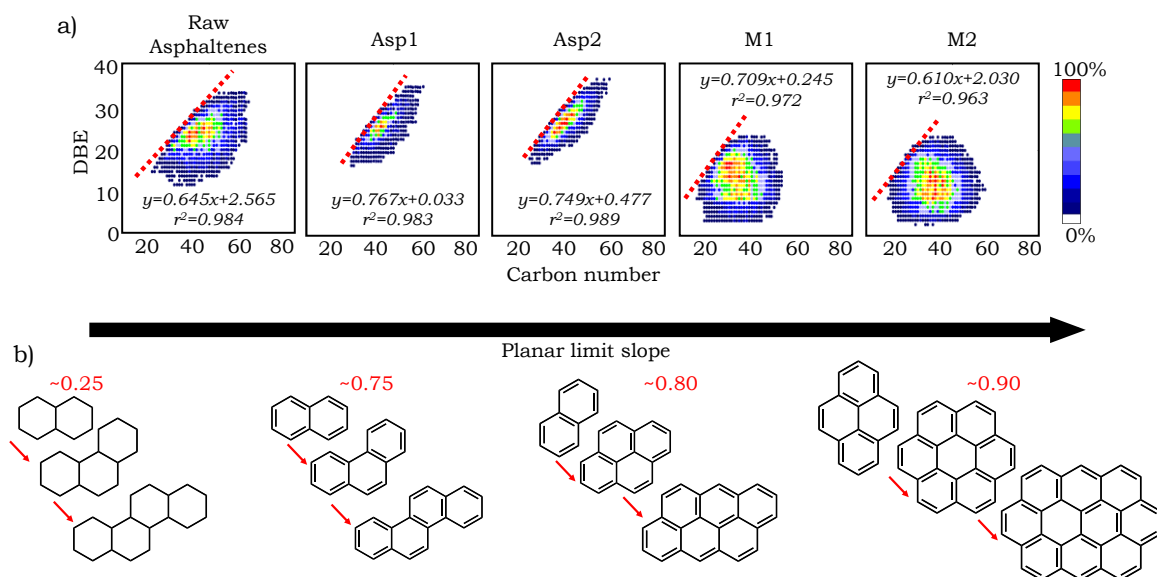
While studying the molecular composition of a Venezuelan vacuum residue by theoretical calculations and FT-ICR MS, Zhang *et al.* (Liu, Xu, et al. 2010) reported that S_1 species with DBE distribution between 3 and 25 are probably thiophenic, while sulfides must have DBE values below 5. In another report, Liu *et al.* (Liu et al. 2011; Wang et al. 2015) using FT-ICR MS observed that sulfide compounds in petroleomes are usually located in the low DBE region ($DBE < 4$) while sulfur in thiophenic functionalities is part of the molecules with DBE values of > 4 . Also, Hauser *et al.* (Hauser et al. 2014) indicated that a thermal cracking of sulfides is more probable than the conversion of thiophenic functionalities. Along these lines, sulfur species in produced maltenes, of the S_1 type, may consist mostly of thiophenic structures.

Figure 3.6 suggest abundant building blocks based on mono-, di-, and tri (and plus)-benzothiophenic structures, with alkylation up to 30 methylene units in accordance

with Figure 3.4. A possible sulfur-containing building block could be benzothiophene (DBE=6). The addition of one and two fused aromatic rings to benzothiophene produces dibenzothiophene (DBE=9) and benzonaphthothiophene (DBE=12), respectively. S_1 compounds with DBE=7 are likely benzothiophenes with one fused cycloalkane ring. Along these lines, the S_1 class species with DBE=10 correspond to dibenzothiophenes with one fused cycloalkane ring.

Additional structural information, in the form of the planar aromatic limits, can be derived from the DBE versus carbon number plots—Figure 3.7. Defined as the lines resulting from connecting maximum DBE values at a given carbon number (Hsu, Lobodin, et al. 2011), the planar limit (particularly the slope of the lines) has been used for structural elucidation of fossil hydrocarbons (Cho et al. 2011). Generally speaking, the planar limit slope corresponds to ~ 0.25 for saturated cyclic compounds, ~ 0.75 for catacondensed aromatic structures and ~ 0.90 for pericondensed aromatic structures. The maximum value for the slope, according to the “90% rule”, is 0.90; this means that the maximum DBE value for a fossil hydrocarbon cannot exceed the 90% of its carbon content. Table 3.3 includes the slope of the planar limits for the HC, S_1 , O_1 , N_1 , and N_2 compound classes in raw, residual asphaltenes (Asp1 and Asp2) and produced maltenes (M1 and M2). For classes S_1 , O_1 , N_1 , and N_2 , the planar limit slopes are significantly higher for the residual asphaltenes than for the raw material, indicating a general structural change toward more condensed aromatic architectures in the residual asphaltic material for these families. Importantly, compounds with the highest relative abundance in residual asphaltenes (in Figure 3.4) are clustered toward the planar aromatic limit, suggesting the prevalence of highly condensed aromatic species with little alkyl substitution after upgrading. Also, O_1 compounds in Asp1 and Asp2 samples are the most condensed structures with the highest planar limit slopes. This fact suggests that hydroconversion produces O-containing residual asphaltenes that are more structurally condensed when compared with the thermal cracking products.

Figure 0.7. a). Planar aromatic limits in DBE versus carbon number plots; b). Planar limit slopes as result of molecular structure.



On the other hand, class HC in maltenes, besides being the most abundant compound class in samples M1 and M2, presents planar limit slopes having a drastic decrease in comparison with the original asphaltenes. As discussed above, produced maltenes are enriched with hydrocarbons having low DBE values and high content of $-\text{CH}_2$ units —Figure 3.4. Considering planar limit slopes of 0.575 and 0.595 for M1 and M2, lower than the slope for original asphaltenes of 0.685, compounds in the maltene fraction may have molecular architectures consisting of a combination of alkyl chains with aromatic or naphthenic units linearly added, as previously indicated by Cho and coworkers (Cho et al. 2011). Here, the analysis of planar limit slopes could give a hint regarding the structure of the original asphaltenes. For example, the observed decline in planar limit slopes for the HC family, in contrast with the original asphaltenes, indicates that, after upgrading, the HC family has a more linear architecture than the starting material. Structurally speaking, this observation hints the existence of island-type structures from which linear structures (maltenes) and condensed aromatic cores (residual asphaltenes) arise after upgrading.

Table 0.3. Planar aromatic limit slopes for raw asphaltenes, residual asphaltenes and produced maltenes.

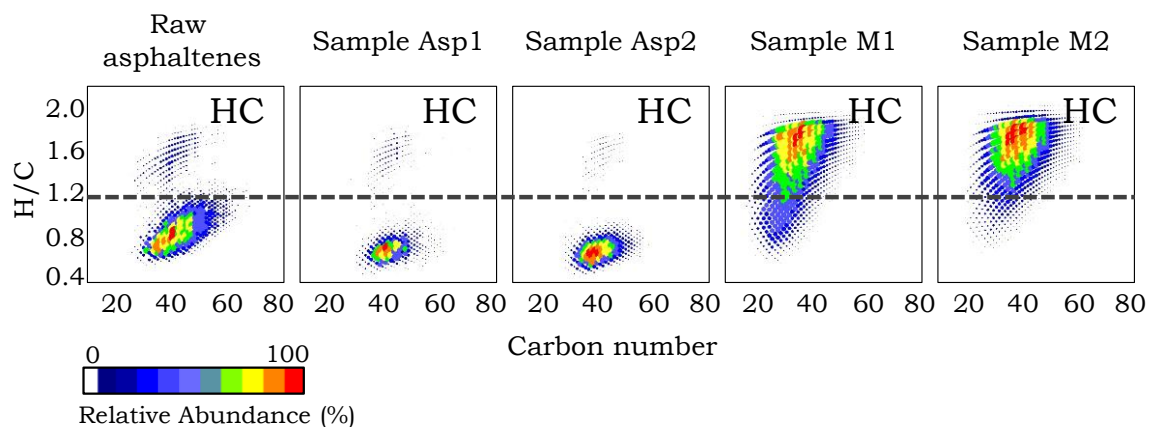
Class	Raw asphaltenes	Asp1	Asp2	M1	M2
HC	0.671	0.675	0.696	0.575	0.595
S ₁	0.614	0.635	0.684	0.694	0.560
O ₁	0.645	0.767	0.749	0.709	0.610
N ₁	0.595	0.711	0.646	0.658	0.645
N ₂	0.578	0.764	0.667	0.444	---

Interestingly, the behavior of the N₁ class in maltenes is contrary to that of the HC class. For the N₁ class, it is observed increased planar limit slopes of 0.658 and 0.645 for samples M1 and M2, respectively, in contrast with the original asphaltene planar limit slope of 0.595. The behavior is similar for residual asphaltenes with slopes of 0.711 and 0.646 for Asp1 and Asp2, respectively. The increment in planar limit slope indicates that, after upgrading, the N₁ compound family in maltenes has a higher condensed architecture than the starting material; structurally speaking, this observation only agrees with an original structure (from which the N₁ family in maltenes arises) having archipelago architecture. However, these conclusions are not as straightforward for compound classes S₁ and O₁. Although, for S₁ and O₁ classes in residual asphaltenes, planar limit slopes always increase, in comparison with the original asphaltene sample, which suggests a shift toward pericondensed aromatic architectures, no definite conclusion can be drawn about the architecture of these families in maltenes.

An alternative method to display compositional information for complex mixtures is provided by the van Krevelen diagrams. In this regard, several groups of researchers have analyzed supercomplex hydrocarbons mixtures using two- and three-dimensional van Krevelen plots. These molecular representations, which are constructed by plotting H/C versus O/C (or N/C or S/C) atomic ratios, are extremely useful in interpreting FT-IRC MS data. They have also been used for visual analysis of petrochemical samples from diverse geological origins, maturity, and processing, and for other complex mixtures, such as natural organic matter, lignin, and humic

acids (Islam et al. 2012; Kim et al. 2003). Strictly speaking, the HC class cannot be displayed in a van Krevelen diagram; however, a modified version of the diagram can be built in terms of H/C ratios *versus* carbon number. Figure 3.8 shows the H/C *versus* carbon number plots for the HC class present in the raw asphaltenes, and in the residual asphaltenes (Asp1, Asp2) and the produced maltenes (M1, M2) after upgrading. In contrast with raw asphaltenes showing H/C ratios between 0.60 and 1.20, residual asphaltenes exhibit the lowest H/C ratios in all samples, with values ranging from 0.50 to 0.85 for the hydrotreated samples and from 0.46 to 0.85 for the thermal-cracked samples. Also, it is possible to visualize that the carbon number is reduced, from 30–65 in the raw asphaltenes, to 33–55 for the hydrotreated and 32–58 for the thermal-cracked residual asphaltenes. In this sense, raw asphaltenes consist of more alkylated molecules (~35 methylene units), when compared with the residual asphaltic compounds (with ~18 methylene units). This observation means the upgrading procedures dramatically transform the feedstock, leaving behind highly aromatic and relatively small molecules with fewer alkyl units in their structures as residues. Merdrignac *et al.* (Gauthier et al. 2008; Merdrignac, A. A. Quoineaud, and Gauthier 2006) used NMR to study molecular changes in asphaltenes upon hydroconversion processes. Lower CH₂/CH₃ ratios were found in residual asphaltenes, compared with the feedstock, suggesting that remaining asphaltenes consist of aromatic cores with fewer CH₂ units, which indicates alkyl chain length reduction.

Figure 0.8. H/C versus carbon number plots for the HC class in raw asphaltenes, and residual asphaltenes (Asp1 and Asp2) and maltenes (M1 and M2) after upgrading.



The maltene fraction, produced after upgrading, exhibits the highest H/C ratios in all samples, with values ranging from 0.80 to 1.82 for the hydrotreated and from 0.95 to 2.00 for the thermal cracked. Also, the carbon number is reduced from 30–65 in the raw asphaltenes, to 20–50 in the hydrotreated and 20–55 in the thermal cracked maltenes, which suggests that maltenic products are smaller and more saturated than the asphaltene feedstock.

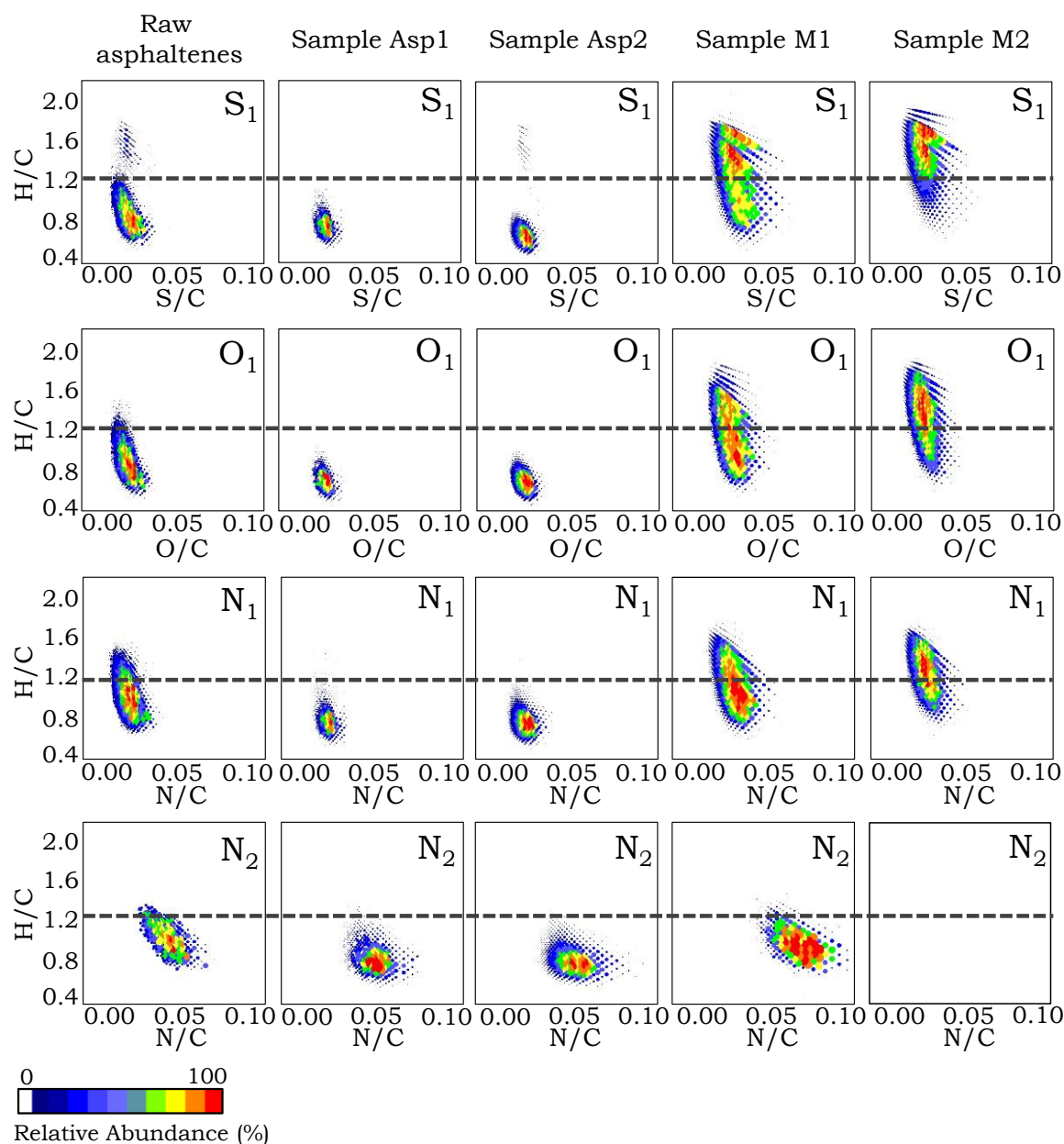
Interestingly, by combining information on planar limit slopes and van Krevelen diagrams, it is possible to strengthen some previous conclusions regarding structural information for the HC class. For example, let us consider the original asphaltene sample with a high abundance of molecular species around H/C ratio of 0.8, the residual asphaltenes with abundant species with H/C ratio of 0.6 and the maltenes with high abundance of species with H/C 1.8. The HC family in maltenes from the upgrading process exhibits H/C ratios much higher than the original or residual asphaltenes, indicating highly saturated, and probably linear, structures composed of naphthenic units with high alkyl content, in agreement with our previous observation of low planar limits for this compound class. The high linearity and

saturation of this fraction could imply its provenance mostly from island-type structures in the original feedstock.

Concerning compound classes containing heteroatoms, Figure 3.9 shows the van Krevelen diagrams for classes S_1 , O_1 , N_1 , and N_2 in the raw feedstock, and in the residual asphaltenes (Asp1, Asp2) and maltenes (M1, M2) after upgrading. In these diagrams, homologous series of compounds correspond to diagonals intersecting in $H/C=2$ with alkylation increasing from right to left along the line. Clearly the heteroatom containing families in maltenes share some of the compositional space of the original asphaltenes. However, in samples M1 and M2, we observe an increase in signal abundance for species with high H/C ratios, in contrast with the original asphaltene sample. This effect is more pronounced in S_1 -, N_1 -, and O_1 -containing maltenes from thermal cracking, and particularly for the S_1 class. Interestingly, heteroatom containing maltenes that have high H/C content or are more saturated (upper left corner of the van Krevelen diagrams for samples M1 and M2) are “bigger”, in terms of number of carbon atoms, than the same species that have a low H/C content (lower right corner of the diagrams for M1 and M2), and are less saturated and “smaller”. On the other hand, when compared with the HC class compositional space (Figure 3.8), the S_1 , N_1 , and O_1 -containing maltenes are shifted to lower H/C ratios, meaning that heteroatom-containing species have a more aromatic nature than the HC class in maltenes. This behavior also supports the conclusions derived from the analysis of the DBE vs carbon number plots (see Figures 3.4 and 3.6), where it is pointed out that S-containing species could be homologous series of mono-, di-, tri-, and tetra-benzothiophenes. However, it is important to highlight that S_1 , O_1 , and N_1 molecules in maltenes exhibit higher H/C ratios than the raw asphaltenes, also with a dramatic decrease in carbon number. For example, the average carbon number in the starting material is ~ 50 (N/C or $O/C=0.020$), whereas for maltenes after hydroconversion, the carbon number is ~ 30 (N/C or $O/C=0.033$) and for maltenes after thermal cracking, the carbon number is ~ 38 (N/C or $O/C=0.026$).

Along the same lines, the N₂ family, which is detected in maltenes after hydroconversion but not in maltenes after thermal cracking, has H/C ratios quite similar to the asphaltene feedstock. However, in this family, the carbon number is dramatically reduced from an average of ~40 (N/C=0.050), in the starting material, to an average of ~25 (N/C=0.080) in hydroconversion maltenes. Regarding the molecular composition of the residual asphaltenes, van Krevelen diagrams confirm that hydroconversion and thermal cracking yield asphaltic material with higher hydrogen deficiency than the feedstock, which could be originated from dealkylation and dehydrogenation reactions on peripheral naphthenic rings in the asphaltene core structure, as discussed previously. As with the previous HC class, van Krevelen diagrams also support the conclusions reached when analyzing planar limit slopes for heteroatomic classes. For example, for the N₁ family, we observe a great abundance of molecular species at a H/C ratio of ~1.1 for raw asphaltenes, abundant species with a H/C ratio of 0.6 for residual asphaltenes, and maltenes with high abundance of species have a H/C ratio of 1.2. The N₁ family in maltenes from the upgrading process exhibits H/C ratios that are quite similar to those of the original asphaltenes. The only explanation for this observation is the existence of archipelago structures that upon cracking would produce fragments with the same H/C ratios as the original structure. These resulting structures are more “condensed” than the original molecule and, hence, would exhibit higher planar limits than the parent structure, just as we reported in Table 3.3.

Figure 0.9. Van Krevelen diagrams for classes HC, S₁, O₁, N₁, N₂ in raw asphaltenes, and residual asphaltenes (Asp1 and Asp2) and maltenes (M1 and M2) after upgrading.



Although the effect of hydroconversion and thermal cracking on the asphaltene feedstock generally is similar for most compound classes, O₁ and S₁ families do not exhibit the same behavior, and we believe they undergo different reaction pathways

in both processes. In the case of thermal cracking, after the process, the planar limit slope decreases from 0.614 to 0.560 and from 0.645 to 0.610 for S_1 and O_1 families, respectively, while H/C ratios increase notably, in comparison with the hydroconversion process.

These results suggest that thermal cracking produces highly saturated S_1 - and O_1 -containing compounds with structures probably consisting of fused linear naphthenic units primarily. On the other hand, hydroconversion yields S_1 and O_1 classes with higher planar limit slopes than the starting material—from 0.614 to 0.694 for S_1 class and from 0.645 to 0.709 for O_1 class, and lower H/C ratios than the thermal cracking products. This observation means that hydroconversion induces the production of less saturated maltenic compounds (for the S_1 and O_1 families) with a more-condensed molecular architecture, when compared with the thermal cracking products.

At this point, it is important to highlight the usefulness of combining information derived from DBE *versus* carbon number plots and van Krevelen diagrams. Separately, these graphs illustrate molecular features such as DBEs, the number of methylene units in the structure, H/C ratios, and carbon numbers; however, together, they are extremely useful to establish reactivity trends of compound families under hydroconversion and thermal cracking processes, as well as structural features of the asphaltene feedstock. As stated above, after upgrading, highly saturated HC with a slightly condensed architecture are produced. On the other hand, upgrading yields N_1 maltenic compounds with a higher degree of molecular condensation and H/C ratios that are similar to those of the starting material; however, S_1 and O_1 classes seem to undergo different reaction pathways. These results indicate the existence of diverse architectures whose transformation, by several routes, results in the products that we observe. Several reports indicate that the transformation of asphaltenes under hydroconversion or thermal cracking conditions begins with the loss of alkyl side chains from the island-type aromatic structures, followed by

modification of peripheral naphthenic rings in the aromatic core. On the other hand, some groups report archipelago-type structures undergoing breakage of the aliphatic linkages between the aromatic moieties to give lighter products than the starting asphaltic compounds. In this sense, the molecular composition of the residual asphaltenes and the produced maltenes in our experiments suggest the coexistence of island-type and archipelago-type structures in the raw asphaltene sample. This observation is not new and has been previously reported by Podgorski *et al.* (Podgorski et al. 2013), who performed FT-ICR MS/MS analysis of asphaltenes and found that their molecular structures display island-type and archipelago-type structural motifs.

Figure 0.10. Hypothetical reaction pathways of model raw asphaltenes under hydroconversion or thermal cracking conditions: an archipelago-type structure producing maltenic products.

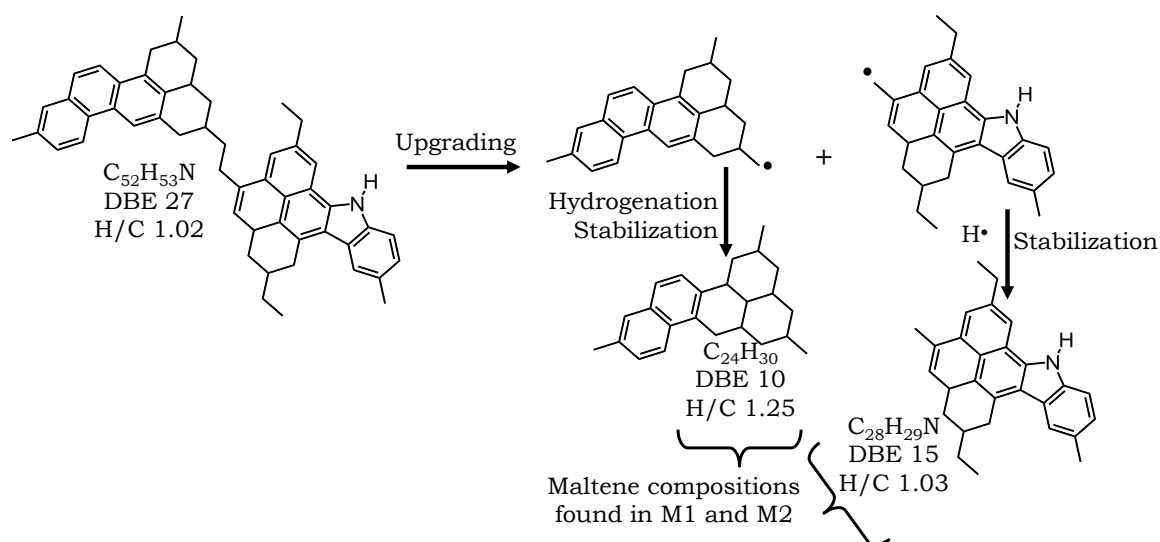
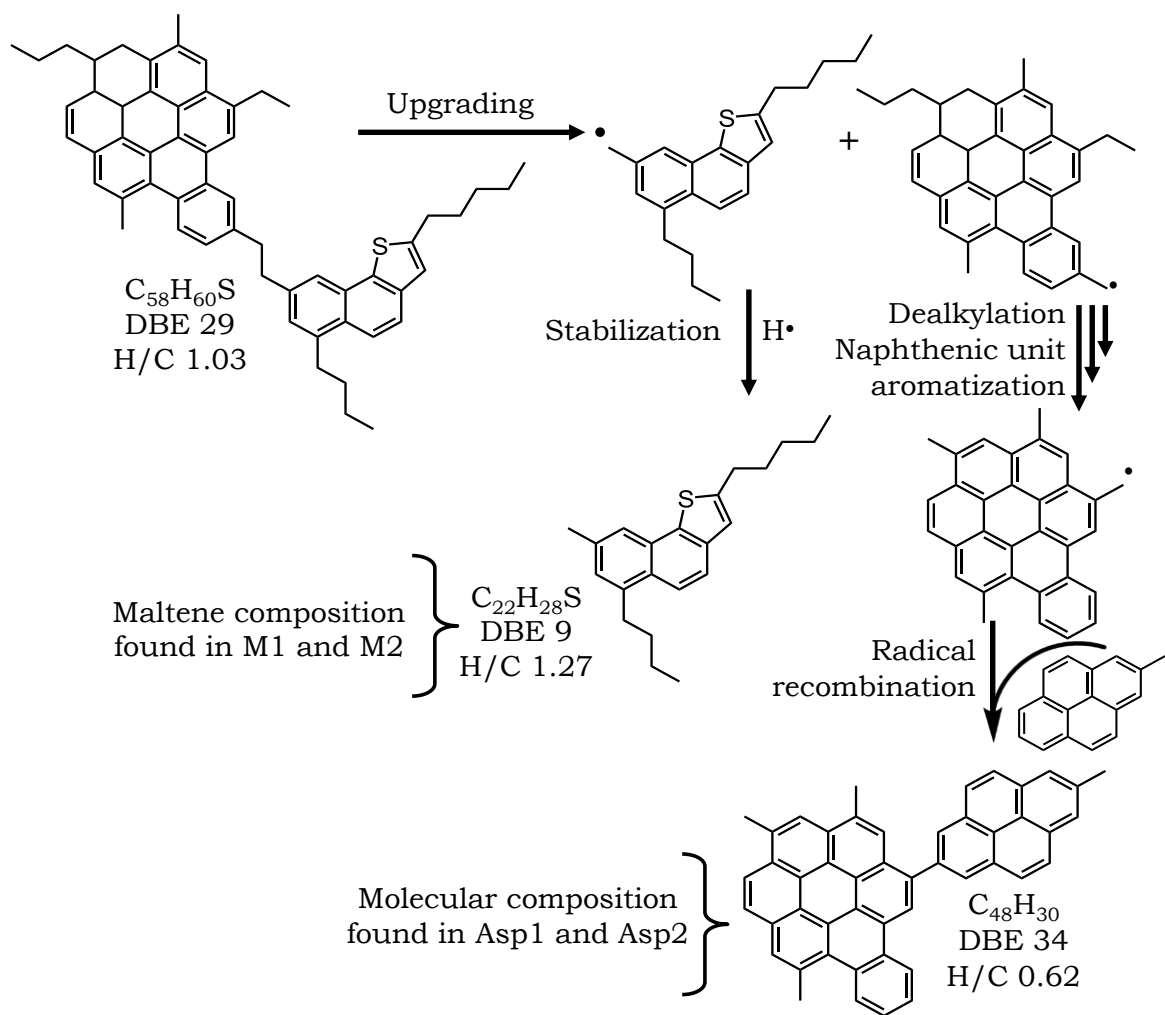


Figure 0.11. Hypothetical reaction pathways of model raw asphaltene under hydroconversion or thermal cracking conditions for an island-type structure with dibenzothiophene substituent, producing molecular compositions present in residual asphaltene and produced maltene.



We hypothesize that, under hydroconversion and thermal cracking conditions, highly stable heteroatomic island-type structures undergo a loss of alkyl side chains and aromatization of peripheral naphthenic rings. As a result, residual asphaltene show increased DBE values and a dramatic decrease in alkylation, while the produced maltene have low DBE values, abundant methylene units, and high H/C ratios, when compared to the original asphaltene. According to our observations,

compound classes HC and S₁ in produced maltenes (see Figures 3.4, 3.6 and 3.9) could originate through this process. On the other hand, heteroatomic archipelago-type structures undergo excision of linkages between aromatic moieties, producing mostly lighter heteroatom-containing maltenic compounds with a slight increase in H/C ratios, and structures with a relative high number of methylene units and reduced DBE and carbon numbers, when compared with the raw asphaltenes. Compound classes N₁ and N₂ in produced maltenes (Figures 3.4 and 3.5) could arise from this process, while compound class O₁ exhibits an intermediate behavior.

Figures 3.10 and 3.11 depicts, solely with an illustrative purpose, the possible reaction routes of the raw asphaltenes transformation to produce maltenic compounds and residual asphaltenes, under the assumptions exposed above. All structures in Figure 3.10 and 3.11 have a molecular composition that is contained within the petroleomes of Figures 3.4, 3.8 and 3.9.

3.5 CONCLUSIONS

Sorting high-resolution mass spectrometry data in compound class distributions is useful to observe general trends in compositional changes when asphaltenes undergo upgrading processes. More specific information is derived from plots of DBE versus carbon number. These graphs suggest that, under thermal cracking and hydroconversion conditions, asphaltenes produce two types of compounds. One consisting of highly condensed and very stable aromatic cores with little alkyl substitution, classified as residual asphaltenes (insoluble in *n*-heptane); and another composed of molecules with lower DBE values and lower carbon numbers than the feedstock, classified as maltenes (soluble in *n*-heptane). Also, from these plots, structural information is also derived in the form of planar limit slopes, which are related to the degree of condensation of the molecular structure. In this sense, the maltenic fractions after upgrading have hydrocarbons (HC class) with low aromatic

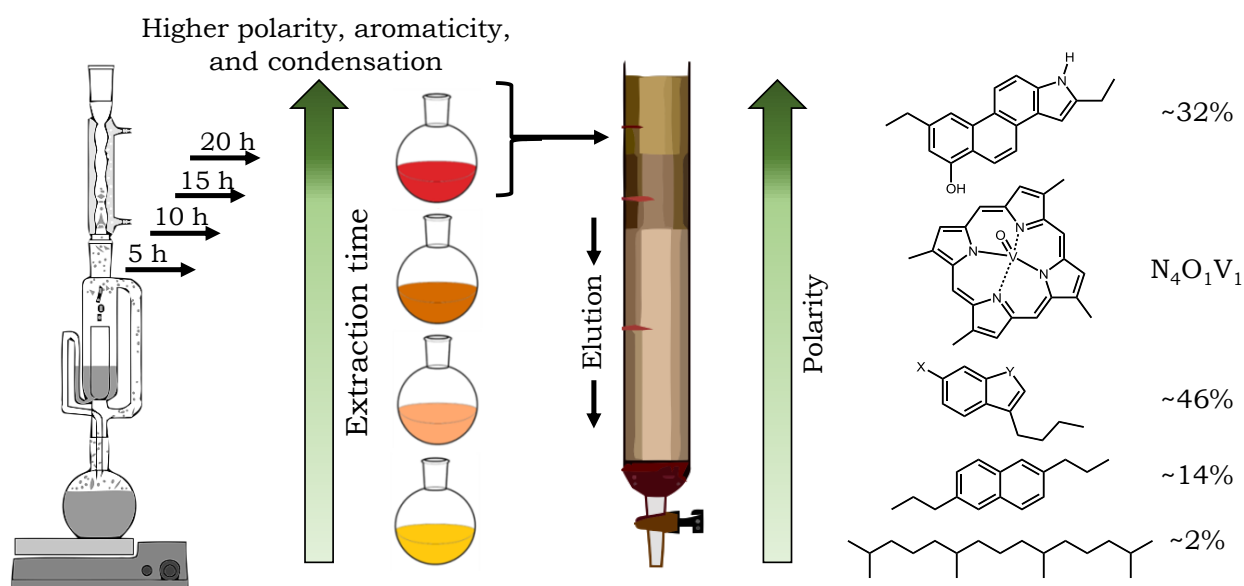
condensation. In contrast, N₁-containing structures seem to be more condensed. On the other hand, residual asphaltenes invariably exhibit a shift toward more-condensed architectures.

Additionally, we can also have some insight into the original asphaltene structure as the petroleomes of the residual asphaltenic and maltenic fractions after upgrading reflect the starting material composition. For instance, it is possible to hypothesize that some raw asphaltenes with island-type structures, having “arms” or side chains made of alkyl units or mono-, di-, or tri-benzothiophene units, undergo upgrading to form the HC and S1 compounds that are present in maltenes. These two classes exhibit high saturation, decreased structural condensation, lower carbon numbers, and dramatic decrease in aromaticity than both the original and the residual asphaltenes. Similarly, one can conclude that N₁-containing compounds in produced maltenes are exclusively derived from archipelago-type structures. All of our observations suggest a parent asphaltene composed of a mixture of island-type and archipelago-type motifs.

4. EXPLORING ASPHALTENE OCCLUSIONS THROUGH HIGH RESOLUTION MASS SPECTROMETRY

*Redrafted from *Martha L. Chacón-Patiño, Silvia J. Vesga-Martínez, Cristian Blanco-Tirado, Jorge A. Orrego-Ruiz, Andrea Gómez-Escudero, and Marianny Y. Combariza. Accepted for publication in Energy and Fuels.*

Figure 0.1. Graphical abstract for Exploring asphaltene occlusions through High-Resolution Mass Spectrometry.



4.1 ABSTRACT

In this contribution, we use high-resolution mass spectrometry to unveil the molecular composition of occluded compounds inside Colombian asphaltenes macrostructures. We use Soxhlet extraction, with *n*-heptane, coupled with asphaltene maceration to obtain four fractions enriched with chemical compounds closely related to asphaltenes. We focused our efforts on the fraction enriched with

strongly occluded compounds, used normal phase column chromatography to fractionate it and atmospheric pressure photoionization coupled to Fourier transform ion cyclotron resonance mass spectrometry to obtain a detailed molecular description. Our results indicate that strongly-occluded compounds are by themselves a complex mixture, consisting mostly of saturated compounds including molecular formulas corresponding to biomarkers, alkyl aromatics with high heteroatom content (up to four heteroatoms), vanadyl porphyrins, and highly aromatic species which we believe are low-molecular weight asphaltenes transferred to the *n*-heptane during the extraction process. We consider this information valuable because analysis of occluded compounds gives us a more thorough molecular description of asphaltenes; besides knowledge of compounds closely related to asphaltenes could not only improve deasphalting processes in pilot plants, but also will help to find new geochemical biomarkers occluded within asphaltenes.

4.2 INTRODUCTION

Currently the definition of asphaltenes is based on their solubility behavior in *n*-alkanes *n*-C₅ to *n*-C₇ (Sheu and Storm 2013); however, there have been efforts to establish a molecular definition of asphaltenes based on structure and function (Snape and Bartle 1985; Speight 2004; Speight et al. 1982). Early works began in the 1950's, with the development of standard methodologies to precipitate asphaltenes from crude oils and bitumen. Current efforts are focused on unraveling asphaltenes' molecular architecture with the goal of predicting reactivity of heavy oils in upgrading processes (Purcell et al. 2010), adsorption of asphaltenic material on mineral surfaces (Adams 2014), and asphaltene deposition (Rogel et al. 2015).

Asphaltenes negatively impact production, transport and refining of heavy crude oils, as direct consequence of their particular molecular characteristics (Akbarzadeh et

al. 2007; Buenrostro-Gonzalez et al. 2001). Asphaltenes are ultra-complex mixtures (Mckenna et al. 2013) with a wide diversity of functional groups that promote non-covalent associations, through acid-base interactions (between carboxylic acids and pyridine groups), hydrogen bonding, formation of coordination complexes (due to the presence of vanadium and nickel), generation of hydrophobic pockets (by van der Waals interactions between alkyl and naphthenic moieties), and π - π stacking (due to interactions between parallel aromatic cores). Although individually weak, synergism between these associations result in the formation of strongly associated structures, or aggregates, that exist even at low concentrations (~ 50 $\mu\text{g/mL}$) and relative high temperatures (~ 70 $^{\circ}\text{C}$). These aggregates are the direct cause of the historical controversies and limitations in asphaltenes' characterization. As an example we have the evolution of asphaltenes' molecular weights from the range of 10^4 – 10^5 amu, observed by gel permeation chromatography and vapor pressure osmometry (Acevedo et al. 1985) to weights between 200–1700 amu, detected by molecular diffusion measurements and two-step laser mass spectrometry (Groenzin and Mullins 2000; Pomerantz et al. 2008).

Flocculation of asphaltene nanoaggregates and subsequent precipitation driven by addition of *n*-alkanes to crude oils always cause occlusion of compounds within the aggregates (Gray et al. 2011). Strausz and co-workers reported that successive cycles of redissolution and reprecipitation of Athabasca asphaltenes, in toluene and *n*-heptane respectively, resulted in decreased asphaltene recovery because of the release of occluded compounds to the *n*-C₇ phase (Strausz et al. 2006). Six cycles of redissolution and reprecipitation resulted in the loss of 22 wt% from the asphaltic material; however, no constant weight was reached in these experiments, suggesting the decrease of *n*-heptane insolubles to be permanent. Loss of recovered asphaltenes is expected upon redissolution and reprecipitation due to a more efficient partitioning of asphaltenes with the solvent (Strausz et al. 2006), to the release of low-polarity and low-molecular weight asphaltenes to the *n*-heptane phase, and to the fact that aggregates can occlude some compounds such as

sterane biomarkers (Chupeng et al. 2009), polyaromatic hydrocarbons (Derakhshesh, Bergmann, and Gray 2013), C₂₇–C₃₁ hopanes (Liao et al. 2006), and resins (Acevedo et al. 1985; Zhao et al. 2010). Some of these compounds, particularly the low-polarity asphaltene subfractions, appear to act as stabilizers of asphaltene dispersions with dissolution rate constants in heptane–toluene mixtures four times higher than the original asphaltene sample (Kaminski et al. 2000).

The existence of occlusions supports the idea of porous asphaltene nanoaggregates in solution (Derakhshesh et al. 2013). Extensive reports indicate that asphaltene occlusion protected trapped biomarkers from thermal degradation during petroleum diagenesis (Ekweozor 1984; Gray et al. 2011; Liao et al. 2006). Experimental data suggest that asphaltene aggregates are stable at high temperatures, up to 300 °C (Tanaka et al. 2004), as a result, occluded compounds inside asphaltene macrostructures should be also stable even at 300 °C (Peters, Walters, and Moldowan 2005). A typical procedure to release occluded compounds from asphaltenes is based on oxidation with H₂O₂/CH₃COOH, which selectively oxides occluded compounds inside the aggregates without altering peripheral alkyl side-chains of asphaltenes (Liao et al. 2004). Liao and co-workers used gas chromatography (GC) to analyze the occluded compounds in Venezuelan asphaltenes released by oxidation with H₂O₂/CH₃COOH and found biomarkers such as pristane and phytane and saturated hydrocarbons with carbon numbers up to 40 (Liao et al. 2004). In another contribution, Gray *et al.* (Derakhshesh et al. 2013) performed occlusion tests for pyrene and phenanthrene inside Athabasca asphaltenes dissolved in toluene, which were further precipitated by the addition of *n*–heptane. GC analysis demonstrated that occluded pyrene and phenanthrene were detected in asphaltene precipitates at a concentration of 6 w%. Besides demonstrating that asphaltene aggregates in toluene solutions exhibit open porous structures, this report also indicates asphaltene precipitation as an inadequate method for sample preparation. Observation of simple polyaromatic hydrocarbons such as pyrene and phenanthrene, which on account of their solubility and structure

should not be present in the *n*-C₇ insoluble fraction, demonstrate occlusion formation in asphaltenes. Along these lines, Acevedo *et al.* (Acevedo *et al.* 2009) also demonstrated that asphaltenes occlude compounds through guest–host interactions. The authors used LDI-TOF mass spectrometry to characterize the extracted occluded–compounds from *para*-nitrophenol asphaltene fractions, by dissolution tests in *n*-heptane, and determined that trapped compounds are enriched in petroporphyrins and resins.

Nowadays high-resolution mass spectrometric techniques, such as Fourier transform ion cyclotron resonance mass spectrometry (FT-ICR MS) combined with soft ionization techniques have become fundamental for the analysis of super complex mixtures like heavy oils, vacuum residues and asphaltenes (Marshall and Rodgers 2004; McKenna, Blakney, *et al.* 2010; Podgorski *et al.* 2013; Rodgers and Marshall 2007; Rodgers *et al.* 2005). Only the high resolving power and the mass accuracy, offered by FT-ICR MS, allow identification and reliable elemental formula assignment for more than 10,000 different chemical species found in crude oil (Lababidi *et al.* 2013). FT-ICR MS analysis makes it possible to classify petroleum components in compound classes: all the molecules having the same number and type of heteroatoms. In each compound class, chemical species are organized in homologous series: all components with the same number of rings and double bonds (double bond equivalents – DBE), but different content of CH₂ units (alkylation degree). Commonly, FT-ICR MS data is presented in isoabundance–contoured plots of DBE *versus* carbon numbers, where the relative abundance is normalized within each class. Using these plots, Rodgers and co-workers (McKenna *et al.* 2013) defined the asphaltene compositional space as containing molecular compositions with DBE values between 20 and 40, and carbon numbers of 30 and 60, which means H/C ratios below 1.1. These authors also demonstrated that asphaltenes are an extension of the maltene compositional space to higher degrees of aromaticity instead of higher carbon numbers. In another contribution (Podgorski *et al.* 2013), Rodgers and coworkers, using infrared multiphoton dissociation (IRMPD) in FT-ICR

MS experiments, gave insights about asphaltene's core structures and presented evidence of the coexistence of archipelago and island motifs in a sample of C₅-insoluble materials (Ruiz-Morales 2002).

In this contribution we explore the molecular features of strongly occluded compounds isolated from the asphaltene fraction of a Colombian heavy oil (API 12). We employed four successive Soxhlet extraction (*n*-heptane) - maceration steps to obtain four hydrocarbon fractions enriched in compounds strongly occluded inside *n*-C₇ asphaltenes' macroporous structures. We used APPI FT-ICR mass spectrometry to access the molecular compositions of the four fractions and found that weakly occluded compounds consist mostly of alkyl aromatics of the family class HC. On the other hand, strongly occluded compounds are complex mixtures of heteroatom containing families such as N₁, N₁O₁, N₁O₁S₁, N₁S₁, O₁, O₂, O₁S₁, O₁S₂, and S₁, having relatively high abundance of molecular compositions clustered at the aromatic limit. Due to the complexity of the strongly-occluded fraction we subfractionated it using normal phase column chromatography, and collected six extracts. HR-MS results indicate the existence of saturated compounds with low heteroatom content, alkyl aromatics with high heteroatom content, vanadyl porphyrins and highly aromatic compounds which we believed are low molecular weight asphaltenes transferred to the *n*-heptane. Along these lines, we provide experimental evidence indicating that deep understanding of asphaltene occlusion could help to enhance the current molecular picture of asphaltenes.

4.3 EXPERIMENTAL SECTION

4.3.1 Materials. Toluene (Tol), *n*-heptane (*n*-C₇), *n*-hexane (*n*-C₆), dichloromethane (DCM), and methanol (MeOH) Chromasolv® solvents (99.9%) were purchased from Sigma Aldrich (St. Louis, MO, USA). Silica gel 60 (0.015–0.040 mm) was purchased from Merck Millipore (Darmstadt, Germany). All solvents and reagents were used as received.

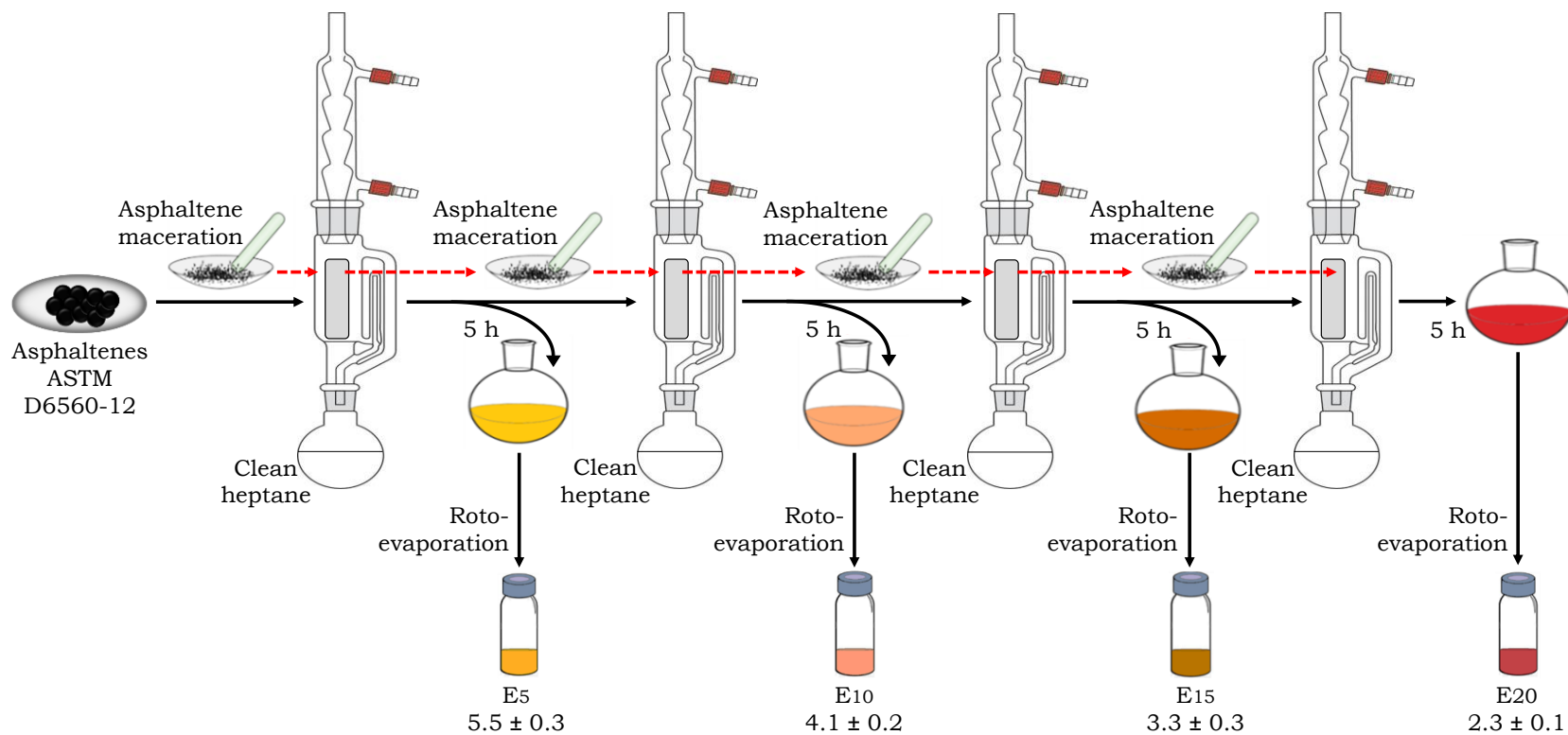
4.3.2 Sample preparation. Asphaltenes were isolated from a Colombian heavy crude oil (API=12) using the ASTM standard methodology D6560–12 with slight variations. In short, 400 mL of *n*-heptane were added dropwise to 10 g of crude oil, under sonication (Branson Ultrasonics, Danbury, CT, 22 kHz, and 130 W). After that, the mixture was heated at 90 °C under reflux during 60 minutes. Subsequently, the mixture was allowed to stand overnight. Asphaltenes were collected by filtration (Whatman grade 42) and placed in a Soxhlet apparatus with *n*-heptane, until the washing solvent was clear (~72 hours). Asphaltenes were recovered by dissolution in hot toluene (~98 °C), which was finally removed to produce solid asphaltenes. 1 g of clean C₇ asphaltenes was crushed using an agate mortar and placed on a Soxhlet apparatus with *n*-heptane as the washing solvent. After five hours of solvent recirculation the *n*-heptane with extracted hydrocarbons was taken and kept for analysis. The remnant solid sample was again pulverized by maceration and placed on the Soxhlet system for extraction with clean *n*-heptane for another 5 hours. This operation was repeated two more times until completing a total of 20 hours of washing. Four different *n*-heptane extracts were gathered in this fashion as depicted in Figure 4.2. The four extracts were rotoevaporated, weighted, labeled E₅, E₁₀, E₁₅, and E₂₀, and kept for FT-ICR mass spectrometric analysis.

4.3.3 Column chromatography fractionation. Fraction E₂₀ was subjected to further fractionation by normal phase column chromatography using as mobile phase a standard elutropic series designed to separate saturates, alky-aromatic and polar compounds in petrochemical samples (Chacón-Patiño et al. 2015a; Vela et al. 1995) 50 mg of fraction E₂₀ were mixed with silica gel and placed at the top of the column (length 30 cm, diameter 2.5 cm). The column was eluted sequentially with *n*-C₆, *n*-C₆: Tol (1:1), Tol, Tol: DCM: MeOH (30:9:1), Tol: DCM: MeOH (10:9:1), and DCM: MeOH (9:1). Fractions were dried with N₂ and weighted. This procedure was repeated several times in order to collect enough sample for UV-Vis spectroscopy and mass spectrometry analysis.

4.3.4 UV-Vis spectroscopy. Samples were diluted in *n*-C₇: Tol (1:1) to a concentration of 50 ppm and analyzed using an UV-vis spectrometer, Genesys10S (Thermo Scientific, Billerica, MA, USA).

4.3.5 APPI FT-ICR mass spectrometry. FT-ICR mass spectrometry analysis was performed on a Solarix 15 T mass spectrometer (Bruker Daltonics, Billerica, MA) with an APPI source, fitted with a Krypton photoionization lamp (10.6 eV), and operated in positive (+) mode. The front and back trapping voltages in the ion cyclotron cell were set at +0.70 V and +0.50 V, respectively. The ICR cell was previously calibrated using a NaTFA solution (*m/z* from 200 to 1200). Each mass spectrum was recorded by the accumulation of 100 scans of time-domain transient signals in 4 mega-point time-domain data sets.

Figure 0.2. Extraction protocol for occluded compounds in *n*-C₇ asphaltenes including weight percentages of the extracted fractions.



All mass spectra were internally recalibrated using a homologous series of alkylated compounds using the Data Analysis Software 4.2 (Bruker Daltonics, Billerica, MA). Mass spectra were further recalibrated by walking recalibration algorithms using the Composer software 1.5.0 (Sierra Analytics, Modesto, CA, USA). Resolving power at m/z 400 higher than 600,000 for all mass spectra, and mass accuracy of 1.0 ppm, provided unambiguous molecular formula assignments for singly charged molecular ions with relative abundance above 1%.

4.4 RESULTS AND DISCUSSION

Figure 4.2 shows the procedure used for extraction of occluded compounds from asphaltenes using n -C₇. We modified the ASTM D6560–12 standard procedure to extract occluded compounds by increasing the asphaltene sample surface area in contact with the solvent through maceration using an agate mortar. The extraction process was carried out in four separated stages until completing 20 hours of solvent recycling. After this, time we could not collect enough sample for MS analysis, so we deemed this the final point of the extraction process. However, it is important to note that the asphaltene washing process could go on indefinitely, as Strausz (Strausz et al. 2006) have determined from redissolution and reprecipitation processes using n -C₇ in Athabasca asphaltenes.

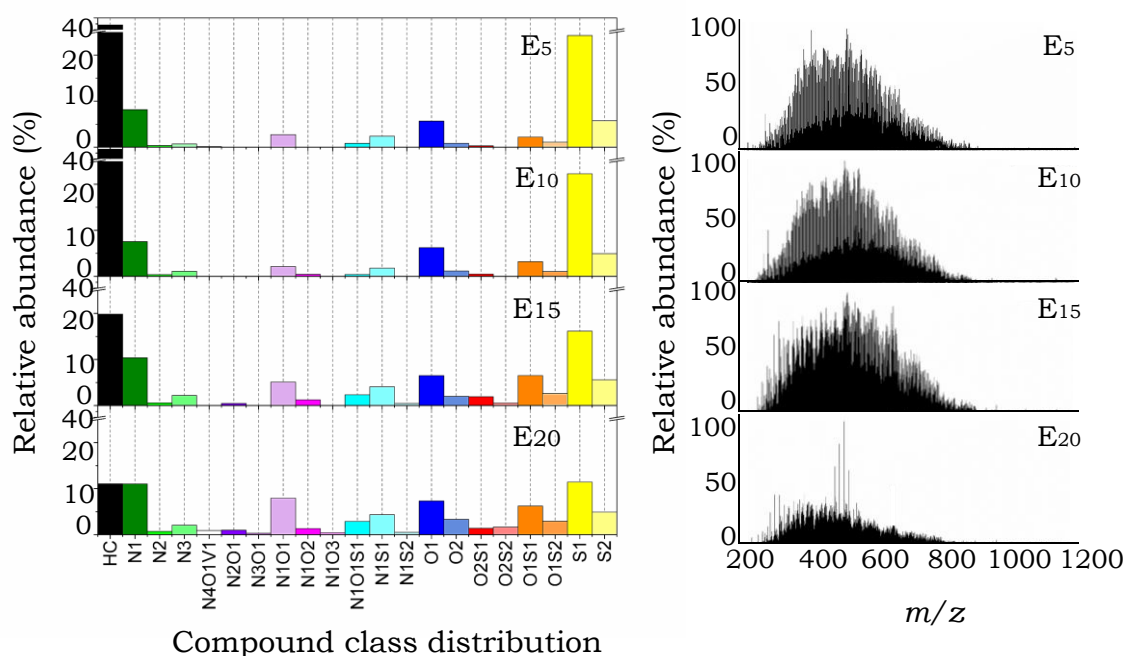
We used HRMS to track n -C₇ extract composition in order to determine appropriate sampling times. For instance, during the first ten hours of extraction fractions E₅ and E₁₀ are enriched with family class HC (DBE<15) in high abundance. From ten to fifteen hours of extraction (E₁₅) the sample is enriched with heteroatom-containing compounds and exhibits a transition in aromaticity: the molecular compositions start shifting towards the aromatic limit. At the last stage of the extraction, fraction E₂₀, we observe the highest heteroatom content and compositions clustered at the aromatic limit. We will discuss HR–MS data more extensively in the next section of the paper.

Although fractions E₅ to E₁₅ are interesting, we focused our attention on fraction E₂₀, which contains *n*-C₇ soluble compounds strongly bound to asphaltenes. We extracted saturates, alkyl aromatics and polars from fraction E₂₀, with the purpose of achieving a more complete characterization by APPI FT-ICR mass spectrometry. Figure 4.2 also contains information about the mass balance of the occluded compounds extracted from the *n*-C₇ asphaltenes. The weight percentages of fractions E₅, E₁₀, E₁₅ and E₂₀ are 5.5, 4.1, 3.3 and 2.3, respectively for a total amount of ~15.2% w of extracted hydrocarbons. Some reports by Strausz, Gray *et al.* (Derakhshesh *et al.* 2013; Strausz *et al.* 2006) point out that the quantity of occluded *n*-C₇ solubles in asphaltenes' aggregates can be up to 50%.

Figure 4.3 presents compound class distributions and molecular weight distributions for fractions E₅, E₁₀, E₁₅ and E₂₀. In general, all fractions contain the same fourteen compound classes, detected both as radical cations and protonated molecules: HC, N₁, N₁, N₃, N₁O₁, N₁O₁S₁, N₁S₁, O₁, O₂, O₂S₁, O₁S₁, O₁S₂, S₁ and S₂. The main compositional differences between the fractions lie in significant variations in relative abundances as the extraction progresses. For instance, fractions E₅ and E₁₀ contain around 55% of hydrocarbons without heteroatoms, or class HC, while the compound classes with heteroatoms in fractions E₁₅ and E₂₀ goes up to ~80% and ~90%, respectively. We hypothesize that occluded compounds relate with asphaltene aggregates through multiple intermolecular interactions such as hydrogen bonding between polar moieties like phenols, carboxylic acids, and weak nitrogen bases; acid base interactions between pyridine or amine moieties and carboxylic acids; and assembly of naphthenic or alkyl groups in hydrophobic pockets through van der Waals interactions. High relative abundance of compound classes such as HC and S₁ during the first stages of the extraction process indicates lack of functionalities able to establish strong interactions with the polar moieties of asphaltenes. It is important to highlight that sulfur is more polarizable than oxygen and nitrogen, and hence, S–hydrogen bonds are weaker. On the other hand, as the extraction proceeds (see Figure 4.3), the fractions are enriched in poly–heteroatomic

compounds, which we believe are strongly occluded inside asphaltenes because of a high chance of interacting through polar interactions, with the active moieties of asphaltenes. For instance, fraction E₂₀ contains compound classes: N₂O₁, N₃O₁, N₁O₂, N₁O₃, N₁S₂, O₂S₂ and N₄O₁V₁, which illustrates its high heteroatom content as well as its compositional similarity to asphaltenes. We have previously reported (Chacón-Patiño et al. 2015) that Colombian asphaltenes contain up to ~90% of compound classes having heteroatoms, along with vanadyl porphyrins or class N₄O₁V₁. To this extent, in terms of compound class distribution, fraction E₂₀ appears to be reflective of asphaltenes' molecular composition, in agreement with Strausz who proposed that successive cycles of redissolution and reprecipitation of asphaltenes could transfer some low-molecular weight asphaltenes to the *n*-heptane.

Figure 0.3. Compound class distributions and molecular weight distributions for fractions E₅, E₁₀, E₁₅ and E₂₀, measured by (+) APPI FT-ICR mass spectrometry.



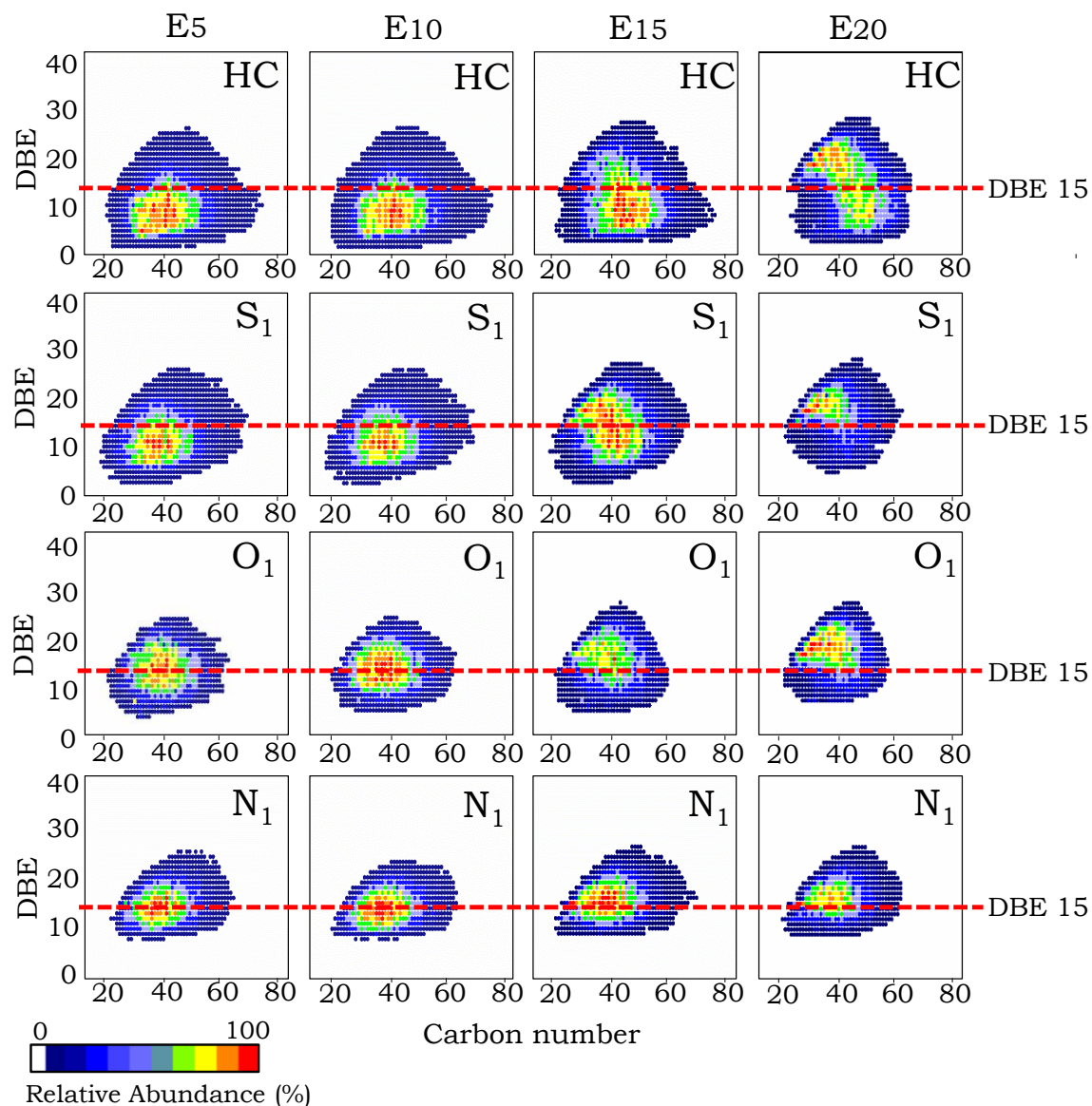
Regarding metal complexes, APPI coupled to FT-ICR mass spectrometry has made possible the direct identification of nickel and vanadium porphyrins in vacuum residues and asphaltenes, as well as in unfractionated heavy oils (McKenna et al. 2009; Qian et al. 2008). Many reports indicate that concentrations of metal complexes are higher in high-boiling cuts and asphaltenic deposits (Adams 2014; Dechaine and Gray 2010; Liu et al. n.d.; Qian et al. 2008, 2010). Thus transfer of vanadyl porphyrins from the asphaltene sample to the maltene phase, as observed in fraction E₂₀, could be consequence of extracting compounds with high heteroatom content. The supramolecular assembly model for asphaltene aggregation, reported by Gray and co-workers (Gray et al. 2011), suggests that vanadyl porphyrins promote formation of metal complexes through axial coordination between vanadium and basic nitrogen; this association is part of the set of intermolecular interactions responsible of asphaltenes' aggregation tendency (Stoyanov et al. 2010). Thus, the presence of molecules with *N*-containing moieties (e. g. in pyridinic or amine groups) in fraction E₂₀, with high relative abundance, could be responsible for the solubility of vanadyl porphyrins in *n*-heptane.

Additionally, molecular weight distributions are illustrative of the unique molecular composition of fraction E₂₀. Fractions E₅, E₁₀ and E₁₅ exhibit similar MWDs, ranging approximately from *m/z* 220 to *m/z* 920 and centered around *m/z* 550. On the other hand, the MWD of fraction E₂₀ starts approximately at *m/z* 220 and ends at *m/z* 800. The distribution center is shifted towards lower *m/z* values -approximately *m/z* 400- and a group of prominent peaks around *m/z* 500, corresponding to vanadyl porphyrins is easily discerned. This molecular distribution is comparable to one reported for Colombian asphaltenes in previous works (220<*m/z*<980, centered around *m/z* 600). However, in this case, the MWD of the fraction E₂₀ presents high abundance of low molecular weight species (~*m/z* 400).

Plots of double bond equivalents (DBE) *versus* carbon numbers (C#) provide a closer look at the molecular information contained in FT-ICR mass spectra. Figure

4.4 presents the isoabundance-contoured plots of DBE *versus* carbon number for classes HC, S₁, O₁ and N₁ present in fractions E₅, E₁₀, E₁₅ and E₂₀, where the relative abundance is normalized within each compound class.

Figure 0.4. Isoabundance-contoured plots of DBE *versus* carbon numbers for classes HC, S₁, O₁ and N₁ in fractions E₅, E₁₀, E₁₅ and E₂₀.



As stated above, fractions E₅ and E₁₀ have comparable molecular compositions with the following features: classes HC and S₁ exhibit similarities with high abundance of

compounds with DBEs between 3 and 15, and carbon numbers between 30 and 50. Classes O₁ and N₁ present high abundance of compositions with DBEs from 10 to 20 and carbon numbers from 30 to 50. Increased DBE values while the carbon content remains unchanged implies an increase in aromaticity. Along these lines, the carbon number/DBE ratios (C#/DBE) are used as aromaticity indicators (Purcell et al. 2010) with low C#/DBE ratios meaning increased aromaticity and *vice versa*. For instance, the C#/DBE ratios for coronene (C₂₄H₁₂), anthracene (C₁₄H₁₀), toluene (C₇H₈), and butyl-toluene (C₁₁H₁₆) are 1.26, 1.40, 1.75, and 2.75, respectively. Table 4.1 includes C#/DBE ratios for fractions E₅–E₂₀. For fraction E₅, compound classes HC, S₁, O₁, and N₁ have weighted averages of C#/DBE ratios of 3.82, 3.12, 2.81, and 2.60, respectively. Fraction E₁₀ presents, to some extent, an increase in aromaticity; thus weighted averages of C#/DBE ratios are 3.48, 3.02, 2.81, and 2.56 for the same classes HC, S₁, O₁, and N₁, respectively. The observation of compound classes O₁ and N₁ with higher aromaticity than HC and S₁ families, is in agreement with several reports related to high-resolution MS analysis of crude oils, asphaltenes and coals. According to these reports, heteroatoms such as N and O could be part of aromatic moieties in pyridinic, pyrrolic, phenolic and furanic compounds (Chacón-Patiño et al. 2015; Omais et al. 2013; Jeremiah M. Purcell et al. 2007). Thus, it is possible to conclude that fractions E₅ and E₁₀ have HC and S₁ compounds with high degree of saturation, while N₁ and O₁ compounds in the same fractions are more aromatic.

Table 4.1. C#/DBE ratios for classes HC, S₁, O₁, and N₁ in fractions E₅–E₂₀.

	HC	S ₁	O ₁	N ₁
E ₅	3.82	3.12	2.81	2.60
E ₁₀	3.48	3.02	2.81	2.56
E ₁₅	3.41	2.77	2.58	2.42
E ₂₀	3.02	2.37	2.32	2.30

Fraction E₁₅, in contrast to fractions E₅ and E₁₀, exhibits distribution maxima shifted towards the aromatic limit, which implies a transition in aromaticity. The aromatic limit, also called planar limit, is defined as the line originated from linking maximum

DBEs' values at given carbon numbers, in contoured plots of DBE *versus* carbon number. Clustering of compositions along the planar limit, hints a molecular structure enriched in aromatic cores with little content of carbon atoms in alkyl-side chains. In general, classes S₁, O₁ and N₁, of the fraction E₁₅ present petroleomes with high abundance of compositions along DBEs 10 to 20, and carbon numbers between 25 and 50. The HC class' petroleome extends abundantly along DBEs 5 to 15, presenting a slight shift toward higher carbon numbers: from 35 to 60. However, Figure 4.4 also shows an important feature of fraction E₁₅: the molecular compositions in this fraction are starting to shift towards the planar limit, which is particularly noticeable for heteroatom-containing molecules (S₁, O₁ and N₁). In addition, C#/DBE ratios also indicate higher aromaticity in this fraction, with values of 3.41, 2.77, 2.58, and 2.42, for classes HC, S₁, O₁, and N₁, respectively. Finally, compound families in fraction E₂₀ exhibit the strongest shift towards the planar limit. The petroleomes for this sample show high abundance of compositions with DBE values between 15 and 25 and carbon numbers between 30 and 45. The weighted averages of C#/DBE ratios, the lowest of the four extracted fractions, are 3.02, 2.37, 2.32, and 2.30 for compound classes HC, S₁, O₁, and N₁, respectively.

Coming back to the planar limit, it is important to understand that the slope has implications in molecular architecture: a higher slope suggests increased structural condensation. In this sense, samples with predominantly pericondensed structures, resulting from the non-linear addition of benzene units, have petroleomes with planar limit slopes near to 0.90; while samples with catacondensed structures, originated by linear addition of benzene units, have petroleomes with planar limit slopes around 0.75. Likewise, molecular architectures made up of linearly added naphthenic units have planar limit slopes near 0.25 (Cho et al. 2011). In a previous work, there are reported values for the planar limit slopes for Colombian asphaltene samples ranging between 0.57–0.76. Table 4.2 contains the planar limit slopes for compound classes HC, S₁, O₁, and N₁ found in fractions E₅, E₁₀, E₁₅, and E₂₀. There is a clear trend between the time of extraction and the planar limit slope, which

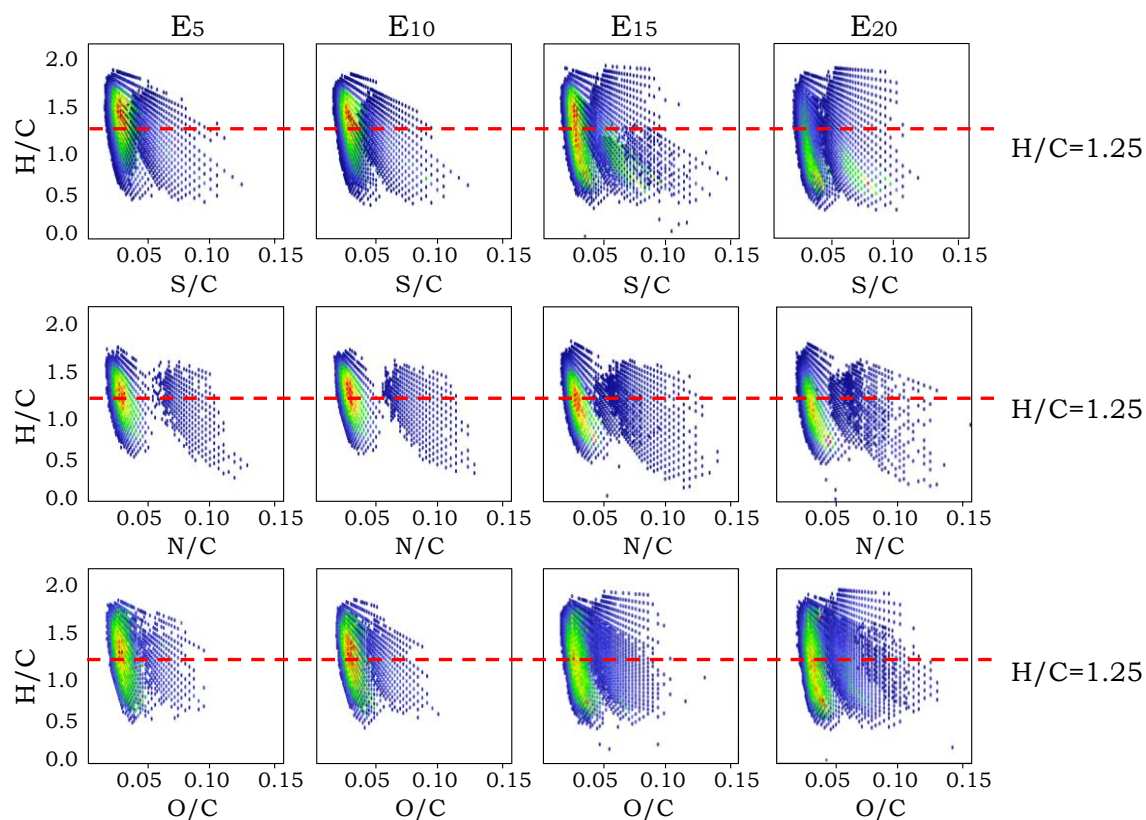
increases as the extraction progresses. Fraction E₂₀ has the highest planar limit slopes and hence, the highest structural condensation along the four fractions, which suggests that the most occluded fraction have some common molecular features with asphaltenes.

Table 4.2. Planar limit slopes for compound classes HC, S₁, O₁, and N₁ in fractions E₅, E₁₀, E₁₅, and E₂₀.

	HC	S ₁	O ₁	N ₁
E ₅	0.549	0.538	0.579	0.458
E ₁₀	0.549	0.581	0.588	0.526
E ₁₅	0.561	0.607	0.621	0.530
E ₂₀	0.608	0.614	0.628	0.558

Interestingly, in every fraction, N₁ compounds exhibit the lowest planar limit slopes and the HC class presents the highest ones. At first sight, this result seems to be in contradiction with the conclusions resulting from the #C/DBE ratios, however, it is important keep in mind that the value of the planar limit slope is a direct result of the molecular architecture. Along this lines, higher planar limit slope and lower aromaticity (or higher #C/DBE ratio) hint HC compounds consisting of a single aromatic core (pericondensed) and long peripheral alkyl side chains; in this sense, higher planar limit slopes could be originated from island type molecular architectures with high condensation. On the other hand, higher aromaticity –or lower #C/DBE ratios for the class N₁, with the lowest planar limit slopes, could suggest archipelago motifs in this family class; such a structure containing several aromatic moieties, interconnected by alkyl bridges, which will show low #C/DBE ratios and low condensation.

Figure 0.5. From top to bottom, Van Krevelen diagrams for S-, N-, and O-compounds in fractions E₅, E₁₀, E₁₅, and E₂₀.



Van Krevelen diagrams are an additional strategy for analyzing high-resolution mass spectrometry data; these diagrams result from plotting H/C *versus* O/C (or N/C or S/C) atomic ratios. Several authors have taken advantage of these plots for graphical differentiation of fossil hydrocarbons from different geological origin and maturity (Wu et al. 2004), molecular comparison between asphaltenes precipitated with different procedures (G. Klein, Kim, Rodgers, A. Marshall, et al. 2006), unraveling the compositional changes in asphaltenes after hydroconversion and thermal cracking processes (Chacón-Patiño et al. 2015), and determining the molecular features responsible of asphaltenes' adsorption on mineral surfaces (Chacón-Patiño et al. 2015a). Figure 4.5 displays the van Krevelen diagrams for the fractions E₅, E₁₀, E₁₅ and E₂₀. The van Krevelen diagrams of the fractions E₅ and E₁₀ show high

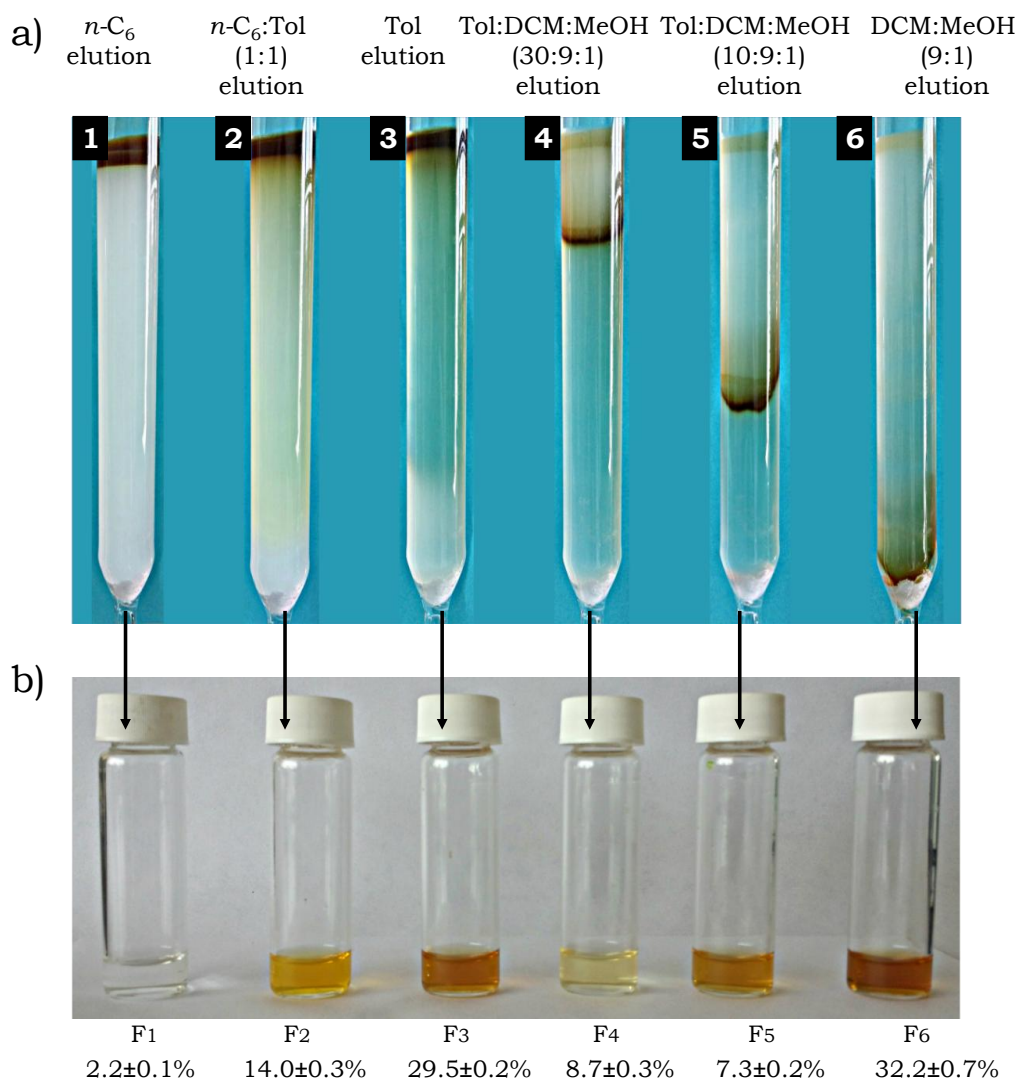
abundance of molecular compositions with H/C ratios above 1.30; while, the fraction E₁₅ has molecular compositions slightly shifted towards lower H/C ratios with an increase of compounds of H/C ratios around 1.25. Finally, the fraction E₂₀ presents the lowest H/C ratios for classes S₁ and N₁ with the highest relative abundances around 1.1. In this regard, Rodgers and co-workers have provided evidence that asphaltenes have characteristic H/C ratios below 1.1 (Mckenna et al. 2013). Along these lines, the high heteroatom content, clustering of compositions along the planar limit, enrichment with molecular species with high aromaticity, H/C ratios around 1.1, and characteristic MWD indicate that fraction E₂₀ is composed (to some extent) by low-molecular weight compounds closely related, structurally, to asphaltenes.

Nowadays a deep understanding of petroleum-derived samples, by high-resolution mass spectrometry, is possible with the use of fractionation methodologies. Several reports have demonstrated that fractionation of fossil hydrocarbons by strategies such as SARA separation (Cho et al. 2012; G. C. Klein et al. 2006), thin layer chromatography (Chacón-Patiño et al. 2015a; Smith et al. 2014), and high performance liquid chromatography (Lababidi et al. 2013; Podgorski et al. 2013), improve the compositional space accessibility in FT-ICR MS analysis. In other words, HR-MS analysis of petroleum fractions allows observation of molecular compositions, which are not accessible when analyzing the whole sample. Observation of additional molecular compositions can be explained from two points of view: fractionation promotes concentration of low abundance compounds present in the unfractionated sample, or elimination of molecules with high ionization efficiencies that mask low ionization efficiency compounds.

To obtain more information on compounds closely related to asphaltenes, fraction E₂₀ was subjected to an additional separation step using normal phase column chromatography with silica as solid support. For this purpose a standard elutropic series of *n*-hexane, toluene, and dichloromethane-methanol 9:1, routinely employed for extracting saturates, alkyl-aromatics, and polars from oil samples, was

used (Bisht et al. 2013; Vela et al. 1995). The additional separation step allowed us to obtain six subfractions from sample E₂₀ (labeled F_n, with n=1–6). These fractions were eluted with the following solvent mixtures: *n*-C₆, *n*-C₆:Tol (1:1), Tol, Tol:DCM:MeOH (30:9:1), Tol:DCM:MeOH (10:9:1), and DCM:MeOH (9:1), which have values of eluent strength on silica (ϵ°) of 0.00, 0.15, 0.29, 0.34, 0.38, and 0.47, respectively. Based on the elutropic series employed, the characteristics of the stationary phase, and the visual progression of the elution process (Figure 4.6a), it is expected subfractions F₁, F₂, and F₃ to be enriched with molecules interacting weakly with the polar SiO₂ surface but strongly with the less polar eluents. In this sense, these fractions should contain low polarity alkyl aromatic compounds with large saturated moieties having steric hindrance around the polar functionalities. Shifting eluents, to the protic mixture Tol:DCM:MeOH, causes desorption of material strongly adsorbed at the top of the column as seen in Figure 4.6a. The mixture Tol:DCM:MeOH is able of interacting, through hydrogen bonding, with the silanol groups at the silica surface hence displacing polar compounds attached to the stationary phase exclusively through hydrogen bonds. It is expected subfractions F₄ and F₅ to have compounds interacting to some extent, with the silica surface; this implies alkyl aromatics with polar functionalities available for hydrogen bonding. Finally F₆, eluted with the most polar eluent mixture (DCM:MeOH), must contain compounds with pyridinic, carboxylic, phenolic and other accessible polar groups able to freely interact with the silica surface (Chacón-Patiño et al. 2015a; Huo et al. 2006; Touchstone 1992). Gravimetric data (Figure 4.6b) indicates that approximately 6% of the sample is irreversibly adsorbed on the stationary phase. Extensive reports on irreversible adsorption of petroleum compounds on mineral surfaces such as alumina, silica, and montmorillonite are available; it is also widely known that asphaltenes adsorb strongly, and some of them irreversibly, on SiO₂ (Adams 2014; Chacón-Patiño et al. 2015a). Molecules having a relative weak interaction with the SiO₂ particles (subfractions F₁, F₂ and F₃) comprise 46% w of the sample, while 48% w of the sample corresponds to eluted compounds interacting strongly with the stationary phase.

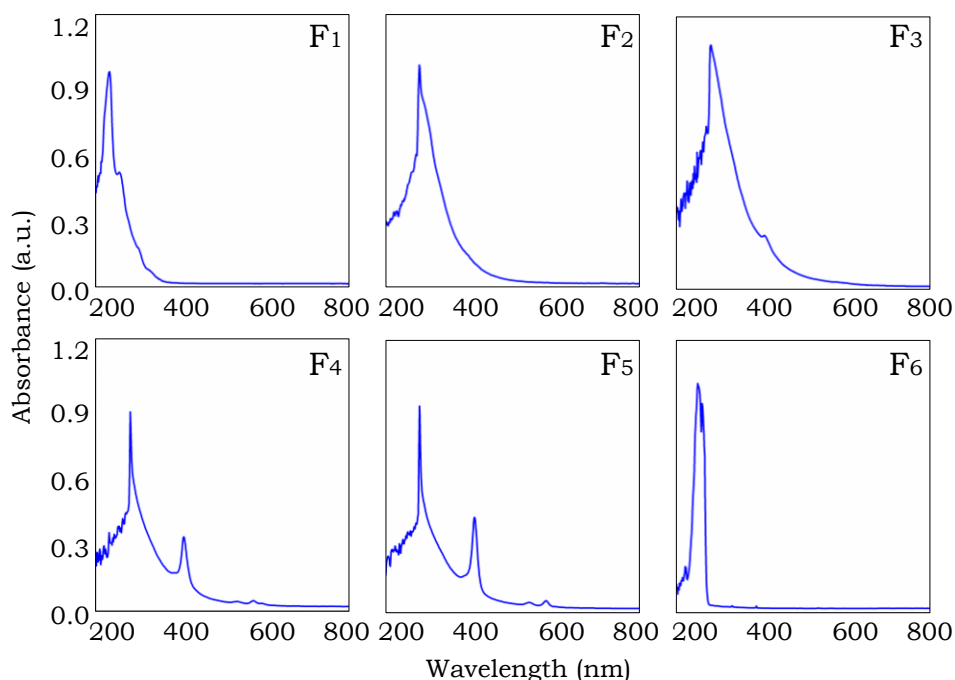
Figure 0.6. Column chromatography of sample E₂₀ including weight percentages of the eluted subfractions.



UV-Vis spectroscopy is indicative of the chemical differences among the sub-fractions. Figure 4.7 shows the UV-vis spectra of samples F₁ through F₆. Fraction F₁, eluted with *n*-hexane, displays an absorption band below 300 nm with maxima around 225 nm and 256 nm. This band is characteristic of hydrocarbons with thiol, sulfide, and disulfide functionalities (Nicodem, Guedes, and Correa 1998).

The advance of the elution process yields sub-fractions F₂ and F₃ with UV-Vis bands shifted towards higher λ — $\lambda_{\text{max}} \sim 300$ which is indicative of sample enrichment with chromophores and aromatic moieties (Yokota et al. 1986). UV-Vis spectra of subfractions F₄ and F₅ present the Soret band, at approximately 408 nm, characteristic of vanadyl porphyrins (Ali et al. 1993b; Qian et al. 2008). It is well-known that petroporphyrins are highly associated to polar crude oil fractions such as asphaltenes; and complexation interactions take place between vanadium or nickel and molecules having N or O functionalities. In this sense, it is possible to suggest high abundance of heteroatomic species in the sub-fractions F₄ and F₅, supporting the presence of metalloporphyrins (Xu et al. 2005; Yang, Hamza, and Czarnecki 2004). Finally, subfraction F₆ exhibits a narrow band between 230–262 nm, peaking at 254 nm, which corresponds to electronic transitions in conjugated aromatics or polyaromatics (El-Bassoussi et al. 2010; Nicodem et al. 1998).

Figure 0.7. UV-Vis spectra of subfractions F₁₋₆.



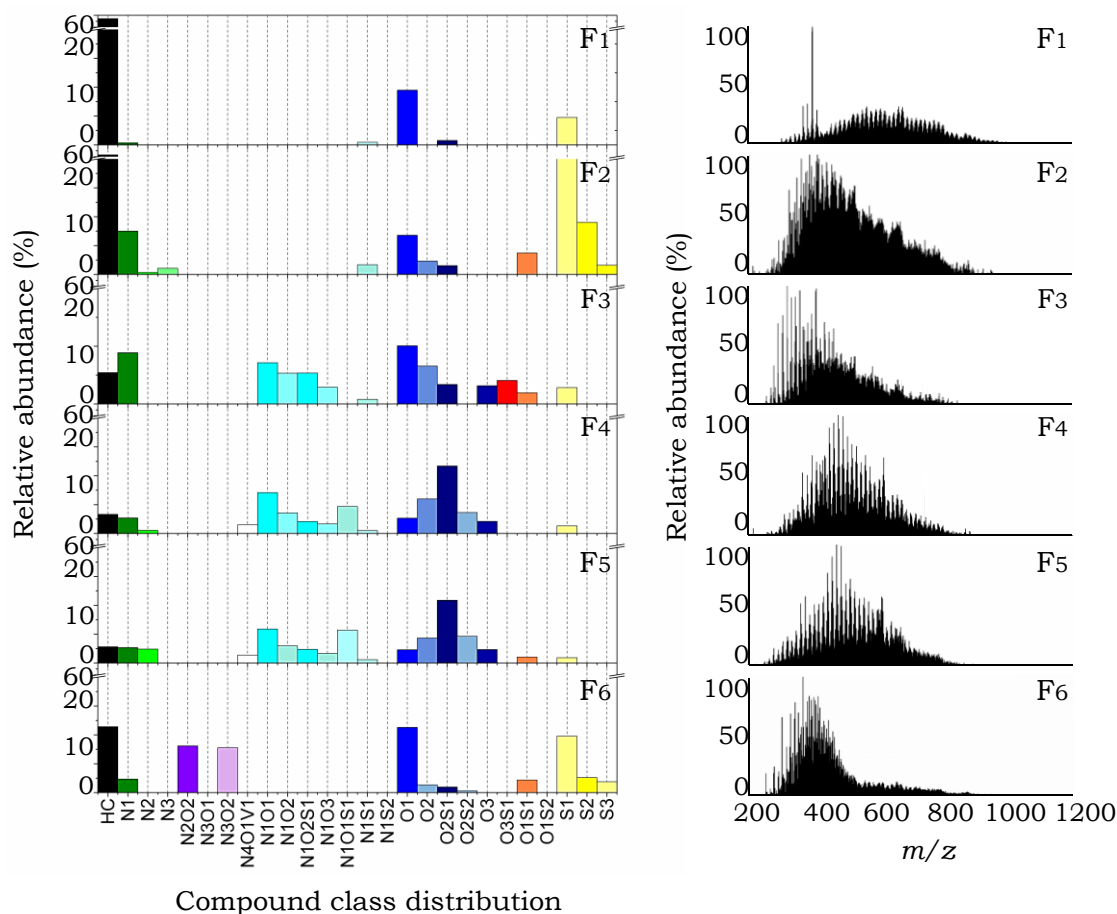
FT-ICR mass spectrometry analysis of samples F₁₋₆ reveals detailed molecular information. Figure 4.8 displays molecular weight distributions and compound class

distributions of samples F₁₋₆ and Figure 4.9 shows the contoured plots of DBE *versus* carbon number. As expected sample F₁, eluted with *n*-hexane, exhibits predominantly compound classes HC (59.5%), O₁ (9.5%) and S₁ (5.7%), with little abundance of classes N₁, N₁S₁ and O₂S₁. MWD for F₁ ranges from *m/z* 300 to 1000 and the mass spectrum reveals prominent signals around *m/z* 400, particularly at *m/z* 396.375046 and *m/z* 410.390665. These signals correspond to molecular compositions with DBE 6 and molecular formulas C₂₉H₄₈ and C₃₀H₅₀; the later could correspond to squalene. Early works (Rubinstein, Spyckerelle, and Strausz 1979) showed that the asphaltenic material is a valuable source of geochemical information, and occlusions can contain biomarkers such as pristane and phytane, squalene (C₃₀H₅₀), steranes and hopanes (Ekweozor 1984). As the most common triterpenoid compound in all life forms, squalene is the precursor of important groups of biomarkers, like steranes and hopanes, and it has been widely found in crude oil asphaltenes (Ekweozor 1984). In our case the protective environment of the macro-porous asphaltene's structures could prevent thermochemical alteration of labile biomarkers such as squalene (Gray et al. 2011). Additionally, Figure 4.9 shows the saturated nature of the subfraction F₁, whose petroleome extends along DBE 4–15 and carbon numbers 30–60. Weighted averages of C#/DBE ratios for this subfraction are: 6.10, 4.50, 4.70, and 2.65 for the classes HC, S₁, O₁ and N₁, respectively. These results indicate that fractionation expands the observable compositional space, enabling the detection of highly saturated compounds, which were not detected when analyzing the whole Fraction E₂₀.

As the polarity of the eluent increases, heteroatom content and DBE in the fractions change. Sample F₂, eluted with *n*-hexane:toluene, consists mostly of compound class HC (35%) and molecular compositions with at least one heteroatom (65%) corresponding to N₁, N₂, N₃, N₁S₁, O₁, O₂, O₁S₁, S₁, S₂ and S₃. It is important to note that the toluene dipole moment and its aromatic character make possible the elution of compounds with higher DBEs, specifically alkyl–aromatics with high heteroatom content. As Figure 4.9 indicates, petroleomes of sub-fraction F₂ exhibit well-defined

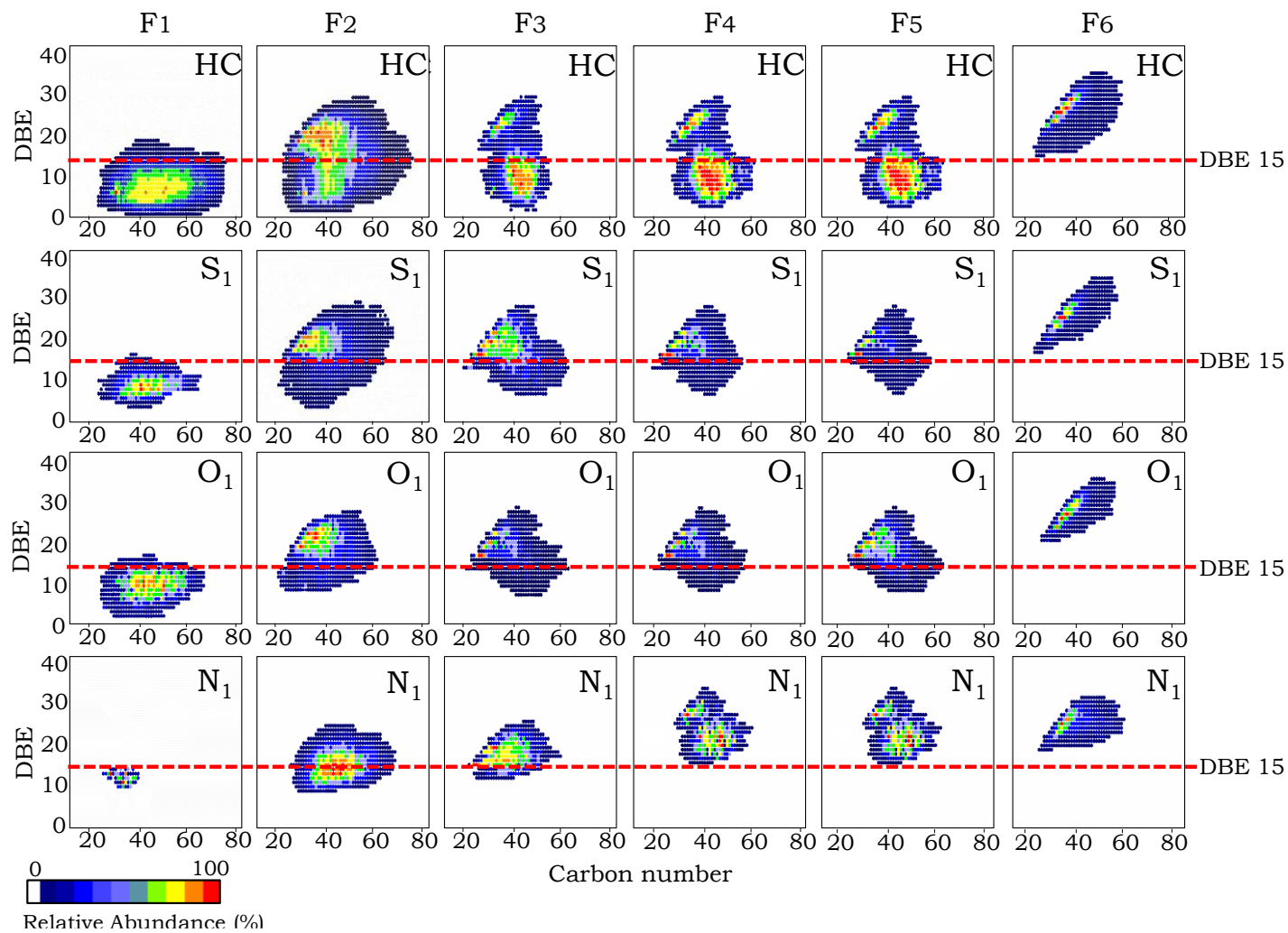
planar limits, which is also characteristic of aromatic fractions from heavy crude oils (Cho et al. 2012, 2011).

Figure 0.8. Compound class distributions and molecular weight distributions for samples F₁₋₆.



In this fashion, petroleomes of subfraction F₂ extend along carbon numbers 25–50, and the following DBEs ranges: for the class HC, DBEs 2–25; for the classes S₁ and O₁, DBEs 15–25; and for the class N₁, DBEs 10–20.

Figure 0.9. Contoured plots of DBE *versus* carbon number for subfractions F₁₋₆, resulted from the column chromatography separation carried out on the fraction E₂₀.



Along these lines, one could say that the most abundant HC alkyl–aromatics in subfraction F₂ could have at least five fused aromatic rings (DBE 15), and up to 25 carbon atoms in alkyl–side chains. These DBE and C# translate into weighted averages of C#/DBE ratios of 2.88, 2.24, 2.16, and 2.84 respectively, indicating higher aromaticity, when compared to the sub–fraction F₁.

As the chromatographic separation proceeds subfractions F₃, F₄, and F₅ are eluted with Tol, Tol:DCM:MeOH 30:9:1, and Tol:DCM:MeOH 10:9:1. These subfractions are enriched in alkyl–aromatic compounds with high heteroatom content of the type: HC, N₁, N₂, N₁O₁, N₁O₂, N₁O₃, N₁O₁S₁, N₁O₂S₁, N₁S₁, O₁, O₂, O₃, O₂S₁, O₂S₂, and S₁. The mass spectrum of subfraction F₃ features prominent peaks at low m/z values, between $250 < m/z < 400$, corresponding to both radical and protonated cations of N₁ compounds. Contoured plots of DBE *versus* carbon number for class N₁ reveal the high aromatic character of this compound class in subfraction F₃ (Figure 4.9). It is evident that the distribution of N₁ compounds in subfraction F₃ is shifted toward higher DBE values, when compared with subfraction F₂. Interestingly the most abundant N₁ compositions in F₃ are clustered at the planar limit. Careful examination of the N₁ class indicates higher aromaticity than the previous two subfractions: DBE values have increased while carbon numbers decrease. The weighted average of C#/DBE ratio for the class N₁ is 2.06, while for the classes HC, S₁, and O₁ are 1.65, 2.11, and 2.12, respectively.

Classes HC, S₁, and O₁ in subfraction F₃ display a particular feature, which is more evident for the class HC, the petroleome for these compound classes has two well–defined regions separated by a compositional “gap”: one corresponding to maltene–like distributions, located along DBEs 2–18 and carbon numbers 30–55; and another one corresponding to more aromatic species or asphaltene–like molecules, whose compositions are clustered in a sort of planar limit “domain”. As discussed before, fraction E₂₀ consist of a mixture of low–molecular weight asphaltenes and maltenes with high heteroatom content. It is believed that

heteroatom-containing maltenes are responsible for transferring low-MW asphaltenes into the *n*-heptane phase, as a consequence of strong interactions between polar and aromatic moieties in those maltenes with the asphaltene core (Chang and Fogler 1994; Wang and Buckley 2003). In a similar way, Rodgers and co-workers reported the molecular composition of a high-boiling fraction from a Middle Eastern heavy oil, and observed petroleomes having three regions: the asphaltene domain, the maltenic space and the gap between them (Podgorski et al. 2013). The compositional gap may be attributed to molecular species that do not ionize efficiently.

Subfraction F₃ shows high abundance of classes O₁, O₂, and O₃. Generally, these moieties are associated with carboxylic acids; however, here they are eluted with toluene, which might seem counterintuitive. Even though an extra effort is needed to ascertain their molecular structure, for instance by combining HR-MS and specific fractionation techniques (Lobodin et al. 2015; Rowland et al. 2014), several reports suggest that carboxylic acids in crude oils are characterized by their low aromaticity, exhibiting DBEs between 2–7 (Lalli et al. 2015; Rowland et al. 2014; Vaz et al. 2013). With the evidence at hand, we hypothesize that oxygen in subfraction F₃ must be part of aromatic moieties, which is in agreement with elution with toluene.

On the other hand, and as anticipated from the UV-Vis spectroscopy analysis, the class N₄O₁V₁ (vanadyl porphyrins) was detected in subfractions F₄ and F₅ (Figure 4.8). It is possible to hypothesize that the presence of abundant N- and O-containing species in sub-fractions F₄ and F₅ “solvates” the vanadyl porphyrins, via complexation interactions. In both sub-fractions, the most abundant homologous series of vanadyl porphyrins correspond to etio-porphyrins with DBE=17 (McKenna et al. 2009). The mass spectra of subfractions F₄ and F₅ present similar distributions, starting at *m/z* 250 and ending at *m/z* 850, and centered on *m/z* 480. These subfractions have weighted average of C#/DBE of 1.65, 2.10, 2.12, and 1.82 for the

classes HC, S₁, O₁, and N₁, respectively; and their petroleomes also present the compositional gap between the maltene and the asphaltene zones.

The experimental evidence suggests that subfractions F₃, F₄ and F₅ are a mixture of occluded maltenes and highly aromatic species, probably low-molecular weight asphaltenes. Since asphaltenes are soluble in toluene, elution with this solvent (to obtain subfraction F₃) releases low-molecular weight asphaltenes interacting weakly with the stationary phase. Increasing the eluent's polarity, as in the mixture Tol:DCM:MeOH, promotes the elution of molecular species interacting strongly with the silica surface. The Tol:DCM:MeOH mixture forms a layer on the SiO₂ surface via hydrogen bonds, displacing molecular species interacting strongly with silica. Seeing that N-containing and high aromatic hydrocarbons interact strongly with minerals such as SiO₂ and Al₂O₃, via classical and non-classical hydrogen bonds, respectively, the observation of high-DBE N-compounds in subfraction F₅ is validated. Finally, sub-fraction F₆, eluted with DCM:MeOH (9:1), exhibits a broad bimodal MWD extending between 220 < *m/z* < 800. The F₆ fraction show compound classes HC, N₁, N₂O₂, N₃O₂, O₁, O₂, O₁S₁, O₂S₁, O₂S₂, S₁, S₂, and S₃; this distribution is very similar, in terms of compound classes, to that of sub-fraction F₂. However, from the point of view of DBE *versus* carbon number sub-fraction F₆ is unique because it features petroleomes extending throughout DBEs 18–30 and carbon numbers between 25–60, with molecular compositions clustered along the planar limit. For F₆ the weighted averages of C#/DBE ratios are 1.33, 1.30, 1.31, and 1.50 for the classes HC, S₁, O₁, and N₁, respectively, indicating the highest aromaticity of all tested fractions.

In previous works, it is reported the petroleomes of Colombian asphaltenes, by APPI FT-ICR mass spectrometry, consisting of abundant compositions clustered along the planar limit region, extending along DBEs 20–35 and carbon numbers between 30–65. In another report, Rodgers and co-workers stated that the asphaltene's compositional space extends along DBEs 20–35 and carbon numbers between

30–65 with relative high abundance of compositions along the planar limit. In this sense, the compositional space of sub-fraction F_6 seems to complement the asphaltene's petroleome. In other words, subfraction F_6 is made up of low-molecular weight asphaltenes. The mixture DCM:MeOH allows the elution of highly polar molecular species with abundant heteroatoms, high aromaticity, and low steric hindrance that interact strongly with the SiO_2 surface.

Table 4.3. Planar limit slopes for the classes HC, S_1 , O_1 , and N_1 , in sub-fractions F_{1-6} .

	HC	S_1	O_1	N_1
F_1	0.527	Undef.	Undef.	Undef.
F_2	0.726	0.617	0.715	0.346
F_3	0.742	0.659	0.719	0.456
F_4	0.739	0.750	0.723	0.471
F_5	0.748	0.737	0.721	0.480
F_6	0.791	0.765	0.741	0.639

Finally, analysis of planar limits slopes suggests that as the separation progress, the fractions become enriched with condensed structures (Table 4.3). For instance, the HC compound class in sub-fraction F_1 presents the lowest planar limit slope (0.527) for the class; the planar limit increases as the extraction proceeds, reaching a maximum of 0.791 in fraction F_6 . This increase suggests selective extraction of more condensed structures as the eluent increases in polarity, which makes sense considering that only highly aromatic and condensed structures with low steric hindrance will be retained by the SiO_2 support and could be only eluted with the most protic solvent mixture. If this is the case, a high planar limit slope should be expected for the HC family in F_6 , which in fact is observed. Another interesting observation comes from analysis of the N_1 compound family. The N_1 class has the lowest planar limit slopes for all the fractions, from 0.346 in F_2 to 0.639 in F_6 , suggesting the lowest structural condensation. However, as one goes from F_5 to F_6 the change in slope for the N_1 class is dramatic: from 0.480 to 0.639. This suggests that N-containing structures with high molecular condensation display the strongest interaction with the silica in fraction F_6 (Parida et al. 2006); and once again only the highest polarity

eluent is able to extract them. In addition, these compounds were not observable in fraction E₂₀, only after fractionation they were detected which indicates that separation techniques are fundamental to solve the molecular puzzle of petroleum. It is also important to highlight that these planar limit slopes satisfy the 90% rule, which states that the maximum DBE value for a petroleum hydrocarbon cannot exceed the 90% of its carbon number. Some groups working on asphaltenes' characterization by FT-ICR MS have reported planar limit slopes between 0.600–0.900 (Hsu, Lobodin, et al. 2011; Podgorski et al. 2013; Purcell et al. 2010). As for Colombian asphaltenes, in a previous work we reported 0.600–0.670 for the studied classes here (Chacón-Patiño et al. 2015).

4.5 CONCLUSIONS

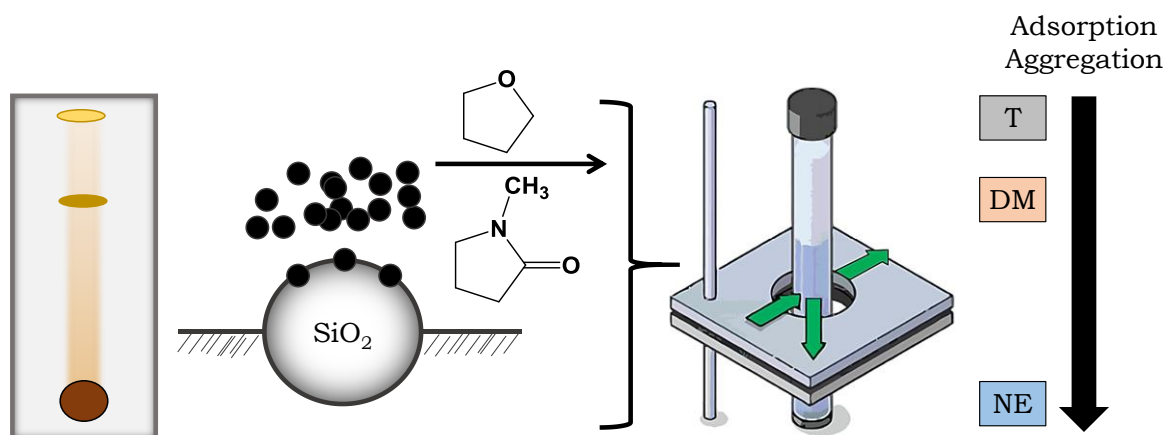
Asphaltene precipitation by addition *n*-heptane promotes occlusion of a wide variety of compounds, including saturated hydrocarbons and alkyl aromatics with high heteroatom content. As the Soxhlet extraction proceeds the fractions become more aromatic and with increased structural condensation. The strongly occluded fraction (E₂₀), further fractionated by column chromatography, is a combination of ~2% of saturated hydrocarbons with little structural condensation and some heteroatom-containing compounds (mostly HC, S₁ and O₁ classes); ~14% of alkyl aromatics with up to 3 heteroatoms (classes S₁, S₂, S₃, O₁, and O₂, among others); ~46% of alkyl aromatics with high heteroatom content (e.g. classes N₁O₃ and N₁O₂S₁); few vanadyl porphyrins; ~32% of low molecular weight asphaltenes with high degree of structural condensation and compound families with up to 5 heteroatoms (e.g. N₃O₂); and ~6% of compositions irreversibly adsorbed on the stationary phase. Although *per definition* asphaltenic species are classified as the *n*-heptane insoluble fraction of petroleum, the presence of "small" asphaltenes in the *n*-heptane phase is not inexplicable, since Strausz and others have reported the transfer of asphaltenes-like compounds to the maltenic phase. Thus, the experimental evidence presented in

this contribution suggests what others have said: the asphaltene concept must be revisited. Commonly, the word asphaltenes brings to mind highly condensed aromatic structures, in most of the cases with island architecture. However, looking our results in the light of the supramolecular model of asphaltene aggregation, it is possible to say that asphaltenes not only consist of aromatic bridged structures having a wide diversity of functional groups, they also include alkyl aromatics with high heteroatom content, which will also contribute to stabilize asphaltenes' aggregates. In this sense, the new definition of asphaltenes must take into account molecular features such as H/C ratio, functional groups, degree of condensation and molecular architecture.

5. THE ROLE OF MOLECULAR COMPOSITION IN ASPHALTENE AGGREGATION

*Redrafted from: *Martha L. Chacón-Patiño, Cristian Blanco-Tirado, and Marianny Y. Combariza. Submitted for publication in Energy and Fuels*

Figure 0.1 Graphical abstract for *The Role of Molecular Composition in Asphaltene Aggregation*.



5.1 ABSTRACT

Asphaltene aggregation and deposition are one of the most challenging issues in crude oil production, transporting, compatibility, and refinery. Thus, understanding the molecular basis of the aggregation phenomena could guide the rational design of methodologies that help to solve this problem. In this contribution, we use the Turbiscan Lab to follow the stability of the dispersions of high-performance thin layer chromatography asphaltene subfractions. For this purpose, three asphaltene samples were fractionated on HPTLC SiO₂ plates, in alkyl-aromatics, polars, and non-eluted compositions. Results of the model aggregation tests of the asphaltene

samples and the subfractions allow concluding that asphaltene adsorption and aggregation behavior are closely related. In this sense, the same molecular features responsible for asphaltene adsorption on mineral surfaces seem to play important roles in asphaltene aggregation. In other words, a synergy between aromaticity, N and O content, and polar group accessibility is the crucial factor in both adsorption and aggregation processes. Besides demonstrating the correlation between polarity and aggregation, it is shown that simply asphaltene fractionation on SiO₂ separates challenging asphaltenes (more prone to aggregate) from stable asphaltenic compositions.

5.2 INTRODUCTION

As the worldwide oil supply shifts towards heavy crude oils, research in asphaltene chemistry is increasing dramatically (Akbarzadeh et al. 2007). Even though, along all the SARA petroleum fractions, asphaltenes are probably the most investigated, but certainly the least understood components (Mullins et al. 2012). Starting with their inaccurate definition, rephrased as the crude oil components insoluble in *n*-alkanes such as *n*-heptane or *n*-pentane, but soluble in aromatic solvents such as toluene or benzene, asphaltenes are ultra-complex and polydisperse (Strausz et al. 1992). It is the interpretation of asphaltenes as a solubility class rather than as a compound class, the cause of misunderstandings in their molecular characteristics (Yokota et al. 1986). For instance, for more than five decades –from the 1950s to 2000s, asphaltenes were viewed as big island-type structures composed of at least fifteen fused aromatic rings with long alkyl side chains, having molecular weights up to 100,000 g/mol. It is important to bear in mind that this molecular sketch of asphaltenes does not meet the known chemistry of petroleum (Gray et al. 2011; Mullins 2011).

Recently, the molecular picture of asphaltenes has been changing, and the new proposed perspective is in agreement with the experimental attributes of the $n\text{-C}_7$ insoluble species (Ancheyta, Trejo, and Rana 2010b; Gray et al. 2011; Schuler et al. 2015). Molecular behavior such as strong aggregation, (Gray et al. 2011; Murgich 2002) adsorption on mineral surfaces (e.g. silica, quartz, and alumina), occlusion inside asphaltene macrostructures (Derakhshesh et al. 2013; Liao et al. 2004), porosity of asphaltene aggregates (Derakhshesh et al. 2013), formation of films at oil–water interfaces (Clingenpeel et al. 2015), interactions with resins and surfactants (Andersen and Speight 2001), and asphaltene upgrading products (Chacón-Patiño et al. 2015b; Karimi et al. 2011; Rueda-Velásquez et al. 2013) support the newborn molecular view of asphaltenes. In this sense, the new description of asphaltenes suggests an ultra-complex mixture without a particular molecular motif, but including bridged aromatic moieties with multiple functionalities accessible for multiple intermolecular associations. In this sense, acid–base interactions between carboxylic acids and nitrogen bases, hydrogen bonding promoted by hydroxyl and amine groups, metal complexes through axial coordination between nitrogen bases and vanadium or nickel, association of saturated moieties without heteroatoms to produce hydrophobic pockets, and $\pi\text{--}\pi$ stacking between aromatic cores are the molecular interactions responsible for asphaltene aggregation (Gray et al. 2011; Murgich 2002). All these associations combine following a supramolecular assembly model to produce strong macroaggregates. Unluckily, it is the strong asphaltene aggregation and the subsequent deposition, one of the most challenging issues in production facilities, transporting systems, crude oil compatibility and refinery processes (Juyal et al. 2005; Mendoza de la Cruz et al. 2009).

Regarding the causes of asphaltene deposition, it is important to have into account several points: first, dead crude oils consist of liquids (saturates, aromatics, and resins) and dissolved solids (the asphaltenes). Like in any dissolution, several variables affect the solubility of the solids. In crude oils, asphaltene solubility is

affected by temperature, pressure drop and compositional changes in the petroleum matrix. Second, at extremely low concentrations in toluene (model conditions), below $\sim 50 \mu\text{g/mL}$, asphaltene already exist as dimers and trimers. Beyond $\sim 50 \mu\text{g/mL}$ these dimers and trimers start producing nanoaggregates. At concentrations higher than $200 \mu\text{g/mL}$, these nanoaggregates go through further association to form clusters, and at this point, slightly changes in any of the variables may induce asphaltene deposition (McKenna et al. 2013; Morimoto et al. 2015). In crude oils (real samples), the picture is less clear, because of the presence of saturates, aromatics, and resins. In some heavy oils, asphaltenes can exist as a stable dispersion at even extremely high concentrations without precipitating. In fact, several heavy oils present up to 15 wt% of asphaltenes and do not exhibit asphaltene deposition issues in production processes. On the other hand, in light crude oils with little asphaltene content $>1\%$, asphaltene precipitation is one of the main concerns in production facilities. The reason is simple, heavy oils usually contain more aromatics and resins; both fractions enhance asphaltene solubility; otherwise, light crude oils are enriched with saturates, compounds decreasing asphaltene stability (Andersen and Speight 2001; Koots and Speight 1975; Speight 2004).

Along these lines, one of the main objectives of crude oil production and transporting is to avoid asphaltene deposition, which is only possible by a comprehensive understanding of the structure and composition of crude oils. In the early 2000s, Petroleomics emerged as the counterpart of genomics, but applied in petroleum chemistry, being aimed to predict crude oil behavior based on the detailed knowledge of the molecular composition (Hsu, Hendrickson, et al. 2011; Rodgers et al. 2005). This young branch of analytical chemistry is based on Fourier transform ion cyclotron resonance mass spectrometry (FT-ICR MS), an essential tool that makes possible the elemental assignment of the thousands of compositions present in a single sample of crude oil (Marshall and Rodgers 2004). In previous works, we reported on the fractionation of asphaltenes by high-performance thin layer chromatography (HPTLC) and the FT-ICR mass spectrometry characterization of

the asphaltene subfractions (Chacón-Patiño et al. 2015a). Correlations between asphaltene adsorption degree and molecular features such as aromaticity, heteroatom/C ratios, and degree of alkylation (or steric hindrance) were concluded. For instance, HPTLC fractionation of purified asphaltenes yielded three individual fractions, with retention factors of 0, ~ 0.69 , and ~ 0.92 . Atmospheric pressure photoionization coupled to FT-ICR MS showed that polar non-eluted compounds, strongly adsorbed on the SiO_2 surface, exhibit molecular compositions with N_nO_o and O_o compound classes, the lowest H/C ratios (< 0.90) along all sub-fractions and little steric hindrance (< 11 carbon atoms in alkyl side-chains). On the other hand, the weakest retained compounds or toluene-eluted, consist mostly of S-containing compositions, presenting higher saturation degree ($0.8 < \text{H/C} < 1.6$) and higher steric hindrance (up to 25 carbon atoms in alkyl side-chains). Our earlier report suggests a synergy between aromaticity, steric hindrance, and heteroatom content as the crucial factor in asphaltene adsorption on mineral surfaces (Chacón-Patiño et al. 2015a). Interestingly, comprehensive reviews on asphaltene adsorption suggest that the same forces driving asphaltene interaction with mineral surfaces must play important roles in asphaltene aggregation and subsequent deposition. Thus, in this contribution, Turbiscan Lab was used to follow the stability of asphaltenes' dispersions made up of the thin layer chromatography asphaltene subfractions from three Colombian crude oils, with distinctive API grades (8.2, 15.0, and 28.0). It was found that the three asphaltene subfractions exhibit a particular behavior in aggregation tests. The most polar or the strongest adsorbed compounds, presenting little steric hindrance and the lowest H/C ratios, exhibit the strongest tendency towards aggregation. On the other hand, the dispersions of the lowest polarity asphaltene subfractions, present the highest stability. Even though, the medium polarity asphaltene subfraction exhibits higher stability when compared with the aggregation of the whole asphaltene sample. Based on the molecular composition of the HPTLC subfractions, achieved by FT-ICR MS and previously published, it was established that aromaticity, N and O content, and polar group accessibility are the crucial parameters in asphaltene aggregation. This knowledge is valuable from two

points of view: we have demonstrated that asphaltene adsorption and aggregation are intimately correlated, and we proved that selective separation of the most polar and hence the most problematic asphaltenes is possible. In this sense, this knowledge could guide the rational design for the selective extraction of challenging asphaltenes from the crude oil matrix.

5.3 EXPERIMENTAL SECTION

5.3.1 Materials. Toluene (Tol), *n*-heptane (*n*-C₇), *n*-hexane (*n*-C₆), dichloromethane (DCM), tetrahydrofuran (THF), *N*-methylpyrrolidone (NMP), and methanol (MeOH) Chromasolv[®] solvents (99.9%) were purchased from Sigma-Aldrich (St. Louis, MO, USA). High-performance thin layer chromatography SiO₂ plates 60 sorbents (particle size of 4–8 μm with aluminum backing and pore size of 60 Å) were purchased from Merck Millipore (Darmstadt, Germany).

5.3.2 Asphaltene samples. Asphaltenes were isolated from three Colombian crude oil samples: extra-heavy, heavy and medium crude oils, with respective API grades of ~8.2, ~15.0, and ~28.2, labeled as crude oils #1, #2, and #3. We followed the ASTM standard methodology D6560–12 with slight modifications. Thus, crude oil/*n*-heptane ratio was selected at 1/40. For instance, 400 mL of *n*-C₇ were added dropwise (1 mL/minute) to 10 g of crude oil, under sonication (Branson Ultrasonics, Danbury, CT, 22 kHz, and 130 W) at 60 °C. After that, the mixture was refluxed for 90 minutes at 95 °C, and then it was allowed to stand for 12 hours. Subsequently, asphaltenes were collected by filtration (Whatman filter paper grade 42), placed in a Soxhlet apparatus, washed with hot *n*-heptane (average temperature at the thimble was 60 °C), until the cleaning solvent was clear (~72-150 hours). Asphaltenes were recovered by dissolution in hot toluene (~98 °C), which was finally dried under N₂ flow to produce solid asphaltenes.

5.3.3 Asphaltene purification. Asphaltene purification was carried out following a method for extracting occluded hydrocarbons, previously described in chapter 4. In short, 1 g of clean *n*-C₇ asphaltenes was macerated using an agate mortar until reaching a particle size of 38 µm (400 mesh) and placed in a Soxhlet apparatus equipped with *n*-heptane. After five hours of solvent recirculation, the *n*-heptane with the extracted hydrocarbons was taken, kept for further experiments and replaced for clean *n*-heptane. Meanwhile, the remnant asphaltene sample was again macerated and subjected to Soxhlet extraction with clean *n*-heptane for another 5 hours. This operation was repeated two more times until completing 20 hours of cleaning. After this washing time, no significant material was extracted from the asphaltene sample (>0.1 mg/200 mL heptane).

5.3.4 Asphaltene fractionation by High-Performance Thin Layer Chromatography and asphaltene recovery. The HPTLC fractionation method is previously described in Chapter 2 and reported. In brief, 2 µL of a purified asphaltene stock solution (5000 ppm) was directly applied to presaturated HPTLC SiO₂ plates. Asphaltene fractionation was performed by sequential elution of the purified sample using an elutropic series of solvents to separate saturates, alkyl aromatics, and polars in petrochemical samples. The elutropic series consists of *n*-hexane, toluene, and 90/10 v/v of DCM/ MeOH. The elution process was carried out several times to collect enough sample for the aggregation tests.

In a previous report, we determined the optimized conditions for asphaltene recovery from the silica plates.(Martha L. Chacón-Patiño et al. 2015a) Thus, the stationary phase with the asphaltene subfractions was removed from the aluminum plate and combined with THF/NMP 90/10 v/v and sonicated for 60 minutes. After that, the mixture was centrifuged for 20 min at 4000 rpm (SiGMA 4- 16KS, Osterode am Harz, Germany). Finally, the supernatant was collected, dried under N₂ flow and weighted. This procedure was carried out several times to collect enough material for the aggregation tests.

5.3.5 Aggregation tests. The aggregation tests were performed under model conditions; that is in mixtures of *n*-heptane and toluene (HepTol). We prepared toluene solutions of the whole asphaltenes, the samples WD (resulting from desorbing unfractionated asphaltenes from the SiO₂ plates) and the HPTLC subfractions from the three Colombian crude oils, at 1 mg/mL. Asphaltene solutions were mixed with *n*-heptane 70% v/v, vortexed at 1000 rpm for 3 seconds and placed immediately in the Turbiscan Lab (Formulaction, L'Union, France). The aggregation behavior of each sample was followed during 90 minutes; every test was carried out per triplicate. Additional aggregation studies were performed on the non-fractionated asphaltenes sample, spiked aromatics and resins from the respective crude oils.

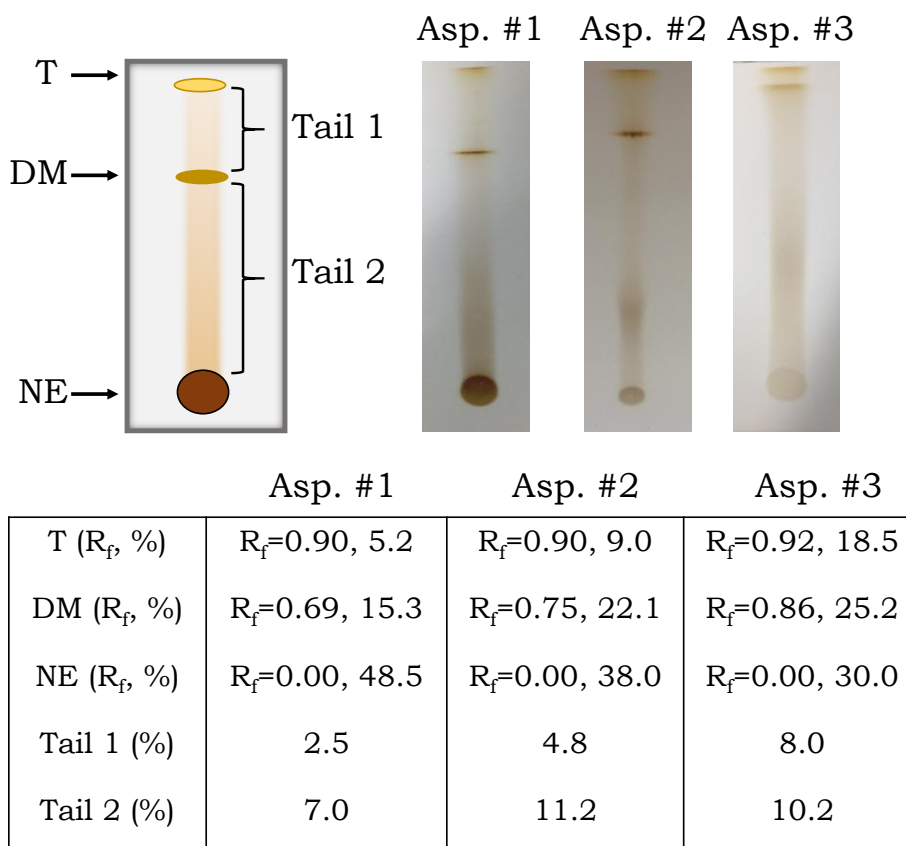
5.4 RESULTS AND DISCUSSION

Separation of asphaltenes on the HPTLC surface is a direct consequence of the intermolecular interactions occurring between the sample, the SiO₂ surface, and the mobile phases. The surface of silica consists of various kind of active groups: silanols –OH are referred as polar functionalities, responsible for solute retention by interaction through hydrogen bonds. On the other hand, siloxanes [-O-Si-O-] are known for being hydrophobic, without significant effects on analyte retention by polar interactions (Nawrocki 1991). Along these lines, the high polarity of the silica surface is a consequence of high content of silanols, in vicinal, geminal or single configurations, and also hydrated silanols –complexes consisting of SiOH-H₂O when the silica is hydrated (Shen et al. 1980). The active groups of the SiO₂ surface can interact strongly with several functionalities found in petroleum compounds, such as carbonyl carbons, fused aromatic moieties, basic nitrogen (pyridine or amine groups), and metal complexes such as vanadyl and nickel porphyrins (Adams 2014; Zahabi et al. 2012). Regarding the mobile phases, we used a standard method for

separating petroleum derived samples into saturates, alkyl aromatics and polars. Consequently, elution with *n*-C₆, having a with eluent strength $\epsilon_0=0$, did not promote the migration of significant amount of material on the SiO₂ surface. This result is in agreement with the asphaltene definition and moreover, supports the purification process described in detail in Chapter 4.

The second eluent solvent is toluene, which presents an eluent strength (ϵ_0) of 0.29, and causes the migration of an asphaltene fraction with retention factors around $Rf_1=0.90\pm0.02$, $Rf_2=0.90\pm0.04$, and $Rf_3=0.92\pm0.03$, for the asphaltenes #1, #2, and #3, respectively. Gravimetric results of the fractionation processes, presented in Figure 5.2, indicate an inverse relationship between the API grades of the crude oil, from where the asphaltenes were extracted, and the amount of material eluted with toluene. It is important to have into account that compounds in T fraction likely consist in alkyl-aromatics species with polar moieties presenting high steric hindrance, interacting weakly with the SiO₂ surface, but strongly with the aromatic eluent toluene. Thus, this observation suggests that asphaltenes from the medium crude oil present higher amount of “weak polar” compositions, when compared with asphaltenes from the extra heavy crude oil. There are few reports in literature about the comparison between the molecular compositions of asphaltenes from light, medium, heavy and extra-heavy crude oils. Even though, the few existing reports do not suggest clear compositional differences among asphaltenes from different geological origins and their correlations with aggregation (Trejo et al. 2004).

Figure 0.2. High-performance thin-layer chromatography fractionation of asphaltenes from three different Colombian crude oils, with their respective retention factors (R_f) and the weight percentages.



Meanwhile, elution with DCM/MeOH, with eluent strength of 0.47, yields a fraction with medium affinity for the SiO_2 surface. Specifically, DM subfractions of asphaltenes #1, #2, and #3 have $R_{f1}=0.69$, $R_{f2}=0.75$, and $R_{f3}=0.86$, respectively; and the gravimetric results indicate an inverse relationship between the amount of material in DM fractions and the API grades of the crude oils. According to the mixture of solvents used for the elution of this fraction, the chemistry of the SiO_2 surface and the molecular composition we previously reported (Chacón-Patiño et al. 2015a) we hypothesize that DM fraction consist of alkyl-aromatic species with high content of heteroatoms, organized in compound families such as N_nO_o , which must exhibit a molecular structure characterized by the accessibility of the polar

functionalities for interacting with the SiO₂ surface exclusively through hydrogen bonds (Badjic and Kostic 2000; Badjić and Kostić 2001). Finally, all the asphaltene samples yielded a non-eluted subfraction, with a retention factor of zero. Interestingly, gravimetric results suggest a direct correlation between crude oil API grades and the amount of material in this fraction. Specifically, for the asphaltenes #1, #2, and #3, NE fractions consist of 48.5 wt%, 38.0 wt%, and 30.0 wt% of the sample. Non-eluted fraction is characterized by high polarity compositions, presenting the lowest H/C ratios and least steric hindrance when compared with the subfractions T and DM.

There are few reports concerning the molecular characterization of asphaltene subfractions from different crude oils. For instance, Kaminski *et al.* (Kaminski et al. 2000) fractionated asphaltenes from diverse geological origins, by differential solubility. For this purpose, asphaltenes were dissolved in dichloromethane and precipitated by sequential addition of different amounts of *n*-heptane (70%, 75%, 80%, and 90%). The asphaltene fraction precipitated with the least volume of *n*-C₇ corresponds to the most polar species and vice-versa. Kaminski and co-workers reported that the most polar asphaltene subfractions are distinguished by high content of metals such as V, Fe, and Ni, when compared with the other subfractions and the whole asphaltenes. Importantly, re-dissolution test applied to the asphaltene subfractions indicated an inverse relationship between polarity and dissolution rates, suggesting that polarity has an important role in asphaltene aggregation. This report supports our earlier findings regarding the important role of polarity in both adsorption and aggregation.

Thus, we selected the Turbiscan Lab for following the aggregation behavior of the dispersions of the whole asphaltenes, the samples WD and the HPTLC subfractions because of its suitability for studying the stability of concentrated and opaque solutions (Mengual et al. 1999). In few words, the Turbiscan tool works upon a turbidimetric principle, measuring the transmitted light and the backscattering of the

dispersions. Containing a pulsed near infrared light source ($\lambda=850$ nm), which moves up and down along the sample cell to acquire data from transmission and backscattering every 40 μm (Figure 5.3a), the Turbiscan measures the stability, by detecting the changes in backscattering or transmission, as a result of particle size variation or particle migration. The intensity of backscattering or transmission is a function of the amount of the dispersed particles (Mengual 1999; Zequan, Xue, and Qiang 2014).

Figure 0.3. a) Turbiscan principle and backscattering profiles for b) a stable crude oil solution, c) O/W emulsion going under coalescence, d) W/O emulsion from extra heavy crude oil upon creaming, e) W/O emulsion from medium crude oil going through precipitation, and f) asphaltene flocculation in HepTol 70:30 v/v.

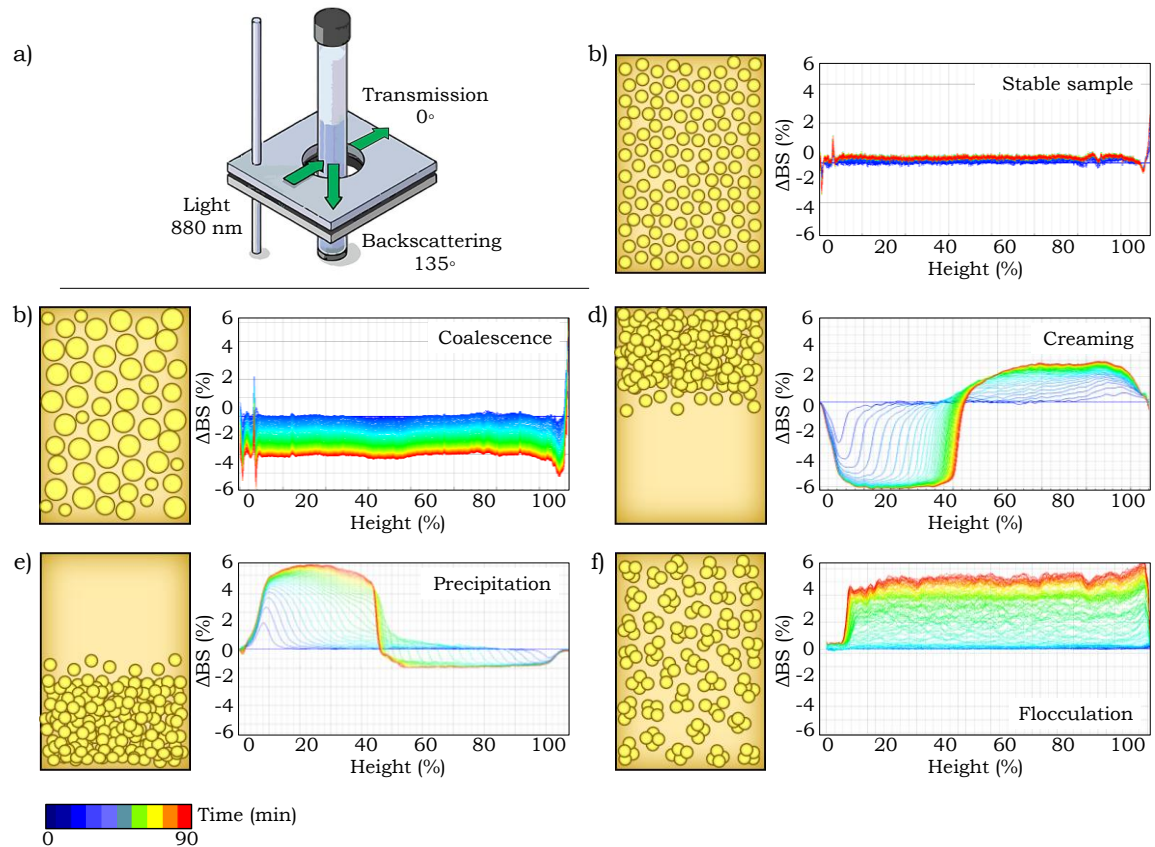


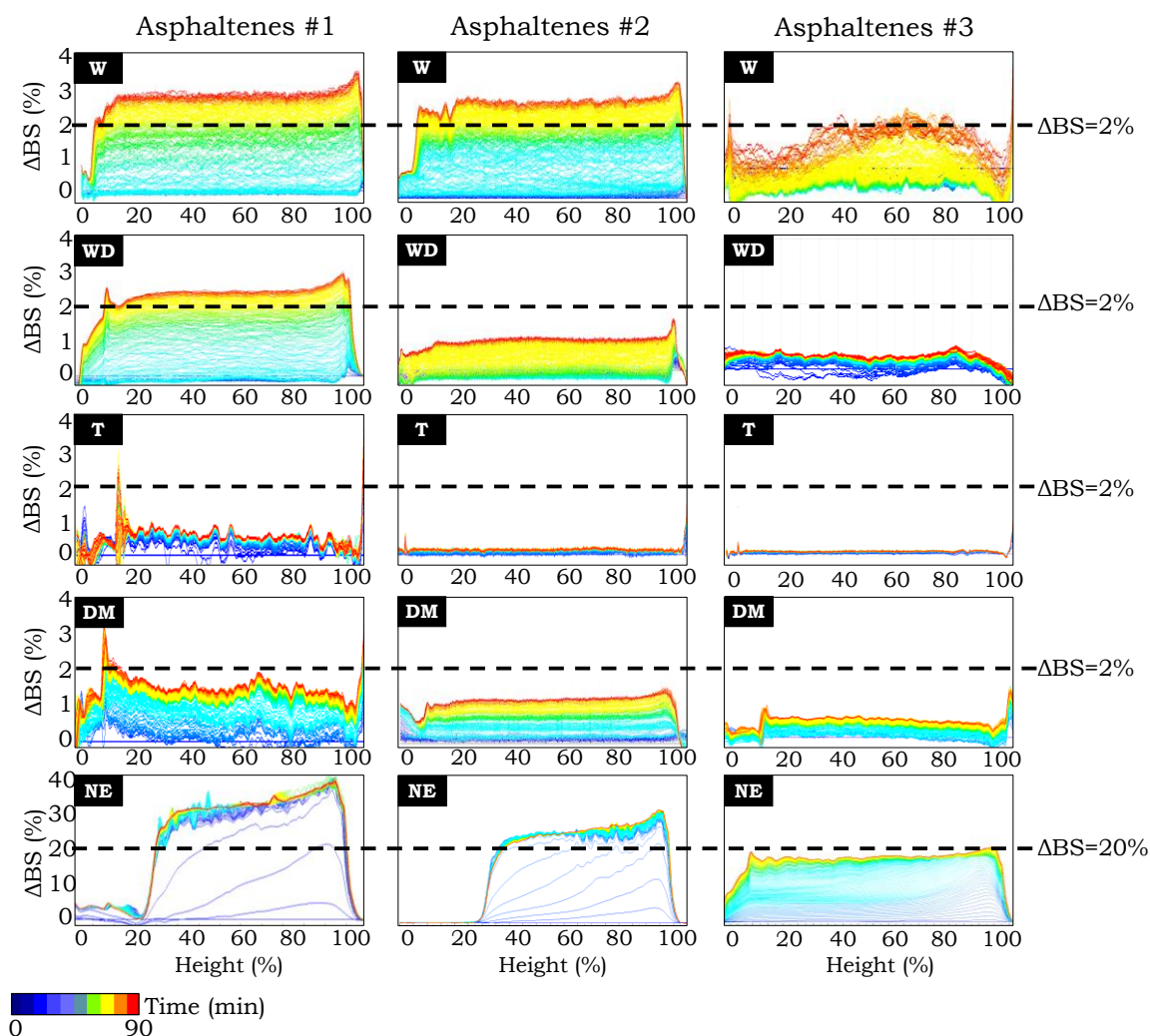
Figure 5.3 illustrates the Turbiscan principle and some examples of the backscattering profiles of studied Colombian petroleum dispersions. Importantly, X axis corresponds to the sample vial height which we are presenting normalized; the Y axis denotes the variation of the backscattering compared to the first scan. Each line in the profile corresponds to one scan, whose color is function of time. For instance, a sample of dehydrated medium Colombian crude oil (API=28.2), diluted in toluene at a concentration of 10 mg/mL, does not exhibit variation in the backscattering profile over time (Figure 5.3b). On the other hand, Figure 5.3c displays the backscattering profile for a synthetic oil-in-water emulsion from the same crude oil; without any kind of surfactant added, the stability of such emulsions is usually poor; thus, the oil droplets will collide and the collisions will lead to droplet fusion or coalescence, decreasing particle concentration and hence declining backscattering. Figure 5.3d and 5.3e presents the evolution of the backscattering of a natural water-in-oil emulsion from the medium and extra-heavy crude oils (API=28.2 and 8.2, respectively), after the addition of a commercial crude oil demulsifier (DEMTROL™). In these cases, creaming (Figure 2d) and sedimentation (Figure 2e) are driven by the density difference between water and crude oil; usually, unresolved emulsion droplets accumulate at the oil-water interface producing a rag layer. Finally, Figure 5.3f illustrates the backscattering profile for flocculation phenomena; this profile corresponds to the aggregation of Colombian asphaltenes at 1.0 mg/mL in a HepTol mixture of 70:30 v/v. In this example, an increase of particle diameter rises the backscattered light.

As stated before, aggregation test for whole asphaltene samples and the subfractions were carried out in toluene asphaltene solutions at 1 mg/mL, and the addition of 70% v/v of *n*-heptane. Figure 3 displays selected backscattering profiles for the aggregation tests of the whole #1, #2, and #3 asphaltenes, the samples WD, and the respective HPTLC subfractions. Starting with the unfractionated asphaltene samples (W), their backscattering profiles are typical for samples undergoing flocculation (Juyal et al. 2012), with maximum ΔBS up to 3.1% for the asphaltenes

from the heaviest crude oil, and 2.2% for the asphaltenes from the lightest crude oil. The aggregation behavior of the whole asphaltene sample desorbed from the silica surface (samples WD) is quite interesting. Apparently, by simple adsorption-desorption of the asphaltenes from the SiO₂ particles, it is possible the removal of chemical species that may play crucial roles in asphaltene aggregation. Several authors have claimed the use of mineral sorbents, such as particles of neutral Al₂O₃, and SiO₂-based sorbents for removing specific compositions from asphaltenes. For instance, Murrell *et al.* (Gould *et al.* 1983; Grenoble *et al.* 1984) demonstrated the efficient adsorption of basic asphaltenes using alumina-silica sorbents. Additionally, comprehensive reviews in asphaltene adsorption and catalyst in refinery processes indicate that solid materials, having Lewis and/or Bronsted acid sites are useful for removing basic asphaltenes from dehydrated heavy oils. Importantly, it is believed that molecular compositions having *N*-basic moieties adsorb on acidic isolated, or single, silanols. Along these lines, one could expect that WD samples exhibit a decreased concentration of basic species, and based on the results of Figure 5.4, one could hypothesize that removal of such species translates into stabilization of the asphaltene dispersions.

As we previously reported, the main molecular differences between the whole asphaltenes and WD samples stand in H/C ratios and content of CH₂ units (or steric hindrance). We reported selective retention of highly aromatic and sizable compounds (H/C ratios below 1.0 and carbon number between 20-50), with little steric hindrance (-up to ~13 CH₂ units in alkyl side chains on the core structures). We hypothesize that selective retained compositions have active functionalities, such as basic nitrogen and acidic oxygen, structurally accessible for strong intermolecular interactions (hydrogen bonding and acid-base associations). Furthermore, the little content of CH₂ units, which could translate into minimum steric hindrance, could induce effective pi-stacking between the aromatic cores. In this sense, selective adsorption of such compositions results in the enhanced stability of the dispersions of the desorbed species.

Figure 0.4. Backscattering profiles for whole #1, #2, and #3 asphaltenes, WD samples, and the respective HPTLC subfractions.



Regarding the HPTLC subfractions, there is a clear trend between polarity (-or retention on silica) and aggregation: more polarity translates into more aggregation. At this regard, Adams (Adams 2014) suggested that the same driving forces in asphaltene adsorption on mineral surfaces must play important roles in asphaltene aggregation. Basically, we reported that a synergy between low H/C ratios (or high aromaticity), few content of methylene units (or steric hindrance) and accessibility of the active functionalities is the crucial factor in asphaltene adsorption, and hence, in

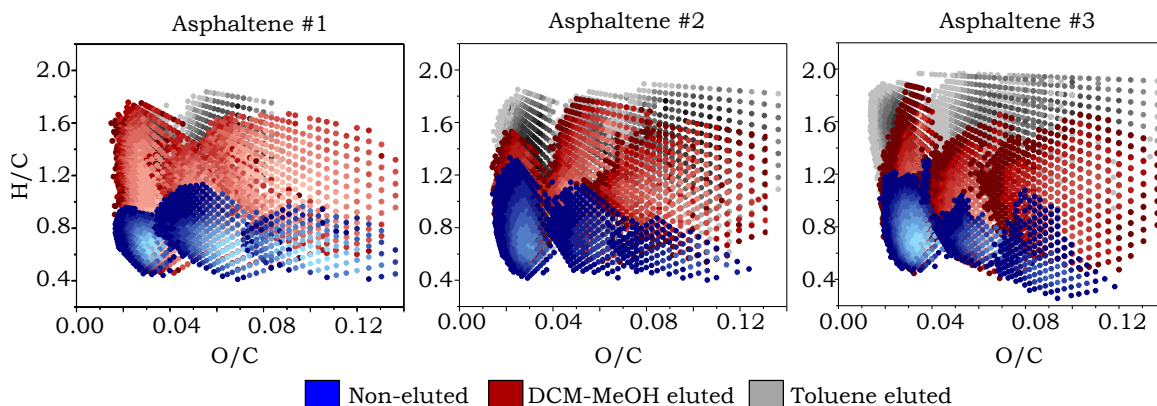
asphaltene aggregation. Consequently, the aggregation behavior of the HPTLC asphaltene subfractions support the ideas explained above. Starting with the least polar fraction, or toluene eluted compositions from asphaltene #1, the average ΔBS is up to 1%, suggesting an increased stability when compared with the whole asphaltenes and the samples WD. Moreover, T fractions from asphaltenes #2 and #3 exhibit lower aggregation tendencies, suggesting some relationship between API grades (-or the chemistry of the crude oil matrix) and asphaltene composition. In Chapter 2, it is reported that toluene eluted compositions consist of alkyl-aromatics enriched in S-functionalities, presenting compound families such as $N_nO_oS_s$, N_nS_s , O_oS_s and S_s . According to our report, this fraction is distinguished because of its high H/C ratios ($0.8 < H/C < 1.6$) when compared with the whole asphaltenes ($0.4 < H/C < 1.6$) and methylene content up to twenty-five CH_2 units. Although high content of N and O in T fraction could induce strong polar interactions, alkyl-side chains may play important role in polar group accessibility. Moreover, several reports suggest low affinity of sulfur functional groups toward silica surfaces as a result of low S-polarizability, which culminate in weaker hydrogen bonds when compared with oxygen (Li et al. 1994). Also, sulfur may be present in sulfide form, having significant steric effects which could block effective intermolecular interactions.

To give a clear discussion about the role of molecular composition in the aggregation of asphaltene subfractions, Figure 5.5 illustrates the general molecular composition of the HPTLC fractions from purified asphaltenes samples #1, #2, and #3, in terms of aromaticity (H/C ratios), homologous series length (steric hindrance) and O/C ratios (molecular size). It is important to have into account that Kilpatrick and co-workers demonstrated that H/C ratios correlate well with asphaltene aromaticity. Thus, according to the Figure 5.5ⁱⁱ, asphaltenes presenting the lowest H/C ratios hence the highest aromaticity, and the shortest homologous series thus the lowest steric hindrance and the highest polar group accessibility, exhibit the strongest

ⁱⁱ These are part of the results we previously published: *Energy & Fuels*, 29 (3), 1323-1331.

adsorption on SiO₂ particles and the strongest aggregation. On the other hand, asphaltenes presenting a molecular composition with higher steric hindrance and lower aromaticity, would be less prone to aggregate and adsorb on mineral surfaces.

Figure 0.5. Molecular composition in terms of van Krevelen diagrams of the HPTLC fractions of the three asphaltene samples.



As the retention factor decreases (stronger adsorption), aggregation trend increases. Along these lines, DM fractions are more unstable when compared with the T fractions. At this point, it is important to remind the nature of the eluent used for obtaining DM fractions. The mixture CH₂Cl₂/MeOH has two important features: it has the highest polarity of the elutropic series and consists of a protic mixture, which can form a layer on the SiO₂ because of its suitability for interacting with the silanols through hydrogen bonds. In this sense, the eluent mixture DCM/MeOH can displace polar components in the asphaltene interacting with the silica exclusively through hydrogen bonding. In fact, DM composition is characterized by high content of N- and O- atoms organized in compound classes such as N₁, N₃, N₁O₁, N₃O₁, N₃O₂, having the highest content of CH₂ units in alkyl side chains –up to 35 CH₂ units. Thus, we could argue that polar moieties in DM fraction may have a high tendency to interact through hydrogen bonds, which is possible only if the polar groups are accessible or located in peripheral domains. In this sense, we hypothesize that polar interactions drive the aggregation of this subfraction. At this regard, Kilpatrick *et al.*

(Spiecker, Gawrys, and Kilpatrick 2003) suggested that the aggregation of low aromaticity asphaltene fractions (presenting H/C ratios around 1.4) is prompted by hydrogen bonding between polar functionalities, which must be organized in peripheral moieties in the asphaltene's structure for worthwhile polar associations.

Finally, subfractions NE present the strongest aggregation along all samples. The backscattering profiles show ΔBS up to 38%, 38.9%, and 20.2% for asphaltenes #1, #2, and #3, respectively. According to our previous report, this fraction is composed of highly aromatic asphaltenes ($0.6 < H/C < 1.2$), with little steric hindrance (up to 12 CH_2 units in alkyl side chains), and high content of N and O, in compound families such as O_n , N_nO_o , and O_o . Compared with the backscattering profiles for the subfractions T and DM, and the whole asphaltenes, the stability of NE asphaltene dispersions is extremely low. We hypothesize that this fraction aggregates following the supramolecular assembly model for asphaltene aggregation, proposed by Gray *et al.* (Gray et al. 2011), who suggested strong asphaltene aggregation as a result of multiple intermolecular interactions, i.e. pi-stacking, hydrogen bonding, acid-base interactions, complexation of heavy metals such as vanadium, nickel and iron by pyridinic groups, and even though, association of alkyl side chains in hydrophobic pockets.

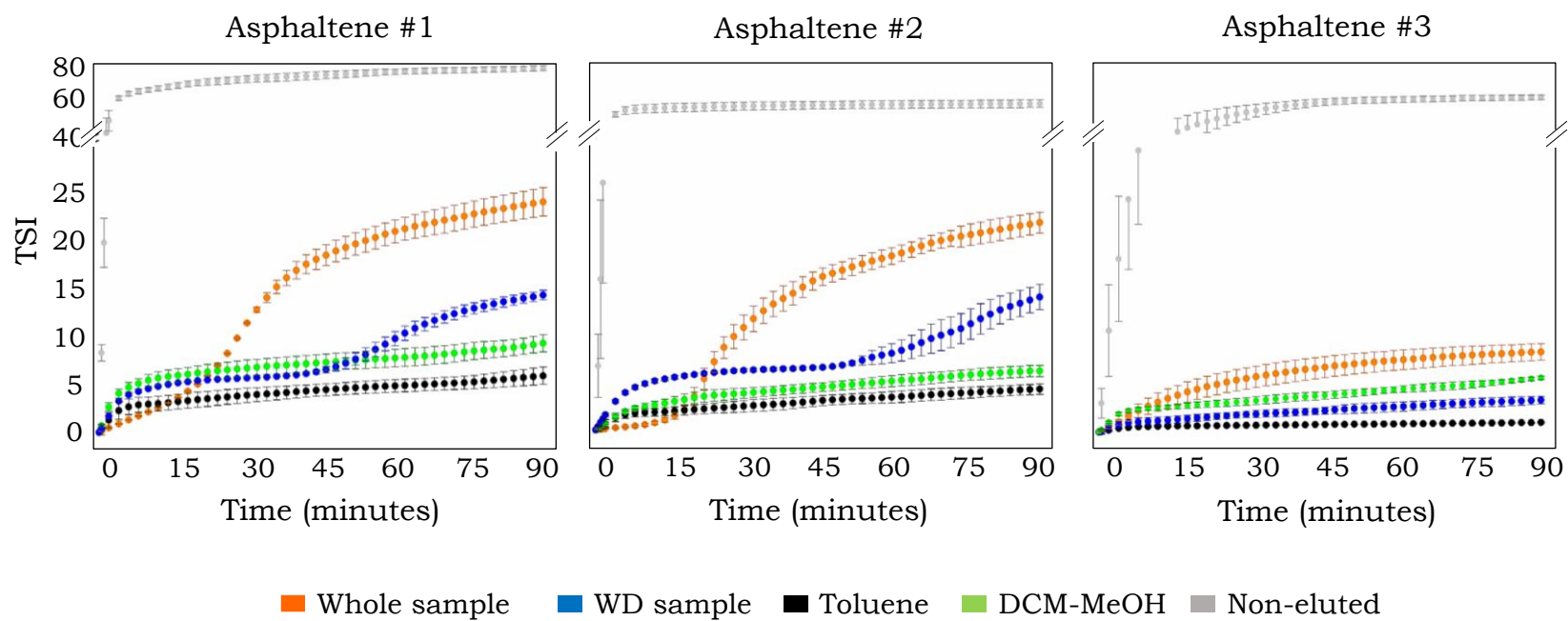
Comparing the stability of asphaltene dispersion by visual inspection of the backscattering profiles, sometimes is tedious and could not let to conclusive observations. To solve this problem, we have used the Turbiscan Stability Index – TSI, which is a relative measurement of the stability of the dispersions and is a parameter derived from the variations in the rate of the backscattering intensity of the sample, relative to the original. Low TSI values indicate stability and few chances of phase separation, while high TSI values are indicative of instability and high probability of phase separation. The TSI is calculated following the equation 1.

$$TSI = \sqrt{\frac{\sum_{i=1}^n (x_i - x_{BS})^2}{n-1}} \text{ (Equation 5.1)}$$

Where χ_i is the average backscattering for each scan, χ_{BS} is the average backscattering for the first scan and n is the number of scans. Figure 5.6 presents the stability plots for the dispersions of the whole asphaltenes, WD samples, and the HPTLC subfractions, in terms of the global Turbiscan stability index in function of time, with the respective error bars. Figure 5.6 enables observation of clear trends in the aggregation behavior of the asphaltenes samples and the subfractions. Starting with the whole asphaltenes, one could conclude that the aggregation in model HepTol solutions, of the whole asphaltenes from extra-heavy and heavy crude oils (#1 and #2) is much higher (TSI maxima= 24.5 and 23.2, respectively), when compared with the aggregation of the asphaltenes from the medium crude oil (TSI maxima=7.1). According to the structural continuity of petroleum, one could expect that asphaltenes from lighter crude oils, exhibit similar molecular parameters than the crude oils matrix. In this sense, asphaltenes from light petroleum could present more aliphatic moieties, being friendlier with paraffinic solvents when compared with the asphaltenes from extra-heavy crude oils (Altgelt and Boduszynski 1992; Boduszynski and Altgelt 1992; Boduszynski 1987, 1988).

On the other hand, asphaltene dispersions WD reveal an aggregation pattern much lower when compared with the original asphaltenes, presenting maximum TSI values of 13.0, 12.8 and 6.2, for asphaltenes #1, #2, and #3, respectively. Moreover, the behavior of the HPTLC subfractions is quite interesting: TSI profiles prove that the most “problematic” asphaltenic species are concentrated in the non-eluted fraction.

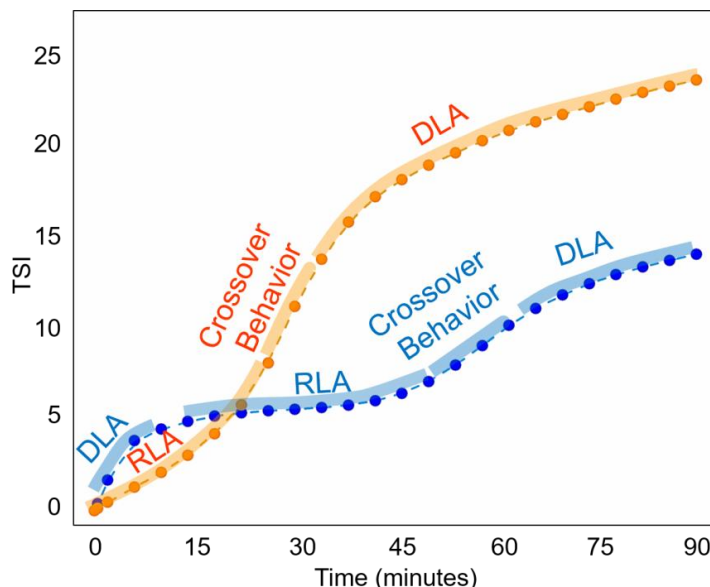
Figure 0.6. Aggregation profiles for whole asphaltenes, WD samples and the HPTLC subfractions in terms of the Turbiscan Stability Index.



For instance, the dispersion of the NE subfraction from asphaltene #1 presents a maxima TSI value of ~ 80.5 , while the dispersion of DM and T asphaltene subfractions are much more stable, with maximum TSI values of 7.5 and 4.0, respectively. The trend is quite similar for asphaltene #2, with TSI values of 57.0, 6.1 and 4.2 for NE, DM, and T dispersions, respectively; and asphaltene #3 presenting TSI values of 60.8, 3.3 and 1.2 for subfractions NE, DM, and T respectively.

An interesting feature of the aggregation behavior of the whole asphaltene and the samples WD is the shape of the TSI curves, which clearly present two well-differentiated trends. Several authors have found two possible mechanisms of asphaltene aggregation, widely known as *diffusion-limited aggregation* (DLA) which is fast, and *reaction-limited aggregation* (RLA) which is slow (I.K. Yudin et al. 1998). Since the TSI values are a result of the variation of the backscattering, and the changes of the backscattered light, in aggregation processes, reflect on the increase of the particle size, we could also use the DLA and RLA mechanisms for describing the aggregation of our samples. Figure 5.7 presents the fitting curves for the aggregation of the whole and WD sample from asphaltene #1. The DLA or fast aggregation is governed only by the diffusion of the particles; in theory, every collision between asphaltene particles could conclude in their aggregation. On the other hand, in the RLA regimen, not all the collisions lead to particle sticking; the aggregation process proceeds throughout slow interactions between the particles (Yudin and Anisimov 2007). Yudin reported a crossover behavior between RLA and DLA regimes. At the initial stage of the aggregation, RLA mechanism is dominant; at some point, after a particular aggregate size, DLA mechanism takes place. Fitting curves in Figure 5.7 suggest that asphaltene aggregation in HepTol solutions is similar to that of typical colloids.

Figure 0.7. Examples of the DLA and RLA mechanisms for asphaltene aggregation. We include the fitting curves for the TSI curves for the whole and WD samples from asphaltenes #1.



Going back to the Figure 5.6, it is clear that whole asphaltenes #1 and #2 exhibit an aggregation behavior governed by both RLA and DLA regimes. On the other hand, whole asphaltenes #3 seem to aggregate only through DLA mechanism. This observation suggests that molecular composition has some important implications in the asphaltene aggregation trend. Yudin demonstrated the mechanism of asphaltene aggregation in hydrocarbon solutions, as strongly dependent phenomena on the concentration. Asphaltene solutions below critical aggregate concentration, exhibit DLA mechanisms, while higher concentration lets on RLA trends. Interestingly, the dispersions from all the HPTLC subfractions follow a DLA mechanism. However, in our case, all the aggregation tests were carried out at the same conditions (1000 mg/mL and 70/30 v/v HepTol) which suggest an important role of the molecular structure in the asphaltene aggregation trend. Although all HPTLC subfractions follow a DLA mechanism, the aggregation rate of the NE dispersions is at least seven times higher, in terms of the TSI, than the DM and T dispersions' rates.

Regarding the role of the molecular composition in asphaltene aggregation, Kilpatrick and co-workers have made important contributions. They reported the fractionation of asphaltenes by differential precipitation (Gawrys, Blankenship, and Kilpatrick 2006) and suggested that the same intermolecular forces do not drive the aggregation of the asphaltenes from different crude oils. In other words, based on H/C, O/C, and N/C atomic ratios, they concluded that the aggregation of some asphaltenes is driven by aromatic associations (π -stacking) and dispersion interactions, while asphaltenes from other sources seem to aggregate through hydrogen bonding between polar functionalities. Also, in another report, Kilpatrick *et al.* (Spiecker *et al.* 2003) demonstrated that the most unstable asphaltenes, or the asphaltene fraction precipitated with the minimum amount of *n*-heptane, consist of the most aromatic and the most polar species. Moreover, redissolution test showed that the most unstable asphaltene fraction is less soluble in methylene chloride, when compared with the whole asphaltenes. The least soluble asphaltene fractions required increasing of the solvent temperature, in order to achieve total redissolution, suggesting hydrogen bonding as the propulsive molecular interaction for the aggregation. In this sense, we conclude that hydrogen bonding may have an important role in the aggregation of the NE dispersions.

Along these lines, we could also hypothesize that in the whole sample, the most stable asphaltene compositions play an important function in the stabilization of the most “problematic” asphaltenes. Thus, T and DM asphaltenes appears to cooperatively solvate the most polar and most aromatic asphaltenes —NE species, in the unfractionated asphaltene sample, through disruption of hydrogen bonding and strong polar interactions. This is the reason why the whole asphaltene samples exhibit a maximum TSI value up to 5 times lower than the TSI of the NE dispersions. Along these lines, the thin layer chromatography fractionation lets on the selective separation of highly stable asphaltenes, leaving behind, in the non-eluted fraction, the chemical species most prone to precipitate.

At this point, it is possible to draw an important conclusion: the molecular differences among the three asphaltene samples lie on the weight percentages of the non-eluted, DM, and T fractions. Along these lines, the most stable asphaltene is enriched in weak-polar compositions, eluted with toluene. On the other hand, the most unstable asphaltene is enriched in non-eluted species characterized by the highest aromaticity and lowest steric hindrance.

5.5 EFFECT OF CRUDE OIL MATRIX IN ASPHALTENE STABILITY

In addition, aggregation tests on the whole asphaltenes, spiked with aromatics and resins were carried out to understand the role of crude oil matrix in deposition issues. Figure 5.8 presents the aggregation trends of the asphaltenes #1, #2, and #3 in presence of 100 wt% aromatics (red dots) and 100 wt% resins (blue dots). It is clear that aromatics by themselves do not have anything to bear in stabilization of asphaltene dispersions, specifically for asphaltenes #2 and #3, where the error intervals suggest lack of conclusive results. Regarding asphaltenes #1, addition of 100 wt% of aromatics decreases the maxima TSI value from 24.3 to 20.8, which may be construed as stabilization. On the other hand, TSI curves demonstrate that resins play an important role in asphaltene stabilization, highly evident for asphaltene #1 and #2. Several authors have pointed out the important role of SARA resins in the stability of asphaltene dispersions (Acevedo et al. 1985; Koots and Speight 1975; Murgich 2002; Sedghi and Goual 2010). Since a structural point of view, the most polar and aromatic resins create a layer on asphaltenes aggregates, enhancing the stability in crude oil solutions (Sedghi and Goual 2010). Phenomenologically, resins affect aggregation and precipitation: certain kinds of resins can increase the stability of asphaltenes, affecting the amount of asphaltene precipitation by common standard methods, and can decrease the size of asphaltene aggregates in small-angle X-ray scattering studies (Andersen and Speight 2001).

Figure 0.8. Effect of SARA aromatics and resins on the stability of whole asphaltene dispersion.

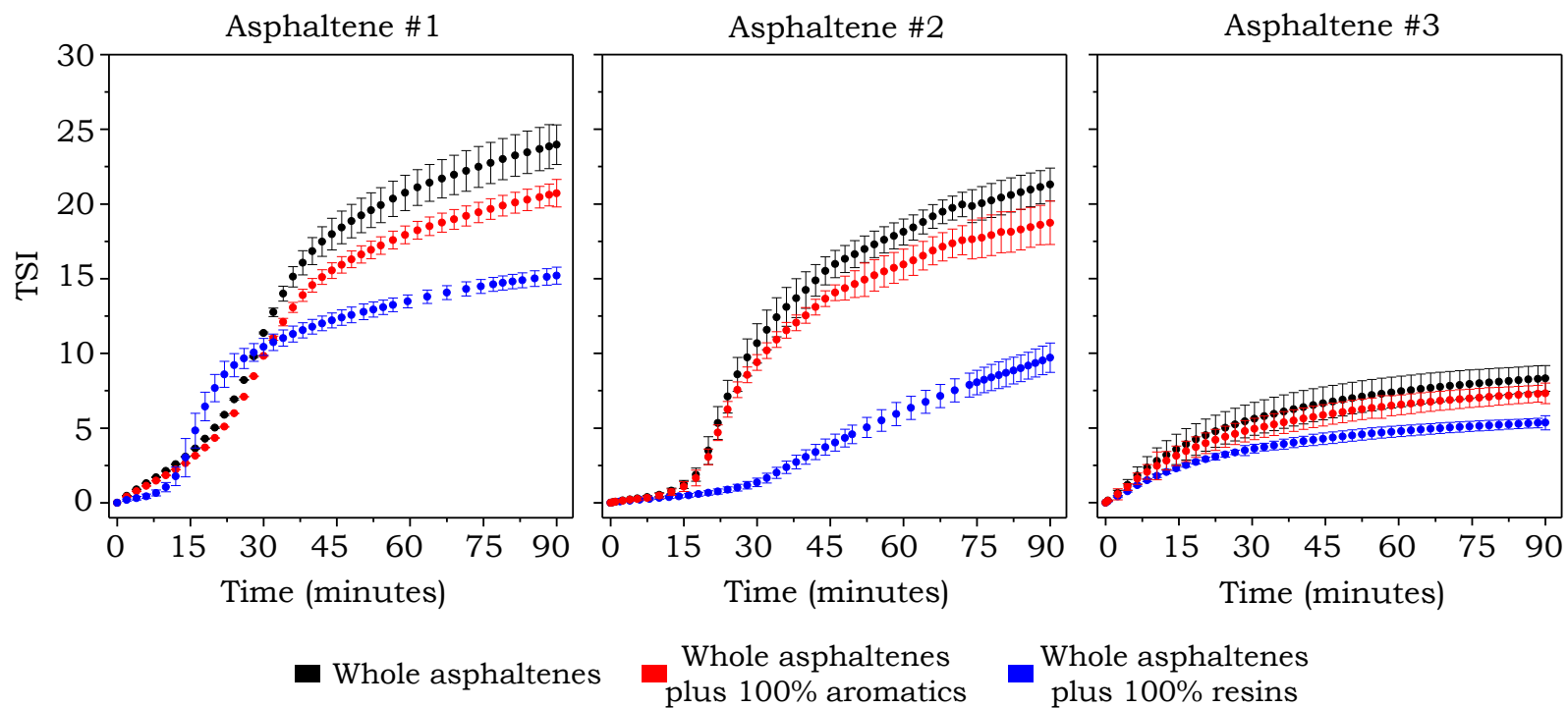
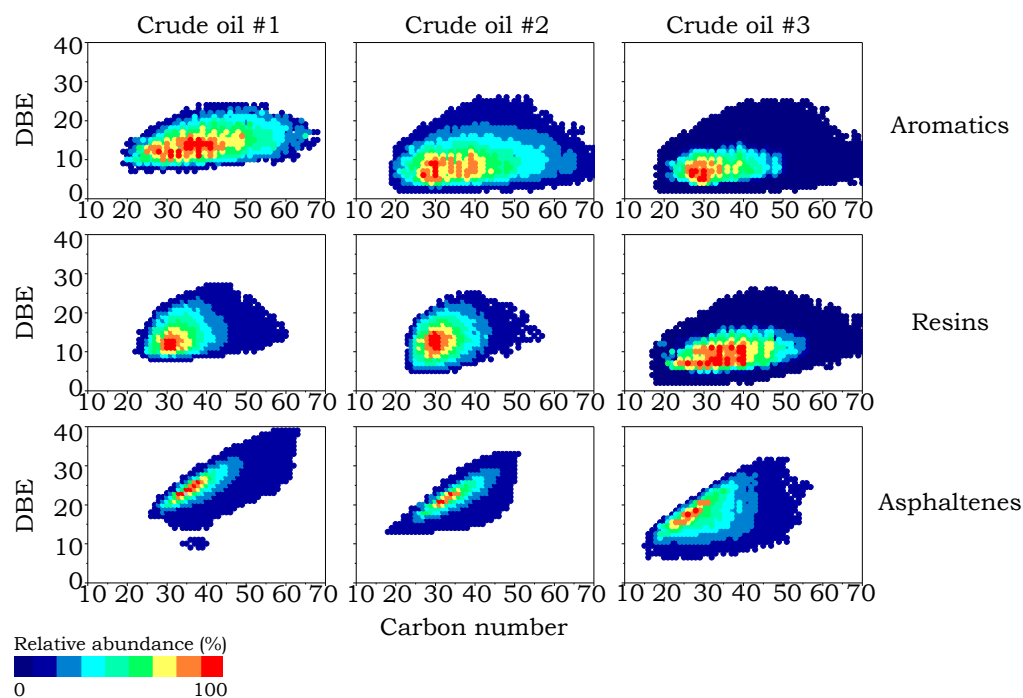


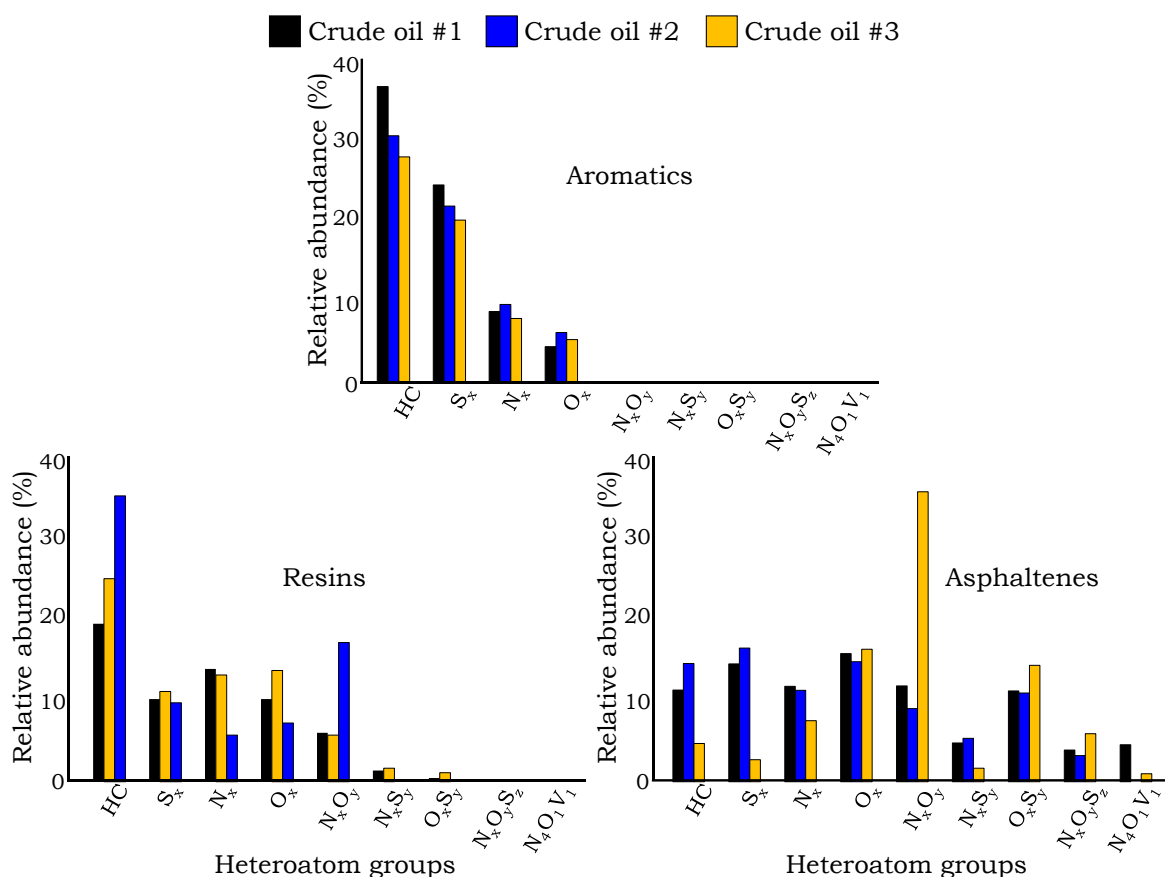
Figure 5.9 includes the molecular composition, in terms of DBE and carbon number distributions for the aromatics, the resins and the whole asphaltenes from each crude oil. Aromatics are featured by low DBE values (between 2-10 for the most abundant compositions from crude oils #2 and #3) and long homologous series (having up to 50 carbon atoms in alkyl side chains). Interestingly, SARA resins from crude oils #1 and #2 represent an intermediate compositional state between asphaltenes and aromatics. For these samples, the highly abundant compositions start shifting towards the planar aromatic limit, the DBE values for the most abundant species are between 10-25 and the homologous series are longer than asphaltenes' but shorter than aromatics'. In terms of compound class distribution (Figure 5.10), resins #1 and #2 are more polydisperse than aromatics, but less polydisperse than asphaltenes. Interestingly, resins from crude oil #3 have a molecular composition highly similar to aromatics #3, which could explain its poor effect on the stabilization of asphaltenes #3.

Figure 0.9. Molecular composition of aromatics, resins and asphaltenes from crude oils #1, #2, and #3.



Simplification of petroleum composition to four types of compounds could explain why only some types of resins stabilize asphaltenes. In a stable sample of crude oil, the most aromatic and polar molecules —asphaltenes (represented with *A* in Figure 5.11) are nanometric solids (or nanoaggregates) dispersed in the oil by resins (represented by *R*). The asphaltene-resin dispersion is dissolved in the crude oil matrix by small aromatics (represented by *a*) which can be seen as solvents. On the other hand, saturates (represented by *s*) that are non-solvents. This model is known as the Wiehe Solubility model, represented in the Figure 5.11 (Wiehe and Kennedy 2000).

Figure 0.10. Compound class distribution in terms of heteroatom groups for aromatics, resins and asphaltenes from crude oils #1, #2, and #3.



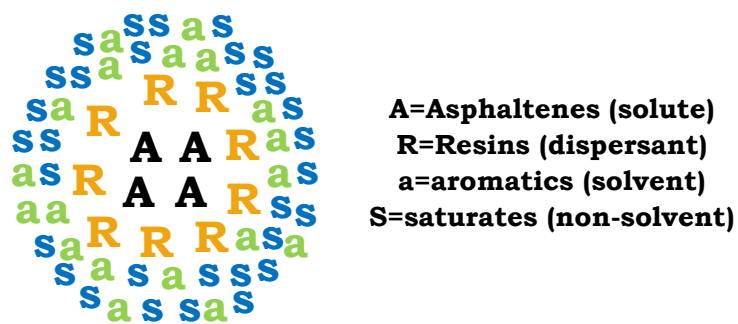
It has been widely reported that asphaltenes are dispersed in the crude oil in a delicate equilibrium, which can be easily disrupted by the addition of saturates or by the removal of resins. As figure 5.11 shows, because of aromatics can be seen as solvent, only the addition of aromatics, under the concentration of *n*-heptane used in the aggregation test of this contribution, does not cause significant stabilization of asphaltenes. However, the addition of natural dispersants –Resins, enhance the stability of the studied dispersions.

The particular molecular composition of resins #1 and #2, having an intermediate compositional space between asphaltenes and aromatics, is the direct cause of its suitability for enhancing asphaltene stability. Several authors have reported that polar and aromatic moieties of resins, interact directly with asphaltenes nanoaggregates. Along these lines, acid-base interactions between basic nitrogen and carboxylic acids, classic hydrogen bonds between polar functionalities, non-classic hydrogen bonds between aromatic moieties and i.e. hydroxyl groups and pi-stacking between aromatic cores are the intermolecular associations responsible for coupling between resins and asphaltenes (Murgich 2002). In these kind of interactions, non-polar and aliphatic regions of resins are oriented towards the crude oil matrix; this phenomenon blocks further associations between asphaltenes nanoaggregates, decreasing the chances of clustering. This sort of dispersion is known as stabilization by steric effect (Strausz et al. 2006). Along the same lines, aliphatic and non-polar moieties interact effectively with the solvent (aromatics) and non-solvent (saturates), which helps to keep the solids (the asphaltenes) in solution.

Additionally, the petroleomes presented in the Figure 5.9 and the progression of heteroatom content with aromaticity (Figure 5.10), state the remarkable continuity in the structure of Petroleum. Boduszynski, in the late 1970's, without using any sophisticated analytical technique such as nuclear magnetic resonance or FT-ICR mass spectrometry, claimed that petroleum is a continuum with regard to composition, molecular weight, aromaticity and heteroatom content as the function

of the boiling point. Now, it is possible to see glimpses of a remarkable structural continuity, which supports the solubility models previously reported.

Figure 0.11. Wiehe solubility model for petroleum.



5.5 CONCLUSIONS

We demonstrated that asphaltene adsorption on mineral surfaces and aggregation in model solutions are closely related. The same molecular features, and hence, molecular interactions, responsible for asphaltene adsorption on SiO_2 particles, have important functions in asphaltene aggregation and hence, deposition. Along these lines, we conclude that besides high content of heteroatoms like N and O, the aggregation is boosted by high aromaticity (or low H/C ratios) and little steric hindrance (or polar group accessibility). Moreover, we demonstrated the utility of the SiO_2 surface chemistry in the separation of aggregation-prone asphaltenes and stable asphaltenic composition. By simple elution of asphaltenes on SiO_2 , through polar, protic and aromatic eluent mixtures, it is possible to promote the selective migration of non-problematic asphaltenes, leaving behind, in the non-eluted fraction, challenging asphaltenes which could induce serious deposition problems.

BIBLIOGRAPHY

Abu Tarboush, Belal J. and Maen M. Husein. 2012. "Adsorption of Asphaltenes from Heavy Oil onto in Situ Prepared NiO Nanoparticles." *Journal of colloid and interface science* 378(1):64–69.

Acevedo, S., B. Mendez, A. Rojas, I. Layrisse, and H. Rivas. 1985. "Asphaltenes and Resins from the Orinoco Basin." *Fuel* 64:1741–47.

Acevedo, S., MA Ranaudo, G. Escobar, Luis Gutiérrez, and Pedro Ortega. 1995. "Adsorption of Asphaltenes and Resins on Organic and Inorganic Substrates and Their Correlation with Precipitation Problems in Production Well Tubing." *Fuel* 74(4):595–98.

Acevedo, Socrates, Josmelith M. Cordero T, Herve Carrier, Brice Bouyssiere, and Ryszard Lobinski. 2009. "Trapping of Paraffin and Other Compounds by Asphaltenes Detected by Laser Desorption Ionization–Time of Flight Mass Spectrometry (LDI–TOF MS): Role of A1 and A2 Asphaltene Fractions in This Trapping." *Energy & Fuels* 23(2):842–48.

Adams, Jeramie J. 2014. "Asphaltene Adsorption, a Literature Review." *Energy & Fuels* 28(5):2831–56.

Agrawala, Mayur and Harvey W. Yarranton. 2001. "An Asphaltene Association Model Analogous to Linear Polymerization." *Industrial & Engineering Chemistry Research* 40(21):4664–72.

Akbarzadeh, Kamran et al. 2007. "Asphaltenes — Problematic but Rich in Potential." *Asphaltenes — Problematic but Rich in Potential. Schlumberger Oilfield Review* 22–

43.

Ali, Mohammad F., H. Perzanowski, a. Bukhari, and Adnan a. Al-haji. 1993a. "Nickel and Vanadyl Porphyrins in Saudi Arabian Crude Oils." *Energy & Fuels* 1(5):179–84.

Ali, Mohammad F., H. Perzanowski, a. Bukhari, and Adnan a. Al-haji. 1993b. "Nickel and Vanadyl Porphyrins in Saudi Arabian Crude Oils." *Energy & Fuels* 1(5):179–84.

Altgelt, K. H. and M. M. Boduszynski. 1992. "Composition of Heavy Petroleums .3. An Improved Boiling-Point Molecular-Weight Relation." *Energy & Fuels* 6(1):68–72.

Alvarez, Elena et al. 2011. "Pyrolysis Kinetics of Atmospheric Residue and Its SARA Fractions." *Fuel* 90(12):3602–7.

Ancheyta, J., G. Centeno, F. Trejo, and G. Marroquin. 2003. "Changes in Asphaltene Properties during Hydrotreating of Heavy Crudes." *Energy & fuels* 17(6):1233–38.

Ancheyta, Jorge, Fernando Trejo, and Mohan Singh Rana. 2010a. *Asphaltenes: Chemical Transformation during Hydroprocessing of Heavy Oils*. CRC Press.

Ancheyta, Jorge, Fernando Trejo, and Mohan Singh Rana. 2010b. "Definition and Structure of Asphaltenes." Pp. 1–86 in *Asphaltene Chemical Transformation during Hydroprocessing of Heavy Oils*. CRC Press.

Andersen, Simon I. and James G. Speight. 2001. "Petroleum Resins: Separation, Character, and Role in Petroleum." *Petroleum Science and Technology* 19(September 2013):1–34.

Badjic, JD and NM Kostic. 2000. "Unexpected Interactions between Sol-Gel Silica Glass and Guest Molecules. Extraction of Aromatic Hydrocarbons into Polar Silica from Hydrophobic Solvents." *The Journal of Physical Chemistry B* 104:11081–87.

Badjić, Jovica D. and Nenad M. Kostić. 2001. "Behavior of Organic Compounds Confined in Monoliths of Sol-gel Silica Glass. Effects of Guest-host Hydrogen Bonding on Uptake, Release, and Isomerization of the Guest Compounds." *Journal of Materials Chemistry* 11(2):408–18.

Badre, Sophie, Cristiane Carla Goncalves, Koyo Norinaga, Gale Gustavson, and Oliver. C. Mullins. 2006. "Molecular Size and Weight of Asphaltene and Asphaltene Solubility Fractions from Coals, Crude Oils and Bitumen." *Fuel* 85(1):1–11.

Benkhedda, Z., P. Landais, J. Kister, J. M. Dereppe, and M. Monthieux. 1992. "Spectroscopic Analysis of Aromatic Hydrocarbons Extracted from Naturally and Artificially Matured Coals." *Energy & fuels* 6(2):166–72.

Bisht, Harender et al. 2013. "Efficient and Quick Method for Saturates, Aromatics, Resins, and Asphaltenes Analysis of Whole Crude Oil by Thin-Layer Chromatography – Flame Ionization Detector." *Energy & Fuels* 27:3006/3013.

Boduszynski, M. M. and K. H. Altgelt. 1992. "Composition of Heavy Petroleum. 4. Significance of the Extended Atmospheric Equivalent Boiling-Point (Aebp) Scale." *Energy Fuels* 6(1):72–76.

Boduszynski, Mieczyslaw M. 1987. "Composition of Heavy Petroleum. Molecular Weight, Hydrogen Deficiency, and Heteroatoms Concentration as a Function of Atmospheric Equivalent Boiling Point up to 1400°F." *Energy & Fuels* 1(1):2–11.

Boduszynski, Mieczyslaw M. 1988. "Composition of Heavy Petroleum. 2. Molecular Characterization." *Energy & Fuels* 2(5):2–11.

Buenrostro-gonzalez, Eduardo, Henning Groenzin, Carlos Lira-galeana, and Oliver C. Mullins. 2001. "The Overriding Chemical Principles That Define Asphaltenes." *Energy & fuels* 15(13):972–78.

Calemma, V. et al. 1995. "Structural Characterization of Asphaltenes of Different Origins." *Fuel and Energy Abstracts* 36(3):192.

Carbognani, Lante and Miguel Orea. 1999. "Studies on Large Crude Oil Alkanes. I. High Temperature Liquid Chromatography." *Petroleum Science and Technology* 17(1-2):165–87.

Chacón-Patiño, Martha L., Cristian Blanco-Tirado, Jorge a. Orrego-Ruiz, Andrea Gómez-Escudero, and Marianny Y. Combariza. 2015a. "High Resolution Mass Spectrometric View of Asphaltene–SiO₂ Interactions." *Energy & Fuels* 29(3):1323–31.

Chacón-Patiño, Martha L., Cristian Blanco-Tirado, Jorge a. Orrego-Ruiz, Andrea Gómez-Escudero, and Marianny Y. Combariza. 2015b. "Tracing the Compositional Changes of Asphaltenes after Hydroconversion and Thermal Cracking Processes by High-Resolution Mass Spectrometry." *Energy & Fuels* 29(10):6330–41.

Chacón-Patiño, Martha L., Cristian Blanco-Tirado, Jorge Armando Orrego-Ruiz, Andrea Gómez-Escudero, and Marianny Y. Combariza. 2015. "Tracing Asphaltene's Compositional Changes after Hydroconversion and Thermal Cracking Processes by High-Resolution Mass Spectrometry." *Energy & Fuels* 29(10):6330–41.

Chakhmakhchev, Alexander and Noriyuki Suzuki. 1995. "Saturate Biomarkers and Aromatic Sulfur Compounds in Oils and Condensates from Different Source Rock Lithologies of Kazakhstan, Japan and Russia." *Organic Geochemistry* 23(4):289–99.

Chang, Chia-Lu and H. Scott Fogler. 1994. "Stabilization of Asphaltenes in Aliphatic Solvents Using Alkylbenzene-Derived Amphiphiles. 1. Effect of the Chemical

Structure of Amphiphiles on Asphaltene Stabilization.” *Langmuir* 10(6):1749–57.

Chiaberge, Stefano et al. 2009. “Investigation of Asphaltene Chemical Structural Modification Induced by Thermal Treatments.” *Energy & Fuels* 23:4486–95.

Cho, Yunju, Young Hwan Kim, and Sunghwan Kim. 2011. “Planar Limit-Assisted Structural Interpretation of Saturates/aromatics/resins/asphaltenes Fractionated Crude Oil Compounds Observed by Fourier Transform Ion Cyclotron Resonance Mass Spectrometry.” *Analytical chemistry* 83(15):6068–73.

Cho, Yunju, JG Na, NS Nho, SH Kim, and Sunghwan Kim. 2012. “Application of Saturates, Aromatics, Resins, and Asphaltenes Crude Oil Fractionation for Detailed Chemical Characterization of Heavy Crude Oils by Fourier Transform Ion Cyclotron Resonance Mass Spectrometry Equipped with Atmospheric Pressure Photoionization.” *Energy & Fuels* 26:2558–65.

Chupeng, Yang, Zewen Liao, Lvhui Zhang, and Patrice Creux. 2009. “Some Biogenic-Related Compounds Occluded inside Asphaltene Aggregates.” *Energy and Fuels* 23(2):820–27.

Clementz, David M. 1976. “Interaction of Petroleum Heavy Ends with Montmorillonite.” *Clays and clay minerals* 24:312–19. Retrieved August 13, 2014.

Comisarow, Melvin B. and Alan G. Marshall. 1976. “Theory of Fourier Transform Ion Cyclotron Resonance Mass Spectroscopy. I. Fundamental Equations and Low-Pressure Line Shape.” *The Journal of Chemical Physics* 64(1):110.

Corilo, Yuri E. et al. 2010. “Petroleomics by ESI(±) FT-ICR MS.” *Analytical Chemistry* 82(10):3990–96.

Dechaine, GP and MR Gray. 2010. “Chemistry and Association of Vanadium Compounds in Heavy Oil and Bitumen, and Implications for Their Selective

Removal.” *Energy & Fuels* 24(3):2795–2808.

Derakhshesh, Marzie, Alexander Bergmann, and Murray R. Gray. 2013. “Occlusion of Polyaromatic Compounds in Asphaltene Precipitates Suggests Porous Nanoaggregates.” *Energy and Fuels* 27(4):1748–51.

Dudášová, Dorota, Sébastien Simon, Pål V. Hemmingsen, and Johan Sjöblom. 2008. “Study of Asphaltenes Adsorption onto Different Minerals and Clays.” *Colloids and Surfaces A: Physicochemical and Engineering Aspects* 317(1-3):1–9.

Dunn, James a., John B. MacLeod, Ron D. Myers, and Roby Bearden. 2003. “Recycle of Vanadium and Nickel-Based Catalysts in a Hydroconversion Process.” *Energy & Fuels* 17(1):38–45.

Ekweozor, Chukwuemeka M. 1984. “Tricyclic Terpenoid Derivatives from Chemical Degradation Reactions of Asphaltenes.” *Organic Geochemistry* 6(0):51–61.

El-Bassoussi, A. A., M. H. M. Ahmed, S. M. El Sayed, J. S. Basta, and E. S. K. Attia. 2010. “Characterization of Some Local Petroleum Residues by Spectroscopic Techniques.” *Petroleum Science and Technology* 28(5):430–44.

Fenn, J. B., Matthias Mann, C. K. Meng, S. F. Wong, and C. M. Whitehouse. 1989. “Electrospray Ionization for Mass Spectrometry of Large Biomolecules.” *Science (New York, N.Y.)* 246(4926):64–71.

Ford, C. D., S. A. Holmes, L. F. Thompson, and D. R. Latham. 1981. “Separation of Nitrogen Compound Types from Hydrotreated Shale Oil Products by Adsorption Chromatography on Basic and Neutral Alumina.” *Analytical Chemistry* 53(33):831–36.

Fyvie, TJ, H. Matis, GA O'neil, and JA Osaheni. 2012. "Methods and System for Removing Impurities from Heavy Fuel." *US Patent 8,088,277*.

Gauthier, Thierry, Pierre Danial-Fortain, Isabelle Merdrignac, Isabelle Guibard, and Anne Agathe Quoineaud. 2008. "Studies on the Evolution of Asphaltene Structure during Hydroconversion of Petroleum Residues." *Catalysis Today* 130:429–38.

Gawrys, Keith L., George a. Blankenship, and Peter K. Kilpatrick. 2006. "On the Distribution of Chemical Properties and Aggregation of Solubility Fractions in Asphaltenes." *Energy and Fuels* 20(2):705–14.

Gray, Mr, Farhad Khorasheh, and Se Wanke. 1992. "Role of Catalyst in Hydrocracking of Residues from Alberta Bitumens." *Energy & Fuels* 6:478–85.

Gray, MR, RR Tykwinski, JM Stryker, and Xiaoli Tan. 2011. "Supramolecular Assembly Model for Aggregation of Petroleum Asphaltenes." *Energy & Fuels* 25:3125–34.

Grenoble, DC, RB Long, and LL Murrell. 1984. "Separating Basic Asphaltenes Using Transition Metal Oxide Acid Catalysts." *US Patent 4,424,114*.

Groenzin, Henning and Oliver C. Mullins. 2000. "Molecular Size and Structure of Asphaltenes from Various Sources." *Energy & Fuels* 14(12):677–84.

Gross, Jürgen H. 2011. *Mass Spectrometry: A Textbook*. Springer Science & Business Media, 2011.

Guan, Shenheng and Alan G. Marshall. 1995. "Ion Traps for Fourier Transform Ion Cyclotron Resonance Mass Spectrometry: Principles and Design of Geometric and Electric Configurations." *International Journal of Mass Spectrometry and Ion*

Processes 146-147:261–96.

Hauser, André, Faisal AlHumaidan, Hassan Al-Rabiah, and Mamun Absi Halabi. 2014. "Study on Thermal Cracking of Kuwaiti Heavy Oil (Vacuum Residue) and Its SARA Fractions by NMR Spectroscopy." *Energy & Fuels* 28(7):4321–32.

Hsu, Chang S., Christopher L. Hendrickson, Ryan P. Rodgers, Amy M. McKenna, and Alan G. Marshall. 2011. "Petroleomics: Advanced Molecular Probe for Petroleum Heavy Ends." *Journal of Mass Spectrometry* 46(4):337–43.

Hsu, Chang S., Vladislav V. Lobodin, Ryan P. Rodgers, Amy M. McKenna, and Alan G. Marshall. 2011. "Compositional Boundaries for Fossil Hydrocarbons." *Energy and Fuels* 25:2174–78.

Hughey, C. a., C. L. Hendrickson, R. P. Rodgers, a. G. Marshall, and K. Qian. 2001. "Kendrick Mass Defect Spectrum: A Compact Visual Analysis for Ultrahigh-Resolution Broadband Mass Spectra." *Analytical Chemistry* 73(19):4676–81.

Hughey, Christine a, Ryan P. Rodgers, and Alan G. Marshall. 2002. "Resolution of 11,000 Compositionally Distinct Components in a Single Electrospray Ionization Fourier Transform Ion Cyclotron Resonance Mass Spectrum of Crude Oil." *Analytical chemistry* 74(16):4145–49.

Huo, Cheng et al. 2006. "Synthesis and Assembly with Mesoporous Silica MCM-48 of platinum(II) Porphyrin Complexes Bearing Carbazyl Groups: Spectroscopic and Oxygen Sensing Properties." *Inorganic chemistry* 45(12):4735–42.

Hurff, S. J. and M. T. Klein. 1983. "Reaction Pathway Analysis of Thermal and Catalytic Lignin Fragmentation by Use of Model Compounds." *Ind. Eng. Chem. Fundam.* 22(1981):426–30.

Islam, Annana, Yunju Cho, Arif Ahmed, and Sunghwan Kim. 2012. "Data Interpretation Methods for Petroleomics." *Mass Spectrometry Letters* 3(3):63–67.

Jackson, George, Alan G. Marshall, Christopher L. Hendrickson, and George S. Jackson. 1998. "Fourier Transform Ion Cyclotron Resonance Mass Spectrometry : A Primer." *Mass Spectrometry Reviews* 17(November):1–35.

Jansen, Tim et al. 2014. "Simulation of Petroleum Residue Hydroconversion in a Continuous Pilot Unit Using Batch Reactor Experiments and a Cold Mock-Up." *Industrial & Engineering Chemistry Research* 53(41):15852–61.

Jiménez Mateos, J. M. and J. L. G. Fierro. 1996. "X-Ray Photoelectron Spectroscopic Study of Petroleum Fuel Cokes." *Surface and Interface Analysis* 24:223–36.

Juyal, Priyanka et al. 2005. "Joint Industrial Case Study for Asphaltene Deposition." *Energy & Fuels* 27(4):1899–1908.

Juyal, Priyanka, Vickie Ho, Andrew Yen, and Stephan J. Allenson. 2012. "Reversibility of Asphaltene Flocculation with Chemicals." *Energy and Fuels* 26:2631–40.

Kaminski, Thomas J., H. Scott Fogler, Nick Wolf, Piyyarat Wattana, and Anurag Mairal. 2000. "Classification of Asphaltenes via Fractionation and the Effect of Heteroatom Content on Dissolution Kinetics." *Energy and Fuels* 14(1):25–30.

Karimi, Arash et al. 2011. "Quantitative Evidence for Bridged Structures in Asphaltenes by Thin Film Pyrolysis." *Energy and Fuels* 25:3581–89.

Kauppila, Tiina J. et al. 2002. "Atmospheric Pressure Photoionization Mass Spectrometry. Ionization Mechanism and the Effect of Solvent on the Ionization of Naphthalenes." *Analytical Chemistry* 74(21):5470–79.

Kawai, Hiroiku and Fumio Kumata. 1998. "Free Radical Behavior in Thermal Cracking Reaction Using Petroleum Heavy Oil and Model Compounds." *Catalysis Today* 43:281–89.

Kendrick, Edward. 1963. "A Mass Scale Based on CH=14.0000 for High Resolution Mass Spectrometry of Organic Compounds." *Analytical biochemistry* 35(13):2146–54.

Kilgour, David P. A., C. Logan Mackay, Patrick R. R. Langridge-Smith, and Peter B. O'Connor. 2012. "Appropriate Degree of Trust: Deriving Confidence Metrics for Automatic Peak Assignment in High-Resolution Mass Spectrometry." *Analytical Chemistry* 84:7431–35.

Kim, Sunghwan, Robert W. Kramer, and Patrick G. Hatcher. 2003. "Graphical Method for Analysis of Ultrahigh-Resolution Broadband Mass Spectra of Natural Organic Matter, the van Krevelen Diagram." *Analytical chemistry* 75(20):5336–44.

Klein, GC, Sunghwan Kim, RP Rodgers, AG Marshall, et al. 2006. "Differences between Pressure-Drop and Solvent-Drop Asphaltenes Determined by Electrospray Ionization Fourier Transform Ion Cyclotron Resonance Mass Spectrometry." *Energy & fuels* 20(9):1965–72.

Klein, GC, Sunghwan Kim, RP Rodgers, Alan G. Marshall, and Andrew Yen. 2006. "Mass Spectral Analysis of Asphaltenes. II. Detailed Compositional Comparison of Asphaltenes Deposit to Its Crude Oil Counterpart for Two Geographically Different Crude Oils by ESI FT-ICR MS." *Energy & fuels* 20(12):1973–79.

Klein, Geoffrey C., Annelie Angstro, Ryan P. Rodgers, and Alan G. Marshall. 2006. "Use of Saturates / Aromatics / Resins / Asphaltenes (SARA) Fractionation To Determine Matrix Effects in Crude Oil Analysis by Electrospray Ionization Fourier Transform Ion Cyclotron Resonance Mass Spectrometry." *Energy & Fuels* 20(10):668–72.

Knotnerus, J. A. N. 1967. "Constitution of Asphaltic Bitumen. Characterization of Bitumens by a Combination of Pyrolysis, Hydrogenation, and Gas-Liquid Chromatography." *Ind. Eng. Chem. Prod. Res. Dev.* 6(1):43–52.

Koots, J. Ann. and James G. Speight. 1975. "Relation of Petroleum Resins to Asphaltenes." *Fuel* 54(720):179–84.

Lababidi, Sami, Saroj K. Panda, Jan T. Andersson, and Wolfgang Schrader. 2013. "Direct Coupling of Normal-Phase High-Performance Liquid Chromatography to Atmospheric Pressure Laser Ionization Fourier Transform Ion Cyclotron Resonance Mass Spectrometry for the Characterization of Crude Oil." *Analytical chemistry* 85(20):9478–85.

Lalli, Priscila M., Yuri E. Corilo, Steven M. Rowland, Alan G. Marshall, and Ryan P. Rodgers. 2015. "Isomeric Separation and Structural Characterization of Acids in Petroleum by Ion Mobility Mass Spectrometry." *Energy & Fuels* 29(6):3626–33.

Li, Maowen, Stephen R. Larter, Daniel Stoddart, and Malvin Bjoroy. 1992. "Liquid Chromatographic Separation Schemes for Pyrrole and Pyridine Nitrogen Aromatic Heterocycle Fractions from Crude Oils Suitable for Rapid Characterization of Geochemical Samples." *Analytical Chemistry* 64(14):1337–44.

Li, Maowen, Steve R. Larter, Yeuge B. Frolov¹, and Malvin Bjoroy. 1994. "Adsorptive Interaction between Nitrogen Compounds and Organic And/or Mineral Phases in

Subsurface Rocks.” *Journal of High Resolution Chromatography* 17(4):230–36.

Liao, Zewen et al. 2004. “Adsorption/Occlusion Characteristics of Asphaltenes: Some Implication for Asphaltene Structural Features.” *Energy & Fuels* 19(1):180–86.

Liao, Zewen et al. 2006a. “Saturated Hydrocarbons Occluded inside Asphaltene Structures and Their Geochemical Significance, as Exemplified by Two Venezuelan Oils.” *Organic Geochemistry* 37(3):291–303.

Liao, Zewen et al. 2006b. “Saturated Hydrocarbons Occluded inside Asphaltene Structures and Their Geochemical Significance, as Exemplified by Two Venezuelan Oils.” *Organic Geochemistry* 37(3):291–303.

Liu, Dong et al. 2013. “Investigation on Asphaltene Structures during Venezuela Heavy Oil Hydrocracking under Various Hydrogen Pressures.” *Energy & Fuels* 27(7):3692–98.

Liu, He et al. n.d. “Characterization of Vanadyl and Nickel Porphyrins Enriched from Heavy Residues by Positive-Ion Electrospray Ionization FT-ICR Mass Spectrometry.” *Energy Fuels* 29(8):4803–13.

Liu, Peng, Chunming Xu, et al. 2010. “Characterization of Sulfide Compounds in Petroleum: Selective Oxidation Followed by Positive-Ion Electrospray Fourier Transform Ion Cyclotron Resonance Mass Spectrometry.” *Analytical Chemistry* 82(15):6601–6.

Liu, Peng, Quan Shi, et al. 2010. “Molecular Characterization of Sulfur Compounds in Venezuela Crude Oil and Its SARA Fractions by Electrospray Ionization Fourier Transform Ion Cyclotron Resonance Mass Spectrometry.” *Energy & Fuels*

24(9):5089–96.

Liu, Peng et al. 2011. “Distribution of Sulfides and Thiophenic Compounds in VGO Subfractions: Characterized by Positive-Ion Electrospray Fourier Transform Ion Cyclotron Resonance Mass Spectrometry.” *Energy and Fuels* 25(7):3014–20.

Lobodin, Vladislav V, Priyanka Juyal, Amy M. McKenna, Ryan P. Rodgers, and Alan G. Marshall. 2014. “Silver Cationization for Rapid Speciation of Sulfur-Containing Species in Crude Oils by Positive Electrospray Ionization Fourier Transform Ion Cyclotron Resonance Mass Spectrometry.” *Energy & fuels* 28:447–52.

Lobodin, Vladislav V., Priyanka Juyal, Amy M. McKenna, Ryan P. Rodgers, and Alan G. Marshall. 2014. “Lithium Cationization for Petroleum Analysis by Positive Ion Electrospray Ionization Fourier Transform Ion Cyclotron Resonance Mass Spectrometry.” *Energy & Fuels* 28(11):6841–47.

Lobodin, Vladislav V., Alan G. Marshall, and Chang Samuel Hsu. 2012. “Compositional Space Boundaries for Organic Compounds.” *Analytical Chemistry* 84:3410–16.

Lobodin, Vladislav V., Winston K. Robbins, Jie Lu, and Ryan P. Rodgers. 2015. “Separation and Characterization of Reactive and Non-Reactive Sulfur in Petroleum and Its Fractions.” *Energy & Fuels* 151006111527009.

Lu, Hong et al. 2014. “Molecular Characterization of Sulfur Compounds in Some Specieal Sulfur-Rich Chinese Crude Oils by FT-ICR MS.” *Science China Earth Sciences* 57(6):1158–67.

Marczewski, AW and Marta Szymula. 2002. “Adsorption of Asphaltenes from Toluene on Mineral Surface.” *Colloids and Surfaces A: Physicochemical and Engineering Aspects* 208:259–66.

Marshall, AG and RP Rodgers. 2004. "Petroleomics: The next Grand Challenge for Chemical Analysis." *Accounts of Chemical Research* 37:53–59.

Masters, C. D., D. H. Root, and E. D. Attanasi. n.d. "Resources Constraints in Petroleum Production Potential." *Science* 253:146–53.

McKenna, Amy M., Gregory T. Blakney, et al. 2010. "Heavy Petroleum Composition. 2. Progression of the Boduszynski Model to the Limit of Distillation by Ultrahigh-Resolution FT-ICR Mass Spectrometry." *Energy & Fuels* 24(5):2939–46.

McKenna, Amy M. et al. 2013. "Heavy Petroleum Composition. 3. Asphaltene Aggregation." *Energy & Fuels* 27(3):1246–56.

Mckenna, Amy M., Alan G. Marshall, and Ryan P. Rodgers. 2013. "Heavy Petroleum Composition. 4. Asphaltene Compositional Space." *Energy & fuels* 27:1257–67.

McKenna, Amy M., Jeremiah M. Purcell, Ryan P. Rodgers, and Alan G. Marshall. 2009. "Identification of Vanadyl Porphyrins in a Heavy Crude Oil and Raw Asphaltene by Atmospheric Pressure Photoionization Fourier Transform Ion Cyclotron Resonance (FT-ICR) Mass Spectrometry." *Energy & Fuels* 23(4):2122–28.

McKenna, Amy M., Jeremiah M. Purcell, Ryan P. Rodgers, and Alan G. Marshall. 2010. "Heavy Petroleum Composition. 1. Exhaustive Compositional Analysis of Athabasca Bitumen HVGO Distillates by Fourier Transform Ion Cyclotron Resonance Mass Spectrometry: A Definitive Test of the Boduszynski Model." *Energy & Fuels* 24(5):2929–38.

Mead, W. L. 1968. "Field Ionization Mass Spectrometry of Heavy Petroleum Fractions." *Analytical Chemistry* 40(4):743–47.

Mendoza de la Cruz, José L., Francisco J. Argüelles-Vivas, Víctor Matías-Pérez, Cecilia De Los a. Durán-Valencia, and Simón López-Ramírez. 2009. "Asphaltene-Induced Precipitation and Deposition During Pressure Depletion on a Porous Medium: An Experimental Investigation and Modeling Approach." *Energy & Fuels* 23(11):5611–25.

Mengual, O. 1999. "TURBISCAN MA 2000: Multiple Light Scattering Measurement for Concentrated Emulsion and Suspension Instability Analysis." *Talanta* 50(2):445–56.

Mengual, Olivier, Gérard Meunier, Isabelle Cayre, Katia Puech, and Patrick Snabre. 1999. "Characterisation of Instability of Concentrated Dispersions by a New Optical Analyser: The TURBISCAN MA 1000." *Colloids and Surfaces A: Physicochemical and Engineering Aspects* 152:111–23.

Merdrignac, Isabelle, AA Quoineaud, and Thierry Gauthier. 2006. "Evolution of Asphaltene Structure during Hydroconversion Conditions." *Energy & fuels* 20(9):2028–36.

Merdrignac, Isabelle, Anne Agathe Quoineaud, and Thierry Gauthier. 2006. "Evolution of Asphaltene Structure during Hydroconversion Conditions." *Energy and Fuels* 20(9):2028–36.

Milton, K. 1999. "A Hypothesis to Explain the Role of Meat-Eating in Human Evolution." *Evolutionary Anthropology Issues News and Reviews* 50:11–21.

Morimoto, Masato et al. 2015. "Asphaltene Aggregation Behavior in Bromobenzene Determined By Small-Angle X-Ray Scattering." *Energy & Fuels* 29(9):5737–43.

Mullins, Oliver C. 2011. "The Asphaltenes." *Annual review of analytical chemistry* 4:393–418.

Mullins, Oliver C. et al. 2012. "Advances in Asphaltene Science and the Yen–Mullins Model." *Energy & fuels* 26:3986–4003.

Murgich, Juan. 2002. "Intermolecular Forces in Aggregates of Asphaltenes and Resins." *Petroleum science and technology* 20:983–97.

Nawrocki, J. 1991. "Silica Surface Controversies, Strong Adsorption Sites, Their Blockage and Removal. Part I." *Chromatographia* 31:177–92.

Nicodem, David E., Carmen L. B. Guedes, and Rodrigo J. Correa. 1998. "Photochemistry of Petroleum." *Marine Chemistry* 63(1-2):93–104.

Oh, Kyeongseok, Terry A. Ring, and Milind D. Deo. 2004. "Asphaltene Aggregation in Organic Solvents." *Journal of Colloid and Interface Science* 271(1):212–19.

Omais, Badaoui, Nadège Charon, Marion Courtiade, Jérémie Ponthus, and Didier Thiébaud. 2013. "A Novel Analytical Approach for Oxygen Speciation in Coal-Derived Liquids." *Fuel* 104:805–12.

Parida, Sudam K., Sukalyan Dash, Sabita Patel, and B. K. Mishra. 2006. "Adsorption of Organic Molecules on Silica Surface." *Advances in colloid and interface science* 121(1-3):77–110.

Pernyeszi, Timea and Imre Dékány. 2001. "Sorption and Elution of Asphaltenes from Porous Silica Surfaces." *Colloids and Surfaces A: Physicochemical and Engineering Aspects* 194:25–39.

Plancher, H., S. M. Dorrence, and J. C. Petersen. 1977. "IDENTIFICATION OF

CHEMICAL TYPES IN ASPHALTS STRONGLY ADSORBED.” *Association of Asphalt Paving Technologists* 46:151–75.

Podgorski, David C. et al. 2013. “Heavy Petroleum Composition. 5. Compositional and Structural Continuum of Petroleum Revealed.” *Energy & Fuels* 27(3):1268–76.

Pomerantz, Andrew E., Matthew R. Hammond, Amy L. Morrow, Oliver C. Mullins, and Richard N. Zare. 2008. “Two-Step Laser Mass Spectrometry of Asphaltenes.” *Journal of the American Chemical Society* 130(23):7216–17.

Pomerantz, Andrew E., Matthew R. Hammond, Amy L. Morrow, Oliver C. Mullins, and Richard N. Zare. 2009. “Asphaltene Molecular-Mass Distribution Determined by Two-Step Laser Mass Spectrometry.” *Energy and Fuels* 23(3):1162–68.

Purcell, Jeremiah M. et al. 2010. “Stepwise Structural Characterization of Asphaltenes during Deep Hydroconversion Processes Determined by Atmospheric Pressure Photoionization (APPI) Fourier Transform Ion Cyclotron Resonance (FT-ICR) Mass Spectrometry.” *Energy & Fuels* 24(4):2257–65.

Purcell, Jeremiah M., Christopher L. Hendrickson, Ryan P. Rodgers, and Alan G. Marshall. 2007. “Atmospheric Pressure Photoionization Proton Transfer for Complex Organic Mixtures Investigated by Fourier Transform Ion Cyclotron Resonance Mass Spectrometry.” *Journal of the American Society for Mass Spectrometry* 18(9):1682–89.

Purcell, Jeremiah M., Ryan P. Rodgers, Christopher L. Hendrickson, and Alan G. Marshall. 2007. “Speciation of Nitrogen Containing Aromatics by Atmospheric Pressure Photoionization or Electrospray Ionization Fourier Transform Ion Cyclotron Resonance Mass Spectrometry.” *Journal of the American Society for Mass Spectrometry* 18:1265–73.

Qian, Kuangnan, Kathleen E. Edwards, John H. Diehl, and Larry a Green. 2004. "Fundamentals and Applications of Electrospray Ionization Mass Spectrometry for Petroleum Characterization." (14):1784–91.

Qian, Kuangnan, Kathleen E. Edwards, Anthony S. Mennito, Clifford C. Walters, and J. Douglas Kushnerick. 2010. "Enrichment, Resolution, and Identification of Nickel Porphyrins in Petroleum Asphaltene by Cyclograph Separation and Atmospheric Pressure Photoionization Fourier Transform Ion Cyclotron Resonance Mass Spectrometry." *Analytical Chemistry* 82(1):413–19.

Qian, Kuangnan, Anthony S. Mennito, Kathleen E. Edwards, and Dave T. Ferrughelli. 2008. "Observation of Vanadyl Porphyrins and Sulfur-Containing Vanadyl Porphyrins in a Petroleum Asphaltene by Atmospheric Pressure Photonization Fourier Transform Ion Cyclotron Resonance Mass Spectrometry." *Rapid communications in mass spectrometry : RCM* 22(14):2153–60.

Ramírez, Fernando Alvarez and Yosadara Ruiz Morales. 2013. "Island versus Archipelago Architecture for Asphaltenes: Polycyclic Aromatic Hydrocarbon Dimer Theoretical Studies." *Energy and Fuels* 27:1791–1808.

Robb, D. B., T. R. Covey, and a P. Bruins. 2000. "Atmospheric Pressure Photoionisation: An Ionization Method for Liquid Chromatography-Mass Spectrometry." *Analytical Chemistry* 72(15):3653–59.

Robbins, Winston K. 1998. "Quantitative Measurement of Mass and Aromaticity Distributions for Heavy Distillates 1 . Capabilities of the HPLC-2 System." *Journal of Chromatographic Sciences* 36(September):457–66.

Rodgers, RP, TM Schaub, and AG Marshall. 2005. "Petroleomics: MS Returns to Its

Roots.” *Analytical Chemistry* 77(1):20A – 27A.

Rodgers, Ryan P. and Alan G. Marshall. 2007. “Petroleomics: Advanced Characterization of Petroleum-Derived Materials by Fourier Transform Ion Cyclotron Resonance Mass Spectrometry (FT-ICR MS).” Pp. 63–93 in *Asphaltenes, Heavy Oils, and Petroleomics*, edited by O. C. Mullins, E. Y. Sheu, A. Hammam, and A. G. Marshall. Springer.

Rogel, Estrella, Michael Moir, and Matthias Witt. 2015. “Atmospheric Pressure Photoionization and Laser Desorption Ionization Coupled to Fourier Transform Ion Cyclotron Resonance Mass Spectrometry To Characterize Asphaltene Solubility Fractions: Studying the Link between Molecular Composition and Physical Behav.” *Energy & Fuels* 29:4201–9.

Rowland, Steven M., Winston K. Robbins, Yuri E. Corilo, Alan G. Marshall, and Ryan P. Rodgers. 2014. “Solid-Phase Extraction Fractionation To Extend the Characterization of Naphthenic Acids in Crude Oil by Electrospray Ionization Fourier Transform Ion Cyclotron Resonance Mass Spectrometry.” *Energy & fuels* 28:5043–48.

Rubinstein, I., C. Spyckerelle, and O. P. Strausz. 1979. “Pyrolysis of Asphaltenes: A Source of Geochemical Information.” *Geochimica et Cosmochimica Acta* 43:1–6.

Rueda-Velásquez, Rosa I., Howard Freund, Kuangnan Qian, William N. Olmstead, and Murray R. Gray. 2013. “Characterization of Asphaltene Building Blocks by Cracking under Favorable Hydrogenation Conditions.” *Energy and Fuels* 27:1817–29.

Ruiz-Morales, Yosadara. 2002. “HOMO-LUMO Gap as an Index of Molecular Size and Structure for Polycyclic Aromatic Hydrocarbons (PAHs) and Asphaltenes: A

Theoretical Study. I.” *Journal of Physical Chemistry A* 106(46):11283–308.

Sabbah, Hassan, Amy L. Morrow, Andrew E. Pomerantz, and Richard N. Zare. 2011. “Evidence for Island Structures as the Dominant Architecture of Asphaltenes.” *Energy and Fuels* 25:1597–1604.

Savage, Phillip E. and Michael T. Klein. 1988. “Asphaltene Reaction Pathways. 4. Pyrolysis of Tridecylcyclohexane and 2-Ethyltetralin.” *Industrial & Engineering Chemistry Research* 27(8):1348–56.

Savage, Phillip E., Michael T. Klein, and Simon G. Kukes. 1985. “Asphaltene Reaction Pathways. 1. Thermolysis.” *Industrial & Engineering Chemistry Process Design and Development* 24(4):1169–74.

Savage, Phillip E., Michael T. Klein, and Simon G. Kukes. 1988. “Asphaltene Reaction Pathways. 3. Effect of Reaction Environment.” *Energy & Fuels* 2(12):619–28.

Schaub, Tanner M. et al. 2005. “Speciation of Aromatic Compounds in Petroleum Refinery Streams by Continuous Flow Field Desorption Ionization FT-ICR Mass Spectrometry.” *Energy & Fuels* 19(9):1566–73.

Schuler, Bruno, Gerhard Meyer, Diego Peña, Oliver C. Mullins, and Leo Gross. 2015. “Unraveling the Molecular Structures of Asphaltenes by Atomic Force Microscopy.” *Journal of the American Chemical Society* 137(31):9870–76.

Schwager, I., WC Lee, and TF Yen. 1977. “Molecular Weight and Association of Coal-Derived Asphaltenes.” *Analytical Chemistry* 49(14):2363–65.

Sedghi, Mohammad and Lamia Goual. 2010. “Role of Resins on Asphaltene Stability.” *Energy and Fuels* 24(11):2275–80.

Senko, M. W., J. D. Canterbury, S. Guan, and a G. Marshall. 1996. "A High-Performance Modular Data System for Fourier Transform Ion Cyclotron Resonance Mass Spectrometry." *Rapid communications in mass spectrometry : RCM* 10:1839–44.

Shen, JH, AC Zettlemoyer, and K. Klier. 1980. "Water on Silica and Silicate Surfaces. 6. Sodium Form of Type-Y Synthetic Zeolite." *The Journal of Physical Chemistry* 84:1453–59.

Sheu, Eric Y. and David A. Storm. 2013. "Colloidal Properties of Asphaltenes in Organic Solvents." Pp. 1–50 in *Asphaltenes: fundamental and applications*, edited by M. Subirana and E. Y. Sheu. Springer Science & Business Media.

Shi, Quan et al. 2010. "Distribution of Acids and Neutral Nitrogen Compounds in a Chinese Crude Oil and Its Fractions: Characterized by Negative-Ion Electrospray Ionization Fourier Transform Ion Cyclotron Resonance Mass Spectrometry." *Energy & Fuels* 24(7):4005–11.

Shirokoff, John W., Mohammad N. Siddiqui, and Mohammad F. Ali. 1997. "Characterization of the Structure of Saudi Crude Asphaltenes by X-Ray Diffraction." *Energy & Fuels* 11(3):561–65.

Smith, Donald F. et al. 2014. "Direct Analysis of Thin-Layer Chromatography Separations of Petroleum Samples by Laser Desorption Ionization Fourier Transform Ion Cyclotron Resonance Mass Spectrometry Imaging." *Energy & Fuels* 28(10):6284–88.

Snape, Colin E. and Keith D. Bartle. 1985. "Further Information on Defining Asphaltenes in Terms of Average Structural Properties." *Fuel* 64(3):427–29.

Speight, J. G. 2004. "Petroleum Asphaltenes - Part 1: Asphaltenes, Resins and the Structure of Petroleum." *Oil and Gas Science and Technology* 59(5):467–77.

Speight, J. G., R. B. Long, T. D. Trowbridge, and N. J. Linden. 1982. "On the Definition of Asphaltenes." *Am. Chem. Soc., Div. Pet. Chem., Prepr.* 27(3):268–75.

Spiecker, P. Matthew, Keith L. Gawrys, and Peter K. Kilpatrick. 2003. "Aggregation and Solubility Behavior of Asphaltenes and Their Subfractions." *Journal of Colloid and Interface Science* 267(1):178–93.

Stenson, Alexandra C., Alan G. Marshall, and William T. Cooper. 2003. "Exact Masses and Chemical Formulas of Individual Suwannee River Fulvic Acids from Ultrahigh Resolution Electrospray Ionization Fourier Transform Ion Cyclotron Resonance Mass Spectra Molecular Formulas Have Been Assigned for 4626 Individual Mass Measurements." *Analytical Chemistry* 75(6):1275–84.

Stoyanov, Stanislav R. et al. 2010. "Computational and Experimental Study of the Structure, Binding Preferences, and Spectroscopy of nickel(II) and Vanadyl Porphyrins in Petroleum." *The journal of physical chemistry. B* 114(6):2180–88.

Strausz, Otto P., Thomas W. Mojelsky, and Elizabeth M. Lown. 1992. "The Molecular Structure of Asphaltene: An Unfolding Story." *Fuel* 71(12):1355–63.

Strausz, Otto P., Manuel Torres, Elizabeth M. Lown, Imre Safarik, and Juan Murgich. 2006. "Equipartitioning of Precipitant Solubles between the Solution Phase and Precipitated Asphaltene in the Precipitation of Asphaltene." *Energy and Fuels* 20(5):2013–21.

Syunyaev, R. Z., R. M. Balabin, I. S. Akhatov, and J. O. Safieva. 2009. "Adsorption of Petroleum Asphaltenes onto Reservoir Rock Sands Studied by Near-Infrared (

NIR) Spectroscopy†.” *Energy & fuels* 23(8):1230–36.

Tachon, Nadine, Farid Jahouh, Michel Delmas, and Joseph H. Banoub. 2011. “Structural Determination by Atmospheric Pressure Photoionization Tandem Mass Spectrometry of Some Compounds Isolated from the SARA Fractions Obtained from Bitumen.” *Rapid communications in mass spectrometry* 25(18):2657–71.

Takagi, Shinsuke et al. 2002. “High-Density Adsorption of Cationic Porphyrins on Clay Layer Surfaces without Aggregation: The Size-Matching Effect.” *Langmuir* 18(16):2265–72.

Tanaka, Ryuzo et al. 2004. “Characterization of Asphaltene Aggregates Using X-Ray Diffraction and Small-Angle X-Ray Scattering.” *Energy and Fuels* 18(7):1118–25.

Trejo, F., G. Centeno, and J. Ancheyta. 2004. “Precipitation, Fractionation and Characterization of Asphaltenes from Heavy and Light Crude Oils.” *Fuel* 83:2169–75.

Trejo, Fernando, Jorge Ancheyta, and MS Rana. 2009. “Structural Characterization of Asphaltenes Obtained from Hydroprocessed Crude Oils by SEM and TEM.” *Energy & Fuels* 23(5):429–39.

Vaz, Boniek G. et al. 2013. “Assessing Biodegradation in the Llanos Orientales Crude Oils by Electrospray Ionization Ultrahigh Resolution and Accuracy Fourier Transform Mass Spectrometry and Chemometric Analysis.” *Energy & Fuels* 27:1277–84.

Vela, Jesus, VL Cebolla, Luis Membrado, and Jose M. Andrés. 1995. “Quantitative Hydrocarbon Group Type Analysis of Petroleum Hydroconversion Products Using

an Improved TLC-FID System.” *Journal of Chromatographic Science* 33(August):417–25.

Wang, Jianxin and Jill S. Buckley. 2003. “Asphaltene Stability in Crude Oil and Aromatic Solvents - The Influence of Oil Composition.” *Energy and Fuels* 17(6):1445–51.

Wang, Shanshan, Qi Liu, Xiaoli Tan, Chunming Xu, and MR Gray. 2013. “Study of Asphaltene Adsorption on Kaolinite by X-Ray Photoelectron Spectroscopy and Time-of-Flight Secondary Ion Mass Spectroscopy.” *Energy & Fuels* 27:2465–73.

Wiehe, Irwin A. .. 2008. “Thermal Conversion Kinetics.” Pp. 97–179 in *Process chemistry of petroleum macromolecules*. CRC Press.

Wiehe, Irwin A. and Raymond J. Kennedy. 2000. “Oil Compatibility Model and Crude Oil Incompatibility.” *Energy and Fuels* 14(1):56–59.

Wu, Zhigang, Ryan P. Rodgers, and Alan G. Marshall. 2004. “Two- and Three-Dimensional van Krevelen Diagrams: A Graphical Analysis Complementary to the Kendrick Mass Plot for Sorting Elemental Compositions of Complex Organic Mixtures Based on Ultrahigh-Resolution Broadband Fourier Transform Ion Cyclotron Resonance.” *Analytical chemistry* 76(9):2511–16.

Xu, Hai, Guohe Que, Daoyong Yu, and Jian R. Lu. 2005. “Characterization of Petroporphyrins Using Ultraviolet - Visible Spectroscopy and Laser Desorption Ionization Time-of-Flight Mass Spectrometry.” (6):517–24.

Yang, Xiaoli, Hassan Hamza, and Jan Czarnecki. 2004. “Investigation of Subfractions of Athabasca Asphaltenes and Their Role in Emulsion Stability.” *Energy and Fuels* 18(3):770–77.

- Yen, Teh Fu., J. G. Erdman, and S. S. Pollack. 1961. "Investigation of the Structure of Petroleum Asphaltenes by X-Ray Diffraction." *Analytical Chemistry* 33(11):1587–94.
- Yokota, Toshitsugu, Fiona Scriven, Douglas S. Montgomery, and Otto P. Strausz. 1986. "Absorption and Emission Spectra of Athabasca Asphaltene in the Visible and near Ultraviolet Regions." *Fuel* 65(8):1142–49.
- Yudin, I. K. et al. 1998. "Crossover Kinetics of Asphaltene Aggregation in Hydrocarbon Solutions." *Physica A: Statistical Mechanics and its Applications* 251(1–2):235–44.
- Yudin, I. K. et al. 1998. "Mechanisms of Asphaltene Aggregation in Toluene–heptane Mixtures." *Journal of Petroleum Science and Engineering* 20(3-4):297–301.
- Yudin, Igor K. and Mikhail A. Anisimov. 2007. "Dynamic Light Scattering Monitoring of Asphaltene Aggregation in Crude Oils and Hydrocarbon Solutions." Pp. 439–68 in *Asphaltenes, Heavy Oils, and Petroleomics*, edited by O. C. Mullins, E. Y. Sheu, A. Hammami, and A. G. Marshall. Springer.
- Zahabi, Atoosa, Murray R. Gray, and Tadeusz Dabros. 2012. "Kinetics and Properties of Asphaltene Adsorption on Surfaces." *Energy & Fuels* 26(2):1009–18.
- Zequan, L. I. U., Yang Xue, and Zhang Qiang. 2014. "TURBISCAN: History , Development , Application to Colloids and Dispersions." 936:1592–96.
- Zhao, Jing et al. 2010. "Comparative Studies on Compounds Occluded inside Asphaltenes Hierarchically Released by Increasing Amounts of H₂O₂/CH₃COOH." *Applied Geochemistry* 25(9):1330–38.