Observations and sources of carbon and nitrogen isotope ratio variation of pentaerythritol tetranitrate (PETN)

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A B S T R A C T

Isotope ratio analysis allows forensic investigators to discriminate materials that are chemically identical but differ in their isotope ratios. Here we focused on the discrimination of pentaerythritol tetranitrate (PETN), an explosive with military and civilian applications, using carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) isotope ratios. Our goal was to understand some of the factors influencing the isotope ratios of commercially manufactured PETN. PETN was isolated from bulk explosives using preparative HPLC, which reduced chemical and isotopic within-sample variability. We observed isotope ratio variation in a survey of 175 PETN samples from 22 manufacturing facilities, with $\delta^{13}C$ values ranging from −49.7‰ to −28.0‰ and $\delta^{15}N$ values ranging from −48.6‰ to +6.2‰. Both within-sample variability and variation of PETN within an explosive block were much smaller than between-sample variations. Isotopic ratios of PETN were shown to discriminate explosive blocks from the same manufacturer, whereas explosive component composition measurements by HPLC were not able to do so. Using samples collected from three industrial PETN manufacturers, we investigated the isotopic relationship between PETN and its reactants, pentaerythritol (PE) and nitric acid. Our observations showed that $\delta^{13}C$ values of PETN were indistinguishable from that of the reactant pentaerythritol. Isotopic separation between nitric acid and PETN was consistent within each sampled manufacturer but differed among manufacturers, and was likely dependent upon reaction conditions. These data indicate that $\delta^{13}C$ variation in PETN is dependent on $\delta^{13}C$ variation of PE supplies, while $\delta^{15}N$ variation in PETN is due to both nitric acid $\delta^{15}N$ and reaction conditions.

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1. Introduction

The explosive pentaerythritol tetranitrate (PETN, $C_5H_8N_4O_{12}$) is in widespread civilian and military use as a component of detonators, boosters, primers, and various plastic explosive formulations, such as Semtex. Its forms include cord ("det cord"), sheets, demolition blocks, and fillings for munitions, and has obvious potential for criminal use [1–3].

Characterizing recovered explosives is crucial to linking suspects and bombings or thwarted incidents. When chemical and physical characterizations are not sufficient to distinguish samples of a different origin, stable isotope ratios analysis can be used to show significant differences in isotopic composition [4–10]. The ability to differentiate and match explosive samples depends on how between-sample variation relates to within-sample

variability. As an estimate of between-sample variation, previous studies comparing explosive samples that contained a main constituent of PETN found that carbon isotope ratios ranged between −48‰ and −28‰ while the nitrogen isotope ratios ranged between −16‰ and −1‰ [11–13]. Benson et al. [12] reported within-sample variability (i.e., measurement standard deviations) of up to ±0.5‰ in both carbon and nitrogen isotope ratios of explosive samples containing PETN while Pierrini et al. [11] reported measurement standard deviations of up to 3‰ for both carbon and nitrogen isotope ratios in samples of Semtex consisting of RDX/PETN mixtures. Widory et al. [13] reported standard deviations of triplicate measurements of several explosive formulations up to 1.7‰ for carbon and 1.4‰ for nitrogen isotope ratios. However, none of these previous studies isolated PETN from its matrices or other explosive components prior to stable isotope ratio analysis. If the component of interest (PETN) was separated and purified from the bulk sample without introducing error by altering the isotope ratio, within-sample variability could be reduced, further enhancing the power of isotope ratio analysis for differentiating PETN-containing explosive samples.

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The wide range of PETN isotope ratios described by the literature suggests that the isotopic variation in the product may be related to variation in the synthesis and/or raw material between or within manufacturers. Laboratory-scale [14] and industrial-scale [15] synthesis studies on other explosive compounds—such as cyclotrimethylene nitramine (RDX), hexamethylene triperoxide diamine (HMTD) [16], and urea nitrate [17]—have shown that isotopic relationships between products and reactants can be used to infer the isotope ratio of the reactants for a product explosive of interest. We predict similar relationships in the reaction of pentaerythritol (PE, $C_5H_{10}O_4$) and nitric acid to form PETN.

We begin to examine fractionation patterns in the synthesis of PETN by describing the fundamental reaction:

$$C_5H_{12}O_4 + HNO_{3(excess)} \rightarrow C_5H_8N_4O_{12} + H_2O + HNO_{3(residual)} \quad (1)$$
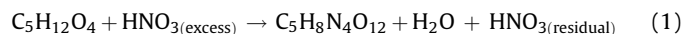
A general description of industrial synthesis is as follows: nitric acid at a concentration of 99–100% is mixed simultaneously with PE into 75–85% nitric acid solution and the nitration is allowed to proceed. During the synthesis reaction, the PE is completely consumed [18] and we predict there will be no difference between the carbon isotope ratio of the PE and PETN because all of the reactant carbon is transferred to the product. However, since a large portion of the nitric acid remains unused, we predict the PETN product will be relatively depleted in the heavy isotope of nitrogen due to isotope fractionation associated with nitration. Existing literature suggests that this is the case: most industrial nitrates have nitrogen isotope ratios between −2‰ and +2‰ [19,20], compared to ranges between −16‰ and −1‰ for PETN [11–13].

Here, we present a carbon and nitrogen isotope ratio survey of 175 PETN samples isolated from explosives produced by 22 different manufacturers. From one manufacturer, we present analyses of PETN isolated from several different blocks (packages) of explosives and subsamples within each block. These data were used to determine if one subsample from a block is sufficient to estimate the isotope ratios of entire block, and also if individual blocks from one manufacturer can be differentiated from other blocks produced by the manufacturer. We also present isotope ratios of products (PETN), waste products (residual nitric acid), and reactants (PE and concentrated nitric acid) collected from synthesis batches at three different manufacturers. Understanding the isotopic relationships between reactants and products in PETN synthesis allows us to explore the factors that cause the observed isotope ratio variation in PETN.

2. Materials and methods

2.1. Explosive sample collections

We obtained explosive samples (0.1–0.5 g) from 22 different manufacturers; some were domestic suppliers while others were foreign. A variety of formulations were sampled to obtain breadth in sampling. While some PETN samples were detonating ('det') cord, other samples were sheet explosive or packaged explosive blocks. Except for the three visited manufacturers, we do not know whether the other 19 manufacturers synthesized their own PETN or purchased it for loading into their particular explosive formulations. To assess variation within explosive blocks (a measure of homogeneity), we obtained seven subsamples (0.1–0.5 g) from each of six distinct packaged blocks representing three different formulations of plastic explosive from a single manufacturer. The seven subsamples were systematically collected from the top face, the four side faces, the bottom face, and the interior of each block. To assess the isotopic links between products and

reactants, we obtained newly manufactured PETN (0.5–1.0 g) following routine synthesis batches from three other manufacturers. During these visits, we also obtained samples (0.5–1.0 g) of the nitric acid and PE reactants used in each of the PETN syntheses as well as the residual nitric acid following completion of PETN synthesis. Several other PE reactants as well as the corresponding synthesized PETN products were collected during one of these visits from archives of one manufacturer, which spanned a 10-year production period.

2.2. Stock chemicals

HPLC-grade acetone, acetonitrile, dichloromethane, hexanes, methanol, and water were purchased from Fisher Scientific. Potassium hydroxide and sodium bicarbonate were purchased from VWR Scientific Products. Working standards for HPLC analysis were calibrated by standard explosive solutions purchased from AccuStandard, Inc. (New Haven, CT, USA), and were mixed in equal amounts of acetonitrile and methanol in concentrations sufficient to bracket the explosive samples.

2.3. Sample preparation

For explosive compounds contained within non-explosive matrices, we subsampled approximately 6 mg of bulk material. For det cord samples, the core was extracted carefully out of a 1 cm section cut open lengthwise. The actual weight was recorded to 1-µg precision using a Sartorius MC-5 microbalance (Sartorius AG, Göttingen, Germany). The subsamples were washed twice with hexanes (2 mL each) and air-dried. For explosives that contained no nitrocellulose, the hexane-insoluble fractions (containing the PETN) were extracted twice with acetone (2 mL each) and the extracts were filtered using a 13-mm diameter, 0.2-µm pore size GHP Acrodisc syringe filter (Pall Gelman Laboratory, Ann Arbor, MI, USA). For explosives that contained nitrocellulose, dichloromethane was used instead of acetone, as nitrocellulose is soluble in acetone and interferes with HPLC preparation. The acetone (or dichloromethane) soluble fractions (containing the PETN) were dried under an air stream.

Some samples collected during manufacturer visits were mixtures of crystallized PETN, water, and residual nitric acid. These were tested for acidity with pH paper and neutralized with saturated sodium bicarbonate solution. The crystallized PETN was separated from the neutralized aqueous phase by centrifugation, washed twice with deionized water, and dried under an air stream. The PETN was dissolved in acetone and extracted from the dried sodium bicarbonate and other acetone-insoluble contaminants by successive dissolution and centrifugation. After the PETN in solution was filtered and air-dried, it was analyzed for purity and explosive component composition using HPLC/UV. Preparative and analytical HPLC were conducted using a Waters HPLC system according to established methods [15]. We produced potassium nitrate from nitric acid samples within a week of sampling according to a previously developed procedure [15]. PE was not processed prior to stable isotope analysis.

2.4. Stable isotope analysis

HPLC-purified PETN, as well as reagent PE, prepared potassium nitrate, and PETN samples that were considered pure (>95%) by analytical HPLC were loaded in duplicate, permitting sufficient sample mass, into 3 mm × 5 mm tin capsules (Costech Analytical, Valencia, CA, USA) at a weight of 2.0 ± 0.2 mg. Carbon and nitrogen isotope ratio analysis was performed concurrently at the Stable Isotope Ratio Facility for Environmental Research (SIRFER) at the University of Utah on a Thermo Finnigan Delta S isotope ratio mass

spectrometer (Bremen, Germany) attached via ConFlo interface to an EA with an autosampler (Carlo Erba, Milan, Italy). EA reactors were packed according to the manufacturer's specifications with chromium oxide and silvered cobaltous/cobaltic oxide in the oxidation reactor and reduced copper wire in the reduction reactor. Magnesium perchlorate removed water upstream from a 3-meter ¼″ packed GC column used to separate N_2 and CO_2 gases.

Stable isotope ratios are expressed in "delta" (δ) notation in ‰ (per mil) relative to a standard, where $\delta = (R_A - R_{Std})/R_{Std}$ and R_A and R_{Std} are the ratios of the rare to abundant isotope (e.g., $^{13}C/^{12}C$ or $^{15}N/^{14}N$) in the sample and the standard, respectively. The international standard used for carbon is Vienna Pee Dee Belemnite (VPDB) and the standard for nitrogen is atmospheric N_2 (Air).

Samples were analyzed alongside an imidazole reference material, which was used to normalize sample $\delta^{13}C$ and $\delta^{15}N$ data to international isotope scales. We also included a pure PETN reference material to monitor instrument stability and estimate long-term analytical uncertainty (given below). We later adopted two-point normalization for $\delta^{13}C$ and $\delta^{15}N$ data using the glutamic acid reference materials USGS40 and USGS41 (available from nist.gov as SRM8573 and SRM8574) [21,22]. Measured isotope ratios for imidazole and previously analyzed samples were corrected using these reference materials.

Statistical analyses were completed using Prism 5.0c (GraphPad Software, Inc., San Diego, CA, USA). All tests were two-tailed and alpha values were set at 0.05. Comparisons between two groups of data or between a group and a theoretical mean were done by t-test; tests were paired when appropriate. Comparisons between more than two groups were performed by one-way ANOVA with Tukey's multiple comparison tests run post hoc. Unequal variances were assessed with Bartlett's test. Correlations were assessed using Pearson's method. Comparisons between slopes and intercepts of regression lines were completed by F test.

3. Results

3.1. The carbon and nitrogen isotope ratio ranges of PETN are large

A plot of $\delta^{13}C$ versus $\delta^{15}N$ values (Fig. 1) revealed large ranges in the surveyed PETN: between −49.7‰ and −28.0‰ for $\delta^{13}C$ and between −48.6‰ and +6.2‰ for $\delta^{15}N$. Long-term analytical uncertainty, estimated by the standard deviations of measurements of a PETN reference material over 22 months, was 0.29‰ for $\delta^{13}C$ ($n = 44$, range of −38.8‰ to −37.0‰) and 0.44‰ for $\delta^{15}N$ ($n = 38$, range of −15.1‰ to −13.2‰). Sampling was not even across the 22 different manufacturers nor formulations (such as det cord and plastic explosive), so we do not attempt to define the expected isotope ratio ranges for PETN associated with specific manufacturers or formulations.

3.2. Within-block carbon and nitrogen isotope ratio variations of PETN are small

We examined the isotope ratio variation within explosive packages ("blocks") by collecting seven subsamples from each of six different blocks from a single manufacturer. The blocks, which were of three different formulations each containing PETN, arrived from two different sources. The $\delta^{13}C$ and $\delta^{15}N$ means and standard deviations from individual blocks are shown in Table 1. To assess the isotopic heterogeneity of the blocks, we compared the variances within a block to those of a PETN reference material. Each block had either equal or less $\delta^{13}C$ variance (Bartlett's; $P < 0.05$) than that of the reference material. Observed $\delta^{15}N$ variances within each explosive block were each less than that of the PETN reference material (Bartlett's; $P < 0.05$). Thus, we
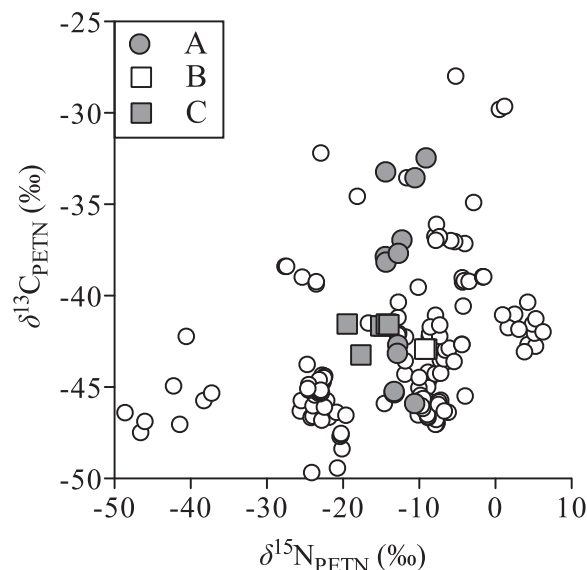

Fig. 1. Scatterplot of pairwise $\delta^{13}C$ and $\delta^{15}N$ values of 175 PETN samples isolated from various explosives, including PETN from visits to manufacturers. PETN from these manufacturers are labeled A–C in the legend. Samples were analyzed in duplicate, permitting sufficient sample mass; data presented are means of replicate analyses.

Table 1

$\delta^{15}N_{PETN}$ and $\delta^{13}C_{PETN}$ means and standard deviations of 7 subsamples taken from 6 individual manufactured explosive blocks. Also shown are the PETN w/w% relative to the bulk sample (by analytical HPLC). The blocks were of three different formulations produced by a single manufacturer and were shipped from two different sources. Values with different superscripts within an analysis were considered significantly different by Tukey's multiple comparison test.

Source	Explosive type	$\delta^{15}N_{PETN}$		$\delta^{13}C_{PETN}$		w/w% PETN	
		Mean	SD	Mean	SD	Mean	SD
1	1	−27.1[a]	0.17	−45.2[a]	0.26	59.7[a]	7.8
1	2	−24.5[b]	0.13	−44.8[a,b]	0.49	70.1[b]	2.0
1	2	−22.6[c]	0.13	−44.5[b]	0.15	71.7[b]	1.6
2	1	−7.4[d]	0.15	−45.9[c]	0.09	66.7[a,b]	11.6
2	2	−9.6[e]	0.15	−45.9[c]	0.19	71.7[b]	1.8
2	3[*]	−8.9[f]	0.07	−46.3[c]	0.28	44.3[c]	2.7

[*] This type of explosive contains RDX as a significant portion of the explosive fraction.

concluded that the variation of isotope ratio values of PETN within an explosive block was sufficiently small that a single subsample from an explosive block would be adequate for its isotopic characterization.

Although one manufacturer made all the blocks, the $\delta^{13}C$ and $\delta^{15}N$ values of PETN between the six blocks had statistically significant differences in $\delta^{13}C$ ($F = 44$, $n = 6$, $P < 0.0001$) and $\delta^{15}N$ (ANOVA, $F = 24,742$, $n = 6$, $P < 0.0001$). Post hoc tests (Table 1) suggested that even though each block could not be discriminated from every other on the basis of $\delta^{13}C$ alone, each block could be discriminated on the basis of $\delta^{15}N$ values alone.

Analytical HPLC observations of PETN content (Table 1) revealed relatively large within-block variation (SD up to 11%, compared to analytical uncertainty of less than 5%). Though quantitative composition analysis suggested differences between blocks (ANOVA, $F = 23$, $n = 6$, $P < 0.0001$), a post hoc test indicated that PETN content alone could not discriminate each of the six different blocks from every other.

To assess whether HPLC purification of PETN affected carbon and/or nitrogen isotope ratios, we compared isotope ratios of unpurified PETN collected directly during three manufacturer

visits to isotope ratios of the same PETN purified by HPLC. The isotope ratio differences were relatively small, though statistically significant (mean difference = 0.095‰ for $\delta^{13}C$, $t = 6.930$, $P < 0.0001$; mean difference = −0.22‰ for $\delta^{15}N$, $t = 8.774$, $P < 0.0001$). These differences were not meaningful for interpretation of data in this study, as they were consistent and within the long-term uncertainty for stable isotope measurements of PETN (i.e., 0.29‰ for $\delta^{13}C$ and 0.44‰ for $\delta^{15}N$). These differences may have been due to impurities in the acetone-soluble raw PETN that was obtained during the visits.

3.3. PETN is indistinguishable in $\delta^{13}C$ from its reactant PE

During the three manufacturer visits, we collected both synthesized PETN and its sole carbon-containing reactant, PE. In each case, the sampled PE was from the same batch of material that was used to synthesize the sampled PETN. The $\delta^{13}C$ values of PETN were positively correlated with the $\delta^{13}C$ values of PE (Fig. 2; Pearson $r = 0.999$, $n = 18$, $P < 0.0001$); and the data matched the 1:1 line; i.e., the slope and intercept for this regression ($\delta^{13}C_{PETN} = 1.008 \times \delta^{13}C_{PE} + 0.36$‰) were not significantly different from those calculated from a line of slope 1 and intercept of 0 ($F = 0.694$, df = 33, $P = 0.41$). We observed no significant difference in $\delta^{13}C$ values between paired PE and PETN ($t = 0.8469$, $n = 18$, $P = 0.41$). One manufacturer (A) provided archived PETN and PE pairs from previous production runs spanning a 10-year period. Variations in $\delta^{13}C_{PETN}$ and $\delta^{13}C_{PE}$ in this manufacturer were much greater than variations in the other two manufacturers (ranges ~12.9‰ versus <1.7‰).

3.4. Nitrogen isotopes are significantly and variably fractionated in PETN synthesis

From three manufacturers, we sampled nitric acid before synthesis ("initial") and after synthesis ("residual"). We also sampled the PETN produced. The $\delta^{15}N$ values of residual nitric acids were more positive than that of the initial nitric acids by 3.2–7.9‰, and $\delta^{15}N$ values of the PETN products were more negative

than that of the initial nitric acid reactants by 5.3–14.4‰ (Table 2). We calculate the isotopic separation (Δ_{PR}, [23]) as the difference between the product and reactant. For PETN, $\Delta_{PR} = \delta^{15}N_{PETN} - \delta^{15}N_{HNO_3(init.)}$. For residual nitric acid, $\Delta_{PR} = \delta^{15}N_{HNO_3(resid.)} - \delta^{15}N_{HNO_3(init.)}$. The isotopic separations were different between the manufacturers; the PETN Δ_{PR} values for Manufacturers A and B were 9‰ more positive than the mean PETN Δ_{PR} value for Manufacturer C, and the residual nitric acid Δ_{PR} values differed between all three manufacturers (Table 2). The small sample count—due to the difficulty in obtaining permission to visit manufacturers—precluded analysis of variance between these separation values. However, we consider these isotope ratio differences significant, as they were more than twice the standard deviations of multiple $\delta^{15}N$ analyses of PETN and residual nitric acid from Manufacturer C (0.45‰ and 0.33‰ respectively, $n = 4$ each).

4. Discussion

4.1. Distinguishing among PETN-based explosive samples is possible because of large between-sample variations in carbon and nitrogen isotope ratios

Fig. 1 shows that it is possible to differentiate samples among the 22 surveyed manufacturers of PETN on the basis of paired $\delta^{13}C$–$\delta^{15}N$ observations. The $\delta^{15}N$ range we observed (−48.6‰ to +6.2‰) is much larger than the range previously described in the literature (i.e., −16‰ to −1‰ [11–13]), suggesting the discriminatory power of $\delta^{15}N$ analysis of PETN may be greater than previously realized. We note that previous studies [11,12] focused on distinguishing among explosives on the basis of bulk stable isotope ratio analyses rather than isolated PETN. Our HPLC observations (Table 1) revealed relatively large within-block variability of PETN content, whereas $\delta^{15}N$ and $\delta^{13}C$ values of PETN within the blocks were homogenous. These results suggest that chemical characterization alone will not be as powerful as isotope ratio analysis for discriminating samples. Since within-sample variabilities of isolated PETN isotope ratios (<0.29‰ for $\delta^{13}C$ and <0.44‰ for $\delta^{15}N$) are much smaller than the overall ranges observed for PETN, we are confident stable isotope ratio analysis of isolated PETN will be useful to discriminate explosives samples in future forensic investigations.

4.2. Carbon isotope ratios of PETN are similar to that of the PE

Understanding the link between products and reactants is an important part of forensic investigations that can make use of information involving the synthesis of an explosive [14–17]. For the PETN synthesis reaction, $\delta^{13}C$ values of the reactants and $\delta^{13}C$ values of the products had a 1:1 relationship, as was expected since the reaction is known to go to completion with a high yield of PETN [18] and no other carbon containing products. These data (see Fig. 2) also show that an explosive manufacturer may acquire PE reactants that vary greatly in $\delta^{13}C$ values over time, directly affecting the $\delta^{13}C$ values of PETN produced at that manufacturer. We conclude that PETN $\delta^{13}C$ variations within a manufacturer are due to changes in the carbon isotope ratios of the PE, not changes in fractionation factors during PETN synthesis. In future sampling schemes that examine between-manufacturer and within-manufacturer variation of carbon isotope ratios in PETN, PE could be collected and analyzed as a proxy for PETN, greatly reducing the difficulty of shipping explosive samples. However, for clandestine manufacture, there exists the possibility for fractionation if the reaction does not go to completion. Future controlled laboratory syntheses may confirm this hypothesis.
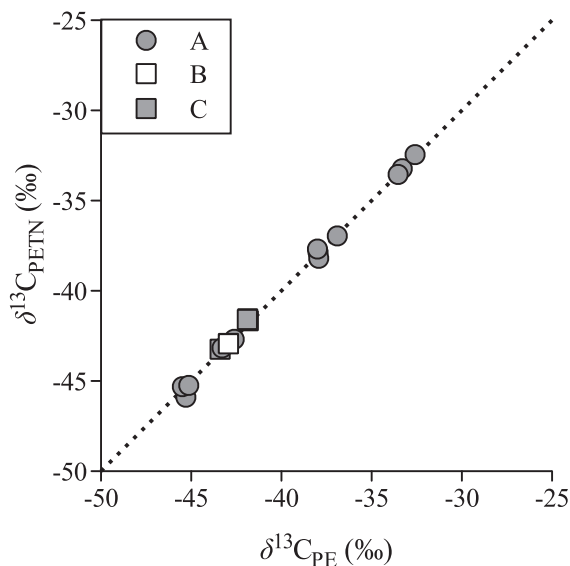

Fig. 2. $\delta^{13}C$ values of PETN as a function of $\delta^{13}C$ values of pentaerythritol (PE) reactant used to make the explosive. The data represent PETN/PE pairs from three manufacturers. The dotted line represents the theoretical 1:1 relationship between $\delta^{13}C$ values of PETN and $\delta^{13}C$ values of pentaerythritol. Samples were analyzed in duplicate, permitting sufficient sample mass; data presented are means of replicate analyses.

Table 2

Reaction conditions (expressed as weight percent nitric acid, HNO_3), isotope ratios of reactant (initial HNO_3) and products (residual HNO_3 and PETN) for PETN synthesis at three factories. Calculated fraction of nitric acid used, f, and isotopic separation between the product and reactant (Δ_{PR}) are shown in the rightmost columns.

Empirical data						Calculations		
Manuf.	w/w% init. HNO_3	w/w% resid. HNO_3	$\delta^{15}N$ init. HNO_3	$\delta^{15}N$ resid. HNO_3	$\delta^{15}N$ PETN	f	Δ_{PR} HNO_3 (resid)	Δ_{PR} PETN
A	99	75	−9.14	−5.97	−14.42	0.478	+3.17	−5.29
B	99	78	−7.41	−1.92	−12.75	0.521	+5.49	−5.34
C	99	83	−0.15	+7.73	−14.51	0.603	+7.88	−14.36

4.3. Nitrogen isotope ratios of PETN and residual nitric acid are related to the initial nitric acid by variable isotopic separations

The variation of PETN and residual nitric acid Δ_{PR} values among manufacturers was likely a result of varying reaction conditions. Though the reaction temperatures and mixing methods were similar, the stoichiometry of the reaction—i.e., the amount of excess nitric acid in the reaction—differed between manufacturers. We calculated the fraction of nitric acid remaining, f, as the molar ratio of the residual nitric acid ($x_{HNO_3(resid.)}$) over the molar ratio of the initial nitric acid ($x_{HNO_3(init.)}$). To transform the nitric acid concentrations (measured in mass fraction; i.e. weight of nitric acid over weight of solution) to mole fraction, we used the relation:

$$x_N = \frac{w \times M_W}{(1 - w) \times M_N + w \times M_W}$$

where x_N is the mole fraction of nitric acid in solution, w is the mass fraction of nitric acid in solution, M_W is the molecular mass of water, and M_N is the molecular mass of nitric acid.

When PETN Δ_{PR} values were plotted as a function of f, we observed a linear relationship between these two parameters ($\Delta_{PR} = -77.4f + 33.0‰$, $r^2 = 0.888$; Fig. 3).

The isotopic separations between the PETN and the initial nitric acid, and between the residual and initial nitric acids (Table 2), become smaller as increasing relative amounts of nitric acid are consumed in the synthesis. It appears that the isotopic

separation depends on the concentration of the remaining nitric acid, which is directly a result of reaction stoichiometry (amount of excess nitric acid). If we extrapolate the prediction to a greater fraction of remaining nitric acid (>0.9), isotopic separation between product PETN and reactant nitric acid becomes quite large. Larger isotopic separations may explain some of the extremely ^{15}N depleted PETN ($-50‰$ to $-40‰$) we observed in this survey (Fig. 1), relative to the typical range of synthetic nitrate ($-2‰$ to $+2‰$, [19,20]).

The reaction was considered a closed system as we measured the concentrations and $\delta^{15}N$ values of the nitric acid entering and leaving the reaction, as well as the $\delta^{15}N$ values of the PETN produced. We assumed there was no side product other than residual nitric acid. Thusly, the residual nitric acid isotope ratio may be calculated by the mass balance equation for the reaction (Eq. (3)).

$$\delta^{15}N_{HNO_3(init.)} = f \times \delta^{15}N_{HNO_3(resid.)} + (1 - f) \times \delta^{15}N_{PETN} \quad (3)$$

We can test the prediction for relating the isotopic separation (Δ_{PR}) between product PETN and reactant nitric acid by combining the mass balance equation with the regression equation from Fig. 3 and solving for the $\delta^{15}N$ values of the residual nitric acid (Eq. (4)).

$$\delta^{15}N_{HNO_3(resid.,predicted)} = 33.0\frac{f - 1}{f} - 77.4(f - 1)$$
$$+ \delta^{15}N_{HNO_3(init.)} \quad (4)$$
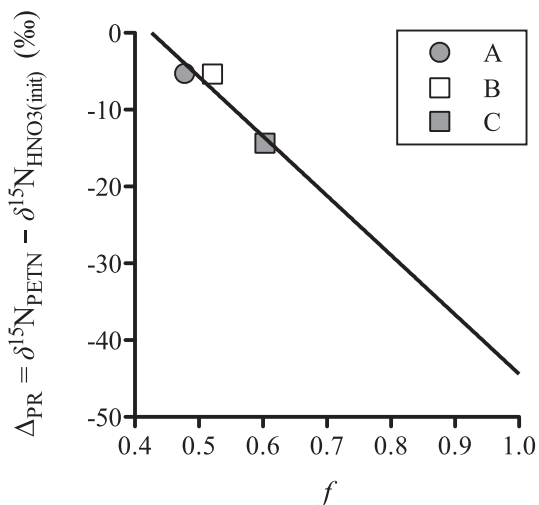
Fig. 3. PETN Δ_{PR} values calculated as $\delta^{15}N_{PETN} - \delta^{15}N_{HNO_3(init.)}$ as a function of f, the fraction of nitric acid used in the reaction. The line is the linear equation $\Delta_{PR} = -77.4f + 33.0‰$, calculated using the method of least squares ($r^2 = 0.888$). Points on the scatterplot represent two sets of PETN and nitric acid for Manufacturer A (two batches), one set for Manufacturer B, and four sets for Manufacturer C (four batches).
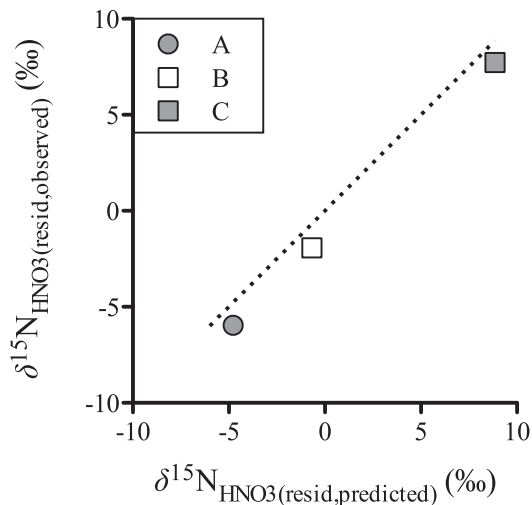

Fig. 4. $\delta^{15}N$ values for residual nitric acid (observed) as a function of predicted $\delta^{15}N$. The prediction is based on the linear regression calculated in Fig. 3 combined with the mass balance equation (Eq. (4)). The dotted line represents the theoretical 1:1 relationship between the observed and predicted $\delta^{15}N$ values. Points on the scatterplot represent residual nitric acid from two batches for Manufacturer A, one batch for Manufacturer B, and four batches for Manufacturer C.

A plot of the observed residual nitric acid δ^{15}N values as a function of the predicted δ^{15}N values is shown in Fig. 4. The slope of the line plotting $\delta^{15}\text{N}_{\text{HNO}_3(\text{resid.,observed})}$ as a function of $\delta^{15}\text{N}_{\text{HNO}_3(\text{resid.,predicted})}$ was not different from that of a 1:1 line ($F = 1.4717$, $P = 0.3489$) but the intercepts were different ($F = 1141.3$, $P < 0.0001$). The observed δ^{15}N values of residual nitric acid were consistently 1.1–1.3‰ more negative than the predicted $\delta^{15}\text{N}_{\text{HNO}_3(\text{resid.})}$ values. The model predicted the residual nitric acid isotope ratio fairly well, showing that mass balance was maintained as expected for a closed system. The prediction's slight overestimation of the ^{15}N enrichment of the residual nitric acid may be due to an unmeasured pool of ^{15}N enriched nitrogen in the system that was potentially incorporated into an unidentified side product.

The regression and mass balance equations for nitrogen isotopes can be used to predict several additional PETN synthesis parameters, given certain information. For example, the nitrogen isotope ratio of residual nitric acid can be calculated given the remaining fraction of residual to starting nitric acid (f) and the nitrogen isotope ratio of either the initial nitric acid or the PETN produced. The fraction of nitric acid remaining after a synthesis, an essential part of the nitration "recipe", may be calculated from any two of the three isotope ratios in the equations: $\delta^{15}\text{N}_{\text{HNO}_3(\text{init.})}$, $\delta^{15}\text{N}_{\text{HNO}_3(\text{resid.})}$, or $\delta^{15}\text{N}_{\text{PETN}}$. Since most known methods of producing PETN involve nitration in an excess of nitric acid, we predict the relationships observed in this study may also apply to clandestine and small-scale PETN synthesis, as well as synthesis on an industrial scale. If information on the synthesis is incomplete, a fully developed nitration fractionation model would still constrain the ranges of nitrogen isotope ratios expected in a PETN sample given the starting nitric acid. Future work should focus on controlled synthesis studies to further understand the effect of reaction conditions on the isotope ratios of synthesized PETN for prediction purposes. These varying conditions may include temperature, reaction rate, and independent variation in initial nitric acid concentration in addition to stoichiometry. Research of this kind is more feasible within laboratory settings than industrial sites.

Acknowledgements

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