Gas chromatography–pyrolysis–isotope ratio mass spectrometry: a new method for investigating intramolecular isotopic variation in low molecular weight organic acids

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Abstract

A new GC–Pyrolysis–IRMS method was developed for the $\delta^{13}C$ determination of carboxyl carbon in low-molecular weight organic acids. By utilizing a palladium-wire reactor at 600 °C with a helium/hydrogen reactant gas, the carboxyl carbon of low-molecular weight organic acids is pyrolytically cleaved and introduced into an IRMS for stable carbon-isotope analysis. The precision of the GC–Py–IRMS method is similar to that of more conventional, combustion-based continuous-flow IRMS techniques and interpretation of isotope-dilution experiments with acetic and octanoic acid shows that the technique is sufficiently accurate for the determination of $\delta^{13}C$ values at natural abundance levels. As a demonstration of this new capability, the carboxyl carbon of low-molecular weight (LMW, C_2–C_6) organic acids generated via hydrous pyrolysis of an oil-prone source rock (the Ghareb Shale) shows $\delta^{13}C$ values consistent with the hypothesis that organic acids readily undergo exchange of their carboxyl carbon with aqueous inorganic carbon. © 2002 Elsevier Science Ltd. All rights reserved.

1. Introduction

Intramolecular carbon isotopic fractionation in organic molecules is a well-theorized, often-inferred and rarely-demonstrated phenomenon in organic geochemistry. Galimov (1973) proposed that thermodynamically ordered isotope distributions in organic compounds were a biological phenomenon. Several subsequent studies have shown that biosynthesis imparts an intramolecular carbon isotope variability within fatty and low-molecular weight organic acids (Vogler and Hayes, 1979; Meinschein et al., 1974; Monson and Hayes, 1980, 1982). More recently, it was proposed that petroleum-associated, low-molecular weight organic acids demonstrated intramolecular isotopic variability where the alkyl carbons reflected

the $\delta^{13}C$ of the source kerogen, while the carboxyl carbons reflected the $\delta^{13}C$ of bicarbonate in equilibrium with the organic acid (Dias, 2000; Franks et al., 2001). In an effort to better understand the chemistry of organic acids in natural systems and provide definitive evidence of intramolecular isotopic fractionation, we set out to develop a technique to determine the $\delta^{13}C$ of carboxyl carbon in C_2–C_8 organic acids in complex aqueous solutions.

The carbon-isotope determination of carboxyl-derived carbon from individual low-molecular weight (LMW) organic acids in a complex solution is not a trivial problem. Two primary chemical methods exist for the $\delta^{13}C$ determination of carboxyl-derived CO_2 for single pure compounds. First, the Schmidt reaction (Wolff, 1946) was used to obtain carboxyl-derived CO_2 from long chain fatty acids (Monson and Hayes, 1980, 1982; Vogler and Hayes, 1979). Second, carboxyl-derived CO_2 was generated by the pyrolysis of purified sodium acetate

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(Gelwicks and Hayes, 1990; Gelwicks et al., 1994, Mein-schein et al., 1974). However, neither of these methods allows for the determination of multiple, individual acids in a complex mixture, and neither is suited to take advantage of the conveniences offered by more modern GC-IRMS analytical techniques.

More recently, Corso and Brenna (1997) described a GC–Py–GC–IRMS system for the position-specific ana-lysis of $\delta^{13}C$ in organic molecules in complex mixtures. Due to the tandem-GC arrangement with a pyrolysis furnace in between the two GCs and a combustion fur-nace at the end of the second GC column, this method allowed for the carbon isotopic determination of pyrolytic fragments from organic compounds. The drawbacks to this arrangement are: (1) the need for a second GC system, and (2) the analysis is possible for only one compound at a time in the mixture of interest.

Our first considered strategy was to measure the $\delta^{13}C$ of the residual alkyl group as a primary amine instead of the carboxyl carbon. The $\delta^{13}C$ of the carboxyl carbon could then be calculated by mass balance from the pre-determined $\delta^{13}C$ of the total acid. From the Schmidt reaction, organic acids are converted to primary amines following loss of the carboxyl carbon (Wolff, 1946). Unfortunately, LMW primary amines are easily lost during concentration due to evaporation of solvent and are difficult to separate with standard chromatographic methods. Primary amines can be converted to alcohols (Austin, 1960; Whitmore and Langlois, 1932) which are less volatile and more chromatographically amenable, however the yields are typically low and isomers (sec-ondary alcohols) are produced from amines with 3 or more carbon atoms.

Maier et al. (1982) demonstrated the gas-phase de-carboxylation of organic acids to give alkane and resi-dual CO_2 by passing the organic acids over a heated palladium catalyst in a hydrogen stream. By using the resolving capability of gas chromatography, by modify-ing a combustion reactor with a palladium catalyst, and by using H_2 as a reactant gas, we propose that the $\delta^{13}C$ value of carboxyl-derived CO_2 from organic acids can be determined using existing continuous-flow stable iso-tope methodology (Ricci et al., 1994; Merritt et al., 1995). This work describes the gas chromatography/ pyrolysis/isotope ratio mass spectrometer (GC–Py–IRMS) system used to determine the $\delta^{13}C$ values of carboxyl-derived carbon from organic acids in complex aqueous solutions.

2. Experimental

2.1. GC–Py–IRMS

The isotopic composition of carboxyl-derived CO_2 from LMW organic acids was determined by continuous-flow, gas chromatography-pyrolysis-isotope ratio mon-itoring mass spectrometry (GC–Py–IRMS). A schematic of the GC–pyrolysis interface is shown in Fig. 1. This new method involves a few simple modifications to the standard combustion reactor and related reaction con-ditions used in more conventional combustion-based IRMS systems (Merritt et al., 1995). The modified reactor consists of four 0.1 mm palladium wires (rather than the copper–platinum or nickel–platinum combination typi-cal of combustion applications) contained within gas-tight ceramic tube (12″ long×1/16″ O.D.×0.7 mm I.D.). After the GC column and before the reactor, a small "trickle" of 100% hydrogen was added to the helium carrier gas stream as a reactant. The reactor was main-tained at 600 °C (rather than 1000 °C more typical of combustion systems). Organic acids passing through the new palladium/hydrogen reactor pyrolyze to produce CO_2 from the carboxyl carbon and residual gaseous alkane (e.g., methane, ethane, propane for acetic, propanoic and butanoic acids, respectively). As with combustion, the resulting CO_2 is carried to the mass spectrometer in the helium stream for subsequent $\delta^{13}C$ analysis (Ricci et al., 1994).

2.2. Isotope-dilution experiments

Proper standardization requires LMW organic acids with carboxyl carbon of known isotopic composition. The $\delta^{13}C$ value of the pyrolysis-derived CO_2 would normally be determined and compared with the known value in order to verify the accuracy and precision of the GC–Py–IRMS technique. Unfortunately, such stan-dards were not available. Instead, to test the system we employed isotope-dilution experiments. These experi-ments determine the linearity of the technique over a wide range of ^{13}C contents, provide a measure of precision, and can provide a good measure of accuracy by comparison of the measured $\delta^{13}C_{carboxyl}$ of the unknown acid with the value obtained by linear regression of isotope dilution results.

In order to bracket the range of organic acids of interest, two isotope dilution experiments were per-formed: one with acetic acid and one with octanoic acid. Briefly, 2 µl of ^{13}C-1-acetic acid (99%) was added to 500 µl of unlabeled acetic acid for an initial fractional abundance of 0.003984 [$\chi = \mu$ label/(µl label + µl non-label)]. Due to the systematic way in which ^{13}C-labeled solutions were transferred from one dilution to another and mixed, the residual volume of the 500 µl syringe needle became important. Through gravimetric deter-mination, this residual volume was determined to be 22.6 µl. So, 272.6 µl of the initial isotopic mixture was added to 250 µl of unlabeled acetic acid to give a second solution with a $\chi = 0.0020782$. Of this second solution, 272.6 µl was added to another 250 µl aliquot of unlabeled acetic acid for a third solution with a $\chi = 0.0010840$.
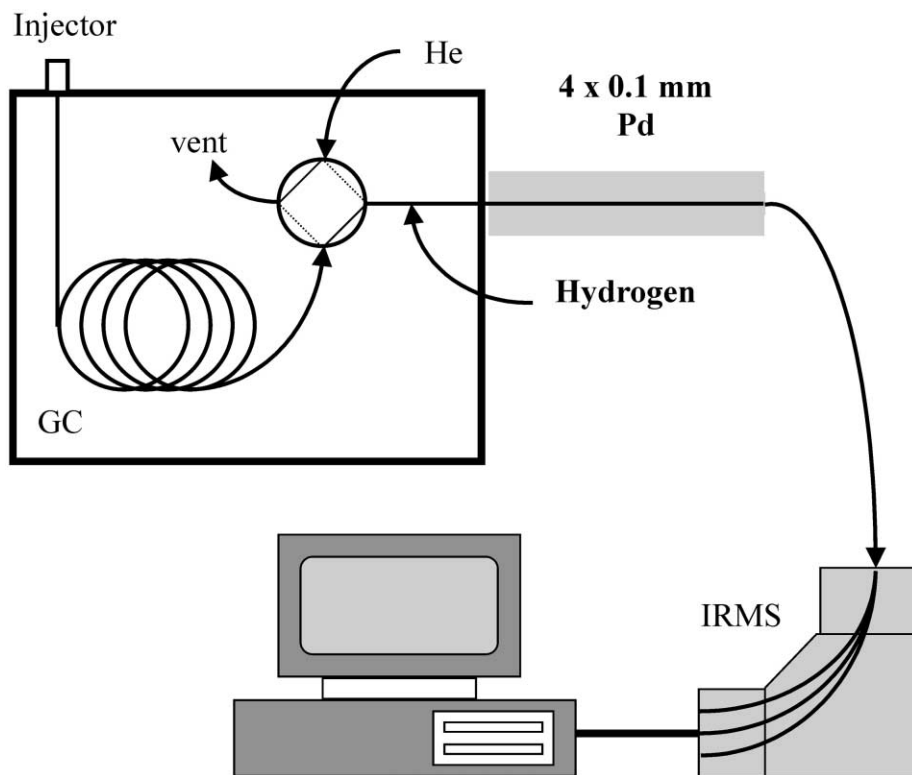
Fig. 1. Schematic of the GC–Pyrolysis–IRMS system. The basic differences from existing GC-combustion interfaces include the lack of the Nafion dryer, the addition of hydrogen to the carrier stream prior to the reactor and the 4 strands of 0.1 mm palladium wire used as a reactor catalyst.

Serial dilution was continued twice more giving two additional solutions with χ of 0.0005655 and 0.0002950. A similar set of solutions was prepared with ^{13}C-1-octanoic acid (99%) and unlabeled octanoic acid. A summary of the calculated χ is provided in Table 1. Each acid mixture was then diluted with distilled-deionized water to a final acid concentration of 0.2 mg/ml.

The ^{13}C content of the carboxyl group for each of the isotopically enriched solutions was determined in replicate ($n=4$) by direct injection. The isotope ratio mass spectrometer system used was a Finnigan-MAT 252. The GC conditions were as follows: FFAP (J&W) column, 30 m×0.53 mm×1 µm; 50 °C hold for 1 min, ramp to 240 °C at 10 °C/min, hold at 240 °C for 10 min; flow rate = 5 ml/min; injector temp = 240 °C; splitless mode for 0.5 min.

2.3. Hydrous pyrolysis generated acids—carboxyl $\delta^{13}C$ determination

As a demonstration of the usefulness of the GC–Py–IRMS technique, the $\delta^{13}C$ of carboxyl-derived carbon from C_2–C_6 organic acids generated from the hydrous pyrolysis of an oil-prone source rock was determined. Briefly, the Ghareb Shale (Dead Sea, Israel) was subjected

Table 1
Average measured ^{13}R and associated ^{13}F for the carboxyl-derived carbon from acetic and octanoic acid at different isotopic dilutions ($n=4$). χ is the fractional amount of ^{13}C-1-acid added per total acid in solution

χ	$^{13}R_{total}$	σ	$^{13}F_{total}$	σ
Acetic acid				
0.0000000	0.0111754	0.0000043	0.0110519	0.0000042
0.0002950	0.0114545	0.0000018	0.0113248	0.0000018
0.0005655	0.0117354	0.0000057	0.0115993	0.0000056
0.0010840	0.0122242	0.0000068	0.0120766	0.0000066
0.0020782	0.0131348	0.0000144	0.0129645	0.0000140
Octanoic acid				
0.0000000	0.0107855	0.0000105	0.0106704	0.0000103
0.0002950	0.0111107	0.0000100	0.0109886	0.0000098
0.0005655	0.0113866	0.0000081	0.0112584	0.0000079
0.0010840	0.0118679	0.0000083	0.0117287	0.0000081
0.0020782	0.0127419	0.0000269	0.0125816	0.0000262

to hydrous pyrolysis conditions at 350 °C for 72 h (Dias, 2000). LMW organic acids were extracted from the residual reactant water and introduced into the GC–Py–IRMS system via solid-phase micro-extraction (SPME,

Dias and Freeman, 1997). The chromatographic conditions were similar to those described above for the isotope-dilution experiments.

3. Results and discussion

3.1. General observations and operating conditions

The GC–pyrolysis interface shown in Fig. 1 has a few unique features that must be addressed. First, the Nafion water trap (a standard feature on GC-combustion systems) was disconnected on the new pyrolysis system. Since no extra water was being generated, none needed to be removed. Additionally, there was concern that the alkanes generated by the pyrolysis might accumulate on or react with the Nafion membrane. No special provisions were made to remove the alkanes produced by the pyrolysis, and were simply allowed to flow into the source with the carrier gas.

The amount of reactant hydrogen added as a "trickle" to the helium carrier stream was also a concern. If too much hydrogen were allowed to flow into the reactor (and therefore, into the source), the excess hydrogen would protonate the CO_2 in the source, producing HCO_2^+ ($m/z = 45$), increasing the 45/44 ratio and thus making the carboxyl-derived carbon dioxide appear isotopically enriched. Too little hydrogen, and the pyrolysis reaction would likely not go to completion. The hydrogen flow level was set by introducing CO_2 into the source via a reference gas injector and monitoring the 45/44 ratio. The hydrogen flow was increased until the 45/44 ratio changed, then the flow was reduced. This allowed for the maximum amount of hydrogen to be introduced into the pyrolysis reactor (and hence the source) without altering the measured 45/44 ratio.

Finally, the reactor temperature was selected to allow for the largest CO_2 signal possible from the pyrolysis of acetic acid. If the temperature was too high or too low, the pyrolysis reaction was less than optimal (as determined by low $m/z = 44$ response). The optimal operation temperature was determined to be 600 °C. Perhaps not coincidentally, this temperature is very close to the 550 °C pyrolysis temperature used by Corso and Brenna (1997) in their GC–pyrolysis–GC configuration. In addition, our reactor performance degraded over about 48 h of continuous use. By heating the reactor overnight (ca. 12 h) to 1000 °C, the efficiency of the reactor was restored. The mechanism of pyrolytic degradation was described by Maier et al. (1982) as a heterogeneous surface-catalyzed reaction. We can only hypothesize that the elevated temperature was required to periodically clean or otherwise reactivate the palladium surface.

This new continuous-flow stable-isotope method has distinct advantages over previous chemical methods of decarboxylation and instrumental methods of isotopic analysis. The hardware changes to existing GC–C–IRMS systems are minimal and the general mode of operation is similar to current GC-combustion methodologies for the $\delta^{13}C$ determination of individual compounds. GC–Py–IRMS allows for the $\delta^{13}C$ determination of carboxyl-derived carbon from a suite of LMW organic acids in complex aqueous solutions. No special purifications, separations, chemical transformations or other derivatizations are required in order to obtain samples suitable for isotopic analyses.

3.2. Isotope dilution experiments

The measured ^{13}R ($= {}^{13}C/{}^{12}C$) and ^{13}C fractional abundance [$^{13}F = {}^{13}C/({}^{13}C + {}^{12}C)$] of the carboxyl-derived carbon from the acetic and octanoic acid isotope-dilution experiments are shown in Table 1. The fractional abundance of the carboxyl position from each sample was calculated from ^{13}R after normalization to our internal reference standard relative to PDB ($^{13}R_{PDB} = 0.0111949$, Stichler, 1995) using

$$^{13}F = {}^{13}R/({}^{13}R + 1) \qquad (1)$$

The average measured fractional abundance ($^{13}F_{unlabel}$, $n = 9$) of the unlabeled acetic and octanoic acid carboxyl carbons were determined to be 0.0110519 ± 0.0000042 and 0.01067404 ± 0.0000103, respectively. In terms of delta notation, this equates to $\delta^{13}Cs$ of -1.74 ± 0.38‰ and -36.57 ± 0.92‰, respectively. The precision of the GC–Py–IRMS technique is similar to that of conventional, combustion-based irm–GCMS analysis, rendering the pyrolysis technique suitable for the determination of $\delta^{13}C$ values at natural isotopic abundance levels.

The accuracy of any isotopic method is normally determined using a reference standard of known, calibrated isotopic composition. Since such a standard was not available, the accuracy of the GC–Py–IRMS method was evaluated by comparison of the above measured fractional abundances to values obtained by linear regression of the fractional abundances measured for the isotopically enriched solutions. The total measured ^{13}F ($^{13}F_{total}$) for a given acid-derived carboxyl carbon is represented by mass balance such that

$$^{13}F_{total} = (\chi \cdot {}^{13}F_{label}) + (1 - \chi){}^{13}F_{unlabel} \qquad (2)$$

where χ is the amount of labeled organic acid added relative to the total organic acid pool and $^{13}F_{label}$ and $^{13}F_{unlabel}$ are the ^{13}C fractional abundances of the label and unlabeled acid carboxyl carbons, respectively. Eq. (2) can be rearranged to give:

$$^{13}F_{total} = {}^{13}F_{label} + \chi({}^{13}F_{label} - {}^{13}F_{unlabel}) \qquad (3)$$

A plot of $^{13}F_{total}$ versus χ is shown for both acetic and octanoic acid in Fig. 2. From Eq. 3, the slope of each plot should equal the difference between $^{13}F_{label}$ and $^{13}F_{unlabel}$ (approx. 0.98) and at the intercepts ($\chi = 0$), $^{13}F_{total}$ equals $^{13}F_{unlabel}$. So, under perfect conditions, the calculated $^{13}F_{total}$ at the intercept should equal the previously measured $^{13}F_{unlabel}$ of the unlabeled acids.

A summary of the linear regressions for the plots in Fig. 2 is presented in Table 2. Using all four ^{13}C-enriched data points for each acid, the r^2 values for the linear fit for both acetic and octanoic acid experiments are better than 0.999, however, the slopes of the 1st order regressions are 0.914 and 0.888, respectively. This is significantly lower than the expected approximate value of 0.98. The intercepts ($\chi = 0$) for both regressions give $^{13}F_{unlabel}$ values of 0.01107159 and 0.01074614 for acetic and octanoic carboxyl carbon, respectively. However, if the most enriched data point for each isotope dilution is excluded from the linear regressions, the slopes improve to 0.948 and 0.934 and the intercepts ($^{13}F_{unlabel}$) improve to 0.01105209 and 0.01071992 for acetic and octanoic acid carboxyl carbons, respectively.

From these data and as is evident by careful inspection of Fig. 2, the isotope dilution results are not perfectly linear. The non-linearity has two primary sources: (1) less than perfect technique used in preparing the isotopic solutions, and (2) non-linear response of the mass spectrometer system with respect to enriched materials. While gravimetric preparation of the isotopic solutions is usually preferred to volumetric preparations, it was decided that the volatility of the organic acids at room temperature might present a more significant problem to the accuracy of the solutions than the accuracy (or lack thereof) of using syringes to prepare the solutions. The syringes used to make the isotope dilutions have an error of ±1%. For a perfect series of isotope dilutions, the average error for the $\delta^{13}C$ value of the isotopic solutions would be about ±0.7‰. The non-linearity of the instrument response to enriched materials using a non-enriched standard is difficult to assess, however since the enrichments were on the order of approximately +170 ‰ relative to our reference CO_2 gas standard, non-linear response at the highest enrichment is likely.

Despite the less-than-perfect linearity described above, the measured $\delta^{13}C$ value of the unlabeled acetic acid carboxyl carbon (-1.74 ± 0.38‰) is in excellent agreement with that determined from linear regression (3 points) of the isotope dilution experiment (-1.72 ± 1.54 ‰). However, the difference between measured and calculated values for octanoic acid carboxyl carbon
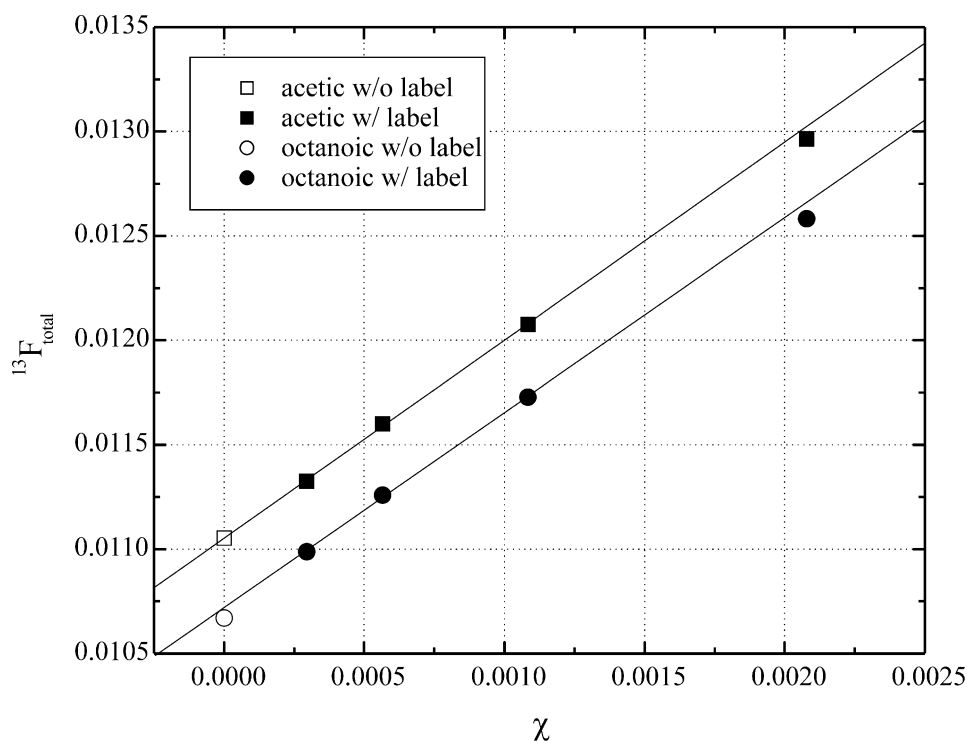

Fig. 2. Plot of the measured ^{13}F of carboxyl-derived carbon in the isotopic solutions versus the fractional amount of ^{13}C-1 label added (χ). The regression lines shown are calculated based on three of the four data points from measurement of the isotopic solutions (i.e. excluding the most enriched solution). At a $\chi = 0$, the calculated $^{13}F_{total}$ for the carboxyl-derived carbon based on the isotopic dilutions should equal the measured $^{13}F_{unlabel.}$.

Table 2

Measured ^{13}R and ^{13}F for the carboxyl-derived carbon of the unlabeled organic acids and the calculated ^{13}R and ^{13}F for the unlabeled acids ($\chi = 0$) based on 3 and 4 point regressions of the isotope dilution data shown in Table 1 and Fig. 2

	$^{13}F_{total}$	σ	$^{13}R_{total}$	σ	$\delta^{13}C$	σ
Acetic acid						
4pts	0.0110716	0.0000158	0.0111955	0.0000158	0.06	1.41
3pts	0.0110521	0.0000172	0.0111756	0.0000172	−1.72	1.54
Unlabeled	0.0110519	0.0000042	0.0111754	0.0000042	−1.74	0.38
Octanoic acid						
4pts	0.0107461	0.0000197	0.0108629	0.0000197	−29.66	1.76
3pts	0.0107199	0.0000165	0.0108361	0.0000165	−32.05	1.48
4pts[a]	0.0107004	0.0000136	0.0108161	0.0000136	−33.83	1.24
Unlabeled	0.0106704	0.0000103	0.0107855	0.0000103	−36.57	0.92

[a] These data are based on a second-order fit to all four enriched data points.

(-36.57 ± 0.92‰ and -32.05 ± 1.48‰, respectively) is less than ideal. It should be noted that if a simple 2nd order fit is used instead of a linear regression, the calculated value for octanoic acid carboxyl carbon improves to -33.83‰ (±1.24). Given the difficulties in preparing perfect isotopic solutions and the uncertainties associated with measuring enriched materials, we are confident in suggesting that the accuracy of the GC–Py–IRMS technique is sufficient for the natural-abundance $\delta^{13}C$ determination of low-molecular weight organic acid carboxyl carbon.

3.3. Intramolecular isotopic variability in organic acids: an example

Intramolecular isotopic fractionation in organic acids is known to arise from biological processes (Vogler and Hayes, 1979; Meinschein et al., 1974; Monson and Hayes, 1980, 1982), thermal decarboxylation (Kharaka et al., 1983) and as a function of thermodynamic equilibrium (Galimov, 1973). However, Dias (2000) and Franks et al. (2001) proposed that intramolecular isotopic variability in organic acids might arise from exchange between the carboxyl group and bicarbonate in formation waters, where the carbon-isotope composition of the total organic acid could be expressed by the following mass balance equation:

$$\delta^{13}C_{total} = \delta^{13}C_{alk}(n_{alk}/n_{total}) + \delta^{13}C_{carb}(1/n_{total}) \quad (4)$$

where n_{total} and n_{alk} are the numbers of carbon atoms in the total acid and the corresponding alkyl group, $\delta^{13}C_{total}$, $\delta^{13}C_{carb}$ and $\delta^{13}C_{alk}$ are the isotopic compositions of the total acid, the carboxyl-derived carbon and the alkyl-derived carbons. As a demonstration of the utility of the GC–Py–IRMS technique, the $\delta^{13}C$ values of carboxyl-derived carbons from LMW organic acids generated from the Ghareb Shale (carbonate) via

hydrous pyrolysis (350 °C/72 h) were determined and are shown in Table 3. Additionally, the $\delta^{13}C$ values of the total organic acids and those of the alkyl-derived carbons are also shown.

The $\delta^{13}C$ values of carboxyl-derived carbons are isotopically enriched relative to the $\delta^{13}C$ value of the total, parent acids by as much as 10.6‰. And, the calculated $\delta^{13}C$ values of the alkyl-derived carbons [from Eq. (4)] are isotopically depleted relative to the $\delta^{13}C$ values of the total acids by as much as 7.9‰.

Fig. 3 plots the $\delta^{13}C$ values of the total acid, carboxyl-derived carbon and alkyl-derived carbon as a function of molecular weight. The carbon-isotope composition of the carboxyl-derived carbon is intermediate of the $\delta^{13}C$ value of the calculated for aqueous carbon dioxide and bicarbonate (Dias, 2000). Additionally, the calculated $\delta^{13}C$ values for the alkyl-derived carbons are generally coincident with the measured $\delta^{13}C$ values of the associated, thermally immature kerogen. As suggested by Franks et al. (2001) and Dias (2000), the alkyl-derived carbons have $\delta^{13}C$ compositions consistent with those of the initial kerogens, while the carboxyl-derived carbons are isotopically similar to the dissolved inorganic carbon.

Table 3

Carbon isotope values for the total acid ($\delta^{13}C_{total}$), carboxyl carbon ($\delta^{13}C_{carb}$) and alkyl carbon ($\delta^{13}C_{alk}$) for C_2–C_6 organic acids generated via hydrous pyrolysis of the Ghareb Shale at 350 °C for 72 h

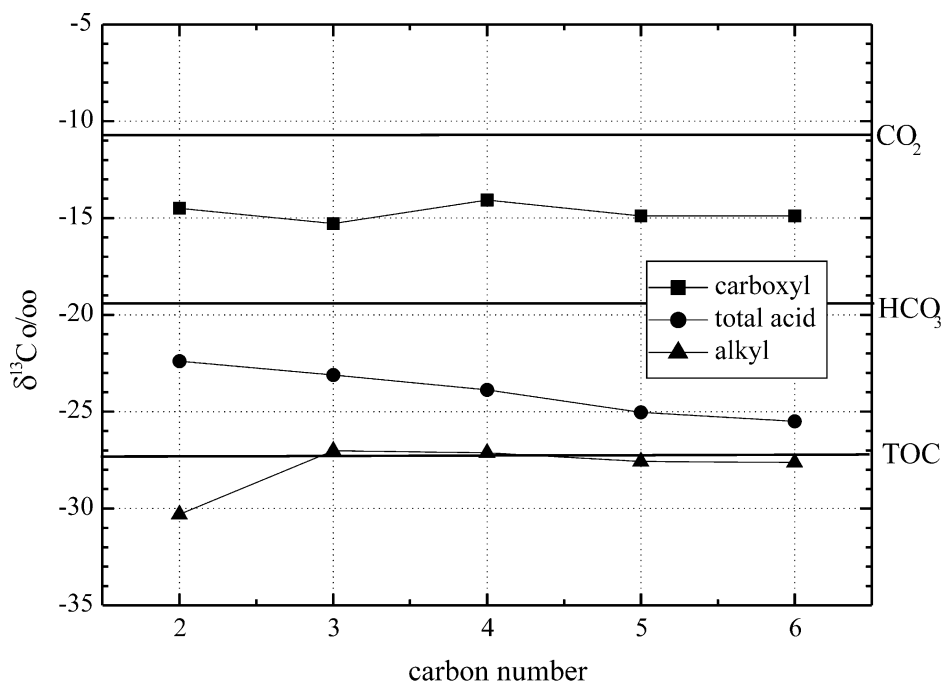| Organic acid	$\delta^{13}C_{total}$	$\delta^{13}C_{carb}$	$\delta^{13}C_{alk}$
Acetic	−22.4	−14.5	−30.3
Propanoic	−23.1	−15.3	−27.0
Butanoic	−23.9	−14.1	−27.1
Pentanoic	−25.0	−14.9	−27.6
Hexanoic	−25.5	−14.9	−27.6

Fig. 3. Measured carboxyl carbon, whole acid and calculated alkyl carbon $\delta^{13}C$ values for C_2–C_6 LMW organic acids generated from the Ghareb shale under hydrous pyrolysis conditions (350 °C/72 h). The line designated "TOC" is the $\delta^{13}C$ of the initial, unaltered kerogen, and the lines designated "CO_2" and "HCO_3^-" are estimations of the $\delta^{13}C$ of these aqueous species at hydrous pyrolysis conditions based on Mook et al. (1974) (Dias, 2000). The "CO_2" line appears more enriched than the "HCO_3^-" line due to an inversion of the fractionation factor at elevated temperatures.

4. Conclusions

The GC–Py–IRMS system described in this work provides stable carbon-isotope determinations of carboxyl-derived carbon from the pyrolysis of low-molecular weight organic acids in complex aqueous solutions for which no special purifications, separations, chemical transformations or other derivatizations are required. Using a reactor of palladium wire operating at 600 °C with a continuous flow of hydrogen the system pyrolyzes organic acids eluting from a GC column in a helium stream to alkane and carboxyl-derived CO_2 which is then analyzed by an IRMS for carbon-isotope composition. The precision of this analysis is similar to that of conventional irm–GCMS analysis that utilizes a combustion system for the oxidation of reduced carbon to CO_2. Based on isotope dilution experiments with acetic and octanoic acids we are confident that the accuracy and precision of the technique are suitable for the natural abundance $\delta^{13}C$ determination of carboxyl-derived carbon from low molecular weight organic acids.

Acknowledgements

We are grateful to Mike Lewan, John Hayes, Stefan Schouten and an anonymous individual for thoughtful, constructive reviews of this manuscript. This work was supported by the ACS Petroleum Research Fund, The Atlantic Richfield Corporation and the United States Geological Survey.

Associate Editor—**L. Schwark**

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