

# Carbon isotope composition of organic compounds produced by abiotic synthesis under hydrothermal conditions

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## Abstract

Although it is widely believed that production of organic compounds by Fischer–Tropsch synthesis and related processes occurs in many geologic environments, unambiguous identification of compounds with an abiotic origin in natural samples has been hampered by a lack of means to discriminate between abiotic compounds and organic matter from biological sources. While isotopic compositions might provide a means to discriminate between biologic and non-biologic sources of organic matter, there are few data presently available to constrain the isotopic composition of compounds produced by abiotic processes in geologic systems. Here, we report results of laboratory experiments conducted to evaluate the isotopic composition of organic compounds synthesized abiotically under hydrothermal conditions. We find the organic products are depleted in  $^{13}\text{C}$  to a degree typically ascribed to biological processes, indicating that carbon isotopic composition may not be a particularly effective diagnostic means to differentiate between biologic and non-biologic sources. Furthermore, our results suggest that the isotopic compositions of reduced carbon compounds found in many ancient rocks that have heretofore been attributed to biological sources could be consistent with an abiotic origin in a hydrothermal setting.

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## 1. Introduction

It has become commonly accepted in the earth and planetary sciences that organic compounds are readily synthesized by the Fischer–Tropsch reaction or related processes during fluid–rock interactions under hydrothermal conditions [e.g., 1–6]. The production of organic compounds by Fischer–Tropsch-type (FTT) synthesis has been invoked to explain the occurrence of hydro-

carbons within igneous rocks and hydrothermal fluids, and has been implicated in a wide variety of geologic processes including the production of methane and hydrocarbons within the deep crust, providing nutrients to microbes in subsurface and hydrothermal environments, and serving as a source of prebiotic compounds on the early Earth [7–20]. In addition, the potential for organic synthesis is one of the principal reasons that submarine hydrothermal systems are considered as a possible site for the origin of life [1–3,6].

Despite the widespread perception that FTT synthesis is a continuous and substantial source of organic matter in submarine hydrothermal systems and elsewhere

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within the Earth's crust, definitive identification of organic compounds with an abiotic origin from natural systems on Earth has been elusive. To date, no compounds from hydrothermal environments other than methane have been demonstrated to have an unambiguous abiotic origin. While organic compounds are prevalent in hydrothermal environments, particularly those covered by sediments, these compounds appear to be predominantly derived from thermal decomposition of biologically derived organic matter [21–23]. Indeed, a major difficulty in being able to recognize the presence of abiotic organic compounds in such systems is the ubiquitous prevalence of biologically derived organic matter that tends to swamp out any signal that might come from abiotic sources.

Recently, it has been proposed that abiotically generated hydrocarbon gases may have isotopic signatures distinctly different from gases derived from biologic sources. In particular,  $C_1$ – $C_4$  hydrocarbons with an apparent abiotic origin from igneous rocks have been found to display a trend of decreasing  $^{13}C$  content with increasing carbon number that is the reverse of the typical thermogenic trend of enrichment in  $^{13}C$  with increasing carbon number [7,9]. Additional support for an abiotic origin for some of these gases comes from their hydrogen isotopic compositions that also exhibit trends that are distinct from the typical thermogenic trend [7]. Although the geologic setting and isotopic composition of these gases is suggestive of an abiotic origin, the production of the isotopic trends under geologic conditions remains to be confirmed by laboratory experiments. Furthermore, the uniqueness of the observed isotopic trends as an abiotic signature is somewhat uncertain since laboratory experiments and field studies of natural gases in sedimentary basins have indicated that reversed carbon isotope trends can also be produced during thermal maturation of biologically derived organic matter under some circumstances [24] or by mixing and other processes during migration [25–27]. At present, it appears unlikely that reversed carbon isotope trends alone provide sufficient evidence to establish an abiotic origin for hydrocarbon gases.

Use of carbon isotope compositions has been particularly prominent in discussions regarding potential biological contributions to the reduced carbon compounds found in rock samples from the early Earth and their implications for dating the emergence of life. Many of the oldest known rocks on Earth have been found to contain reduced carbon compounds (kerogen and graphite) that are depleted in  $^{13}C$  by 20–40% relative to coexisting carbonate [28–34]. Because this depletion is similar in magnitude to that produced by biological

carbon fixation, these compounds have frequently been interpreted as evidence for the evolution of relatively complex life forms prior to 3.5 billion years (Ga) ago [e.g., 29,30,33,34]. Recent reassessments of the geologic context of several of these ancient carbon-bearing rock strata, however, suggest an abiotic origin for the reduced carbon involving either Fischer–Tropsch-type (FTT) processes in a hydrothermal environment or decomposition of iron-rich carbonates under metamorphic conditions [36–40]. Because the isotopic signature of organic compounds produced by abiotic pathways is largely unknown at present, it has remained unclear whether abiotic sources could potentially account for the isotopic compositions of the reduced carbon compounds found in these ancient rocks.

Laboratory studies of isotope fractionation during Fischer–Tropsch-type synthesis could help to resolve uncertainties associated with the identification of abiotic organic compounds. However, studies of isotopic fractionation during abiotic synthesis have been scarce and limited primarily to light hydrocarbon gases [41,42] and/or experimental conditions with no clear application to the inferred geologic environments where abiotic synthesis is thought to occur [43,44]. In order to address this issue, we conducted laboratory experiments to measure the carbon isotope composition of organic compounds synthesized abiotically under hydrothermal conditions. Here, we describe the results of the experiments, and discuss the implications for identification of abiotic organic compounds in geologic systems and the source of carbon compounds in rocks from the early Earth.

## 2. Methods

The experiments were performed by heating an aqueous solution of formic acid (HCOOH) in the presence of Fe powder at 250 °C and 325 bar. Native Fe is unstable under hydrothermal conditions and reacts rapidly with water during heating to form magnetite ( $Fe_3O_4$ ) plus  $H_2$ , resulting in strongly reducing conditions within the reaction vessel. Aqueous formic acid also rapidly decomposes to  $CO_2$  plus  $H_2$  under the experimental conditions [45] and was used as a carbon source to circumvent technical difficulties associated with dissolving large amounts of  $CO_2$  in water.

Experiments were conducted in a flexible-cell hydrothermal apparatus using a gold reaction cell with titanium fittings [45–47]. Prior to the experiments, the reaction cell was heated overnight at 350 °C in air to remove potential organic contaminants. The titanium fittings had previously been heated in air to create an

inert TiO<sub>2</sub> layer on the surface. Numerous previous experiments with the same reaction system have demonstrated that the reactor itself is not catalytic for organic synthesis under the experimental conditions [45,47].

For the experiment, 0.7 g of powdered Fe (200 mesh, >99.9% pure; Johnson Matthey) was combined in the reaction vessel with 37 ml of an aqueous solution containing 125 mmol HCOOH. The reactor was pressurized to 325 bar at room temperature and then heated to 250 °C while maintaining approximately constant pressure (reactor reached final temperature within ~2 h). After 44 h and then again at 86 h, several small fluid samples were removed through a sampling valve into gas tight syringes and analyzed for volatile gases and dissolved organic acids. Additional fluid samples were injected into evacuated 20 ml culture tubes with butyl rubber stoppers for isotopic analyses of volatile hydrocarbons. Prior to use, the butyl rubber stoppers were boiled in 1 N NaOH to remove trace hydrocarbon contaminants. Following the second sample, the reactor was cooled to room temperature, after which ~3 ml of dichloromethane (DCM) was injected into the reaction cell and shaken for ~15 min in order to extract non-volatile organic products.

Quantitative analyses of dissolved H<sub>2</sub>, CO<sub>2</sub>, CH<sub>4</sub>, and volatile hydrocarbons were performed by gas

chromatography (GC) as previously described [45,47]. Organic acids were analyzed by ion chromatography. The DCM extract was concentrated by evaporation at room temperature prior to analysis by gas chromatography–mass spectrometry (GC–MS) for identification of non-volatile products and by gas chromatography with flame ionization detection for quantification. The carbon isotope compositions of individual hydrocarbons were measured by compound specific isotope analysis on a ThermoFinnigan DeltaPlus using a 30 m long, 320 μm internal diameter AT-Q (Alltech) column for separation of volatile compounds and a CP-Sil 5CB column (50 m×0.25 mm i.d., 0.25 μm film thickness) for separation of compounds in the DCM extract. Carbon isotopes are reported in standard delta notation relative to the PDB standard. Analysis of replicate samples indicated that analytical uncertainty was approximately ±1.0‰.

Concentration of the DCM extract resulted in partial loss of some semi-volatile, lower molecular weight hydrocarbons (<~C<sub>11</sub>), but loss of heavier compounds was probably negligible. It is likely that evaporation of these semi-volatile compounds resulted in isotopic fractionation, so that the reported isotopic compositions of the C<sub>9</sub>–C<sub>11</sub> hydrocarbons should be considered provisional. Note that this has no effect on the reported composition of the volatile hydrocarbons (C<sub>1</sub>–C<sub>6</sub>) since

Table 1  
Abundance and isotopic composition of dissolved volatile compounds produced during the hydrothermal experiment

Compound	Sample after 44 h		Sample after 86 h		Total amount synthesized (μmol)*
	Concentration (mmol kg <sup>-1</sup> )	δ <sup>13</sup> C (‰)	Concentration (mmol kg <sup>-1</sup> )	δ <sup>13</sup> C (‰)	
H <sub>2</sub>	411	–	390	–	
ΣCO <sub>2</sub>	47.9	–15.0	41.4	–14.2	
Methane	10.1	–49.0	10.9	–50.3	404.
Ethane	3.1	–44.5	3.3	–44.4	122.
Ethene	0.12	–	0.033	–	1.2
Propane	1.1	–44.6	1.2	–47.1	44.
Propene	0.25	–	0.23	–	8.5
<i>n</i> -Butane	0.64	–	0.64	–43.8	24.
Isobutane	0.02	–	0.021	–	0.78
1-Butene	0.01	–	0.014	–	0.52
<i>n</i> -Pentane	0.25	–	0.26	–49.3	9.6
1-Pentene	0.02	–	0.025	–	0.93
ΣC <sub>6</sub> <sup>†</sup>	0.12	–	0.12	–50.4	4.4
Formic acid <sup>‡</sup>	–	–	0.91	–	34.
Acetic acid	–	–	0.57	–	21.
Propionic acid	–	–	0.028	–	1.0
Butanoic acid	–	–	<0.02	–	<0.7

\*Calculated by multiplying the measured aqueous concentration after 86 h by the total mass of fluid (37 g). <sup>†</sup>*n*-Hexane and other isomers of hexane and hexane were not distinguishable during chromatographic analysis and are reported as a total. <sup>‡</sup>The reported abundances of carboxylic acids include both the acid and acid anion that may have been present in solution.

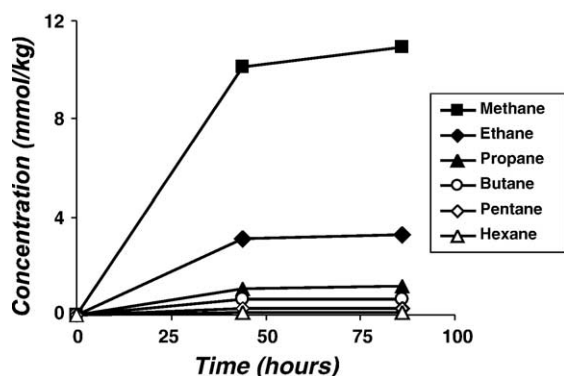


Fig. 1. Concentrations of dissolved hydrocarbons during the hydrothermal experiment.

their isotopic compositions were measured on gas samples obtained prior to the extraction of the heavier compounds with DCM.

### 3. Results

Rapid decomposition of formic acid and reaction of Fe with water resulted in high concentrations of H<sub>2</sub> and CO<sub>2</sub> in the first fluid sample obtained 44 h after initiation of heating (Table 1). Accompanying these compounds was production of substantial amounts of methane and a suite of C<sub>2</sub>–C<sub>6</sub> hydrocarbons composed predominantly of alkanes (Fig. 1; Table 1). The abundances of methane and hydrocarbons remained essentially the same when sampled after 86 h, indicating that the reaction proceeded rapidly during the early stages of the experiment and then slowed, possibly

owing to passivation of the mineral catalysts. Organic compounds in the DCM extract obtained after termination of the experiment were dominated by a homologous series of *n*-alkanes with lesser amounts of *n*-alkenes and *n*-alkan-1-ols (Fig. 2). Relative to straight chain hydrocarbons, branched compounds were present in very low abundance. Quantitative analyses of alkanes and alkanols are listed in Table 2; alkenes were not routinely quantified, but analyses of C<sub>12</sub> and C<sub>17</sub> compounds indicated that the summed total of all alkene isomers was ~30% of the corresponding *n*-alkane of the same carbon number. Solid inorganic products consisted of magnetite and the iron carbonate mineral siderite (FeCO<sub>3</sub>).

Products of FTT synthesis are typically dominated by unbranched alkanes that exhibit a characteristic linear decrease in abundance on a semilog plot of abundance vs. number of carbons, where the absolute value of the slope is referred to as the chain growth probability factor ( $\alpha$ ). The structures and relative abundances of organic compounds produced during the hydrothermal experiment closely resemble those generated by Fischer–Tropsch synthesis and exhibit a typical FTT distribution with an  $\alpha=0.38$  for the light hydrocarbons and alkanols and  $\alpha=0.12$  for heavier hydrocarbons (Fig. 3). The break in slope at carbon number  $\approx 9$  and the drop off in abundance for longer carbon chain lengths are commonly observed during Fe-catalyzed Fischer–Tropsch synthesis [48,49].

With the exception of C<sub>2</sub>–C<sub>4</sub> alkanes, all organic products had measured  $\delta^{13}\text{C}$  values within 1‰ of  $-50.5\text{‰}$  (Fig. 4; Tables 1 and 2). The organic products

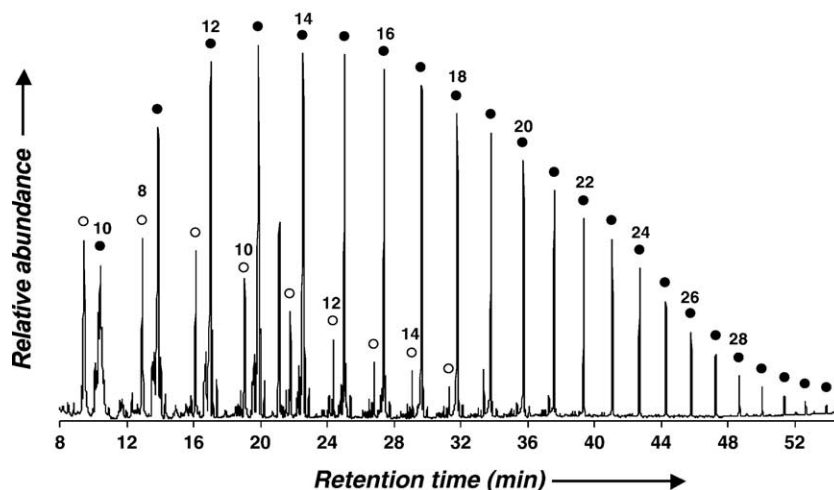


Fig. 2. Total ion chromatogram from GC–MS analysis of the DCM extract. Closed circles identify peaks corresponding to *n*-alkanes, while open circles are *n*-alkan-1-ols. Numbers indicate the carbon chain length of the compounds (only even number peaks labeled). Lower molecular weight hydrocarbons to left of diagram have been partially lost during sample concentration.

Table 2

Abundance and isotopic composition of non-volatile organic compounds in the DCM extract

Compound	Total amount synthesized ( $\mu\text{mol}$ )	$\delta^{13}\text{C}$ (‰)
<i>n</i> -Nonane	0.050*	(-48.6) <sup>†</sup>
<i>n</i> -Decane	0.067*	(-48.8) <sup>†</sup>
<i>n</i> -Undecane	0.075*	(-48.1) <sup>†</sup>
<i>n</i> -Dodecane	0.075	-50.5
<i>n</i> -Tridecane	0.061	-50.7
<i>n</i> -Butadecane	0.047	-50.7
<i>n</i> -Pentadecane	0.036	-51.0
<i>n</i> -Hexadecane	0.028	-50.1
<i>n</i> -Heptadecane	0.023	-50.3
<i>n</i> -Octadecane	0.018	-49.8
<i>n</i> -Nonadecane	0.014	-50.2
<i>n</i> -Eicosane	0.010	-50.1
<i>n</i> -Heneicosane	0.0074	-50.6
<i>n</i> -Docosane	0.0053	-50.5
<i>n</i> -Tricosane	0.0038	-50.3
<i>n</i> -Tetracosane	0.0025	-50.6
<i>n</i> -Pentacosane	0.0015	-50.5
<i>n</i> -Hexacosane	0.0011	-51.0
<i>n</i> -Heptacosane	0.00052	-51.8
<i>n</i> -Octacosane	0.00025	-
1-Hexanol	1.24	-
1-Heptanol	0.52	-47.3
1-Octanol	0.200	-48.5
1-Nonanol	0.083	-48.2
1-Decanol	0.038	-
1-Undecanol	0.017	-

\*The amounts of alkanes  $\leq C_{11}$  reflect depletion during concentration of the extract prior to analysis. <sup>†</sup>The isotopic composition of shorter-chain alkanes may have been effected by evaporation during extract concentration and should be considered as provisional values.

exhibit carbon isotopic contents that are depleted in  $^{13}\text{C}$  by  $\sim 36\%$  relative to dissolved  $\text{CO}_2$  ( $\delta^{13}\text{C} = -14\%$ ) and  $\sim 31\%$  relative to precipitated siderite ( $-19\%$ ). Ethane,

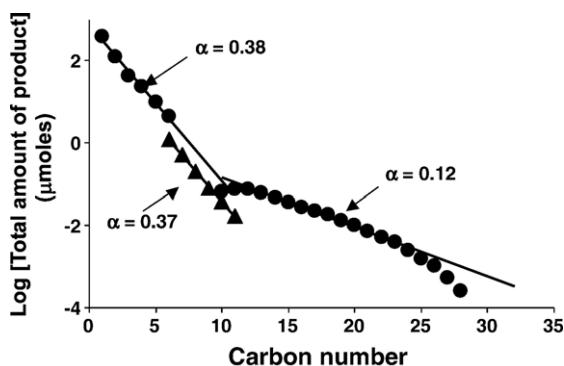


Fig. 3. Abundances of *n*-alkanes (●) and *n*-alkan-1-ols (▲) generated during abiotic organic synthesis under hydrothermal conditions. Lines are linear regressions of the  $C_1$ – $C_6$  alkanes ( $\alpha=0.38$ ;  $r^2=0.99$ ),  $C_7$ – $C_{11}$  alkanols ( $\alpha=0.37$ ;  $r^2=1.00$ ), and  $C_{12}$ – $C_{22}$  alkanes ( $\alpha=0.12$ ;  $r^2=0.99$ ). The abundances of  $C_{10}$  and  $C_{11}$  alkanes reflect partial loss of these compounds during sample treatment.

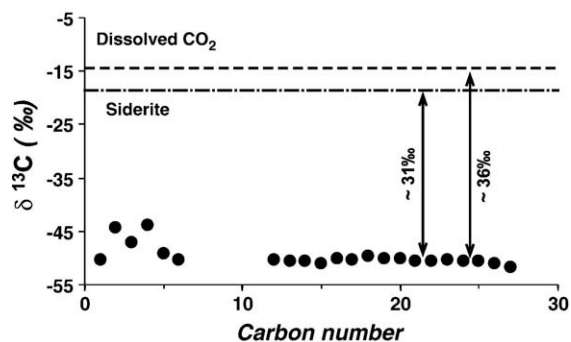


Fig. 4. Carbon isotopic composition of hydrocarbons synthesized under hydrothermal conditions. Horizontal lines represent isotopic composition of dissolved  $\text{CO}_2$  ( $-14.2\%$ ) and precipitated siderite ( $-19.1\%$ ).

propane, and butane were enriched in  $^{13}\text{C}$  relative to other hydrocarbons. The cause of this minor enrichment is difficult to determine with the present data, but may be attributable to additional formation of short-chain *n*-alkanes through reduction of short-chain alkenes or carboxylic acids that were isotopically enriched during initial synthesis or as the result of isotopic exchange with dissolved  $\text{CO}_2$ . For instance, it has been shown experimentally that acetic acid can be enriched in  $^{13}\text{C}$  through exchange of the carboxyl carbon with dissolved  $^{13}\text{CO}_2$  [50,51], and subsequent reduction of the acetic acid to ethane would result in  $^{13}\text{C}$  enrichment of the ethane. We were unable to measure the isotopic composition of the starting formic acid directly. However, since the formic acid was essentially quantitatively converted to  $\text{CO}_2$ , siderite, and methane, it can be inferred from mass balance that the starting isotopic composition of formic acid was about  $-20\%$ .

Previous experimental studies of abiotic synthesis have shown that background sources can contribute significant amounts of organic compounds to hydrothermal experiments, which can lead to erroneous interpretations if they are not accounted for [47,52,53]. In order to evaluate the extent to which organic contaminants may have contributed to the observed products and isotopic signatures in the present study, a second experiment was performed using identical procedures but substituting  $^{13}\text{C}$ -labeled formic acid (99%  $\text{H}^{13}\text{COOH}$ ) as the carbon source. This experiment yielded organic products whose composition and yields were essentially identical to those for the unlabeled experiment, except that all organic carbon compounds identified were composed predominantly of  $^{13}\text{C}$ . For example, Fig. 5 shows a comparison of mass spectra for hexadecane produced in the  $^{13}\text{C}$ -labeled experiment with that of the unlabeled experiment, and the spectra

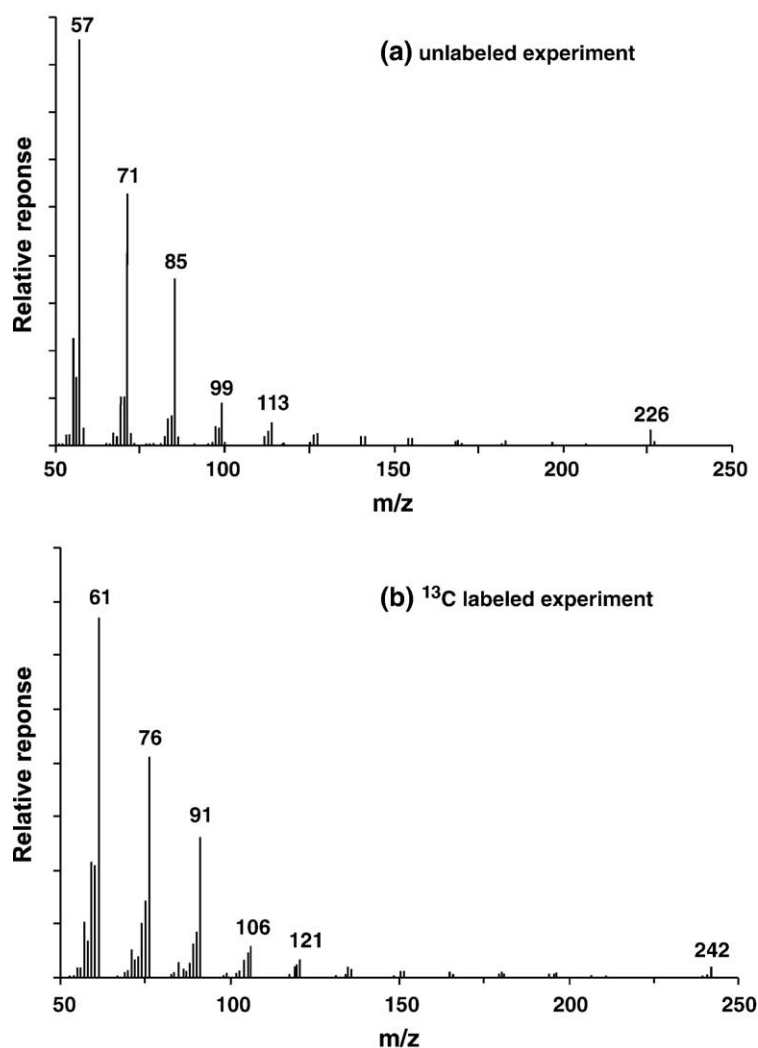


Fig. 5. Comparison of mass spectra for hexadecane ( $C_{16}H_{32}$ ) from the unlabeled and  $^{13}C$ -labeled experiments. The spectrum in (a) is typical of unbranched alkanes and prominent peaks include the molecular ion ( $m/z=226$ ) and mass fragments for  $^{12}C_4H_9^+$ ,  $^{12}C_5H_{11}^+$ ,  $^{12}C_6H_{13}^+$ ,  $^{12}C_7H_{15}^+$ , and  $^{12}C_8H_{17}^+$  ( $m/z=57, 71, 85, 99,$  and  $113,$  respectively). Analogous peaks in the spectrum from the labeled experiment (b) reflect the formation of  $^{13}C$ -labeled compounds, including the molecular ion at  $m/z=242$  (corresponding to  $^{13}C_{16}H_{34}$ ) and  $m/z=61, 76, 91, 106,$  and  $121$  (corresponding to  $^{13}C_4H_9^+$ ,  $^{13}C_5H_{11}^+$ ,  $^{13}C_6H_{13}^+$ ,  $^{13}C_7H_{15}^+$ , and  $^{13}C_8H_{17}^+$ ). Minor peaks at  $m/z=57, 71, 85,$  etc. in (b) indicate the contribution of a small amount of unlabeled hexadecane from background contaminants in the labeled experiment equivalent to  $<10\%$  of the total.

shows that nearly all of the carbon in hexadecane from the labeled experiment consists of  $^{13}C$ . The labeled experiment indicated  $<10\%$  contribution from background sources for heavier compounds, with substantially smaller contributions for lighter hydrocarbons (e.g.,  $<3\%$  for methane and ethane). The results of the labeling experiment indicate that the large majority of organic compounds observed were formed by abiotic synthesis during the course of the experiment, with little or no contribution from background contaminants.

Although the contribution of background sources to the measured isotopic compositions cannot be com-

pletely resolved with the present data, it is very unlikely that the minor contribution of hydrocarbons from background sources had a substantial effect on the isotopic compositions. The results of the labeled experiment indicate that the contribution from background sources is less than 10% for heavier hydrocarbons, and even less for the lighter compounds. Unless the background compounds have a highly unusual isotopic composition (i.e.,  $\delta^{13}C < -100\%$  or  $>0\%$ ), these trace background sources cannot alter the measured isotopic composition by more than a few per mil. Furthermore, any variation in the isotopic

compositions would probably be systematic, so that the trends among different hydrocarbons were unlikely to be affected to any significant extent.

#### 4. Discussion

The structures and relative abundances of organic products observed in the experiment indicate they formed by Fischer–Tropsch-type syntheses under hydrothermal conditions. Fischer–Tropsch synthesis is a surface catalyzed process in which oxidized carbon compounds ( $\text{CO}_2$  or  $\text{CO}$ ) bind to the surface of the catalyst where they undergo stepwise reduction to form methylene groups, which then polymerize at the surface to form longer carbon chains. Chain growth terminates when the alkyl chain thus formed combines with an H atom bound to the surface rather than an additional methylene group. Our previous studies have shown that while synthesis of organic compounds more complex than methane from reduction of  $\text{CO}_2$  may occur readily within a water-saturated vapor phase at the experimental conditions, it does not appear to proceed within a liquid aqueous phase under such conditions [45,47,54]. We therefore infer that organic synthesis in the present experiment likely occurred by the FTT mechanism within  $\text{H}_2$ - and  $\text{CO}_2$ -rich vapor bubbles formed on the surface of the mineral catalyst during heating owing to the relatively low solubility of  $\text{H}_2$  in water [55]. Geologic evidence indicates that conditions sufficiently reducing to produce similar  $\text{H}_2$ - and  $\text{CO}_2$ -rich vapors occur during the early stage serpentinization of ultramafic rocks [56–58] and may also occur locally during interaction of migrating fluids with mafic minerals in sialic rocks [9]. Analogous formation of a separate vapor phase may also occur in seafloor hydrothermal environments owing to boiling at shallow depths or during injection of magmatic volatiles [4].

It is possible that the native Fe initially present in the experiments may have been catalytically active in the synthesis reaction. While native Fe is absent from hydrothermal environments on Earth, many common Fe-bearing minerals in hydrothermal systems, including magnetite and hematite, are known to be effective catalysts for FTT synthesis. The active catalytic sites for these minerals, however, are pockets of native Fe or Fe-carbides that form on their surfaces under reducing conditions [48,49]. Consequently, we expect that our experiments provided catalytic sites very similar to natural environments, albeit the process may have been more efficient in the experiments owing to a higher density of active sites. Additional experiments per-

formed with magnetite as catalyst have indicated that FTT does occur, but lower yields of synthesis products relative to background compounds precluded accurate carbon isotopic analysis of the abiotic products [McCollom, unpublished results].

The isotopic results indicate that the hydrocarbons synthesized were significantly depleted in  $^{13}\text{C}$  relative to the initial carbon source, with a consistent depletion relative to  $\text{CO}_2$  ( $\Delta^{13}\text{C}_{\text{CO}_2}$ -hydrocarbon) of  $\sim 36\%$ . Further experimentation will be required to determine the exact mechanisms responsible for the isotopic fractionations observed here, as well as to evaluate the potential dependence of the fractionation on such factors as the composition of the mineral catalyst, reaction temperature and pressure, and duration of reaction. However, the observation that the long-chain hydrocarbons are characterized by isotopic compositions that are indistinguishable from  $\text{CH}_4$  suggests that significant isotopic fractionation does not occur during the polymerization step of FTT synthesis. In this respect, the isotopic composition of the compounds produced by FTT reactions may differ from those produced during spark-induced polymerization of methane [44] or found in fluids in granitic rocks [7,9] which display depletions in  $^{13}\text{C}$  abundance with increasing carbon number. The  $^{13}\text{C}$  depletions observed during this study are similar in magnitude to equilibrium fractionation between  $\text{CO}_2$  and  $\text{CO}$  at the experimental temperature ( $\sim 35\%$  at  $250^\circ\text{C}$  [59]), suggesting that the bulk of the fractionation may occur during interconversion of  $\text{CO}_2$  and  $\text{CO}$  with little or no additional fractionation during hydrocarbon synthesis.

The magnitude of carbon isotope fractionation observed in the experiment is similar to that produced by many biological processes [60] (Fig. 6). In geologic samples, reduced carbon compounds whose isotopic compositions are depleted by 20–40‰ relative to coexisting carbonates are typically attributed to a biological origin. However, the similarity of  $^{13}\text{C}$  fractionation between inorganic carbon sources and organic matter observed during FTT synthesis in our hydrothermal experiment and typical values for biological fixation suggests that it might not be possible to distinguish between biotic and abiotic sources of reduced carbon based on isotopic composition alone. In metamorphosed rocks where the structural characteristics of the original organic matter have been obscured, unambiguous discrimination between these sources may be particularly difficult. These results suggest that it may be problematic to use carbon isotopic measurements to evaluate the potential contribution of abiotic synthesis to the organic matter found in geologic environments such

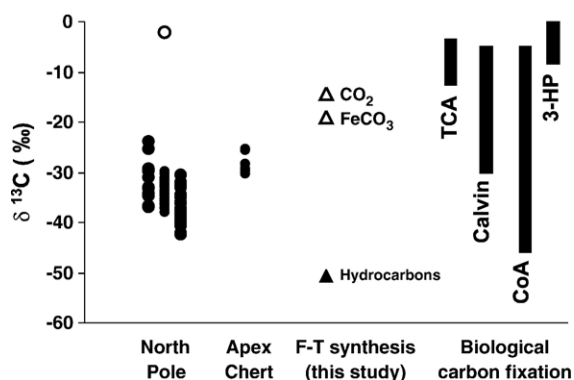


Fig. 6. Comparison of the isotopic fractionation observed for abiotic organic synthesis under hydrothermal conditions in the present study (triangles) with the measured isotopic compositions of organic matter in >3.46 Ga rocks from the Warrawoona Group of Western Australia (circles). Also shown for reference are isotopic fractionations measured in laboratory cultures of prokaryotes employing different carbon fixation pathways [58] (bars show range of observed values). Data for the Warrawoona Group are from the North Pole area [27,30,31] and Apex chert [34]. The open circle reflects the isotopic composition of carbonate minerals coexisting with organic matter in the North Pole area. Carbon fixation pathways include the reductive tricarboxylic acid cycle (TCA), Calvin cycle (Calvin), acetyl-CoA pathway (CoA), and 3-hydroxypropionate cycle (3-HP).

as submarine hydrothermal systems and the deep terrestrial subsurface.

On the other hand, while the data indicate that it may be difficult to differentiate biotic from abiotic organic compounds based on the isotopic composition of bulk samples or even analysis of individual compounds, the results do indicate the possibility that a robust biomarker may exist in the form of analysis of intramolecular carbon atoms within larger lipid compounds. Previous research has shown that alternating carbons within fatty acids and other compounds formed by biosynthetic pathways can have distinctive <sup>13</sup>C signatures that differ by several per mil; for instance, odd-numbered carbon atoms within fatty acid molecules produced by *Escherichia coli* have been found to be depleted by ~6‰ relative to even-numbered carbon atoms in the same compound [61,62]. These differences arise from the biochemical pathway whereby fatty acids are assembled by addition of 2-carbon acetyl units whose isotopic composition differs substantially in their methyl and carboxyl groups.

In contrast, the consistent isotopic fractionation observed in the synthesized carbon compounds in this study, as well as the addition of methylene units in single-carbon increments during FTT synthesis, indicates that organic compounds formed by FTT synthesis will have a constant <sup>13</sup>C composition for all carbon

atoms within an individual compound (i.e., no difference in even- and odd-numbered intramolecular carbon atoms). Analysis of individual carbon atoms within a molecule such as an alkane or alkanolic acid could therefore allow for differentiation between biotic and abiotic sources. Of course, such analysis would require the presence of intact molecules, and consequently might not be particularly useful for mature organic matter where most of the original carbon compounds have been converted to graphite or kerogen. However, this approach might be useful in modern hydrothermal environments or rocks where structural information remains intact.

#### 4.1. Implications for interpretation of carbon isotopic signatures from the early Earth

The presence of reduced carbon compounds in rocks from the early Earth that possess <sup>13</sup>C depletions of the same magnitude as those produced by biological processes has been widely used to infer that biological organisms were present at the time and place where the rocks originally formed. For instance, graphite in >3.8 billion year old rocks from the Isua Greenstone Belt and Akilia Island in southwest Greenland have been identified as biological in origin based exclusively on their carbon isotopic signature, which are depleted by 20–40‰ relative to coexisting carbonates [33,34]. Similarly, carbon isotopes have been used as supporting evidence for a biological origin for putative microfossils in >3.46 billion year old cherts from the Warrawoona Group of Western Australia [28–32]. Carbon compounds in these rocks exhibit isotopic compositions that are generally depleted in <sup>13</sup>C by 25–40‰ relative to coexisting carbonate minerals (Fig. 6). Organic matter in more recent sediments dominated by input from photosynthetic organisms typically exhibits carbon isotope compositions in the range of –25‰ to –30‰ [28], and modern autotrophic prokaryotes can produce <sup>13</sup>C depletions of 0‰ to ~40‰ during carbon fixation [60]. Thus, the isotope compositions of reduced carbon in these ancient rocks are coincident with the range of values resulting from biological carbon fixation, indicating that biological processes could potentially account for their isotope composition.

Over the last few years, however, the biological relevance of both of these occurrences of reduced carbon from the early Earth has been contested on the basis of geological and geochemical observations. There is now compelling evidence that occurrences of graphite from Isua and Akilia in Greenland which had previously



been identified as biological in origin are instead a product of thermal decomposition of siderite ( $\text{FeCO}_3$ ) during rock metamorphism [38–40]. The fact that graphite from this locale possesses a carbon isotopic signature within the range of biological fractionation provides additional evidence that abiotic processes can produce isotopic signatures that may be difficult to differentiate from biotic processes.

Recent field and laboratory studies have likewise indicated a non-biological source for reduced carbon compounds in cherts from the Warrawoona Group. Instead, it has been suggested that these compounds formed by abiotic organic synthesis in a hydrothermal setting by a Fischer–Tropsch-type process [35–37]. However, it has remained unclear whether abiotic sources could explain the isotopic composition of the reduced carbon compounds. The organic compounds produced in the present experiment are depleted in  $^{13}\text{C}$  relative to  $\text{CO}_2$  to the same amount as observed for reduced carbon compounds at Warrawoona (Fig. 6), indicating that the isotopic composition of those compounds could be consistent with an abiotic origin in a hydrothermal setting as suggested by recent studies [35–37].

Reduced carbon compounds in the Warrawoona Group rocks are presently found in silica-rich chert veins lined by barite, a setting that implies a relatively oxidizing environment. Within the context of an abiotic synthesis model, the absence of likely catalysts and the reducing conditions that would be most favorable for organic synthesis in their current setting suggests that the compounds were synthesized deeper in the hydrothermal system under appropriate geochemical conditions and subsequently transported to their present location. Their occurrence in the chert veins can be attributed to precipitation of higher molecular weight organic compounds from migrating hydrothermal fluid owing to conductive cooling or mixing with cold seawater, similar to processes producing accumulations of petroleum in modern seafloor hydrothermal systems [21–23]. This deposition may have occurred contemporaneously with, and for the same reason as, the precipitation of the cherts from dissolved silica in the hydrothermal fluid, or may reflect the overprint of a later stage hydrothermal fluid.

While the carbonaceous matter in the Warrawoona rocks is sometimes described as kerogen, analysis of this material indicates it is composed primarily of disordered or amorphous graphite [30,35]. Disordered graphite with similar characteristics to that in the Warrawoona rocks can arise from abiogenic as well as biogenic sources [e.g., 63]. Although the current composition of

reduced carbon in the Warrawoona rocks is substantially different from the products of FTT synthesis (predominantly alkanes), thermal maturation during metamorphism may have converted the original organic compounds into their present form. The Warrawoona rocks have experienced greenschist facies metamorphism, indicating they have been heated to  $>300\text{ }^\circ\text{C}$  for extended periods of time (probably millions of years). During heating at high temperatures, higher molecular weight alkanes like those that would be trapped in hydrothermal systems will undergo cracking and reforming to polycyclic aromatic hydrocarbons (PAH) in addition to producing  $\text{CO}_2$  and methane. With continued heating, these PAH would condense into disordered carbon like that observed in Warrawoona.

The results of this study as well as other recent studies challenging the biological nature of reduced carbon compounds in Earth's oldest rocks may have substantial implications for interpreting the early rock record with respect to the origin and early evolution of life. Most scientists have come to accept that life on Earth originated prior to 3.8 Ga ago and that complex life capable of photosynthesis had evolved well before 3.5 Ga, based largely on the presence of  $^{13}\text{C}$ -depleted carbon compounds in the oldest known rocks. These dates follow closely on the late heavy meteorite bombardment (3.8–4.0 Ga) that may have sterilized the Earth's surface, implying either a very rapid origin for life or an ability to survive large meteorite impacts in subsurface or deep sea habitats [64]. However, if the isotopic composition of organic matter in the most ancient rocks can be explained by abiotic processes, the possibility must be entertained that complex life was not present on Earth, or at least not widespread, until a much later date, and the early evolution of life may have been a much more gradual process than is presently thought.

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