



# The characteristics of the biomarkers and $\delta^{13}\text{C}$ of *n*-alkanes released from thermally altered solid bitumens at various maturities by catalytic hydrolysis

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## ABSTRACT

Solid bitumen occurs extensively in the paleo-reservoirs of marine sequences in southern China. The fluids in these paleo-reservoirs have usually experienced severe secondary alteration such as biodegradation and/or thermal maturation. The concentrations of extractable organic matter (EOM) in the resulting solid bitumens are too low to satisfy the amount required for instrumental analysis such as GC–MS and GC–IRMS. It is also difficult to get enough biomarkers and *n*-alkanes by dry pyrolysis or hydrous pyrolysis directly because such solid bitumens are hydrogen poor due to high maturities. Catalytic hydrolysis (HyPy) can release much more EOM from solid bitumen at mature to highly over-mature stages than Soxhlet extraction, dry pyrolysis and hydrous pyrolysis. However, whether the biomarkers in hydrolysis products can be used for bitumen-source or bitumen–bitumen correlations has been questionable. In this study, a soft biodegraded solid bitumen sample of low maturity was thermally altered to various maturities in a closed system. HyPy was then employed to release bound biomarkers and *n*-alkanes. Our results show that the geochemical parameters for source and maturity based on biomarkers released from these thermally altered bitumen residues by HyPy are insensitive to the degree of thermal alteration. Furthermore, the maturity parameters are indicative of lower maturity than bitumen maturation products at a corresponding temperature. This suggests that biomarker source and maturity parameters, based on the products of HyPy, remain valid for bitumens which have suffered both biodegradation and severe thermal maturation. The distributions of  $\delta^{13}\text{C}$  of *n*-alkanes in hydrolysis products are also insensitive to the temperature used for bitumen artificial maturation. Hence, the  $\delta^{13}\text{C}$  values of *n*-alkanes in hydrolysis products may also provide useful information in bitumen–bitumen correlation for paleo-reservoir solid bitumens.

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## 1. Introduction

In the marine sequences of southern China, solid bitumens occur extensively in paleo-reservoirs. They can be found outcropped from Permian to Precambrian strata in this area. For example, paleo-reservoir solid bitumens occur extensively in coral reefs of the Silurian strata of Guangxi Province, such as the Dachang paleo-reservoir of Nandan County and the Anran paleo-reservoir of Longlin County (Zhao et al., 2006). Solid bitumens can also be found extensively in Permian coral reefs of Guizhou Province, such as the Bialai paleo-reservoir in Wangmo County, the Shitouzhai paleo-reservoir in Ziyun County and the Ceyang paleo-reservoir in Ceheng County. They also occur in the Cambrian strata in Weng'an County and Tongren County of Guizhou Province. The oldest paleo-reservoirs have been found in the Dengying Formation of the Sinian System, such as the Nanshanping

paleo-reservoir in Cili County of Hunan Province. These paleo-reservoirs have usually experienced multiple secondary alteration events during continuous tectonic orogenies. The biggest and best known paleo-reservoir in southern China is the Majiang paleo-reservoir of Guizhou Province. This reservoir used to cover 2450 km<sup>2</sup> in the Caledonian period (Early Paleozoic), but now covers an area of 800 km<sup>2</sup> (Xue et al., 2007; Liu et al., 2008), mainly in Majiang, Duyun and Danzhai of the South Guizhou Depression. The extent of the original oil reservoir was controlled by both lithological and structural factors and it contained 16 billion tonnes of crude oil before it was destroyed (Wu, 1989). The Majiang paleo-reservoir came into being in the Late Caledonian Period. This area was uplifted after the end of Caledonian Period and the caprock of the paleo-reservoir was eroded, resulting in the loss of light hydrocarbons by evaporation and severe biodegradation (Xue et al., 2007). In the Indosinian Period (Early Mesozoic), the reservoir was rapidly buried and the crude oil began to crack due to high maturity resulting from the deep burial (4–5 km). From the Yanshanian Period (Late Mesozoic) on the reservoir was uplifted,

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outcropped and destroyed in continuous tectonic orogenies. At present, the Majiang paleo-reservoir typically contains only solid pyro-bitumens (most with bitumen reflectance ( $R_b$ )  $\geq 2.0\%$ ) that have been altered by severe thermal maturation (Xue et al., 2007; Fang et al., 2011). The solid bitumens of the Majiang paleo-reservoir are stored mainly in the pores and structural fissures of the carbonates of the Lower-Ordovician Honghuayuan Formation (O<sub>1h</sub>), the intergranular pores of the sandstones of the Lower-Middle Silurian Wengxiang Group (S<sub>1-2wx</sub>) and the pores of the carbonates of the Lower-Permian Maokou Formation (P<sub>1m</sub>). The Cambrian marine shales are thought to be the main source rock of the hydrocarbons in the Majiang paleo-reservoir (Wu, 1989; Xue et al., 2007; Tenger et al., 2008).

Routine molecular geochemical methods are of limited use for studying high maturity paleo-reservoir solid bitumens because of the difficulties of getting enough biomarkers and *n*-alkanes by Soxhlet extraction (Zhao et al., 2006, 2007). Furthermore, the free biomarkers obtained from these solid bitumens by Soxhlet extraction have often been severely altered during thermal maturation. However, many previous studies have indicated that secondary alteration processes such as biodegradation have much less impact on asphaltenes than on hydrocarbons (Rubinstein and Strausz, 1977; Rubinstein et al., 1979; Cassani and Eglinton, 1986). The aliphatic sub-structures attached to polyaromatic nuclei are not altered by biodegradation (Bonnamy et al., 1987). Asphaltenes appear to possess alkyl distributions and biomarker fingerprints inherent to their source (Behar et al., 1984; Jones et al., 1988). Hence, asphaltene pyrolysates have been used successfully as an oil-source correlation tool for severely biodegraded oils (Rubinstein et al., 1979; Behar et al., 1984; Rullkötter and Michaelis, 1990; Wilhelms et al., 1994; Xiong and Geng, 2000). However, although biodegraded bitumens usually contain abundant asphaltenes, it is difficult for solid bitumen of high maturity to yield sufficient amounts of biomarkers by chemical degradation, dry pyrolysis and hydrous pyrolysis methods due to their being hydrogen poor (Sun et al., 2008). Therefore, most studies on the solid bitumens in southern China are limited to obtaining bulk isotopic values, FT-IR spectra and bitumen reflectance (Zhao et al., 2006, 2007). It is challenging to correlate the high maturity solid bitumens to their potential source rocks by these routine methods.

Fixed-bed catalytic hydrolysis (HyPy) developed by Nottingham University is an open system pyrolysis method assisted by a dispersed sulfided molybdenum catalyst. Hydrogen at high pressures (>10 MPa) is used as a carrier gas and this can cleave the biomarkers covalently bound to geo-macromolecules at relatively low temperature (<450 °C; Love et al., 1995). Previous studies (Love et al., 1995, 1998; Rocha et al., 1997; Bishop et al., 1998; Bennett et al., 2004) suggested that HyPy provided a way to release soluble organic matter from geo-macromolecules in amounts suitable for gas chromatographic (GC) and combined gas chromatography-mass spectrometry (GC-MS) analysis (Hsu and Drinkwater, 2001; Hsu, 2003). Many researchers thereafter tried to release biomarkers from kerogens of various maturities (Murray et al., 1998; Brocks et al., 2003; Zhou et al., 2006, 2007; Sun et al., 2008; Lockhart et al., 2008). HyPy is a technique for releasing biomarkers from high maturity geo-macromolecules, especially when they are not only very low in EOM but also very poor in hydrogen. Numerous studies (Murray et al., 1998; Bishop et al., 1998; Bowden et al., 2006; Lockhart et al., 2008) have suggested that the biomarkers released by HyPy from kerogen usually showed lower maturities than those in the free bitumen.

In this research, a biodegraded soft solid bitumen derived from a Cambrian source rock was artificially thermally altered to highly overmature stages to simulate the natural thermal maturation process that the biodegraded bitumens in the Majiang paleo-reservoir have experienced. The covalently bound biomarkers and *n*-alkanes

were released from the thermally altered solid bitumen residues of various maturities by HyPy. The saturated hydrocarbon fractions of both closed system bitumen thermal maturation products and hydrolysis products released from thermally altered bitumens at various maturities were analyzed by GC-MS for the commonly used biomarker parameters. In addition, stable carbon isotopes of *n*-alkanes were measured by combined gas chromatography-isotope ratio mass spectrometry (GC-IRMS). Whether source characterization and bitumen-bitumen or bitumen-source correlations can be made on the basis of the materials released from thermally altered bitumens of various maturities by HyPy is discussed.

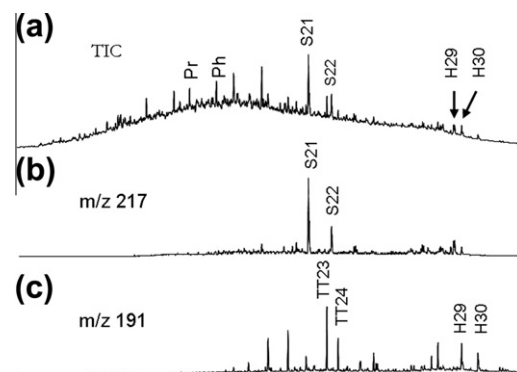
## 2. Experimental

### 2.1. Samples

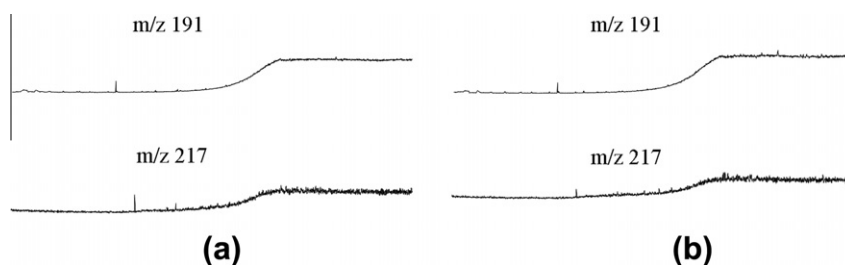
Due to severe thermal maturation, crude oils sourced from the Cambrian source rocks are difficult to find in the Southern Guizhou Depression where the Majiang paleo-reservoir is located. However, biodegraded bitumen and oil seepage which are likely sourced from the Cambrian marine shales can still be found in some outcrops of the Longmen Mountain structure, Sichuan Province due to continuous tectonic uplift since the Indosinian orogeny (Huang et al., 2008; Rao et al., 2008). Thus a soft solid bitumen sourced from the Cambrian marine shales (Huang and Wang, 2008) was collected from outcrop of the Longmen Mountain structure. This soft solid bitumen is believed to resemble the bitumens in the Majiang paleo-reservoir at the end of Late Silurian after the caprock of the reservoir was eroded and the oil altered by both evaporation and biodegradation. The bitumen is depleted of low molecular weight hydrocarbons (Fig. 1), especially *n*-alkanes due to evaporation and biodegradation during exposure to the atmosphere (Huang et al., 2008). The abundances of saturates, aromatics, resins and asphaltenes are 1%, 7%, 32% and 61%, respectively. The biomarkers in the extractable organic matter (EOM) were identified by GC-MS (Fig. 2). The  $\alpha\alpha\alpha 20R/\alpha\alpha\alpha 20S$  C<sub>29</sub> sterane ratio from GC-MS result suggests the bitumen was mildly biodegraded and it is ranked 5 on the Peters and Moldowan (PM) biodegradation scale (Peters and Moldowan, 1993; Hsu, 2003). The bitumen reflectance is 0.58%. Rock-Eval/TOC analysis showed that the TOC is 71.7%,  $T_{max}$  is 436 °C and the hydrogen index is 470 mg/g TOC.

### 2.2. Experimental methods

The solid bitumen was crushed into powder and then sealed in a glass tube under nitrogen gas. The tubes were then heated for 72 h at 350 °C, 380 °C, 400 °C, 430 °C or 460 °C. The equivalent vitrinite



**Fig. 1.** (a) GC-MS total ion chromatograms (TIC) of the saturated fraction in EOM of soft solid bitumen; (b) *m/z* 217 mass chromatograms, displaying sterane distributions; (c) *m/z* 191 mass chromatograms, displaying terpane distributions. Pr = Pristane; Ph = Phytane; S21 = pregnane; S22 = homopregnane; TT23–24 = C<sub>23–24</sub> tricyclic terpanes; H29–30 = C<sub>29–30</sub> 17 $\alpha$ (H) 21 $\beta$ (H) hopanes.



**Fig. 2.** The  $m/z$  191 and 217 mass chromatograms of solvent blanks from HyPy blank experiments and subsequent column chromatography separation: (a) solvent blank ahead of B-350hs; (b) solvent blank ahead of B-460hs.

**Table 1**

Basic information of artificial thermal maturation on bitumen and catalytic hydropyrolysis on altered bitumen residues.

Simulation temp. (°C)	Equivalent Ro (%) of bitumen	ID of bitumen maturation products	ID of hydropyrolysates	Yield of extractable bitumen maturation products ( $C_{15+}$ ) (mg/g TOC)	Yield of hydropyrolysates ( $C_{15+}$ ) (mg/g TOC)
350	1.08	B-350	B-350hs	223.9	58
380	1.49	B-380	B-380hs	140.8	57
400	1.74	B-400	B-400hs	101.3	82
430	2.27	B-430	B-430hs	63.0	71
460	2.86	B-460	B-460hs	45.0	58

reflectance (Ro) values of the heated bitumens can be calculated by Easy %Ro (Sweeney and Burnham, 1990). The calculated equivalent %Ro values are in Table 1. After each thermal simulation, the heated bitumen powder was first recovered by repeated dichloromethane (DCM) washing and sonication at room temperature. The washed bitumen residue was then Soxhlet extracted by a ternary solvent (benzene:acetone:methanol, 5:5:2) for 2 weeks to extract residual EOM. The washed solution and the Soxhlet extract were combined and left to dry at room temperature to calculate the total yield of the extractable maturation products ( $C_{15+}$ ). Then the extractable maturation products ( $C_{15+}$ ) were fractionated into saturate, aromatic, resin and asphaltene (SARA) fractions by column chromatography. For convenience, the original bitumen is termed B-original, and the bitumen thermal maturation products at 350 °C, 380 °C, 400 °C, 430 °C and 460 °C are termed B-350, B-380, B-400, B-430 and B-460, respectively (Table 1).

After removing the extracts, the solid bitumen powders were impregnated with an aqueous solution of ammonium dioxymolybdate  $[(NH_4)_2MoO_4 \cdot 2H_2O]$  to give a molybdenum loading of 5 wt%. They were then vacuum dried for subsequent HyPy experiments involving two steps. In the first step, the vacuum dried solid bitumen residue (500 mg) was placed in a fixed bed and heated resistively from ambient temperature to 300 °C and kept isothermal for 2 min to cleave weaker covalent bonds, leaving stronger bonds for HyPy (Love et al., 1995). The thermally desorbed volatiles at this stage were collected on silica gel in a liquid nitrogen cold trap without further analysis. The whole system was then cooled down to room temperature for replacing the cold trap collecting tube and cleaning the pipes. The second step was for releasing stronger covalently bound compounds. The fixed bed temperature was increased from ambient to 250 °C at 300 °C/min, then increased to 520 °C at 5 °C/min and kept at 520 °C for 5 min. In the whole process, the hydrogen pressure was kept at 15.0 MPa and the flow was kept at 4 l/min to blow the newly formed volatile compounds away quickly (Love et al., 1995). The hydropyrolysates were collected on clean silica gel held in a liquid nitrogen cold trap. After the experiment, the silica gel was extracted with DCM:methanol (93:7). The solvent was left to dry at room temperature and the yield of hydropyrolysates ( $C_{15+}$ ) was calculated. After the solvent was removed under nitrogen flow, about 1 ml DCM was added to dissolve the hydropyrolysates. Then asphaltenes were precipitated by adding excess

*n*-hexane. The maltene fraction was separated into saturate, aromatic and polar fractions by silica–alumina column chromatography eluting with *n*-hexane, toluene and methanol, respectively. For convenience, the hydropyrolysates are termed B-350hs, B-380hs, B-400hs, B-430hs and B-460hs, according to the temperatures at which the soft solid bitumen was thermally altered.

Before each HyPy experiment, the cold trap collecting tube of HyPy system was washed with DCM and then heated at 600 °C for 4 h. A HyPy blank experiment was performed before running real samples. Silica gel in collecting tube of the HyPy blank run (containing no sample) was Soxhlet extracted with DCM:methanol (93:7) and the extract (solvent blank) was concentrated for subsequent column chromatography separations. Then GC–MS analysis was performed on the concentrated solvent blank to make sure that there is no contamination coming from the solvent, HyPy experiment or column chromatography separations, which may influence the biomarker distribution in hydropyrolysates. Fig. 2 displays the  $m/z$  191 and 217 mass chromatograms of saturated fractions of two solvent blanks from HyPy blank experiments ahead of B-350hs (Fig. 2a) and B-460hs (Fig. 2b). No steranes or hopanes are detected in the two solvent blanks.

### 2.3. Instrumental analysis

#### 2.3.1. Gas chromatography–mass spectrometry (GC–MS)

Analysis of the saturate biomarkers was conducted using a Thermo Scientific Trace ULTRA GC coupled with a Thermal Scientific Trace DSQ II mass spectrometer. A DB-1 fused silica capillary column (60 m  $\times$  0.25 mm i.d.  $\times$  0.25  $\mu$ m film thickness) was held at 70 °C for 2 min, ramped to 290 °C at 4 °C/min, and then held at 290 °C for 30 min. Helium was used as carrier gas with a constant flow rate of 1.5 ml/min. The temperature of the injector was 290 °C. The ion source was operated in the electron impact (EI) mode with electron energy of 70 eV. Biomarker parameters were calculated based on the peak area of each compound.

#### 2.3.2. Compound-specific isotope analysis

*n*-Alkanes were isolated from the saturated hydrocarbon fractions by urea adduction similar to that described by Liao et al. (2004, 2009). Stable carbon isotope analysis of individual *n*-alkanes was performed on a GV Isochrom II isotope-ratio mass spectrometry

(IRMS) system interfaced with a Hewlett–Packard 5890 GC. The GC was fitted with a fused silica column (DB-1, 30 m × 0.25 mm i.d.) leading directly into the combustion interface. CO<sub>2</sub> was used as reference gas and was automatically introduced into the IRMS before and after each analysis. The isotope values were calibrated against the reference gas and are reported in per mil (‰) relative to Pee Dee Belemnite (PDB). In order to maintain the reproducibility and accuracy of isotope data, the standard was run between samples and each sample was analyzed at least twice. The reproducibility was typically within a range of ±0.2‰.

### 3. Results and discussion

The amount of extractable bitumen maturation products (C<sub>15</sub>+) decreases from 223.9 mg/g TOC to 45 mg/g TOC with increasing temperature (Table 1) due to more and more severe thermal decomposition. However, the yield of hydropyrolysates released from altered bitumen initially increased with simulation temperature and reached its highest value of 82 mg/g TOC at 400 °C. It then decreased to 58 mg/g TOC at 460 °C (Table 1). Bishop et al. (1998) suggested that the total yield by HyPy is ultimately controlled by thermal maturity and source type. Fig. 3 displays the GC–MS TICs of saturated fractions of bitumen maturation products at various temperatures and those of the corresponding hydropyrolysates.

Fig. 4 shows the m/z 191 and 217 mass chromatograms from GC–MS analysis of the saturate fractions of both the original (unheated) bitumen and bitumen maturation products at different simulation temperatures. The distribution of both terpanes and steranes in the bitumen maturation products changes considerably with simulation temperature. Jones et al. (1988) suggested that pyrolysis of whole bitumen (asphalts) is of limited use because the hydrocarbon distribution in the biodegraded bitumen may obscure the distribution of newly generated hydrocarbons. In addition, the bitumen may contain some heteroatomic species, particularly those with oxygen functional groups, such as steroid acids (Seifert et al., 1972; Rodrigues et al., 2000; Peters et al., 2005) and hopanoid acids (Jaffé and Gallardo, 1993; Watson et al., 2002) in biodegraded bitumen. These acids may be reduced to hydrocarbons and will yield an erroneous signal because these compounds are not actually coming from the macromolecular portion of the bitumen.

Fig. 5 shows the m/z 191 and 217 mass chromatograms of the saturate fractions of the hydropyrolysates. There is virtually no difference between the patterns of triterpanes and steranes for HyPy pyrolysates of bitumens artificially matured at temperatures

between 350 °C and 460 °C. This indicates that the geo-macromolecule structure of asphaltene has had a protective effect on the covalently bound biomarkers (Russell et al., 2004) which were then released by HyPy.

#### 3.1. Source related biomarker parameters in bitumen maturation products and hydropyrolysates

A sterane ternary diagram of C<sub>27</sub>–C<sub>28</sub>–C<sub>29</sub> ααα20R is commonly used to distinguish groups of crude oils from different source rocks or different organic facies of the same source rock (Seifert et al., 1984; Seifert and Moldowan 1986; Peters et al., 2005). It is commonly accepted that C<sub>27</sub> ααα20R steranes exhibit lower biore-sistance than C<sub>28</sub> ααα20R and C<sub>29</sub> ααα20R steranes to biodegradation (Peters et al., 2005). Thus C<sub>27</sub> ααα20R steranes were likely altered more severely than C<sub>28</sub> and C<sub>29</sub> steranes in the original bitumen during biodegradation. When the severely biodegraded bitumen was thermally altered, heavy fractions (asphaltenes and resins) began to crack causing the distributions of C<sub>27</sub>–C<sub>28</sub>–C<sub>29</sub> steranes to change (Fig. 6). However, the distribution of C<sub>27</sub>–C<sub>29</sub> ααα20R regular steranes in hydropyrolysates is much more consistent (Fig. 6). Thus, the distribution of C<sub>27</sub>–C<sub>28</sub>–C<sub>29</sub> sterane in a hydropyrolysate should be a reliable index for source characterization and bitumen–bitumen or bitumen–source correlation.

The ratio of C<sub>23</sub> tricyclic terpane to C<sub>23</sub> and C<sub>24</sub> tricyclic terpanes (TT<sub>23</sub>/(TT<sub>23</sub> + TT<sub>24</sub>)) remains constant in original bitumen and bitumen maturation products at all temperatures (Fig. 7b), consistent with the tricyclic terpanes having high resistance to both biodegradation and thermal alteration (Seifert and Moldowan, 1979; Peters et al., 2005). Fig. 7b also shows that the TT<sub>23</sub>/(TT<sub>23</sub> + TT<sub>24</sub>) ratio remains very stable in hydropyrolysates, all being in the range of 0.60–0.63. Hence, the TT<sub>23</sub>/(TT<sub>23</sub> + TT<sub>24</sub>) ratio is a reliable index in bitumen–source and bitumen–bitumen correlations for solid bitumens that have suffered heavy biodegradation and subsequent severe thermal alteration.

The ratios of C<sub>23</sub> tricyclic terpane to C<sub>30</sub> hopane (TT<sub>23</sub>/H<sub>30</sub>), C<sub>29</sub> norhopane to C<sub>30</sub> hopane ratio (H<sub>29</sub>/H<sub>30</sub>) and gammacerane to C<sub>30</sub> hopane (Gam/H<sub>30</sub>) are commonly used as source related parameters (Peters et al., 2005). C<sub>23</sub> tricyclic terpane, C<sub>29</sub> norhopane and gammacerane are considered to have higher biore-sistance than C<sub>30</sub> hopane (Peters et al., 2005). At severe biodegradation stages, the TT<sub>23</sub>/H<sub>30</sub> ratio is therefore likely increased by preferential removal of hopanes. With the cracking of heavy fractions during artificial maturation, the ratio of TT<sub>23</sub>/H<sub>30</sub> decreases with increasing simulation temperatures (Fig. 7a). However, the ratios within the 5-ring terpanes, namely, H<sub>29</sub>/H<sub>30</sub> and

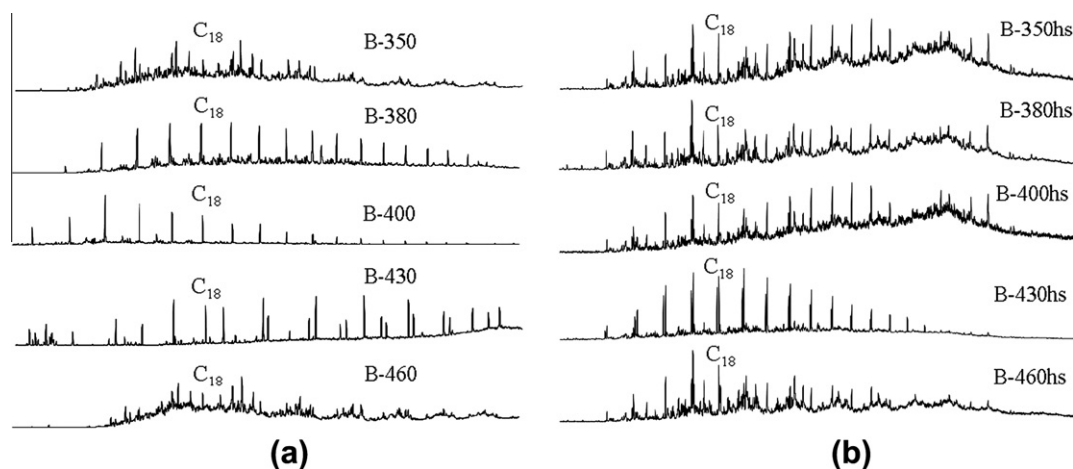
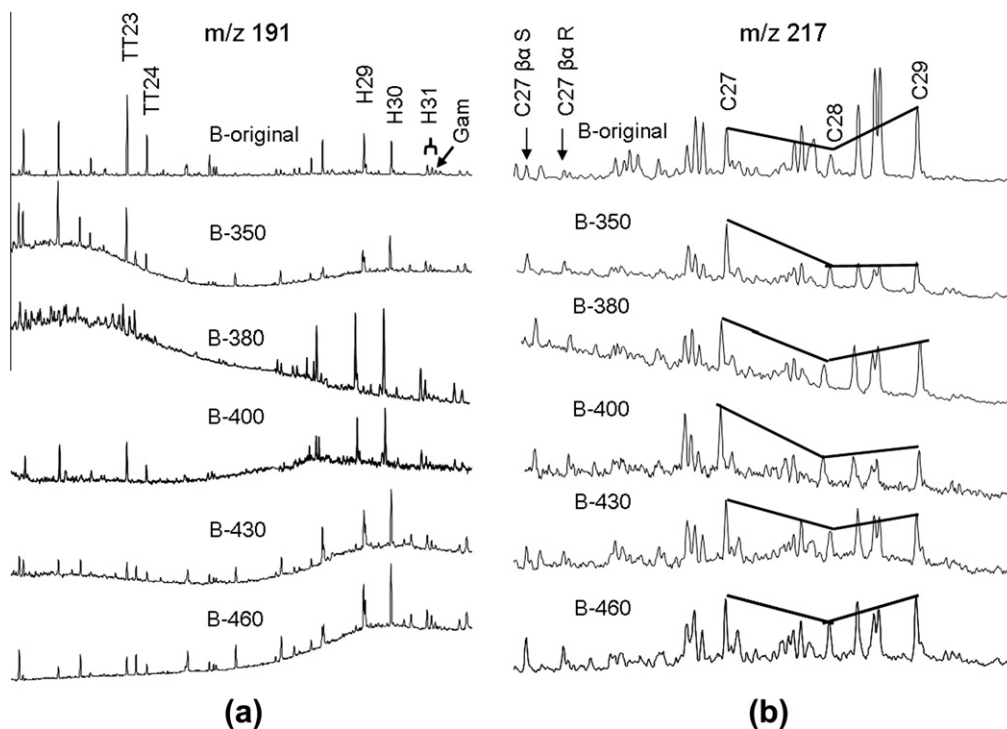


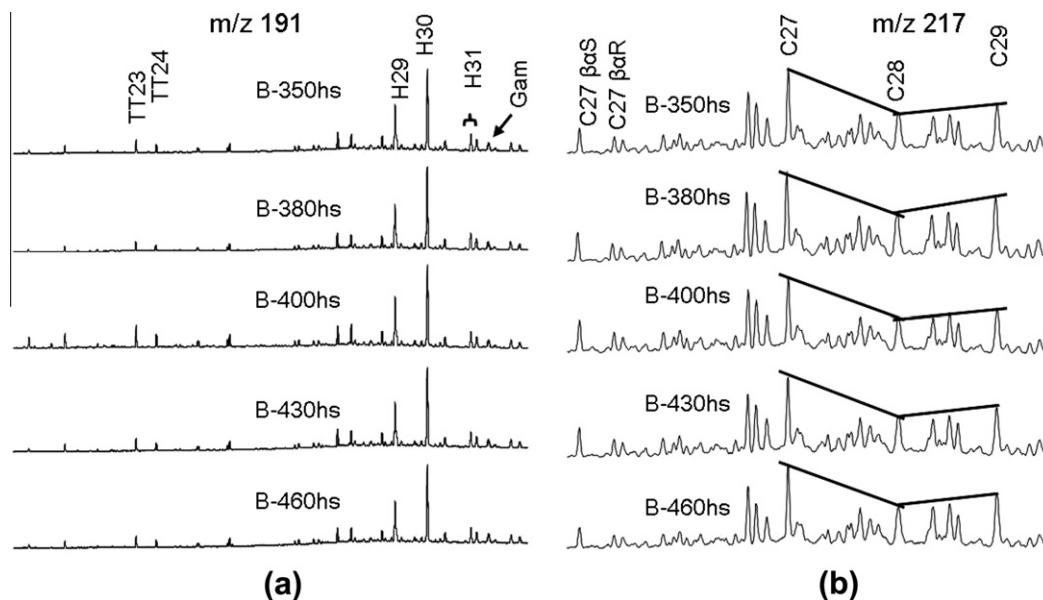
Fig. 3. The GC–MS total ion chromatograms (TIC) of saturated fractions: (a) bitumen maturation products; (b) hydropyrolysates. C<sub>18</sub> = C<sub>18</sub> n-alkane.



**Fig. 4.** GC-MS selected ion chromatograms (SIC) of the saturated fractions in bitumen maturation products: (a)  $m/z$  191 mass chromatograms, displaying terpane distributions; (b)  $m/z$  217 mass chromatograms, displaying sterane distributions. TT23–24 =  $C_{23-24}$  tricyclic terpanes; H29–31 =  $C_{29-31}$  17 $\alpha$ (H) 21 $\beta$ (H) hopanes; Gam = gammacerane; C27  $\beta\alpha S$  =  $C_{27}$   $\beta\alpha 20S$  diacholestane; C27  $\beta\alpha R$  =  $C_{27}$   $\beta\alpha 20R$  diacholestane; C27 =  $C_{27}$   $\alpha\alpha\alpha 20R$  cholestane; C28 =  $C_{28}$   $\alpha\alpha\alpha 20R$  ergostane; C29 =  $C_{29}$   $\alpha\alpha\alpha 20R$  stigmastane.

Gam/ $H_{30}$ , change much less. Our explanation is that there is a high abundance of 5-ring terpanes relative to 3-ring terpanes covalently bound to asphaltene (resin) geo-macromolecule structure in solid bitumen, which can be released by both closed system thermal maturation and subsequent HyPy. Fig. 5 provides strong evidence for this explanation in that the relative abundance of tricyclic terpanes to hopanes is very low in all hydropyrolysates. Fig. 7 shows

that the ratios of TT<sub>23</sub>/ $H_{30}$ ,  $H_{29}$ / $H_{30}$  and Gam/ $H_{30}$  are very consistent in all hydropyrolysates, more stable than the corresponding ratios in the bitumen maturation products at the same simulation temperature. This is because the free biomarkers in the altered bitumen, which may obscure the characteristics of newly generated biomarkers (Jones et al., 1988), were eliminated by the long Soxhlet extraction with ternary solvent before HyPy. Asphaltene



**Fig. 5.** GC-MS selected ion chromatograms of saturated fractions in hydropyrolysates: (a)  $m/z$  191 mass chromatograms, displaying terpane distributions; (b)  $m/z$  217 mass chromatograms, displaying sterane distributions. TT23–24 =  $C_{23-24}$  tricyclic terpanes; H29–31 =  $C_{29-31}$  17 $\alpha$ (H) 21 $\beta$ (H) hopanes; Gam = gammacerane; C27  $\beta\alpha S$  =  $C_{27}$   $\beta\alpha 20S$  diacholestane; C27  $\beta\alpha R$  =  $C_{27}$   $\beta\alpha 20R$  diacholestane; C27 =  $C_{27}$   $\alpha\alpha\alpha 20R$  cholestane; C28 =  $C_{28}$   $\alpha\alpha\alpha 20R$  ergostane; C29 =  $C_{29}$   $\alpha\alpha\alpha 20R$  stigmastane.

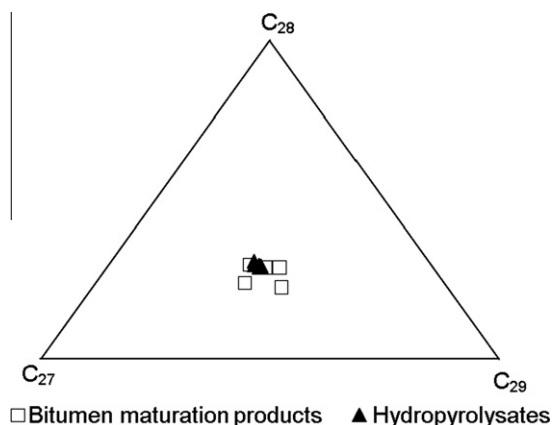


Fig. 6. Sterane ternary diagram of  $C_{27}$ – $C_{28}$ – $C_{29}$   $\alpha\alpha\alpha 20R$  regular steranes in bitumen maturation products and hydropyrolysates.

pyrolysates are known to be useful in oil-source correlation for severely biodegraded oils (Rubinstein et al., 1979; Behar et al., 1984; Rullkötter and Michaelis, 1990; Wilhelms et al., 1994) because asphaltenes possess alkyl distributions and biomarker fingerprints inherent to their source (Behar et al., 1984; Jones et al., 1988).

### 3.2. Maturity related biomarker parameters in bitumen maturation products and hydropyrolysates

Diasterane/sterane ratios are commonly used as source related parameters to distinguish petroleum from carbonate versus clastic source rocks (Peters et al., 2005). However, diasterane/sterane ratios are considered to be affected also by thermal maturity (Peters et al., 2005). At overmature stages, the increase in diasterane/

sterane ratios may be attributed to either the rearrangement of steranes to diasteranes or decomposition of regular steranes, or both (Peters et al., 2005). Fig. 8 shows that the  $C_{27}$  diacholestane to  $C_{27}$  cholestane ratio ( $C_{27} \beta\alpha R/C_{27} \alpha\alpha R$ ) for the artificially matured bitumens increases with increasing temperature. The  $C_{27} \beta\alpha R/C_{27} \alpha\alpha R$  ratio is 0.23 in original bitumen and increases progressively to 0.43 in B-460 (Table 2). By comparison, the  $C_{27} \beta\alpha S$  diacholestane to  $C_{27} \beta\alpha R$  diacholestane ratios ( $C_{27} \beta\alpha S/C_{27} \beta\alpha R$ ) for the hydropyrolysates show no increase with increasing temperature, again suggesting that the molecules are protected from rearrangement or destruction by their incorporation covalently bound macromolecules.

Most maturity parameters are only used in the oil generative window at lower maturity stages because these parameters may cease to change at high maturities (Peters et al., 2005). Many commonly used maturity parameters are based on the apparent isomerization of biomarkers. The isomerization of  $17\alpha$ -hopanes at C-22 occurs earlier than that of regular steranes at C-20 (Peters et al., 2005). Hence, the  $22S/(22S + 22R)$  homohopane isomerization ratios for  $C_{31}$ – $C_{35}$  hopanes are highly specific for the immature to early oil generation stages. The data in Fig. 7 show that the isomerization ratios of  $C_{31}$  hopane ( $C_{31} 22S/(22S + 22R)$ ) in the original bitumen, bitumen thermal maturation products and hydropyrolysates at all simulation temperatures are all about 0.6, very close to the equilibrium point.

The  $C_{29}$  sterane isomer ratios of  $C_{29} 20S/(20S + 20R)$  and  $C_{29} \beta\beta/(\beta\beta + \alpha\alpha)$  are often used as maturity indicators for crude oils. Isomerization at C-20 results in the ratio of  $C_{29} 20S/(20S + 20R)$  increasing progressively from <0.1 to about 0.5 during maturation. The end point of this ratio is thought to be in the range of 0.52–0.55 (Seifert and Moldowan, 1986; Peters et al., 2005). However,  $C_{29} 20S/(20S + 20R)$  in the original bitumen is 0.50, higher than 0.45 in B-350 (Table 2). Peters et al. (2005) suggested that biodegradation can affect the sterane isomers by preferential removal of

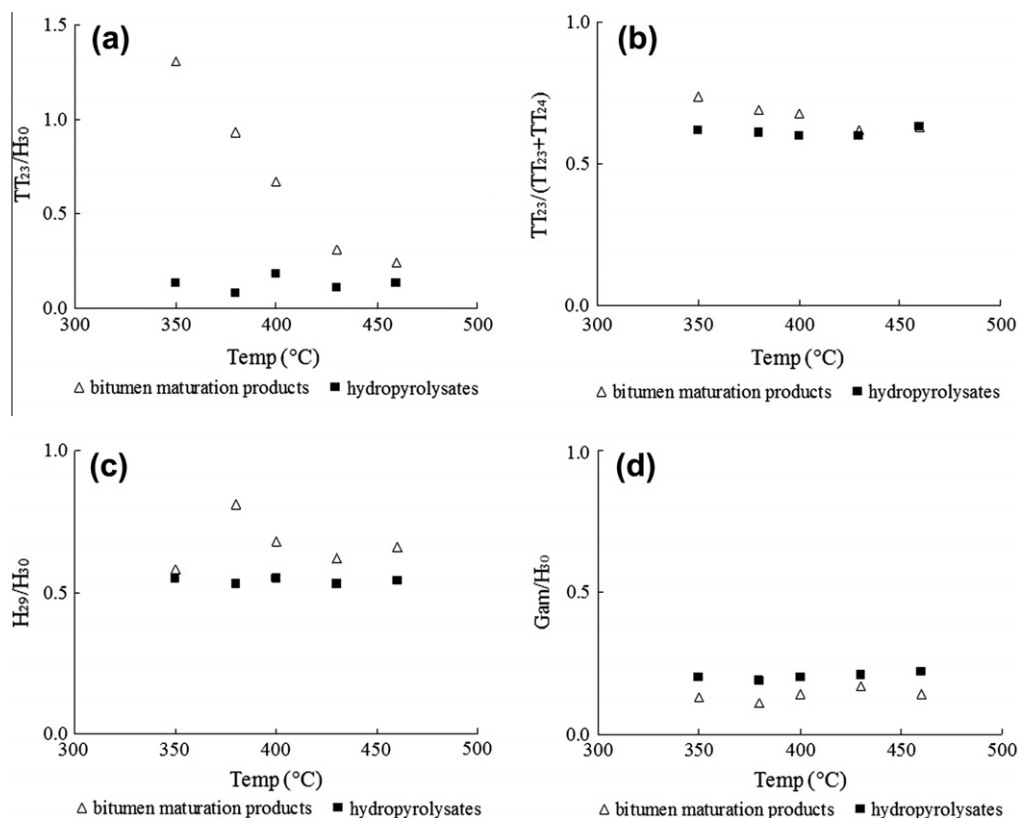
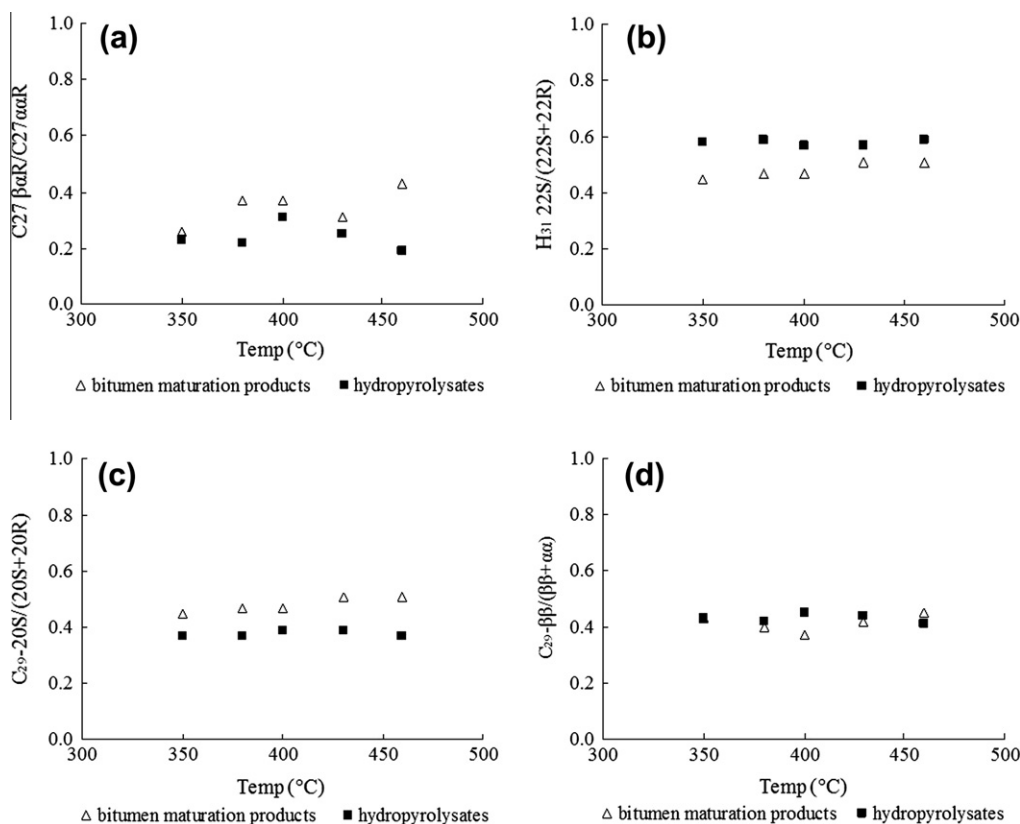


Fig. 7. Variations of source-related biomarker parameters with simulation temperature: (a)  $TT_{23}/H_{30}$ ; (b)  $TT_{23}/(TT_{23} + TT_{24})$ ; (c)  $H_{29}/H_{30}$ ; (d)  $Gam/H_{30}$ .



**Fig. 8.** Variations of maturity-related biomarker parameters with simulation temperature: (a)  $C_{27} \beta\alpha R / C_{27} \alpha\alpha R$ ; (b)  $H_{31} 22S / (22S + 22R)$ ; (c)  $C_{29-20S} / (20S + 20R)$ ; (d)  $C_{29-\beta\beta} / (\beta\beta + \alpha\alpha)$ .

**Table 2**  
Biomarker parameters in bitumen maturation products and hydropyrolysates.

Biomarker parameters	B-original	Bitumen maturation products					Hydropyrolysates				
		B-350	B-380	B-400	B-430	B-460	B-350hs	B-380hs	B-400hs	B-430hs	B-460hs
$C_{27} / (C_{27} + C_{28} + C_{29})$	0.28	0.42	0.35	0.38	0.35	0.32	0.36	0.36	0.37	0.36	0.36
$C_{28} / (C_{27} + C_{28} + C_{29})$	0.21	0.24	0.23	0.30	0.29	0.29	0.30	0.29	0.30	0.31	0.30
$C_{29} / (C_{27} + C_{28} + C_{29})$	0.51	0.34	0.43	0.32	0.36	0.39	0.34	0.34	0.33	0.32	0.35
$C_{27} \beta\alpha R / C_{27} \alpha\alpha R$	0.23	0.26	0.37	0.37	0.31	0.43	0.23	0.22	0.31	0.25	0.19
$H_{31} 22S / (22S + 22R)$	0.59	0.60	0.59	0.61	0.59	0.55	0.58	0.59	0.57	0.57	0.59
$C_{29-20S} / (20S + 20R)$	0.50	0.45	0.47	0.47	0.51	0.51	0.37	0.37	0.39	0.39	0.37
$C_{29-\beta\beta} / (\beta\beta + \alpha\alpha)$	0.57	0.43	0.40	0.37	0.42	0.45	0.43	0.42	0.45	0.44	0.41
$TT_{23} / H_{30}$	2.04	1.31	0.93	0.67	0.31	0.24	0.13	0.08	0.18	0.11	0.13
$TT_{23} / (TT_{23} + TT_{24})$	0.65	0.74	0.69	0.68	0.62	0.63	0.62	0.61	0.60	0.60	0.63
$H_{29} / H_{30}$	1.26	0.58	0.81	0.68	0.62	0.66	0.55	0.53	0.55	0.53	0.54
$Gam / H_{30}$	0.19	0.13	0.11	0.14	0.17	0.14	0.20	0.19	0.20	0.21	0.22

$\alpha\alpha 20R$  steranes, thus  $C_{29} 20S / (20S + 20R)$  may be altered during biodegradation. The data in Fig. 8 indicate that  $C_{29} 20S / (20S + 20R)$  in the bitumen maturation products increases from 0.45 to 0.51 with increasing simulation temperature. The  $C_{29} 20S / (20S + 20R)$  ratio in hydropyrolysates is about 0.37, much lower than its end point. It remains fairly constant in all hydropyrolysates, all in the range of 0.37–0.39. The  $C_{29} 20S / (20S + 20R)$  ratio is much lower in the hydropyrolysates than that in bitumen maturation products at all temperatures (Table 2).

$C_{29} \beta\beta / (\beta\beta + \alpha\alpha)$  ratio is a maturity index thought to be independent of source input. The  $C_{29} \beta\beta / (\beta\beta + \alpha\alpha)$  ratio increases from <0.1 to about 0.7 (Seilfert and Moldowan, 1986; Peters et al., 2005), with the apparent equilibrium point in the range of 0.67–0.71. Peters et al. (2005) suggested that the  $C_{29} \beta\beta / (\beta\beta + \alpha\alpha)$  ratio is slower than the  $C_{29} 20S / (20S + 20R)$  ratio to reach equilibrium during thermal maturation. However,  $C_{29} \beta\beta / (\beta\beta + \alpha\alpha)$  ratios

in the hydropyrolysates are all in the range of 0.41–0.45. There are no systematic changes in  $C_{29} \beta\beta / (\beta\beta + \alpha\alpha)$  ratios in hydropyrolysates with simulation temperature (Fig. 8). These results are consistent with those of Bishop et al. (1998), Murray et al. (1998), Bowden et al. (2006) and Lockhart et al. (2008) who also reported that the isomerization indices of both  $C_{29}$  steranes and  $C_{31}$  hopanes in hydropyrolysates showed lower maturity than those of source rock extracts.

Both the isomerization and decomposition of free homohopanes and steranes in bitumens or oils can lead to the increase in isomerization ratios (Pan et al., 2008). Several previous studies have demonstrated that the increases in isomerization ratios of hopanes and steranes are mainly resulting from the decomposition of biomarkers rather than direct isomerization (Larcher et al., 1988; Dzou et al., 1995; Farrimond et al., 1998). Thus the release of covalently bound homohopanes and steranes from kerogens and other

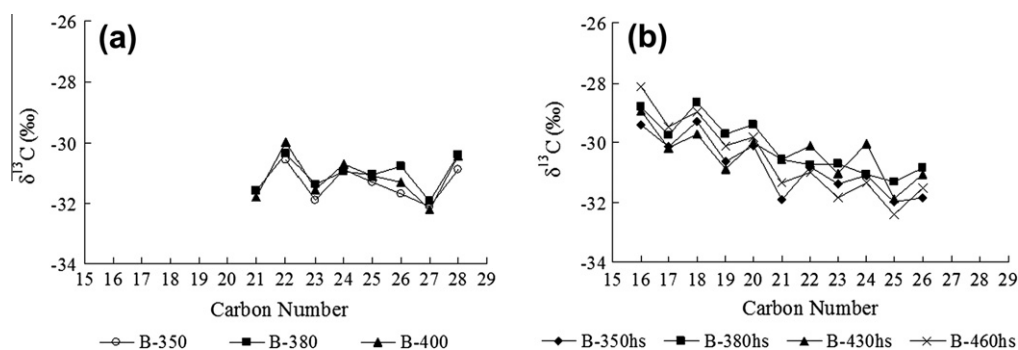


Fig. 9. The  $\delta^{13}\text{C}$  values of *n*-alkanes in saturated fractions: (a) bitumen maturation products; (b) hydropyrolysates.

macromolecules would usually lead to decreasing isomer ratios in favor of immaturity (Lu et al., 1989; Peters and Moldowan, 1991; Bishop et al., 1998). It is considered that the biomarkers released by HyPy are covalently bound to geo-macromolecules of kerogen (Love et al., 1998; Lockhart et al., 2008) and asphaltenes (Russell et al., 2004). The biomarker moieties incorporated within the geo-macromolecular structure of kerogens and asphaltenes are considered to be protected from secondary alterations such as biodegradation (Rubinstein et al., 1979; Behar et al., 1984a; Cassani and Eglinton, 1986; Russell et al., 2004) and thermal maturation (Love et al., 1998; Lockhart et al., 2008) that are common to free biomarkers. In addition, HyPy is a kind of open system pyrolysis using high pressure hydrogen as carrier gas. The volatile biomarkers are blown away by hydrogen flow at high velocity shortly after being released from geomacromolecules (Love et al., 2005). Thus decomposition and isomerization of the biomarkers in hydropyrolysates are dramatically reduced by the protection of high pressure hydrogen and the quick escape from the hot pyrolysis tube. Thus the effect of thermal alteration on both the source related and maturity parameters in hydropyrolysates is significantly reduced.

### 3.3. $\delta^{13}\text{C}$ values of *n*-alkanes in bitumen maturation products and hydropyrolysates

The stable carbon isotopic composition of individual *n*-alkanes has been used for characterizing organic sources (Freeman et al., 1990; Hayes et al., 1990; Rieley et al., 1991), correlating oil with source (Bjørøy et al., 1991, 1994; Peters et al., 2005; Fang et al., 2011) and reconstructing source rock paleoenvironment and paleoclimate (Hayes et al., 1990; Murray et al., 1994; Ruble et al., 1994; Schoell et al., 1994; Wilhelms et al., 1994; Mayer and Schwark, 1999; Sun et al., 2000; Xiong et al., 2003, 2007). Xiong and Geng (2000) compared the isotopic compositions of *n*-alkanes released from asphaltenes of severely biodegraded oils by closed system pyrolysis and suggested it as a viable oil-source correlation method for biodegraded oils.

In this study, the isotopic compositions of *n*-alkanes in bitumen maturation products and hydropyrolysates were measured and compared (Fig. 9). The concentration of *n*-alkanes in bitumen pyrolysates B-430 and B-460 were too low to allow measurement of their isotopic values. Furthermore, it was not possible to measure the isotope values for *n*-alkanes with <21 carbons in any of the bitumen sample. However, *n*-alkane concentrations in the hydropyrolysates were high enough to permit measurement of the isotope values. In the carbon number range of 21–26, the  $\delta^{13}\text{C}$  values of *n*-alkanes in hydropyrolysates are very similar to those in the bitumen maturation products at the same simulation temperatures. The differences between  $\delta^{13}\text{C}$  values of *n*-alkanes with the same carbon number in bitumens vs. hydropyrolysates are

usually <2‰. Such variations (<2–3‰) can be attributed to the difference in maturity (Bjørøy et al., 1992). In both bitumen maturation products and hydropyrolysates, the  $\delta^{13}\text{C}$  values of *n*-alkanes with odd carbon number are slightly lighter than *n*-alkanes with even carbon number. The similarity in the distributions of  $\delta^{13}\text{C}$  values for *n*-alkanes may be considered as an indicator of possible identical source (Liao et al., 2009). The studies by Love et al. (1998) also showed that the distribution of  $\delta^{13}\text{C}$  values of *n*-alkanes in EOM of source rocks was very similar to that in hydropyrolysates. Nevertheless, Jones et al. (1988) and Xiong and Geng (2000) reported some isotopic differences between isotopic values of crude oil alkanes and corresponding asphaltene pyrolysate alkanes. For this reason, oil-source correlations based on the distribution of  $\delta^{13}\text{C}$  values of *n*-alkanes in hydropyrolysates may be less reliable than those based on biomarkers.

## 4. Conclusions

A biodegraded soft solid bitumen of low maturity was thermally altered to various maturities in a closed system at different temperatures to simulate the thermal maturation process of biodegraded bitumens in the Majiang paleo-reservoir. The bitumen maturation products at various temperatures were collected and then HyPy was performed to release biomarkers and *n*-alkanes from the bitumen residues. The commonly used biomarkers in bitumen maturation products and hydropyrolysates were analyzed and the results indicated that the source related biomarker parameters in hydropyrolysates are insensitive to thermal stress. Furthermore, maturity parameters derived from analysis of the hydropyrolysates indicate lower thermal maturity relative to bitumen maturation products, due to the protection effect of macromolecular structure of asphaltenes and the high openness of the HyPy system. The results suggest that biomarkers released from solid bitumen by HyPy can be used to study the source characteristics of solid bitumens and to make bitumen–bitumen and bitumen–source correlations in the Majiang paleo-reservoir and other paleo-reservoirs that had suffered both biodegradation and severe thermal maturation. The distributions of  $\delta^{13}\text{C}$  of *n*-alkanes in hydropyrolysates are very similar to each other and are also similar to those of bitumen matured in a closed system at lower simulation temperatures. The distribution of  $\delta^{13}\text{C}$  of *n*-alkanes in hydropyrolysates may also be useful in bitumen–bitumen correlations for paleo-reservoir solid bitumen.

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