
Compositional variability in bitumens from eolian sedimentary *facies* of Pirambóia formation, Paraná basin, Brazil: characterizing biodegradation

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Resumo

Acumulações de betume foram identificados dentro de facies sedimentares eólicas da Formação Pirambóia, na margem leste da Bacia do Paraná no Brasil. Esta bacia é conhecida por ter acumulações de areia oleosa lavráveis significativas (areias betuminosas), com uma das 19 ocorrências com uma quantidade de óleo estimada em 6 milhões de barris. A variabilidade de composição de uma série deste alcatrão betuminoso de areia retirada de quatro facies sedimentares eólicas identificadas (foresets e bottomset de dunas, interdunas secas e úmidas) foi determinada por meio de técnicas diferentes para caracterizar e avaliar a biodegradação. Resultados obtidos sugerem que betumes de facies interdunas secas sofreram a biodegradação mais grave e as facies bottomset contém o óleo menos biodegradado. Além disso, alguns compostos (C₂₃ terpane tricíclico, C₂₉ hopane, gammacerane e diastereómeros C₂₇) apresentam elevada resistência à degradação microbiana, provando que estes biomarcadores pode ser úteis para análise de biodegradação em óleos altamente biodegradados, tais como betumes. A análise do biomarcador aromático, no entanto, mostra que esta classe de compostos não é útil para a análise de biodegradação amostras de afloramentos e areias de alcatrão, porque eles são mais susceptíveis a outros processos de alteração secundária, tais como lavagem com água. A grande importância deste estudo é o de permitir a estimativa mais precisa da composição química dos betumes de facies eólico levando a melhorar as previsões sobre a intensidade da biodegradação em diferentes partes do reservatório de arenito Pirambóia.

Palavras-chave: biomarcadores de petróleo, óleo pesado, areia alcatrão, Bacia do Paraná

Abstract

Bitumen accumulations were identified within eolian sedimentary facies of Pirambóia Formation, in the eastern margin of Paraná Basin in Brazil. This basin is known to have significant mineable oil sand accumulations (tar sands), with one of the 19 occurrences with amount of oil in place estimated in 6 million barrels. The compositional variability in a series of this tar sand bitumens taken from four identified eolian sedimentary facies (foreset and bottomset of dunes, dry and wet interdune) was determined by different techniques to characterize and evaluate biodegradation. Obtained results suggest that bitumens from dry interdune facies have suffered the most severe biodegradation and the bottomset facies contains the least biodegraded oil. Furthermore, some compounds (C₂₃ tricyclic terpane, C₂₉ hopane, gammacerane and C₂₇ diasteranes) show high resistance to microbial degradation, proving that these biomarkers can be useful to biodegradation analysis in highly biodegraded oils, such as bitumens. Aromatic biomarker analysis, however, show that this class of compounds is not useful for biodegradation analysis of outcrops samples and tar sands, because they are more susceptible to other secondary alteration processes, such as water washing. The major significance of this study is to enable more accurate estimation of the chemical composition of the bitumens from eolian facies leading to improve predictions on biodegradation intensity in different parts of Piramboia sandstone reservoir.

Keywords: petroleum biomarker, heavy oil, tar sand, Paraná Basin.

1. INTRODUCTION

Tar sands are consolidated sandstones filled with highly viscous and extra heavy oil, also called natural bitumen. According Meyer & Witt (1990), natural bitumens are semisolid or solid mixtures of hydrocarbons and as much as 50 percent heterocyclic compounds, composed largely of carbon and hydrogen, but have substituents of nitrogen, oxygen, sulfur and trace metals. Moreover, they are often characterized by fluids that have variable physical and chemical properties, such as viscosity (Koopmans *et al.* 2002). One possible cause of these variable properties is the different extent of secondary alteration processes, such as water washing, bacterial degradation, in reservoir maturation, and fractionation of light and heavy ends due to migration, which represent post-accumulation alteration of petroleum in the reservoir (Seifert & Moldowan 1979, Brooks *et al.* 1988, Palmer 1993, Jones *et al.* 2008, Jia *et al.* 2010).

The production of extra heavy oils and bitumens is likely to increase worldwide as light conventional oil resources are depleted (Koopmans *et al.* 2002). Nowadays, bitumen has a great potential value as a component of global energy resources and it is up to ten times more abundant than conventional oil (Wu *et al.* 2013). The vast deposits of bitumens are common in shallow reservoirs on the flanks of foreland basins in North and South America (Head *et al.* 2003).

A significant mineable oil sands accumulation in Brazil is in the Paraná Basin, which have approximately 19 occurrences in the eastern part, mostly occurring in sandstones of the Triassic Pirambóia Formation. The Anhembi occurrence is apparently the one with higher oil potential with amount of oil in place estimated in 6 million barrels of oil (Thomaz Filho 1982). In the present time, this accumulation has a heavy and high-viscosity oil (near 5° API), with medium-to-high sulfur content (2-3% by weight) (Thomaz Filho *et al.* 2008). These oil properties suggest that this petroleum accumulation has been degraded microbially over geological timescales because biodegradation is a primary control on fluid properties in many heavy oil settings (Larter *et al.* 2012).

Crude oil biodegradation proceeds as a series of changes in the physical and chemical properties of the oil. Many of these changes are characteristics of biodegradation rather than physical removal processes (Head *et al.* 2010). Understanding and interpreting quantitative

parameters for biodegradation evaluation, including geological conditions and biochemical pathways, has been extensively studied (Wilkes *et al.* 2000, Aitken *et al.* 2004, Peters *et al.* 2005, da Cruz *et al.* 2008, da Cruz *et al.* 2011). Although simultaneous removal of different compound classes occurs during biodegradation, the different rates of degradation of various components result in the apparent sequential removal of specific compound classes (Head *et al.* 2003). Typically, bacterial degradation starts from linear and branched alkanes. At very advanced levels of degradation, some characteristic compounds can be generated such as 17 α , 25-norhopanes and naphthenic acids. Aromatic steroid hydrocarbons are among the most persistent compounds and are only removed when the oil is very extensively biodegraded (Peters *et al.* 2005, Head *et al.* 2010).

The relatively systematic changes in chemical composition of petroleum have been used by petroleum geochemists to formulate a number of qualitative biodegradation scales, of which the most widely used was suggested by Peters & Moldowan (1993) that ranges from 0 to 10, with rank 1 indicating a slight biodegradation while rank 10 denotes severe effects. Another way to study biodegradation is using geochemical parameters, that are biomarker ratios obtained by conventional gas chromatography-mass spectrometry (GC-MS), since the biomarkers are widely distributed and easily detected by this technique (Wu *et al.* 2013). Since some these compounds are more resistant to biodegradation than others, some biomarker ratios can be used to estimate and compare the biodegradation degree in different oils (Peters *et al.* 2005).

It has been reported that tricyclic terpanes and gammacerane are more resistant to biodegradation than 17 α -hopanes (Seifert & Moldowan 1979, Brooks *et al.* 1988, Jiang *et al.* 1990, Alberdi *et al.* 2001, Jia *et al.* 2010). It has also been reported that diasteranes are more resistant to biodegradation than regular steranes (Seifert & Moldowan 1979, Brooks *et al.* 1988) and that the steranes isomers susceptibility to microbial degradation is as follows: $\alpha\alpha\alpha$ 20R >> $\alpha\beta\beta$ 20R >> $\alpha\beta\beta$ 20S >> $\alpha\alpha\alpha$ 20S (Peters *et al.* 2005). Another observation from previous studies on polycyclic aromatic hydrocarbons is that alkylphenanthrenes substitution at the 9 or 10 position suggests increased resistance to biodegradation (Rowland *et al.* 1986, Budzinski *et al.* 1995; Huang *et al.* 2004) and that

monoaromatic steroid hydrocarbons appear to be more resistant to microbial degradation than the triaromatic steroid hydrocarbons (Lin *et al.* 1989). Therefore, C₂₃ tricyclic terpane/C₃₀ hopane, gammacerane/C₃₀ hopanes, C₂₇ $\alpha\alpha\alpha$ 20S/(20S + 20R), C₂₈ $\alpha\alpha\alpha$ 20S/(20S + 20R), C₂₉ $\alpha\alpha\alpha$ 20S/(20S + 20R), C₂₇ diasteranes/(C₂₇ diasteranes + C₂₇ steranes), 9-methylphenanthrene/3-methylphenanthrene, 9-methylphenanthrene/2-methylphenanthrene, MA/(MA + TA) (monoaromatic/triaromatic steroids ratio) increases with the biodegradation.

Although reservoir geochemistry has been widely applied to investigations of reservoir continuity, field charging, tar mats in light oil reservoirs, and tubing leakage issues, among others (Kaufman *et al.* 1990, Wilhelms & Larter 1994a, Wilhelms & Larter 1994b, Larter & Aplin 1995), almost no reservoir geochemical studies of bitumen fields have considered the compositional variability of the petroleum among sedimentary facies using biomarker parameters. The Paraná Basin in Brazil has excellent outcrops of tar sand reservoirs to make this kind of study, where oil has been shown to occur in fluvial and eolian sedimentary facies of the lower Pirambóia Formation (Araújo *et al.* 2005, Thomaz Filho *et al.* 2008).

1.1. Paraná Basin

The Paraná Basin is a very large intracratonic basin, which measures over 1,000,000 km² in Brazil, and 600,000 km² in Argentina, Paraguay and Uruguay. In this basin there are tar sands outcrops in eolian reservoir rocks attributed to the Irati-Pirambóia petroleum system (Cerqueira & Santos Neto 1986). Source maturation is the result of heating by dolerite intrusions (diabase bodies) that fed the Lower Cretaceous Serra Geral flood basalts (Thomaz Filho 1982). Hydrocarbon accumulation occurs along fault zones or lineaments, especially near structural highs (Araújo *et al.* 2005).

The tar sands in the Paraná Basin are sandstones impregnated with extra heavy oil (bitumen) and is restricted to eastern margin of the basin. There are about 19 known localities clustered in two areas in São Paulo State. The Betumita Farm area is one of the major extra heavy oil exposition in this State, pouring from

Facies can be defined as a rock unit based on its distinctive lithologic features, including composition, grain size, bedding characteristics, and sedimentary structure. Each facies represents an individual depositional event (Miall 2000). Perhaps different sedimentary facies in the same formation can indicate reservoir compartmentalization, since these facies have variable features, as porosity and lithology. These features can induce the secondary alteration process, and the oil from identically sourced oils may have different composition based on the facies (Garcia *et al.* 2011).

Garcia *et al.* (2011) suggested using SARA analysis that there is correlation between the types of eolian sedimentary facies of Pirambóia Formation and its bitumens characteristics, which were probably influenced by a post-filling process, such as water washing and biodegradation. However, an analysis at the molecular level had not been performed. Thus, the aim of this work was study the compositional variability by biomarkers analyses among bitumens from eolian sedimentary facies in the Pirambóia Formation, Paraná Basin, Brazil. This kind of study may assist in predicting de quality of the mineable oil in tar sand reservoir prior to decision of what facies are better to explore.

sandstones of the Pirambóia Formation. The oil in place was estimated at 6 million barrels based on a mean content of 5.5% of oil in weight (Araújo *et al.* 2005; Thomaz Filho *et al.* 2008).

The Pirambóia Formation is divided in four eolian sedimentary facies (shown in Fig. 1), with different physicochemical characteristics (Caetano-Chang 1997, Martins *et al.* 2014): dune foreset, which have better permo-porosity and dune bottomset, which have intermediate permo-porosity, with both facies formed in high energy settings by the deposition of dunes. Conversely, wet and dry interdune facies, which have the worse permo-porosity, with both facies formed under lower energy settings by the elevation of the groundwater level and show semi-horizontal stratification. The interdune facies have a higher quantity of argillaceous minerals than the other facies (Garcia *et al.* 2011; Antunes *et al.* 2014).

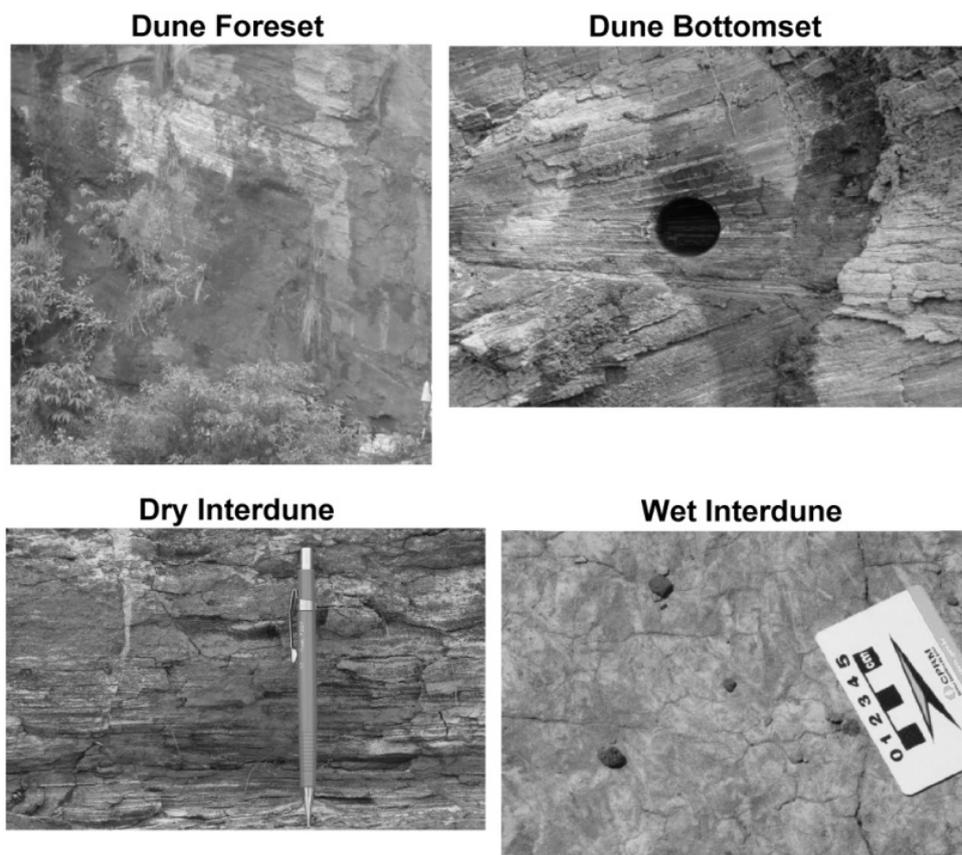


Figura 1- Outcrops of typical sedimentary facies from Pirambóia Formation: dune foreset and dune bottomset, from Betumita Farm; dry interdune from Mina Farm; wet interdune from Guareí, São Paulo State.

2. MATERIALS AND METHODS

2.1 Samples and analytical methods

Rock samples comprise outcrop tar sands of Pirambóia Formation, which were collected during geological fieldwork and cataloged according to their sedimentary facies. They came from four different localities in São Paulo State (Shown in Fig. 2): Mina Farm; Betumita Farm; Bofete Farm; and Guareí. It was selected five samples of each sedimentary facies: dry interdune (S1 to S5); dune foreset (S6 to S10); wet interdune (S11 to S15); and dune bottomset (S16 to S20).

The rock samples were all crushed and pulverized before bitumen extraction with dichloromethane using a Soxhlet system.

2.2 GC-FID and GC-MS analysis

The GC-FID analyses on the saturated hydrocarbon fractions were performed using an Agilent 6890N gas chromatography with flame ionization detector (using synthetic air, H₂ and N₂) equipped with a HP-5 fused silica capillary column (30 m x 0.32 mm x 0.25 µm film

thickness, Agilent, USA). The GC oven was ramped from 40 °C to 320 °C at 2.5 °C min⁻¹, with a final temperature held for 18 minutes. The injector temperature was 280 °C and the FID was set at 340 °C. Helium was used as the carrier gas, with a 1mL/min constant flow. Asphaltenes were precipitated with n-pentane, and then filtrated to obtain the malthe fractions. The malthe fractions were then separated into saturate, aromatic and resin fractions by successive elutions with hexane, hexane/dichloromethane (8:2, v/v), and dichloromethane/methanol (9:1, v/v), respectively, using pre-activated silica gel column chromatography, according to Martins *et al.* (2014). The saturate hydrocarbon fractions were analyzed by GC-FID and GC-MS and the aromatic hydrocarbon fractions were analyzed by GC-MS.

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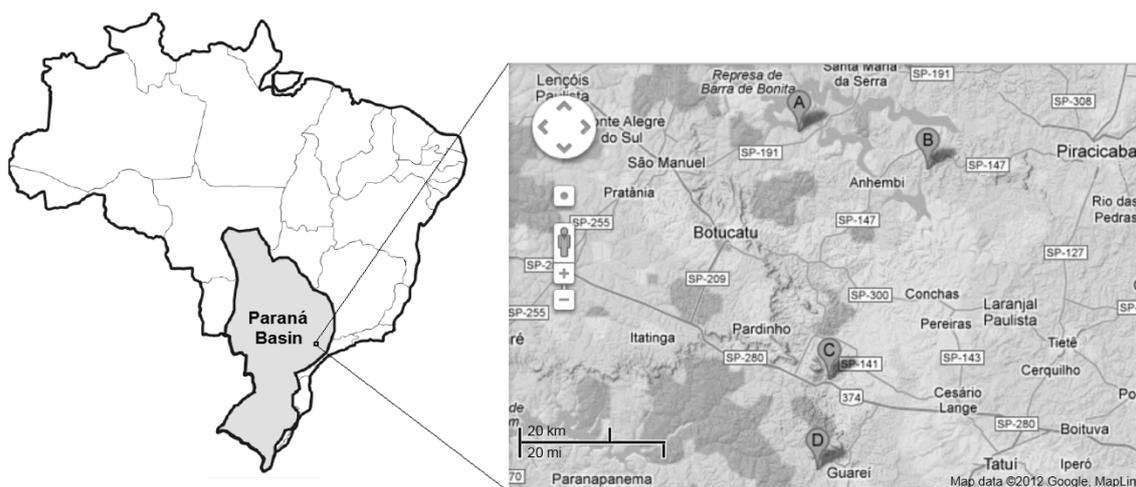


Fig. 2. Location maps of Paraná Basin in Brazil (modified from Anjos & Guimarães 2008), and the study area where the outcrop tar sands were collected (Google Maps): (A) Mina Farm (samples S1 to S4, and S13); (B) Betumita Farm (samples S6 to S10, and S15 to S20); (C) Bofete Farm (samples S11 and S12); (D) Guareí (samples S5 e S14).

Analysis of the saturated and aromatic hydrocarbon fractions were conducted using Agilent 6890N Gas Chromatography, equipped with a DB5MS (30 m x 0.25 mm x 0.25 μm film thickness) coupled to an Agilent 5973-MSD mass spectrometer. The GC oven for the saturated hydrocarbon analysis was isothermally held at 60 $^{\circ}\text{C}$ for 2 min, ramped from 60 $^{\circ}\text{C}$ to 200 $^{\circ}\text{C}$ at 22 $^{\circ}\text{C min}^{-1}$, then isothermally held at 200 $^{\circ}\text{C}$ for 3 min, and finally ramped from 200 $^{\circ}\text{C}$ to 320 $^{\circ}\text{C}$ at 3 $^{\circ}\text{C min}^{-1}$, with the final temperature held for 25 minutes. The GC oven for the aromatic hydrocarbon was isothermally held at 70 $^{\circ}\text{C}$ for

1 min, ramped from 70 $^{\circ}\text{C}$ to 110 $^{\circ}\text{C}$ at 12 $^{\circ}\text{C min}^{-1}$, then isothermally held at 110 $^{\circ}\text{C}$ for 22 min, and finally ramped from 110 $^{\circ}\text{C}$ to 320 $^{\circ}\text{C}$ at 1.5 $^{\circ}\text{C min}^{-1}$, with the final temperature held for 3 minutes. Helium was used as the carrier gas, with a 1mL/min constant flow. The injector and transfer line temperatures were 300 $^{\circ}\text{C}$. The MS was operated with an ionization energy of 70 eV and a source temperature of 230 $^{\circ}\text{C}$. Injection was performed in the splitless mode, with an injected sample of 1 μL . Data were collected in the selected ion monitoring (SIM) mode.

3. RESULTS AND DISCUSSION

3.1. SARA and hydrocarbon analysis

The gross composition (%) of saturated hydrocarbon, aromatic hydrocarbon, resin and asphaltene fractions obtained by precipitation of asphaltenes and by liquid chromatography of maltheane are given in Table 1. The abundances of polar compounds (resins and asphaltenes) are higher than the other fractions in all samples, as expected to bitumens.

In general, samples from the dry interdune facies show a higher content of asphaltenes and resins than samples from the other facies, in agreement with the results obtained by Garcia *et al.* (2011). The S6 sample from the dune foreset facies, and S13 and S15 samples from the wet interdune facies also show higher content of polar compounds. This result suggests highest biodegradation degree for these bitumens, because biodegradation leads to a decrease in saturated hydrocarbon content

(and to smaller decrease in aromatic hydrocarbon content) through oxidation. This biodegradation process produces compounds such as alcohols, phenols and carboxylic acids among others, that become part of altered resin fraction (Head *et al.* 2003, Liao *et al.* 2009). The relative content of asphaltene fraction tends to increase with biodegradation, not only because saturated and aromatic hydrocarbons tend to decrease, but also because some new oxidized compounds could be closely linked with functionalities of resins and asphaltenes and form new molecules of resins and asphaltenes. Those enlarged resins may be precipitated by hexane as newly generated asphaltenes during separation, and the amount of asphaltene fraction will be significantly increased during biodegradation (Liao *et al.* 2009).

Table 1 Gross composition (%) to each fraction obtained by SARA analysis for the bitumens from eolian facies of Pirambóia Formation, Paraná Basin.

Facies	Sample	^a Sat%	^b Aro%	^c Res%	^d Asp%
Dry Interdune	S1	6	2	16	76
	S2	6	2	16	76
	S3	6	3	16	75
	S4	9	3	19	69
	S5	6	1	24	69
Dune Foreset	S6	3	5	20	72
	S7	17	18	35	30
	S8	20	13	39	28
	S9	15	19	36	30
	S10	13	19	36	32
Wet Interdune	S11	11	14	38	37
	S12	12	14	38	36
	S13	5	3	9	83
	S14	9	13	40	38
	S15	3	5	17	75
Dune Bottomset	S16	13	7	59	21
	S17	12	16	10	62
	S18	22	27	17	34
	S19	26	31	13	30
	S20	13	24	30	33

^aSat = Saturated; ^bAro = Aromatic; ^cRes = Resin; ^dAsp = Asphaltene.

The chromatograms obtained by GC-FID analysis (usually called fingerprints) of saturated fractions for all 20 samples reveal absence of n-alkanes and isoprenoids, and very low abundance of cyclic hydrocarbons, mainly those with low molecular weight, as can be observed in the examples shown in Fig. 3 (for more detail, see all chromatograms in Martins *et al.* 2014). The GC-FID chromatograms also reveal a significant unresolved complex mixture (UCM), which appears as a hump in gas chromatograms (Head *et al.* 2010). These results correspond with the typical effects of severe biodegradation, and then all samples

have been heavily degraded by microorganisms. However, the chromatograms obtained in bitumens from the dry interdune facies apparently have the highest UCM and lower abundance of n-alkanes (Fig. 3a) than the bitumens from the other facies (Fig. 3b, c and d), which suggest that these samples are more biodegraded, in agreement with the results obtained by SARA analysis.

Thus, SARA and hydrocarbon analysis seems useful as a prior analysis to evaluate the compositional variability and the biodegradation extent among bitumens samples from tar sands.

3.2. Biomarker analysis

Utilizing GC-MS, we can find out the influence of biodegradation on biomarkers. In this work, geochemical parameters based on biomarker ratios of terpane, sterane, and some aromatic compounds are used to evaluate the

extent of biodegradation of the bitumens from eolian sedimentary facies of Pirambóia tar sands. Some geochemical parameters calculated for the 20 samples are presented in Table 2.

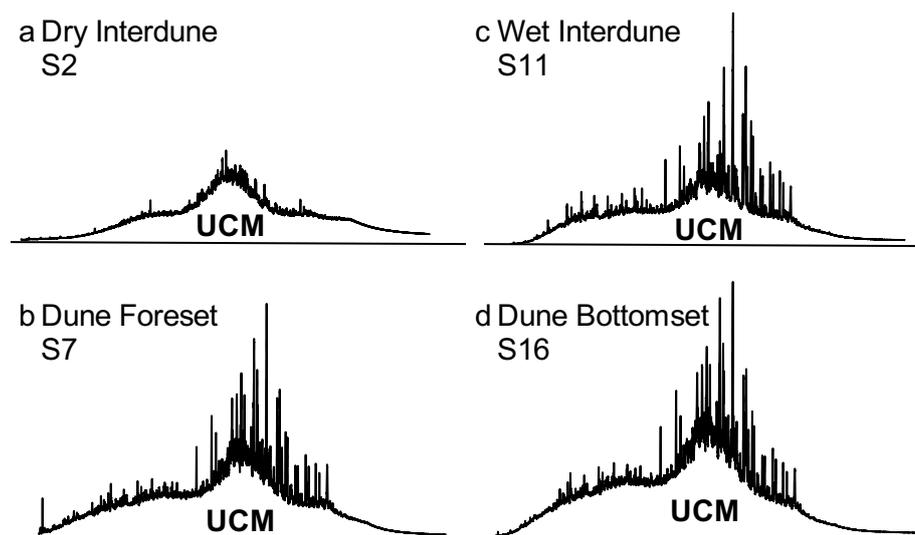


Figure 3. Chromatograms obtained by GC-FID of bitumens from each facies of Pirambóia Formation, Paraná Basin.

3.3. Terpanes

The distributions of terpanes (m/z 191) as obtained by GC-MS analysis of saturated hydrocarbons for one sample from each facies are given in Fig. 4. Tricyclic terpanes from C_{19} to C_{25} are detected, but the complete series was not found in all 20 samples, with the C_{19} , C_{20} and C_{21} tricyclic terpanes not detected in S6, S13 and S15 bitumen samples. The C_{23} tricyclic terpane appears to be the most resistant to biodegradation in the tricyclic terpane class and is present in all samples. Pentacyclic terpanes in most bitumens from Pirambóia tar sands were dominated by C_{29} hopanes, C_{30} hopanes and gammacerane, and the relative intensity of homohopanes generally decreases with an increasing carbon number. Gammacerane presents high resistance to biodegradation, showing high relative intensity in samples where the C_{30} hopane was almost not detected, as in the sample from the interdune facies. The relative intensity of tricyclic terpanes was lower than that of hopanes in all samples, as can be observed in Figure 4.

The highest value of the biomarkers ratios is from sample S14 because it outcrops in a locality that has intense exposition to the meteoric water, since this region is periodically flooded (Fig. 5).

The high resistance of gammacerane, C_{23} tricyclic terpane and C_{29} hopane to severe microbial degradation indicate that these compounds can be used as good biomarkers for

The terpane distribution in Pirambóia tar sands from the dry interdune facies differ from the other facies, as can also be observed in Fig. 4. Although terpanes and steranes are resistant to biodegradation, some studies have shown that they can be degraded under severe conditions of biodegradation (Chosson *et al.* 1991), as can be observed for the S2 sample, from dry interdune facies.

The geochemical parameters C_{23} tricyclic terpane/ C_{30} hopane, gammacerane/ C_{30} hopane, gammacerane/ C_{31} hopanes (S + R), and C_{29}/C_{30} show highest values for samples from the dry interdune facies (A, B, C and D ratios, Table 2). Since it has been reported that C_{23} tricyclic terpane and gammacerane are more resistant to biodegradation than 17α -hopanes (Seifert & Moldowan 1979, Brooks *et al.* 1988, Zhang *et al.* 1988, Jiang *et al.* 1990, Alberdi *et al.* 2001, Jia *et al.* 2010), higher values of these ratios in samples from the dry interdune facies could indicate the more intense biodegradation level to this facies. The correlation among these ratios is showed in Fig. 5.

biodegradation studies specially in tar sands and outcropped samples.

The m/z 191 mass chromatograms of most samples, except samples S2, S5 (dry interdune facies) and S14 (wet Interdune facies), show 25-norhopanes (C_{29}), but in lower abundance to samples from the dry interdune facies. A whole series of 25-norhopanes and 17-nortricyclic terpanes are detected in m/z 177 mass chromatograms for most samples (Fig. 6).

Table 2 Selected geochemical parameters for the bitumens from eolian sedimentary facies of Pirambóia Formation, Paraná Basin.

Facies	Sample	A	B	C	D	E	F	G	H	I	J	L	M	N
Dry Interdune	S1	1.55	3.48	4.23	4.26	1.61	n.d.	n.d.	n.d.	0.66	0.12	0.86	0.78	n.d.
	S2	2.53	3.76	7.02	4.06	n.d.	n.d.	n.d.	n.d.	0.55	0.16	0.82	0.74	n.d.
	S3	0.50	0.89	1.27	1.36	0.20	n.d.	n.d.	0.69	0.57	0.35	0.95	0.80	n.d.
	S4	1.09	2.67	3.90	3.82	n.d.	n.d.	n.d.	n.d.	0.62	0.13	0.90	0.80	n.d.
	S5	0.46	2.12	3.12	1.37	n.d.	0.50	0.51	0.43	0.35	0.39	0.96	0.83	n.d.
Dune Foreset	S6	0.30	0.77	0.92	0.65	0.24	0.51	0.73	0.50	0.45	0.37	0.95	0.82	0.96
	S7	0.24	0.78	0.89	0.67	0.36	0.48	0.62	0.43	0.43	0.35	1.31	1.79	0.74
	S8	0.32	0.70	0.90	0.71	0.44	0.49	0.62	0.43	0.44	0.32	1.20	n.d.	0.92
	S9	0.26	0.73	0.88	0.68	0.31	n.d.	n.d.	n.d.	0.60	0.24	2.20	n.d.	0.65
	S10	0.31	0.64	0.85	0.69	0.31	0.49	0.62	0.44	0.45	0.32	1.27	n.d.	0.62
Wet Interdune	S11	0.26	0.94	1.14	0.66	n.d.	0.53	0.60	0.49	0.41	0.38	0.95	0.62	0.73
	S12	0.30	0.87	1.21	0.65	0.09	0.53	0.61	0.48	0.42	0.37	1.02	0.66	0.78
	S13	0.33	0.66	0.82	0.84	0.31	0.70	n.d.	0.70	0.54	0.28	n.d.	n.d.	n.d.
	S14	3.32	12.44	14.07	8.53	n.d.	n.d.	n.d.	0.86	0.45	0.35	1.17	n.d.	0.46
	S15	0.36	0.76	0.91	0.68	0.26	0.51	0.76	0.47	0.50	0.35	1.00	0.91	0.96
Dune Bottomset	S16	0.35	0.76	0.88	0.72	0.46	0.49	0.63	0.45	0.44	0.34	1.13	1.69	0.90
	S17	0.23	0.69	0.91	0.64	0.34	0.49	0.63	0.43	0.44	0.34	1.37	1.75	0.62
	S18	0.28	0.65	0.87	0.66	0.40	0.48	0.63	0.44	0.45	0.34	1.28	2.43	0.58
	S19	0.21	0.69	0.86	0.66	0.29	0.48	0.63	0.42	0.43	0.35	1.35	1.60	0.61
	S20	0.27	0.63	0.83	0.69	0.25	0.48	0.61	0.44	0.44	0.33	1.31	1.52	0.54

A = C₂₃ tricyclic terpane/C₃₀ hopane; B = gammacerane/C₃₀ hopane; C = gammacerane/C₃₁ hopanes (S + R); D = C₂₉/C₃₀ hopane; E = 25-norhopane/C₃₀ hopane; F = C₂₇ ααα 20S/(20S + 20R); G = C₂₈ ααα 20S/(20S + 20R); H = C₂₉ ααα 20S/(20S + 20R); I = C₂₇ diasteranes/(C₂₇ diasteranes + C₂₇ steranes); J = C₂₉/(C₂₇ + C₂₈ + C₂₉) αββ steranes; L = 9-methylphenanthrene/3-methylphenanthrene; M = 9-methylphenanthrene/2-methylphenanthrene; N = TA/(MA + TA). **n.d. = not detected.

According to many works, such as McCaffrey *et al.* (1996), hopanes may be demethylated by microbial activity in the reservoir, producing the 25-norhopanes. For instance, Cortes *et al.* (2013) identified the presence of the complete 25-norhopanes series in biodegraded oils from the Apiay field, Llanos Basin, Colombia. Accordingly, the 25-norhopanes detected in most samples of this work indicate heavy biodegradation up to level 6 (according to the scale proposed by Peters &

Moldovan, 1993). However, these compounds have lower relative abundances in samples from the dry interdune facies, and could not be detected in the samples S2, S4 and S5. Probably, the biodegradation was so severe in this facies, that the 25-norhopanes were degraded by microorganisms, which can be occur in oxic conditions as was observed in biodegradation experiments performed in the laboratory by Bost *et al.* (2001).

3.4. Steranes

Pregnane, homopregnane, C₂₇ diasteranes, C₂₇, C₂₈ and C₂₉ steranes are detected in most samples collected from Pirambóia Formation. Samples from the dry interdune facies show much lower relative abundances of C₂₇, C₂₈ and C₂₉ steranes than the other facies, with almost no detection of all

isomers of these compounds. Furthermore, it is not detected pregnane and homopregnane in the samples S1, S2, S3 and S4, which belong to this facies (Fig. 7). Among the samples from other facies, the C₂₈ steranes presents lower relative abundances.

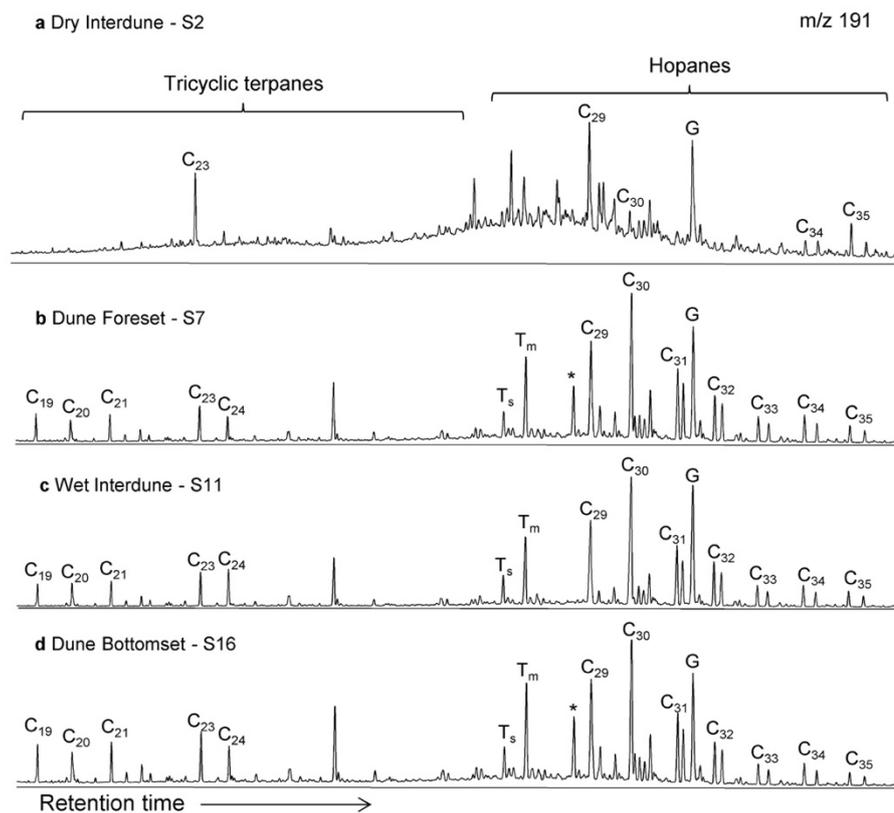


Figure 4 - Mass chromatograms (m/z 191) of saturated hydrocarbons of the tar sands from the eolian sedimentary facies of the Pirambóia Formation, Paraná Basin: (a) dry interdune facies; (b) dune foreset facies; (c) wet Interdune facies; and (d) dune bottomset facies. *Presence of 25-norhopane.

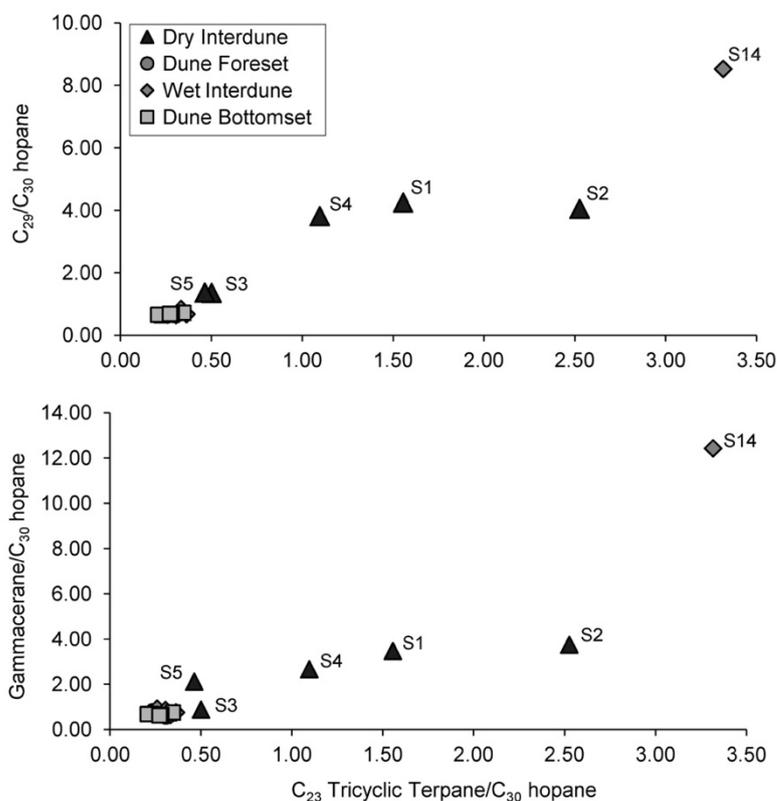


Fig. 5. Biomarker ratios in saturated hydrocarbons of tar sands samples from the eolian sedimentary facies of the Pirambóia Formation, showing the effects of biodegradation.

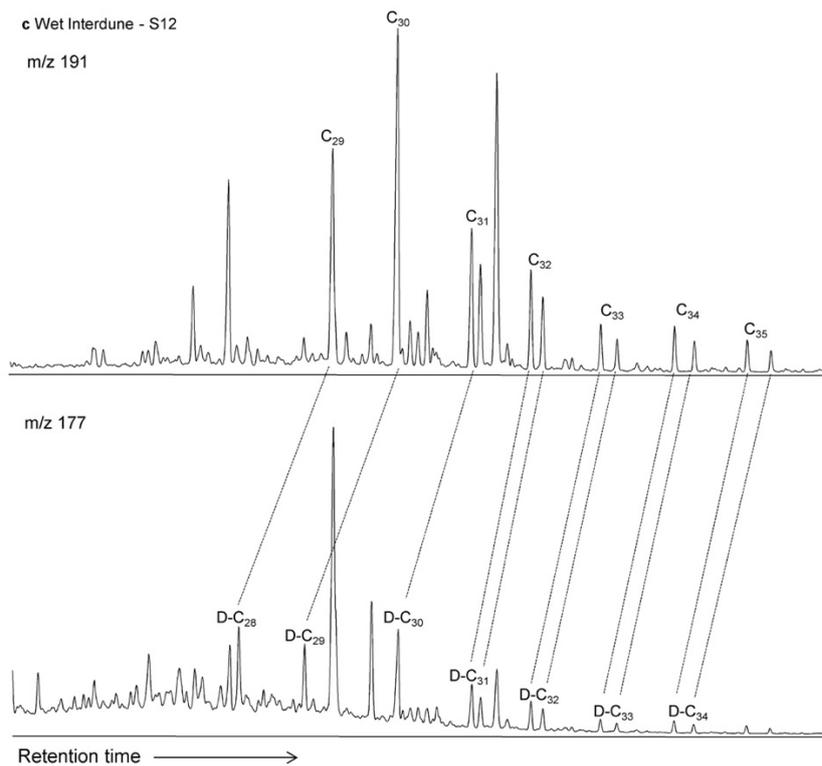


Fig. 6. Mass chromatograms (m/z 191 and 177) of saturated hydrocarbons in tar sand sample S12 from the Pirambóia Formation, Paraná Basin. (C_{28} - C_{34} demethylated hopanes (D) occur in the m/z ion chromatogram of most severely biodegraded oil samples from Pirambóia Formation that still contain a full suite of hopanes, as seen in the m/z 191 ion chromatogram).

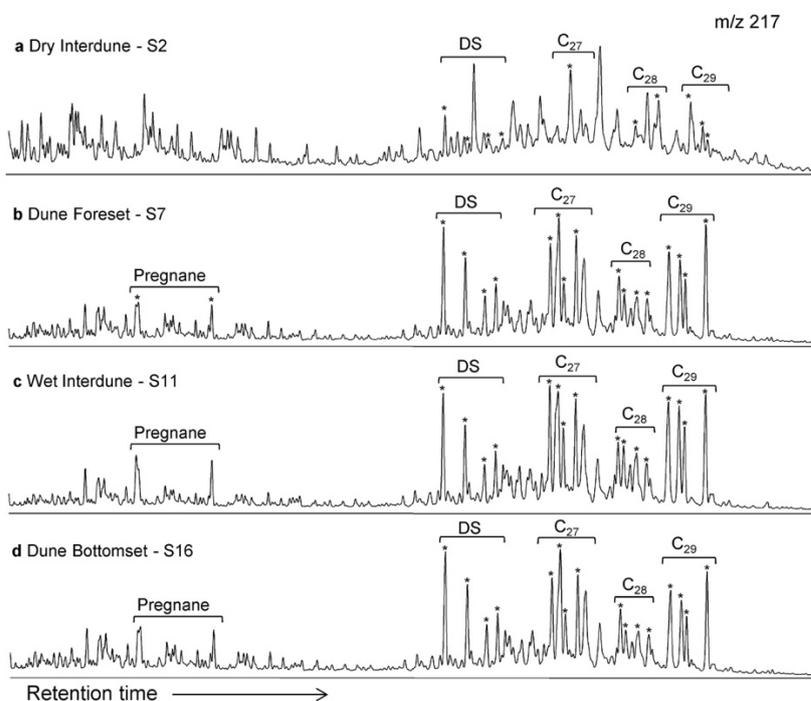


Fig. 7. Mass chromatograms (m/z 217) of saturated hydrocarbons in tar sands from the eolian sedimentary facies in the Pirambóia Formation, Paraná Basin. DS = C_{27} diasteranes (isomers $\beta\alpha$ 20S, $\beta\alpha$ 20R, $\alpha\beta$ 20S, $\alpha\beta$ 20R, respectively); C_{27} = C_{27} Steranes, C_{28} = C_{28} Steranes, C_{29} = C_{29} Steranes (isomers $\alpha\alpha\alpha$ 20S, $\alpha\beta\beta$ 20R, $\alpha\beta\beta$ 20S, $\alpha\alpha\alpha$ 20S, respectively).

In general, steranes isomers susceptibility to microbial degradation is as follows: $\alpha\alpha\alpha$ 20R \gg $\alpha\beta\beta$ 20R \gg $\alpha\beta\beta$ 20S \gg $\alpha\alpha\alpha$ 20S (Peters *et al.* 2005). Therefore, lowest values of ratios C_{27} $\alpha\alpha\alpha$ 20S/(20S + 20R), C_{28} $\alpha\alpha\alpha$ 20S/(20S + 20R) and C_{29} $\alpha\alpha\alpha$ 20S/(20S + 20R) (Table 2; ratios F, G and H, respectively) for the samples from the dune bottomset facies

indicates lower biodegradation levels for the samples from this facies. On the other hand, the $\alpha\alpha\alpha$ 20R steranes isomers were not detected in the samples S1, S2, S3 and S4, which are from the dry interdune facies, accordingly to the higher biodegradation level of samples from this facies. These results can be best observed in the Fig. 8.

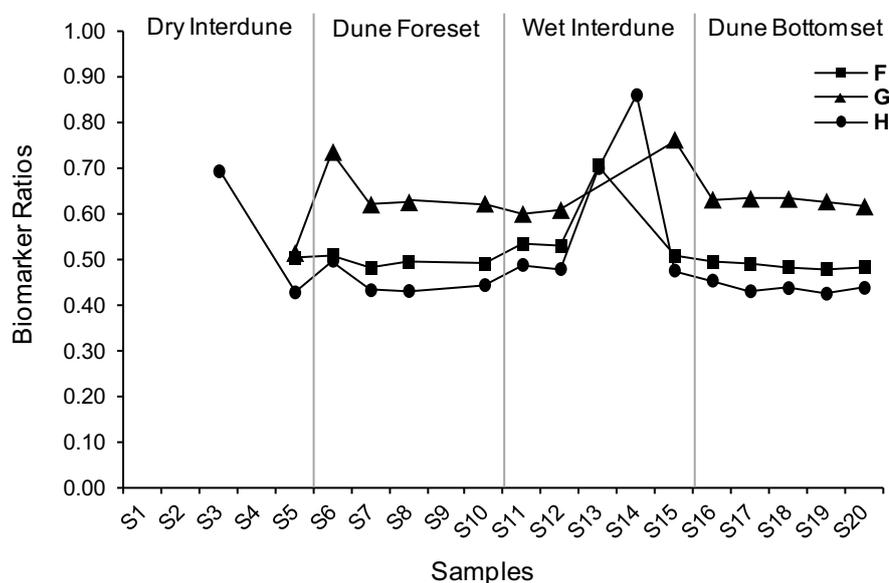


Fig. 8. Comparison of biomarker steranes ratios in tar sands among the eolian sedimentary facies of the Pirambóia Formation. The ratios were calculated as following: F = C_{27} $\alpha\alpha\alpha$ 20S/(20S + 20R); G = C_{28} $\alpha\alpha\alpha$ 20S/(20S + 20R); H = C_{29} $\alpha\alpha\alpha$ 20S/(20S + 20R).

C_{27} diasteranes were detected in all samples collected in the Pirambóia Formation. According to Peters *et al.* (2005), diasteranes are particularly resistant to biodegradation, with evidence that C_{27} - C_{29} steranes are destroyed completely before diasteranes alteration (Seifert & Moldowan 1979, Connan 1984). The C_{27} diasteranes/(C_{27} diasteranes + C_{27} steranes) ratio (I ratio, Table 2) show the highest values for samples from the dry interdune facies, since the oil in this facies is the most biodegraded.

However, the ratio of C_{27} diasteranes $\beta\alpha$ 20R/(C_{27} diasteranes $\beta\alpha$ 20R + C_{27} diasteranes $\beta\alpha$ 20S) of samples from the dry interdune facies show the lowest values. This is not in agreement with previous studies, which founded that biodegradation of diasteranes results in stereoselective loss of C_{27} 13 β ,17 α (H) 20S over C_{27} 13 β ,17 α (H) 20R epimers (Seifert & Moldowan 1979, Peters *et al.* 2005). These results are presented in Fig. 9.

3.5. Aromatics

Aromatic hydrocarbons with a lower number of rings, such as benzene, methylbenzenes, naphthalenes, and methylnaphthalenes are not detected in the tar sands samples from Pirambóia Formation. Conversely, in most samples are detected polycyclic aromatic compounds with a higher number of rings, such as methylphenanthrenes (Fig. 10), monoaromatic and triaromatic steroid hydrocarbons (Fig. 11).

Methylphenanthrenes isomers with methyl substituents at the 9 position show higher relative abundance than the other isomers, except in the samples S11 and S12 (wet interdune). Furthermore, this compounds isomers with methyl substituents at 1 and 2 positions showing lower relative abundance are not detected in samples S8, S9, and S10 samples from foreset, and S13, S14 from wet interdune facies (Fig. 10).

Methylphenanthrenes isomers show very low relative abundance in samples S3 and S4

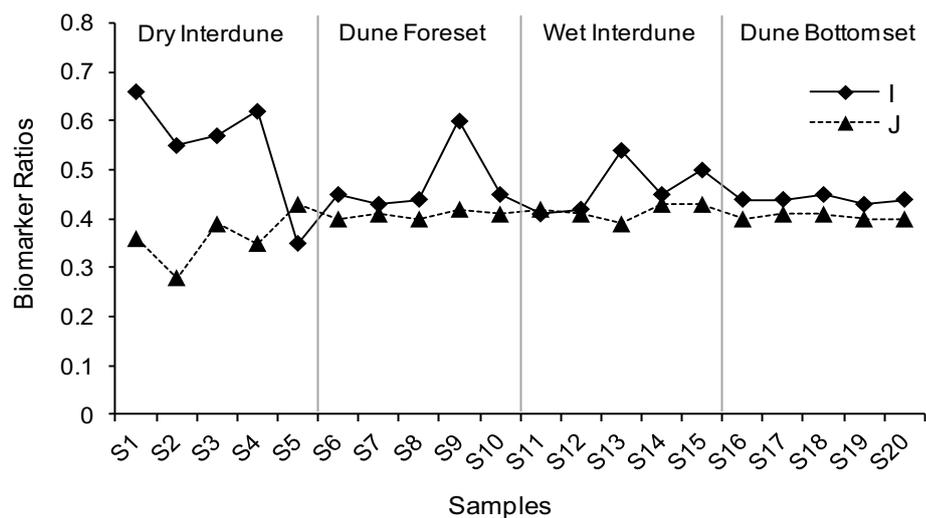


Fig. 9. Comparison of biomarker steranes and diasteranes ratios in tar sands among the eolian sedimentary facies of the Pirambóia Formation. The ratios were calculated as follows: I = C_{27} diasteranes / (C_{27} diasteranes + C_{27} steranes); J = $C_{29} / (C_{27} + C_{28} + C_{29})$ $\alpha\beta$ steranes.

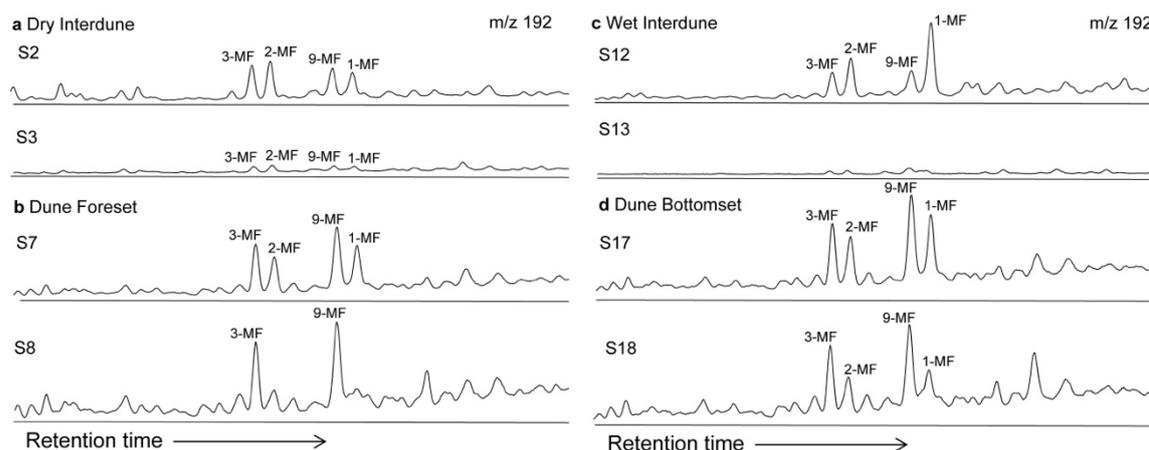


Fig. 10. Mass chromatograms (m/z 192) of aromatic hydrocarbons in tar sands from eolian sedimentary facies in the Pirambóia Formation, Paraná Basin. 3-MF = 3-methylphenantrene; 2-MF = 2-methylphenantrene; 9-MF = 9-methylphenantrene; 1-MF = 1-methylphenantrene.

Monoaromatic steroid hydrocarbons are not detected in samples S1, S2, S3, S4, and S5 from the dry interdune facies, and in S13 sample from the wet interdune facies. Triaromatic steroid hydrocarbons were not detected in samples S4 and S13.

Methylphenanthrene ratios (Table 2), and the mass chromatograms (m/z 192) (Fig. 10) may indicate that 9-methylphenanthrene is more resistant to biodegradation than the other isomers, except in the S11 and S12 samples. Therefore, the 9-methylphenanthrene/3-methylphenanthrene and 9-methylphenanthrene/2-methylphenanthrene ratios would increase when biodegradation increases. However, this was not observed. The

samples from the dry interdune facies, which has the least biodegraded bitumen, shows in general the lowest values of these ratios (Fig. 12). This differential susceptibility to degradation showed by methyl phenanthrenes was also observed in the Athabasca oil sands by Bennett & Larter (2008), in which 9-methylphenanthrene may show selective resistance to degradation than 1-methylphenanthrene, whilst they also seen that 9-methylphenanthrene may be removed prior to 1-methylphenanthrene. These inconsistent patterns of biodegradation are typically observed in petroleum analysis around the world, and difficult the consolidation of biodegradation scales.

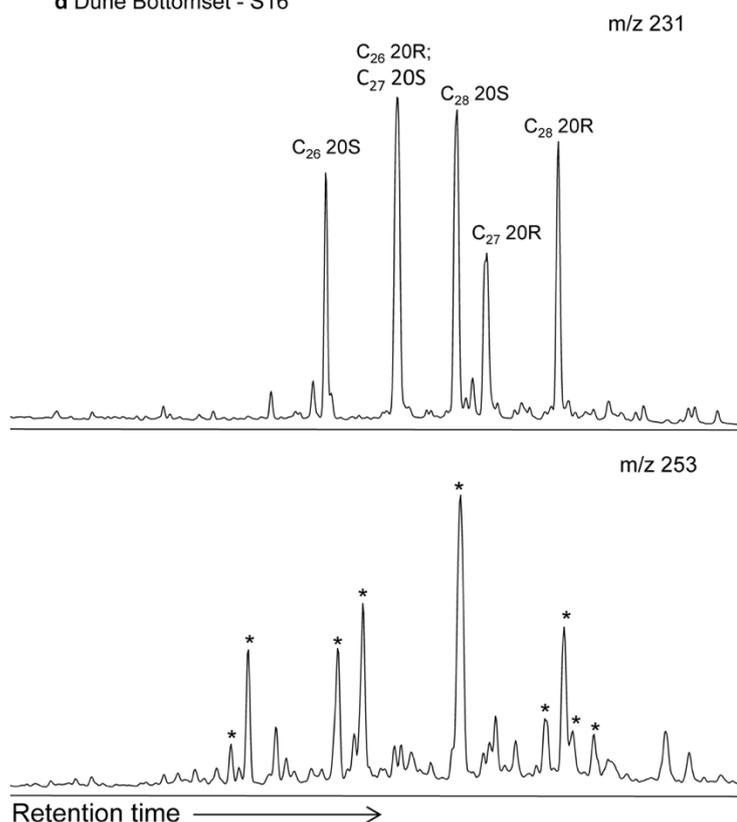


Fig. 11. Mass chromatograms (m/z 231 and 253) of aromatic hydrocarbons in tar sands from eolian sedimentary facies in the Pirambóia Formation.

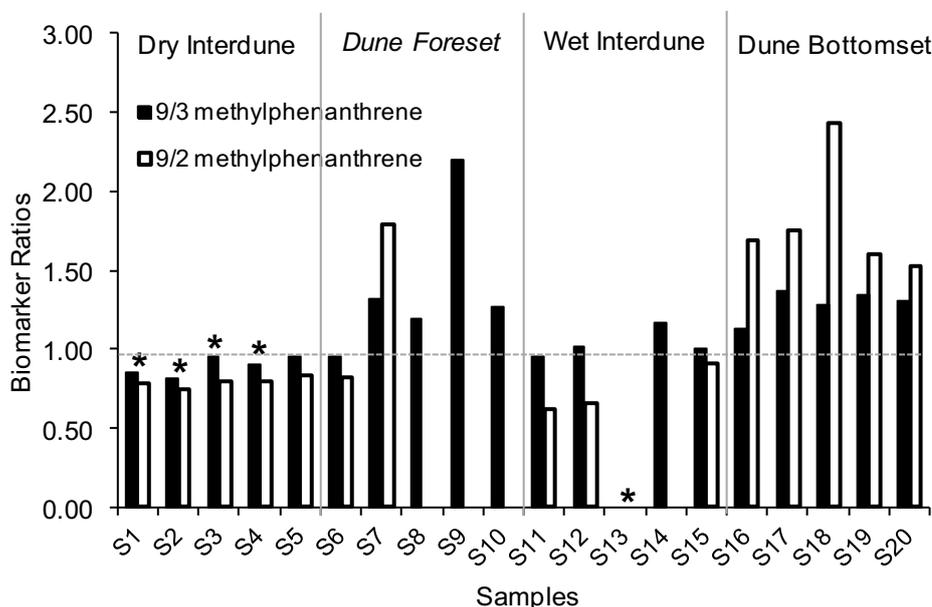


Fig. 12. Biomarker ratios in aromatic hydrocarbons of tar sand samples from the eolian sedimentary facies of the Pirambóia Formation, showing the effects of water washing. *Samples from Mina Farm locality.

Another observation from Fig. 12 is that the tar sand samples collected in the Mina Farm locality (S1, S2, S3, S4 and S13) show ratios values of about one, which indicated that methylphenanthrene isomers were equally depleted. This is an indication that other secondary alteration processes, as water washing, may have occurred. Biodegradation is

usually selective to some isomers (Lin *et al.* 1989), but water washing is not isomer selective (Huang *et al.* 2004). In addition, aromatic compounds are more susceptible to being carried away by meteoric water than saturated hydrocarbons.

Monoaromatic steroid hydrocarbons are not detected in S1, S2, S3, S4, and S5 samples

from the dry interdune facies, and S13 from the wet interdune facies. Triaromatic steroid hydrocarbons were not detected in samples S4 and S13, and corroborate with the suggestion that water washing process occurred on these samples, since these compounds are very

4. CONCLUSIONS

In general, all analysis performed in this work indicate heavy degradation degree to the bitumens in tar sands from Pirambóia Formation, first evidenced by SARA analysis, that showed higher amount of polar compounds to all samples. The GC-FID analysis of saturated fraction also indicates this, showing a complete loss of n-alkanes and isoprenoids, and occurrence of huge UCM in the GC traces. In addition, these analyses indicate that the tar sands in dry interdune facies may have the most biodegraded bitumen among the eolian facies of Pirambóia Formation, Paraná Basin.

Saturated biomarkers ratio analyses confirm that the dry interdune facies contains the most biodegraded bitumen and suggest that the bottomset facies contains the least biodegraded. Furthermore, even with high

resistant to biodegradation. Therefore, it is clear, that aromatic biomarkers are not appropriate to analyze the degree of biodegradation of tar sands and outcrops samples.

biodegradation levels, some compounds like C₂₃ tricyclic terpane, C₂₉ hopane, gammacerane and C₂₇ diasteranes show high resistance, proving that these biomarkers are useful in biomarker analysis of very biodegraded bitumens.

Aromatic biomarker analysis, however, shows that this class of compounds is not useful in biodegradation analysis for rock outcrops and tar sands, since it is more susceptible to other secondary alteration processes, such as water washing.

This work increases our knowledge of the chemical composition of mineable oil in tar sands from different sedimentary facies, potentially improving predictions of bitumen quality in different regions of the reservoir and assisting production decisions.

5. ACKNOWLEDGMENTS

We are grateful to PRH20-ANP, Capes and FAPERJ for financial support, Petrobras/Rede de Geoquímica for providing

the necessary infrastructure to conduct research, Margaret R. Hennon and Prof. Allen Howard (Utah State University) for revising the text.

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