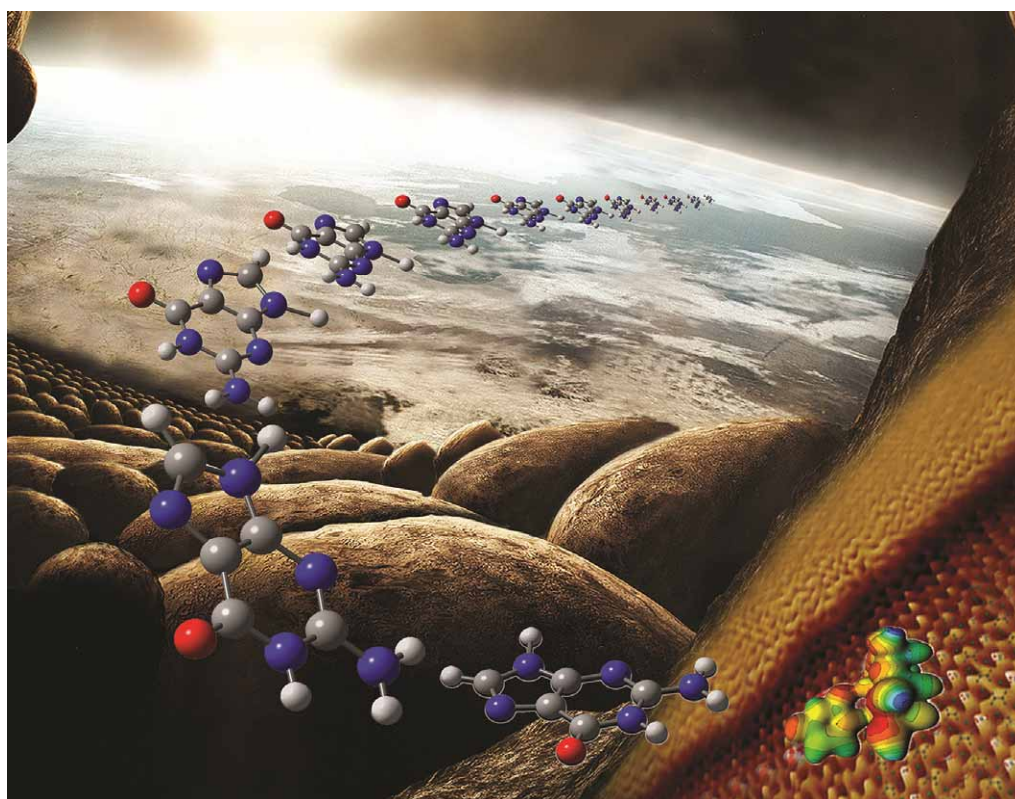


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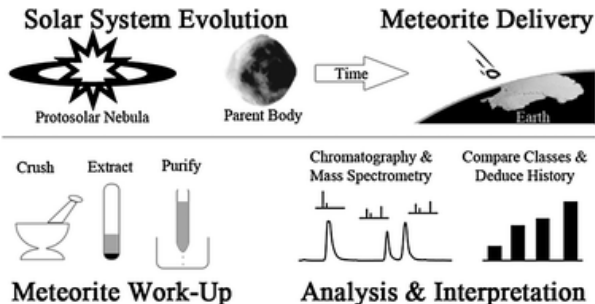
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A tutorial review of analytical methods for extraterrestrial samples and what these analyses can tell us about prebiotic chemistry.



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## TUTORIAL REVIEW

## Understanding prebiotic chemistry through the analysis of extraterrestrial amino acids and nucleobases in meteorites†

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The discoveries of amino acids of extraterrestrial origin in many meteorites over the last 50 years have revolutionized the Astrobiology field. A variety of non-terrestrial amino acids similar to those found in life on Earth have been detected in meteorites. A few amino acids have even been found with chiral excesses, suggesting that meteorites could have contributed to the origin of homochirality in life on Earth. In addition to amino acids, which have been productively studied for years, sugar-like molecules, activated phosphates, and nucleobases have also been determined to be indigenous to numerous meteorites. Because these molecules are essential for life as we know it, and meteorites have been delivering them to the Earth since accretion, it is plausible that the origin(s) of life on Earth were aided by extraterrestrially-synthesized molecules.

Understanding the origins of life on Earth guides our search for life elsewhere, helping to answer the question of whether biology is unique to Earth. This *tutorial review* focuses on meteoritic amino acids and nucleobases, exploring modern analytical methods and possible formation mechanisms. We will also discuss the unique window that meteorites provide into the chemistry that preceded life on Earth, a chemical record we do not have access to on Earth due to geologic recycling of rocks and the pervasiveness of biology across the planet. Finally, we will address the future of meteorite research, including asteroid sample return missions.

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### Introduction

Meteorites are the remnants of asteroids that have fallen to the surface of the Earth. These asteroids are formed in proto-solar nebulae, dense molecular clouds that are in the process



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of star formation. Bits of matter come together to form increasingly larger aggregates, progressing from molecules to dust grains, asteroids, planetesimals, planets and stars, in a process called accretion. A considerable amount of chemistry can occur both in the gas phase and on ice and mineral grain surfaces during the accretion phase, which lasts on the order of millions of years; many of the products of these reactions were thus incorporated into growing asteroids and planetesimals. As these objects grew in size, they experienced secondary (post-accretion) alteration events. Heating caused by impact shocks, radiogenic decay, or other sources occurred on many bodies, resulting in a spectrum of planetesimals ranging from primitive asteroids to planets that experienced significant melting, or differentiation, leading to formation of molten cores. This post-accretionary heating provided additional opportunities for chemical reactions to take place, both directly from thermal energy<sup>1</sup> (thermal metamorphism) and from liquid water (aqueous alteration) resulting from the melting of water ice.<sup>2</sup> Hence, the size and composition of asteroids and related objects provided a diverse suite of environments in which chemistry occurred.

Meteorites recovered on Earth are classified as either 'falls', objects that were observed to fall to the Earth, or 'finds', meteorites whose falls were not observed. As might be expected, most of the known meteorites are finds, rather than falls. Each year, the National Aeronautics and Space Administration (NASA), the Smithsonian Institution, and the National Science Foundation (NSF) send a joint expedition to Antarctica to recover meteorites (the ANtarctic Search for METeorites, or ANSMET). Antarctic meteorites are often less contaminated than meteorites recovered from more hospitable environments; in addition, the typically black meteorites tend to stand out in sharp contrast to the ice and snow of the Antarctic blue ice fields.

Earth-based analyses have led to the classification of at least 45 different types of meteorites that differ in elemental, isotopic, and mineralogical compositions.<sup>3</sup> The most primitive meteorites, carbonaceous chondrites, have been found to be the richest in soluble organic molecules. While many organic compounds of potential importance to the origins of life have been found in meteorites including sugar acids, hydroxy acids, aldehydes, ketones, and amines,<sup>4</sup> this review will focus on amino acids, the monomers of proteins, and nucleobases, the information-containing bases of DNA and RNA.

There are eight distinct groups of carbonaceous chondrites, five of which have been reported to contain extraterrestrial amino acids, and three of which have been determined to contain extraterrestrial nucleobases. The features that distinguish them from each other have been reviewed elsewhere,<sup>3</sup> but data relevant to the analysis of amino acids and nucleobases in meteorites are summarized in Table 1. The type and degree of alteration that took place on a meteorite is indicated on a continuum scale of 1–6, with 1 denoting strong aqueous alteration and 6 indicating strong thermal metamorphism. Generally, the thermal and/or aqueous alteration that asteroids experienced correlates with variations in the abundances and isomeric distributions of amino acids and nucleobases in meteorites, suggesting that secondary post-accretion alteration events significantly influenced asteroid chemistry. Because meteorites

**Table 1** A summary of total amino acid and nucleobase data for five of the eight carbonaceous chondrite groups. Amino acid and nucleobase analyses of CH, CB, and CK meteorites have not yet been reported

Meteorite classification	Example	Predominant alteration type	Amino acids (ppm) <sup>5,6</sup>	Nucleobases (ppb) <sup>7</sup>
CI1	Orgueil	Aqueous	~7	~100
CM2	Murchison	Aqueous	~15	~500
CR2	EET 92042	Aqueous	300	<25
CO3	ALHA 77307	Thermal	~1	nd
CV3	EET 96026	Thermal	~1	nd

likely delivered amino acids, nucleobases, and other biologically relevant molecules to the prebiotic Earth, these alteration effects may have indirectly played an important role in the nature of prebiotic compounds that were available for the origins of life on Earth.

### Meteoritic amino acids

Fifty years ago, Degens and Bajor<sup>10</sup> measured the sugar and amino acid contents of the Bruderheim and Murray meteorites by paper chromatography. Based on differences in abundances and distributions of these compounds between meteorites and terrestrial sediments, it was concluded that these compounds were likely indigenous to the meteorite and abiotic in origin. The proposed abiotic origin was supported by Miller's prior demonstration that amino acids and other organic molecules could be synthesized from mixtures of simple gases abundant in space.<sup>11</sup> In the decades since, improvements in both chromatographic separation techniques and detection methods have allowed the unambiguous identification of more than 80 amino acids ranging from two to nine carbons in carbonaceous meteorites (Table 2).

The observation of so many amino acids in meteorites, most of which are not used in biology, is a strong argument for their extraterrestrial origin. It is also important to analyze the organic composition of terrestrial samples from meteorite recovery sites, such as soil, sand or Antarctic ice, to compare with meteorite samples to rule out terrestrial contamination as the source of the meteoritic amino acids. Many of the non-protein amino acids found in meteorites recovered from Antarctica (listed in Table 2), are not present in the Antarctic ice above a level of 0.01 parts-per-trillion.<sup>6</sup> Most amino acids, including many found in meteorites, are chiral, meaning they possess two non-superimposable mirror image structures, or enantiomers, commonly referred to as left-handed (L) and right-handed (D). While abiotic processes typically produce racemic (50/50) mixtures of L- and D-amino acids, biology uses the L-enantiomer almost exclusively. Thus, another important indicator that amino acids are indigenous to the meteorite is that the chiral proteinogenic amino acids are present as racemic mixtures, similar to the enantiomeric ratios observed in the Miller spark discharge experiments (more detail on chirality is given in the Implications for the Origin of Life section). Finally, comparing the stable isotopic ratios (<sup>13</sup>C/<sup>12</sup>C, <sup>15</sup>N/<sup>14</sup>N and D/H; discussed in more detail below) of meteoritic amino acids to terrestrial amino acids can be a valuable tool for assigning an extraterrestrial origin.

**Table 2** Amino acids that have been detected in meteorites<sup>4,5,8,9</sup>

<b>C2</b> Glycine	<b>C6</b> Leucine
<b>C3</b> Alanine $\beta$ -Alanine Serine Sarcosine	Isoleucine <i>allo</i> -Isoleucine Norleucine Pseudoleucine Cycloleucine 2-Methylnorvaline Pipelic acid 2-Amino-2-ethylbutanoic acid 2-Amino-2,3-dimethylbutanoic acid 3-Amino-2-ethylbutanoic acid <sup>a</sup> 3-Amino-2,3-dimethylbutanoic acid <sup>a</sup> 3-Methylamine-pentanoic acid <sup>a</sup> 4-Aminohexanoic acid 4-Amino-3,3-dimethylbutanoic acid <sup>a</sup> 4-Amino-2-methylpentanoic acid <sup>a</sup> 4-Amino-3-methylpentanoic acid 4-Amino-4-methylpentanoic acid 6-Aminohexanoic acid $\alpha$ -Amino adipic acid $\beta$ -Amino adipic acid 2-Methylglutamic acid other isomers detected, but not identified
<b>C4</b> Threonine <i>allo</i> -Threonine $\alpha$ -Aminobutyric acid $\beta$ -Aminobutyric acid $\alpha$ -Aminoisobutyric acid $\beta$ -Aminoisobutyric acid <i>N</i> -Ethylglycine <i>N,N</i> -Dimethylglycine <i>N</i> -Methylalanine <i>N</i> -Methyl- $\beta$ -alanine Aspartic acid 2,3-Diaminobutanoic acid 2,4-Diaminobutanoic acid 3,3'-Diaminoisobutanoic acid	<b>C7</b> 2-Amino-2,3,3-trimethylbutanoic acid 2-Amino-2-ethyl-3-methylbutanoic acid 2-Amino-2-ethylpentanoic acid 2-Amino-3-ethylpentanoic acid 2-Amino-2,3-dimethylpentanoic acid 2-Amino-2,4-dimethylpentanoic acid 2-Amino-3,3-dimethylpentanoic acid 2-Amino-3,4-dimethylpentanoic acid 2-Amino-4,4-dimethylpentanoic acid <i>allo</i> -2-Amino-2,3-dimethylpentanoic acid <sup>a</sup> <i>allo</i> -2-Amino-3,4-dimethylpentanoic acid 2-Amino-2-methylhexanoic acid 2-Amino-3-methylhexanoic acid <i>allo</i> -2-Amino-3-methylhexanoic acid <sup>a</sup> 2-Amino-4-methylhexanoic acid <i>allo</i> -2-Amino-4-methylhexanoic acid <sup>a</sup> 2-Amino-5-methylhexanoic acid 2-Aminoheptanoic acid $\alpha$ -Aminopimelic acid 1-Aminocyclohexanecarboxylic acid other isomers detected, but not identified
<b>C5</b> Valine Norvaline Isovaline 3-Aminopentanoic acid 3-Amino-2-methylbutanoic acid <i>allo</i> -3-Amino-2-methylbutanoic acid 3-Amino-2,2-dimethylpropanoic acid 3-Amino-2,2-dimethylpropanoic acid 3-Amino-2-ethylpropanoic acid 4-Aminopentanoic acid 4-Amino-2-methylbutanoic acid 4-Amino-3-methylbutanoic acid 5-Aminopentanoic acid Glutamic acid 2-Methylaspartic acid 3-Methylaspartic acid <i>allo</i> -3-Methylaspartic acid <i>N</i> -Methylaspartic acid 4,4'-Diaminoisopentanoic acid	<b>C8</b> Isomers detected but not identified
	<b>C9</b> Phenylalanine Tyrosine other isomers detected, but not identified

<sup>a</sup> Denotes compounds that were only tentatively identified.

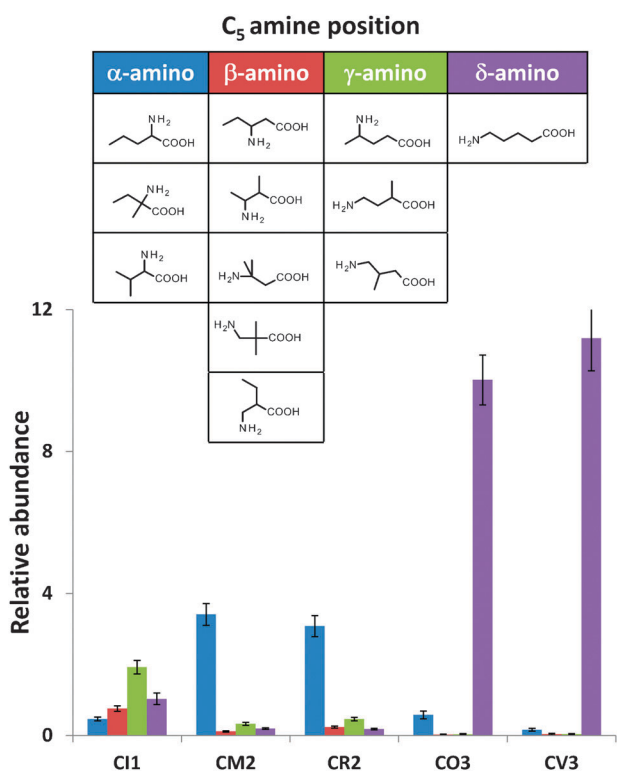
### Molecular diversity in meteoritic amino acids

The Murchison meteorite, which fell in 1969 in southeastern Australia, has been extensively studied for amino acids for several reasons, perhaps the two most important of which are that it has a complex distribution of amino acids and that a tremendous amount of meteorite material (~100 kg) was recovered. In addition to possessing many of the proteinogenic amino acids, Murchison also contains a number of non-proteinogenic amino acids not found in the soil collected from the Murchison meteorite fall site,<sup>12</sup> with  $\alpha$ -amino acids as the predominant isomers (Fig. 1). While the two most abundant amino acids in Murchison are glycine and  $\alpha$ -aminoisobutyric acid (AIB), every possible structural isomer of the three-, four-, and five-carbon primary,

acyclic aliphatic amino acids has been found with the abundances tending to decrease as the carbon number increases. Other structural groups, including *N*-alkyl amino acids, cyclic amino acids,  $\beta$ -amino alkenedioic acids, and diamino acids are also present.<sup>4</sup> It is likely that all isomers are present for the amino acids of higher molecular weight, but because the number of possible isomers grows quickly with each additional carbon (*e.g.*, eight possible enantiomers and isomers of four-carbon amino acids, 23 possible five-carbon amino acid enantiomers and isomers, >100 possible six-carbon amino acids, *etc.*), obtaining authentic standards and achieving chromatographic resolution for each molecule pose significant challenges.

The predominance of  $\alpha$ -amino acids observed in Murchison is not universal among meteorites. In particular, analysis of





**Fig. 1** Five-carbon amino acid distributions by amine position. Abundances have been normalized to the number of possible isomers for  $\alpha$ -,  $\beta$ -,  $\gamma$ - and  $\delta$ -amino acids. The meteorites are Orgueil (CI1), Murchison (CM2), EET 92042 (CR2), ALHA 77307 (CO3) and EET 96026 (CV3).<sup>5,6</sup>

the CI chondrites Ivuna and Orgueil revealed that  $\beta$ -alanine was the most abundant amino acid in CI chondrites.<sup>5</sup> Recent analyses of CV and CO thermally altered chondrites reveal a predominance of straight-chain, amino-terminal acids (*n*- $\omega$ -amino acids).<sup>6</sup> Comparisons of the isomeric distributions of amino acids in the various carbonaceous chondrite groups have yielded important insights into the dominant amino acid-forming chemistries in meteorite parent bodies. More specifically, the five-carbon amino acids have proven remarkably informative, as the number of isomers is small enough that adequate chromatographic separation can be achieved for all five-carbon amino acids, but the available “structure space” allows sufficient molecular diversity to observe differences between meteorites of the different groups (Fig. 1). So while the CM and CR carbonaceous chondrites contain predominantly  $\alpha$ -amino acids, the other three groups (CI, CV, and CO) have very different structural distributions, implying that different formation pathways are active in the various meteorite parent bodies. It has also been shown that the total amino acid abundances are the highest in the most primitive (least altered) CR2 and CR3 chondrites ( $\sim 80$  to 300 ppm) and are significantly lower ( $\sim 0.7$  to 7 ppm) in the more aqueously altered CI, CM, and CR type 1 chondrites<sup>5</sup> as well as in thermally altered CO and CV chondrites.<sup>6</sup>

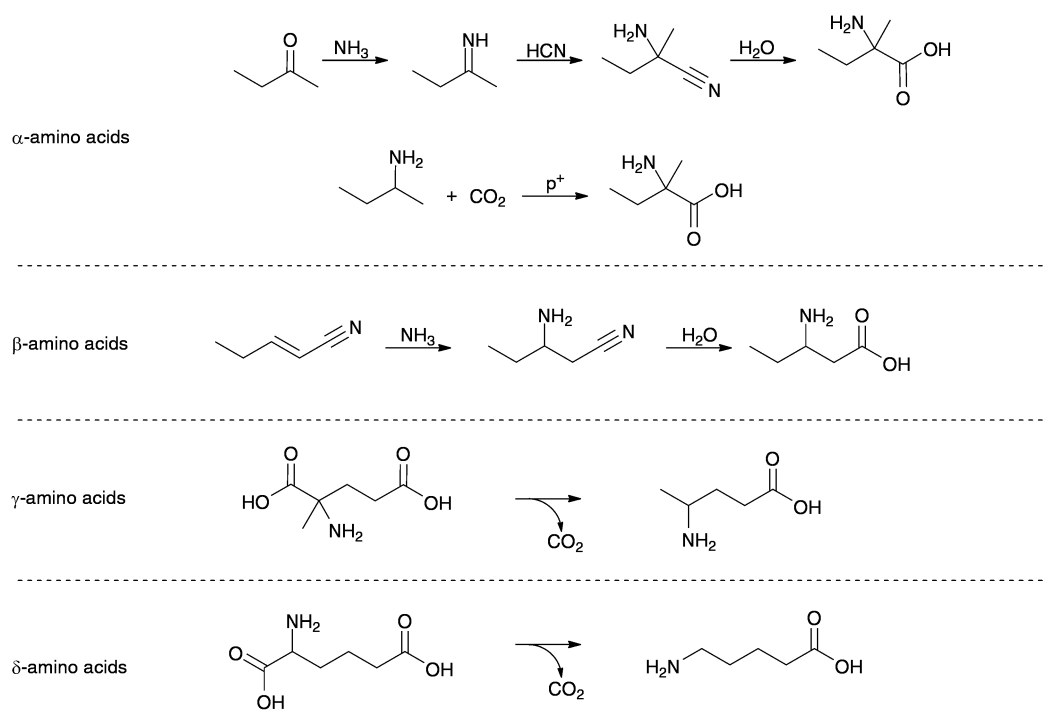
#### Formation pathways to amino acids in meteorites

A number of synthetic routes have been proposed for the amino acids in meteorites based on the observed relative

abundances of various isomers. It is important to note that the analytical work-up of meteorites for amino acids almost always involves aqueous extraction and, often, acid hydrolysis (described in greater detail below). The acid hydrolysis step liberates additional free amino acids, indicating that a significant fraction of these compounds are present in the meteorites as acid-labile precursors. By analyzing the abundances of amino acids in unhydrolyzed fractions of a meteorite sample, a more complete picture of the free amino acids is obtained; however, the extent to which precursor molecules are converted to amino acids during the aqueous extraction is unknown. Regardless of whether these compounds were present in the meteorite or were formed from precursors during aqueous extraction, free amino acids are released from meteorites by the action of water, and were thus likely available for the origins of life on Earth. Elucidation of the formation pathways for meteoritic amino acids has been hindered by a lack of knowledge of the extent to which these molecules exist as free amino acids within the meteorite parent body or are present in as yet unidentified precursor forms.

**$\alpha$ -amino acids.** The discovery of high proportions of  $\alpha$ -amino acids in Murchison correlated well with Miller’s Strecker-cyanohydrin synthesis work, which demonstrated that reactions of aldehydes and ketones with  $\text{NH}_3$  and HCN yielded  $\alpha$ -amino acids (Fig. 2). Because HCN is an abundant molecule in space and ammonia and various carbonyls have been detected in the Murchison meteorite, these reactions are generally accepted as responsible for both  $\alpha$ -amino and  $\alpha$ -hydroxy acids that have been found in Murchison and other aqueously altered meteorites, suggesting that amino acid Strecker synthesis occurred within the meteorite parent body.<sup>13</sup> Reductive amination of  $\alpha$ -keto acids that have been found in meteorites<sup>4</sup> has also been proposed as a route to  $\alpha$ -amino acids. It is also possible to form  $\alpha$ -amino acids outside meteorite parent bodies, such as on ice surfaces.<sup>14,15</sup> This topic is also reviewed in greater detail elsewhere in this issue, so we will only include one example: it has been demonstrated that carboxylation of amines with  $\text{CO}_2$  can occur under proton irradiation (Fig. 2).<sup>16</sup> Alternatively, several studies in the 1960s–1980s demonstrated the formation of amino acids by mineral surface-catalyzed reactions of  $\text{H}_2$ , CO, and  $\text{NH}_3$ .<sup>17</sup> These laboratory reactions, dubbed Fischer–Tropsch type (FTT) syntheses, were shown to produce many of the same  $\alpha$ -amino acids<sup>17</sup> observed in the Murchison meteorite.<sup>18</sup> However, a link between FTT reactions and meteoritic organics has yet to be firmly established,<sup>19</sup> and thus this possibility remains largely speculative.

**$\beta$ -amino acids.**  $\beta$ -amino acids cannot be directly produced by the Strecker-cyanohydrin pathway. Instead, they are believed to have formed by Michael addition of ammonia to  $\alpha$ ,  $\beta$ -unsaturated nitriles (Fig. 2).<sup>20</sup> Like the Strecker-cyanohydrin reactions, these Michael addition reactions also occur in liquid water. The relative proportions of  $\alpha$ - and  $\beta$ -amino acids were likely determined by the respective precursor abundances (aldehydes and ketones relative to unsaturated nitriles) within an asteroid, as well as parent body conditions and differences in reactivity with  $\text{NH}_3$  between the alkene and carbonyl precursors.



**Fig. 2** Formation pathways for amino acids in meteorites.  $\alpha$ -amino acids: The Strecker-cyanohydrin pathway is the generally accepted synthetic route to  $\alpha$ -amino acids inside aqueously altered meteorites. Laboratory simulations have shown that amines can also be carboxylated to  $\alpha$ -amino acids when subjected to radiation in  $\text{CO}_2$ -water ices. This pathway may lead to the formation of amino acids outside of meteorite parent bodies.  $\beta$ -amino acids: The likely synthetic route to  $\beta$ -amino acids in meteorites is the Michael addition of ammonia to  $\alpha$ ,  $\beta$ -unsaturated carboxylic acid precursors.  $\gamma$ - and  $\delta$ -amino acids: Decarboxylation of  $\alpha$ -amino dicarboxylic acids is a possible formation mechanism for  $\gamma$ - and  $\delta$ -amino acids observed in meteorites.

Michael addition on the CI parent body is a plausible explanation for the enhanced levels of  $\beta$ -alanine observed in Orgueil and Ivuna.<sup>5,21</sup>

**$\gamma$ - and  $\delta$ -amino acids.** The origins of  $\gamma$ - and  $\delta$ -amino acids in meteorites are less well understood than their  $\alpha$ - and  $\beta$ -counterparts. One possibility is that they are decarboxylation products of  $\alpha$ -amino dicarboxylic acids (Fig. 2). It has also been proposed that some  $\gamma$ - and  $\delta$ -amino acids found in Murchison could have been produced from the hydrolysis of lactams that have been identified in this meteorite.<sup>22</sup> The source of the lactams, however, has not been established.

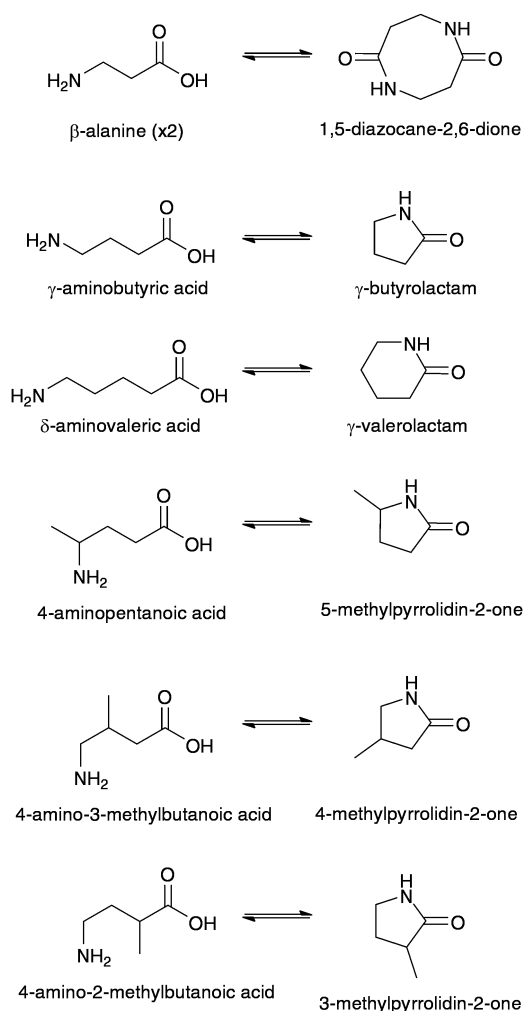
Alternatively, high-temperature/high-pressure (up to 400 °C at 250 atm) hydrothermal reactions of potassium cyanide, formaldehyde and ammonium hydrogen carbonate have been shown to produce straight-chain, amino-terminal acids ( $n$ - $\omega$ -amino acids).<sup>23</sup> Under these hydrothermal conditions, amino acids capable of forming stable lactams, such as  $\gamma$ -amino- $n$ -butyric acid,  $\delta$ -amino- $n$ -valeric acid, and  $\epsilon$ -aminocaproic acid were much more resistant to thermal degradation by decarboxylation (Fig. 3).<sup>24</sup> While the temperatures and pressures experienced within meteorite parent bodies are likely different than the experimental conditions employed in the hydrothermal reactions, the enhanced stability of lactams may explain the relatively high abundances of five-carbon  $\gamma$ - and  $\delta$ -amino acid isomers observed in CI, CO, and CV chondrites (Fig. 1). The observation that CI chondrites were heated much less than CO and CV chondrites may explain the different

amino acid distributions between the three groups.<sup>1,25</sup> While the lactam formed by  $\beta$ -alanine cyclization is likely to be sterically disfavoured as it makes a four-membered ring, it is possible that  $\beta$ -alanine could be protected from decarboxylation by dimerization to 1,5-diazocane-2,6-dione (Fig. 3).

In the laboratory hydrothermal reactions described above, the use of formaldehyde drives the reaction to straight-chain  $\omega$ -amino acids and a similar chemistry may predominate in CO and CV chondrites that also contain predominantly  $n$ - $\omega$ -amino acids; these molecules are then protected from thermal degradation through lactam formation. In CI chondrites, however, conditions favoured the formation of branched chain  $\omega$ -amino acids, which could still be protected from decarboxylation by lactamization (Fig. 3).

### Amino acid analysis

The amino acid suites of meteorites are generally very large. The Murchison meteorite, for example, contains over 100 different amino acids ranging from two to nine carbons, although only 80 or so have been unambiguously identified (Table 2).<sup>9</sup> Many of these amino acids are chiral, meaning they can exist as two stereoisomers. Achieving adequate separation of such complex mixtures to allow identification and quantification poses a significant analytical challenge. While the high molecular diversity of meteoritic amino acids effectively necessitates the use of mass spectrometry for detection, both gas chromatography and liquid chromatography have been used successfully to



**Fig. 3** Three- to five-carbon amino acids found in meteorites capable of forming stable lactams. Lactam formation has been shown to protect amino acids from decarboxylation under hydrothermal conditions. This may explain the distributions of amino acids observed in thermally altered CO and CV chondrites (predominantly *n*- $\omega$ -amino acids including  $\beta$ -alanine,  $\gamma$ -aminobutyric acid and  $\delta$ -aminovaleric acid) and in CI chondrites ( $\beta$ -alanine is the most abundant amino acid, and  $\gamma$ -amino acids are the most abundant five-carbon isomers).

identify and quantitate many amino acids. The respective techniques have inherent trade-offs in terms of sensitivity, chromatographic resolution, and compatible instrumentation, explaining the continued use of both methods today.

#### Amino acid extraction and purification

Amino acids and other water-soluble organics in meteorites are typically isolated by hot-water extraction followed by acid hydrolysis,<sup>26</sup> a procedure that was originally developed for amino acid analyses of lunar samples returned by Apollo<sup>27</sup> and subsequently used to extract extraterrestrial amino acids from the Murchison meteorite.<sup>18,28</sup> Although purification and analytical techniques have improved greatly over the past 40 years, the extraction techniques have remained basically unchanged. Meteorites are crushed to a powder using sterile mortars and pestles (*e.g.*, heated in air at 500 °C for 24 h). Water is added to 0.1–1 g of the meteorite powder in a 2 : 1 to

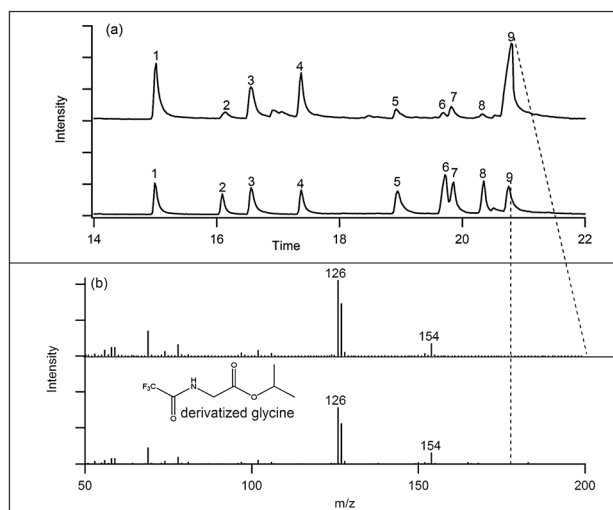
4 : 1 mass ratio and sealed in a glass ampoule for extraction at 100 °C for ~24 h. The resulting extract is decanted from the solid material, which is washed several times. The total extract is dried and half is subjected to acid vapor hydrolysis from ~6 N hydrochloric acid in sealed glass ampoules. The hydrolysis step converts precursor molecules such as amides, nitriles or lactams to free amino acids. The resulting hydrolyzed and unhydrolyzed fractions contain a variety of other water-soluble compounds in addition to amino acids, including carboxylic acids, hydroxy acids, and a variety of cations and anions. To prevent interference with derivatization reactions used for analysis, amino acids are typically purified by cation-exchange chromatography. This removes both non-cationic organic molecules and metal ions, and allows elution of the amino acids in a solution compatible with subsequent analytical techniques. Molecules such as carboxylic acids and hydroxy acids pass through the column and can be purified using anion exchange chromatography as necessary.

**Gas chromatography-mass spectrometry.** Gas chromatography-mass spectrometry (GC-MS) has been used for meteoritic amino acid analyses since the 1970s, and offers very good separation and identification capabilities.<sup>28</sup> Samples are derivatized to enhance amino acid volatility, generally by esterifying the acid group and adding a trifluoroacetyl group to the amine. Enantiomeric separation can be achieved either by creating diastereomers using a chiral alcohol (such as D-2-butanol) in the derivatization reaction, or, more typically, by utilizing a chromatography column that contains a chiral stationary phase (*e.g.*, Chirasil L-Val).<sup>29</sup> Mass spectral detection, typically with a quadrupole mass spectrometer, provides mass-fragmentation data that can be used to identify compounds. Comparison of chromatographic retention times and mass fragmentation with a standard is required for absolute identification of a particular amino acid and to assess contributions of potential interfering compounds. Mass fragmentation patterns and relative retention times can also be used, however, for tentative identification of individual compounds.<sup>30</sup> Fig. 4 shows representative GC-MS data from an acid hydrolyzed hot-water extract of the CR2 chondrite EET 92042. Chromatographic separation of most amino acid enantiomers is achieved, and compound identification is confirmed by comparison of retention times and mass spectral fragmentation patterns with those of known standards.

**Gas chromatography-isotope ratio mass spectrometry.** Compound-specific stable isotopic analysis of individual amino acids can be achieved through gas chromatography coupled with isotope ratio mass spectrometry (GC-IRMS). In this technique, GC is used to separate the compounds in a complex mixture, such as a meteorite extract, as in GC-MS. As each compound elutes, it is directed through reactors for combustion and reduction (for carbon or nitrogen analysis) or pyrolysis (for hydrogen or oxygen analysis). The resulting CO<sub>2</sub>, N<sub>2</sub>, H<sub>2</sub>, or CO gas is then analyzed in a magnetic sector mass spectrometer, and the stable isotopic ratios are calculated.

GC-IRMS was first applied to the analysis of amino acids in carbonaceous chondrites in the 1990s.<sup>31,32</sup> These compound-specific stable isotopic measurements are one important way





**Fig. 4** (a) GC-MS total ion chromatogram from the acid-hydrolyzed, hot-water extract of the CR2 chondrite EET 92042 (top trace) and an amino acid standard (bottom trace). Peaks are identified as: (1)  $\alpha$ -aminoisobutyric acid, (2) D,L-isovaline, (3) D-alanine, (4) L-alanine, (5) D- $\alpha$ -amino-*n*-butyric acid, (6) D-valine, (7) L- $\alpha$ -amino-*n*-butyric acid, (8) L-valine, (9) glycine. (b) Mass spectral data for peak 9 (glycine) in both EET 92042 (top) and the standard (bottom). The meteoritic compound is thus identified as glycine derivatized with trifluoroacetic acid and isopropanol based on the matching mass spectra and chromatographic retention times.

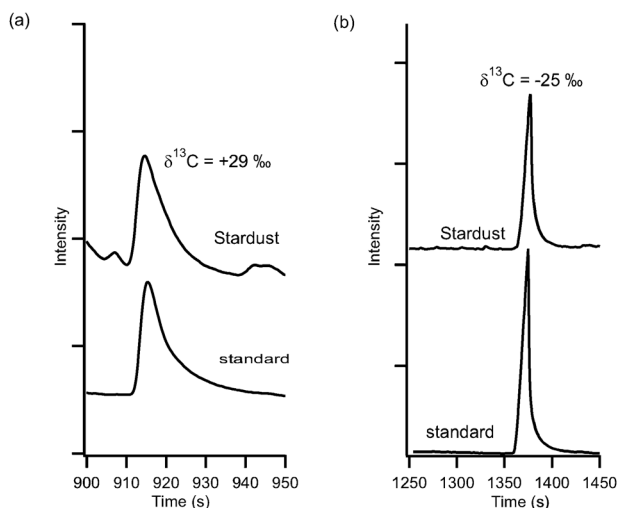
of determining the origin and indigenous nature of meteoritic amino acids. Extraterrestrial amino acids typically are enriched in heavy isotopes relative to terrestrial compounds, reflecting the cold environments in which these compounds or their precursors formed. The range of carbon, nitrogen, and hydrogen isotopic ratios (expressed in  $\delta$  values, indicating the difference in per mil, ‰, between the  $^{13}\text{C}/^{12}\text{C}$ ,  $^{15}\text{N}/^{14}\text{N}$ , or D/H ratio in the sample and the same ratio in a defined terrestrial standard) of individual amino acids in the CM2 Murchison and CR2 GRA 95229 carbonaceous chondrites are shown in Table 3; the terrestrial isotopic ranges for carbon and nitrogen in individual amino acids and for organic hydrogen are shown for comparison. The reference values are: Vienna PeeDee Belemnite (C; VPDB), air (N; AIR) and Vienna standard mean ocean water (D/H; VSMOW).

Compound-specific amino acid measurements have been used to support extraterrestrial origins of both meteoritic amino acids<sup>31</sup> and of glycine detected in comet-exposed material returned by NASA's Stardust mission (Fig. 5),<sup>40</sup> as well as to argue for indigenous excesses of L-enantiomers of certain meteoritic amino acids.<sup>32</sup> The possibility of co-eluting compounds

**Table 3** Compound-specific isotopic ratios for amino acids

Isotope ratio	Terrestrial range <sup>33–35</sup>	Murchison (CM2) <sup>30,32,36,37</sup>	GRA 95229 (CR2) <sup>8,38,39</sup>
$\delta^{13}\text{C}$ (‰ VPDB)	–70 to +11	–6 to +52	–18 to +51
$\delta^{15}\text{N}$ (‰ AIR)	–20 to +30	+37 to +184	+77 to +139
$\delta\text{D}$ (‰ VSMOW)	–270 to +66 <sup>a</sup>	+180 to +3419	+868 to +7245

<sup>a</sup> Deuterium terrestrial range represents organic hydrogen in a range of materials, not just amino acids.



**Fig. 5** GC-IRMS chromatograms from the analysis of comet-exposed material, showing the  $^{12}\text{CO}_2$  peak and carbon isotopic value resulting from the combustion of the GC peak identified as (a) glycine and (b)  $\epsilon$ -amino-*n*-caproic acid, a monomer of Nylon-6. The glycine peak is enriched in  $^{13}\text{C}$ , indicating an extraterrestrial origin, while the lower  $\delta^{13}\text{C}$  value for EACA reflects terrestrial contamination.<sup>40</sup>

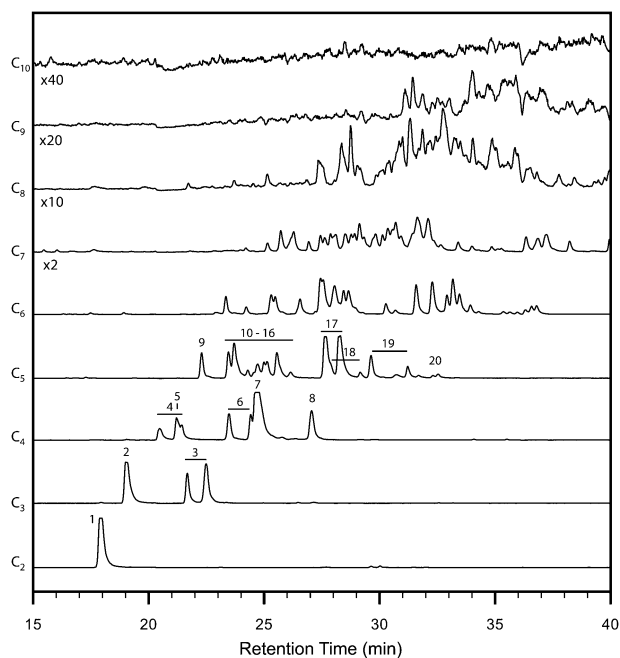
must always be carefully examined, however, to rule out interferences that could affect both isotopic measurements and enantiomeric ratios;<sup>41</sup> the coupling of GC-IRMS instrument suites with quadrupole mass spectrometry to allow simultaneous compound identification by mass fragmentation and isotopic analysis of individual compounds significantly reduces this concern.<sup>40</sup>

Isotopic ratios also suggest links to potential formation mechanisms. Analyses of the carbon isotopic ratios in individual amino acids in the Murchison meteorite revealed correlations between amino acid structure, chain lengths, and isotopic compositions that supported a formation of  $\alpha$ -amino acids through Strecker-cyanohydrin synthesis.<sup>30</sup> Deuterium measurements of these compounds suggested a strong link between  $\alpha$ -methyl,  $\alpha$ -amino acids and cold, pre-accretionary formation environments.<sup>37</sup> The amino acids in CR2 chondrites are more highly enriched in deuterium than those in CM2 chondrites, pointing towards different formation and parent-body chemistry for these meteorite classes.<sup>39</sup>

**Liquid chromatography-mass spectrometry (LC-MS).** GC-MS analysis of meteoritic amino acids has been successfully used for decades. An inherent limitation of this technique, however, is the relatively low sensitivity of GC-MS, with limits of detection on the order of a few pmol. Because meteorites are sample-limited commodities, more sensitive techniques were needed to analyze smaller and less amino acid-rich meteorites such as the aqueously altered Type 1 carbonaceous chondrites. This led to the development of methods for meteoritic amino acid analysis using high-performance liquid chromatography with fluorescence detection (HPLC-FD). The use of HPLC-FD enhances sensitivity by up to  $\sim 1000$ -fold over GC-MS; however, individual amino acids and enantiomeric ratios could only be determined by fluorescence retention times.<sup>42</sup> Co-eluting compounds could interfere with the identification

and quantitation of amino acids in these samples. These limitations were overcome by connecting a time-of-flight or other mass spectrometer in series with the fluorescence detector.<sup>26</sup> This instrument arrangement enables the simultaneous detection of amino acids by both mass and fluorescence, permitting both measurements to be made from the same sample, as well as alleviating complications due to co-eluting compounds provided they are of different masses. Because the sensitivity of the electrospray ionization technique commonly used for liquid chromatography-mass spectrometry is improved with lower flow rates, the use of ultra high performance- and nano-liquid chromatography has reduced the limits of detection of amino acids to fmol levels or lower,<sup>26</sup> permitting the analysis of sub-mg meteorite samples.

In principle, it should be possible to achieve enantiomeric resolution of amino acid enantiomers using a chiral stationary phase. However, satisfactory separation of the complex amino acid mixtures observed in meteorites (see Table 2) has yet to be demonstrated using a chiral liquid chromatography column. Instead, HPLC-based analyses typically achieve enantiomer separation by diastereomer formation using chiral derivatization reagents. For example, *o*-phthalaldehyde and *N*-acetyl-L-cysteine (OPA/NAC) react with primary amines to form fluorescent, diastereomeric amino acid derivatives that are readily resolved using achiral stationary phases (Fig. 6).<sup>43</sup> This technique has been



**Fig. 6** LC-ToF-MS single-ion chromatograms of  $\alpha$ -aminoalkylamino acids in an extract of the Murchison meteorite. Numbered compounds are: (1) glycine; (2)  $\beta$ -alanine; (3) D- and L-alanine; (4)  $\gamma$ -aminobutyric acid; (5) D- and L- $\beta$ -aminoisobutyric acid; (6) D- and L- $\beta$ -amino-*n*-butyric acid; (7)  $\alpha$ -aminoisobutyric acid; (8) D- and L- $\alpha$ -amino-*n*-butyric acid; (9) 3-amino-2,2-dimethylpropanoic acid; (10) D- and L-4-aminopentanoic acid; (11) D- and L-4-amino-3-methylbutanoic acid; (12) D-, L- and *allo*-3-amino-2-methylbutanoic acid; (13) D- and L-3-amino-2-ethylpropanoic acid; (14) 5-aminopentanoic acid; (15) D- and L-4-amino-2-methylbutanoic acid; (16) 3-amino-3-methylbutanoic acid; (17) D- and L-isovaline; (18) D- and L-3-aminopentanoic acid; (19) D- and L-valine; (20) D- and L-norvaline.

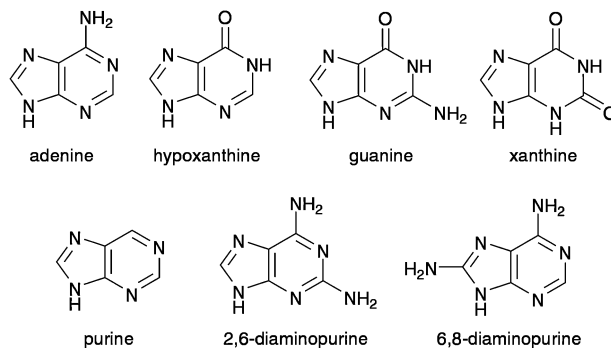
employed to achieve adequate separation of all isomers of primary amino acids up to five carbons in length.

### Nucleobases in meteorites

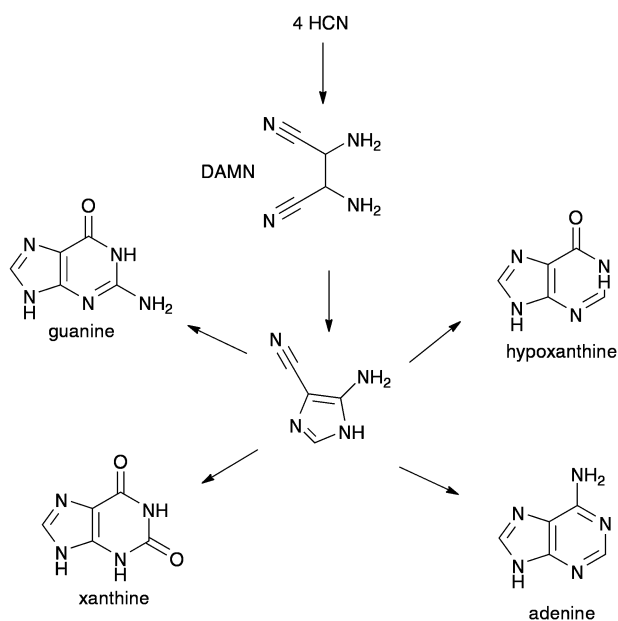
Nucleobases are subunits of nucleotides, the monomers of RNA and DNA, without the sugar and phosphate groups. More specifically, nucleobases are substituted one-ring (pyrimidine) or two-ring (purine) nitrogen heterocyclic compounds that contain the information in RNA and DNA by the arrangement of their hydrogen-bonding groups. In contrast to amino acids, nucleobases have been far less studied in meteorites, in part because of analytical difficulties associated with their lower relative abundances (Table 1). The purines adenine, guanine, hypoxanthine, and xanthine and the pyrimidine uracil have been reported in the Murchison meteorite, as well in as the Murray and Orgueil meteorites.<sup>44,45</sup> These compounds are all commonly found in biology, and determining their origin as terrestrial or extraterrestrial poses a significant challenge. Previous attempts to establish an extraterrestrial origin were complicated by experimental artifacts, low abundances of nucleobases, and the possibility of contamination during sample preparation. In addition, efforts to obtain isotopic data to rule out terrestrial contamination have been complicated by the low abundances of nucleobases in meteorites. Martins and co-workers<sup>45</sup> reported compound-specific carbon isotope values for uracil ( $\delta^{13}\text{C} = +44.5\%$ ) and xanthine ( $\delta^{13}\text{C} = +37.7\%$ ) extracted from the Murchison meteorite.<sup>45</sup> However, the presence of co-eluting  $^{13}\text{C}$ -enriched (extraterrestrial) carboxylic acids in Murchison meteorite could have contributed to the  $\delta^{13}\text{C}$  values measured for uracil and xanthine.<sup>7</sup> Recently, it has been reported that the Murchison meteorite and several other CM chondrites from Antarctica contain a different distribution of nucleobases than are found in biology, including three unusual nucleobase analogs (purine, 2,6-diaminopurine, and 6,8-diaminopurine) that are terrestrially rare or not found on the Earth (Fig. 7).<sup>7</sup> These findings strongly support an extraterrestrial origin for the nucleobases and nucleobase analogs identified in these meteorites.

### Formation mechanisms for nucleobases in meteorites

Several formation mechanisms for meteoritic nucleobases have been proposed. One suggested mechanism for the formation of extraterrestrial purines is by the reaction of hydrogen cyanide



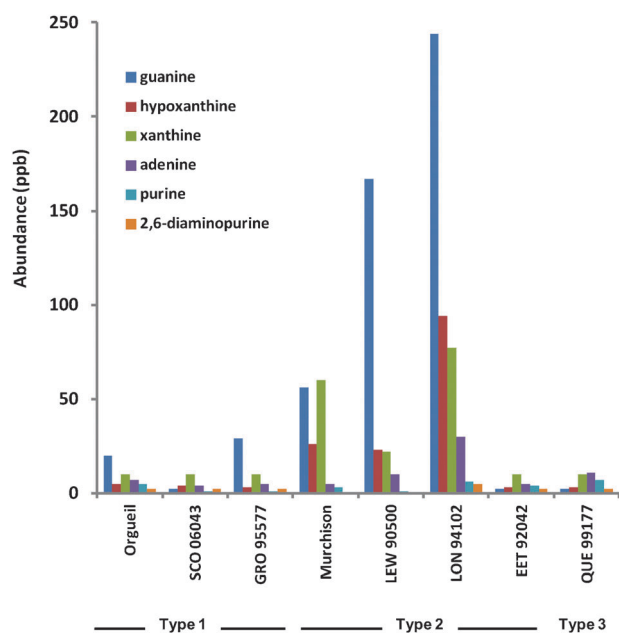
**Fig. 7** Purine nucleobases that have been unambiguously identified in meteorites. Purine and 6,8-diaminopurine are not found in terrestrial biology while 2,6-diaminopurine is rare.



**Fig. 8** Proposed pathway to purine nucleobases from HCN polymerization.<sup>46</sup> The relative concentrations of water, urea and HCN likely determine the distributions of nucleobase products and would vary between meteorite parent bodies.

and ammonia on the parent asteroid, also known as HCN condensation or polymerization.<sup>46</sup> In this reaction, stepwise oligomerization of HCN forms an intermediate molecule called diaminonitrile (DAMN) that can subsequently form intermediates to react with either HCN to form adenine and hypoxanthine or urea to form guanine and xanthine (Fig. 8). Laboratory experiments of  $\text{NH}_4\text{CN}$  and urea were shown to generate the same suite of purines found in Murchison and LON 94102 meteorites (*i.e.*, adenine, guanine, hypoxanthine, purine, 2,6-diaminopurine, and 6,8-diaminopurine).<sup>7</sup> Abiotic synthesis of adenine and purine from dehydration and condensation of formamide has also been proposed.<sup>47</sup> This pathway is initiated by dehydration of formamide to cyanide, and subsequent reactions to generate 2-iminoacetonitrile, which may lead to formation of either purine or adenine based on starting concentrations of ammonium formate and cyanide. Notably, this reaction mimics biological synthesis of purine nucleobases, and is of particular interest for abiotic synthesis of nucleobases on prebiotic Earth, where formamide's low vapor pressure would have allowed a more stable starting material than HCN.

**Distribution of meteoritic nucleobases.** Callahan *et al.*<sup>7</sup> reported the abundances of purines in 11 carbonaceous chondrites, making possible comparisons between nucleobase distributions among CI, CM, and CR chondrites of types 1–3 (Fig. 9). The observed distributions of nucleobases in carbonaceous chondrites presents a dataset that may shed light on which formation mechanism(s) is most plausible for meteorite parent bodies. The CM2 meteorites have by far the most abundant and diverse suite of nucleobases of the meteorites investigated, suggesting they represent environments more conducive to formation and preservation of bases. Within the CM class of carbonaceous meteorites, increasing aqueous



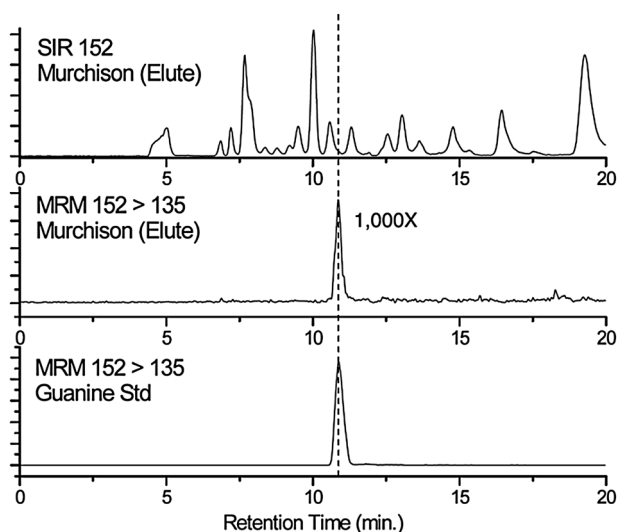
**Fig. 9** Distribution of purine nucleobases in aqueously-altered carbonaceous chondrites. The meteorites are arranged from most to least aqueously altered (type 1, left, to type 3, right).

alteration is coupled with a decrease in overall abundance and diversity of nucleobases. In general, the distribution of nucleobases in the most altered carbonaceous chondrites (Types 1 and 2) was dominated by guanine relative to the other bases, while the least altered meteorites (Type 3) had distributions favoring xanthine, adenine and purine.

### Nucleobase extraction and purification

Nucleobases are less soluble in water than amino acids, and thus are extracted and isolated by different protocols. Traditionally these molecules are extracted from powdered meteorite material in 95% formic acid in sealed glass ampoules at 100 °C for 24 h.<sup>7</sup> Various analytical techniques have been employed to purify the extracts for nucleobase analysis, including multi-step column chromatography and sublimation. Advances in analytical methods such as single-use solid phase extraction (SPE) cartridges have improved nucleobase extraction yields and reduced the possibilities for contamination compounded by multi-step purification processes. Extracts are loaded onto SPE cartridges and rinsed with 5% ammonia in water, followed by methanol. Nucleobases are eluted using 5% formic acid in methanol. The resulting extract is then dried down and can be brought up in solvent appropriate for LC-MS, or derivatized for GC-MS.

**Nucleobase analysis.** Early detections of nucleobases in meteorites were done *via* GC-MS and HPLC-UV absorption.<sup>44</sup> These techniques required extensive purification steps that did not entirely remove compounds that co-eluted during GC-MS measurements or UV-absorbing background that interfered with detection of trace amounts of nucleobases in meteorites. Recent advances in mass spectrometry and analytical techniques have made unambiguous identification and quantification of nucleobases possible. Callahan *et al.*<sup>7</sup> used HPLC



**Fig. 10** Multiple Reaction Monitoring (MRM) using triple quadrupole mass spectrometry allows detection of trace compounds in complex mixtures.<sup>7</sup> The top trace shows the single-ion chromatogram for  $m/z = 152$  (monoprotonated guanine) of a formic acid Murchison meteorite extraction. The middle trace shows MRM monitoring of the 152 to 135 transition that occurs in authentic guanine (bottom trace). The identical chromatographic retention times and parent-to-daughter retention times provide strong evidence that the compound in the meteorite is guanine.

coupled with a triple quadrupole mass spectrometer (LC-QqQ-MS) and HPLC coupled with a Fourier Transform mass spectrometer (LC-FT-MS) to measure the abundance and distribution of nucleobases and nucleobase analogs in 11 different meteorites. QqQ-MS consists of two quadrupoles separated by a collision cell and allows for sensitive detection of characteristic daughter ions produced by fragmentation of individual parent molecules. Using multiple reaction monitoring (MRM), nucleobases were identified on the basis of parent-to-daughter ion transitions specific to each nucleobase along with chromatographic retention times (Fig. 10). Because the detector only measures the targeted masses in MRM, sensitivity is increased. Full fragmentation MS/MS spectra may also be collected for further verification if the target compound is present in sufficient abundance.

FT-MS takes these capabilities a step further, combining the ability to monitor parent masses and multiple fragment masses with high resolution mass measurements, allowing unambiguous assignment of elemental formulae. The FT-MS technique, which measures the mass-to-charge ( $m/z$ ) ratio of ions based on the cyclotron frequency in a fixed magnetic field,<sup>48</sup> allows for extremely high resolving power and is highly suitable for identifying compounds in complex mixtures, such as those found in carbonaceous chondrites.

## Implications for the origin of life

### A source of amino acids and nucleobases

The delivery of complex organic compounds, including amino acids and nucleobases, by carbonaceous chondrites to the early Earth and other planetary bodies could have been an

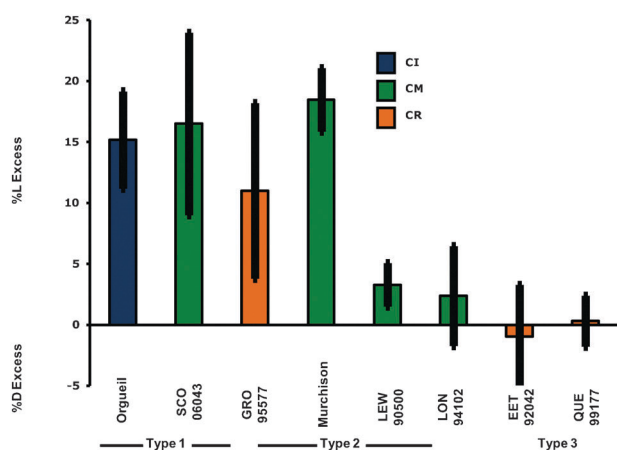
important source of prebiotic compounds required for the emergence of life.<sup>49</sup> In fact, the present day mass flux of carbon-rich (up to 40% carbon by weight) micrometeorites to the Earth is estimated to be  $\sim 40\,000$  tons per year,<sup>50</sup> and the meteoritic input of carbon to the early Earth may have been up to 1000 times higher during the period of heavy bombardment  $\sim 4$  billion years ago. It has been determined that some micrometeorites recovered from Antarctica contain the extraterrestrial amino acid AIB, demonstrating that some amino acids in these particles will survive atmospheric entry heating and can be delivered intact to the Earth,<sup>51,52</sup> in addition to the known amino acid and nucleobase delivery by larger meteorites. Therefore, carbonaceous meteorites and their fragments (micrometeorites and interplanetary dust particles) could have been a major source of organic matter on the prebiotic Earth and other planetary bodies in our Solar System.<sup>53</sup>

### Meteoritic amino acid and the origins of homochirality

The enantiomeric compositions of meteoritic amino acids appear are of particular interest for the origin of life, since these molecules are the monomers of proteins and enzymes common to all life on Earth. The homochirality observed in biological molecules, so-called “left-handed” amino acids and “right-handed” sugars (by analogy with glyceraldehyde), is a property important for molecular recognition processes and is thought to be a prerequisite for life. In contrast to biology, which is dominated by L-amino acids, abiotic processes form racemic amino acids (equal mixtures of L- and D-enantiomers). Therefore, the questions of why and how the nearly exclusive production of one hand of such molecules arose from what were presumably equal mixtures of L- and D-enantiomers in a prebiotic world continue to be a crucial hurdle in understanding the origin of life.

The first amino acid analyses of interior pieces of the Murchison meteorite shortly after its fall found that the chiral amino acids were racemic ( $D/L = 1$ ), indicating that the amino acids were indigenous to the meteorite and very little terrestrial amino acid contamination of the meteorite had occurred.<sup>28</sup> One major discovery was the finding of non-racemic  $\alpha$ -dialkyl amino acids in the CM chondrites Murchison and Murray, with slight to significant L-excesses ranging from 0 to  $\sim 18\%$  of the five-carbon  $\alpha$ -dialkyl amino acid isovaline.<sup>29,54</sup> These results are difficult to explain since the abiotic formation of isovaline and other  $\alpha$ -dialkyl amino acids (*e.g.*, by Strecker synthesis) in carbonaceous meteorites should produce racemic mixtures. In contrast to the  $\alpha$ -hydrogen protein amino acids common to all life on Earth,  $\alpha$ -dialkyl amino acids such as isovaline found in meteorites are not prone to geologically rapid racemization (conversion of one enantiomer to the other) under aqueous or radiogenic conditions; therefore, the initial enantiomeric ratios of these amino acids are more likely to have been preserved since the time of their formation. L-isovaline excesses have since been detected in several different carbonaceous meteorite groups and appear to correlate with the degree of aqueous alteration of the meteorites as inferred from their mineralogy (Fig. 11). These findings suggest that liquid water plays an important role in





**Fig. 11** Enantiomeric excesses (in percent) of isovaline measured in CI, CM, and CR carbonaceous chondrites correlate with the degree of aqueous alteration of the samples.<sup>54</sup>

the amplification of L-isovaline excesses in meteorites and point toward a possible prebiotic contribution to the origin of biological homochirality by the delivery of extraterrestrial organic material from asteroids and comets to the early Earth. The observation that only one amino acid has been found to have a D-excess<sup>55</sup> (*allo*-isoleucine; however, its diastereomer, isoleucine, has only been found in L excess) while at least nine different amino acids have been found with L-excesses accepted as having extraterrestrial origins may indicate that the origin of life on Earth, and possibly elsewhere in our solar system, was biased toward L-amino acid homochirality from the very beginning. Although several mechanisms have been proposed for the initial breaking of chiral symmetry (ref. 55 and elsewhere in this issue), more research is needed in this area.

### Meteoritic nucleobases and the RNA World

Life as we know it is characterized by RNA and DNA, macromolecules that store and transmit information through base-pairing interactions of their nucleobases. Because RNA can serve both the information-containing role of DNA and the catalytic role of proteins, it has been proposed that RNA preceded both DNA and proteins during the origin of life on Earth (the RNA World hypothesis).<sup>56</sup> While laboratory studies have demonstrated the synthesis of nucleobases under conditions potentially relevant to the prebiotic earth,<sup>56</sup> the finding of extraterrestrial nucleobases in meteorites,<sup>7,44</sup> including several that are rare or not used at all in terrestrial biology, provides an important confirmation that these molecules were made abiotically in the evolution of the Solar System and were likely present on the prebiotic Earth. Modern ribosomal and transfer RNAs contain dozens of modified nucleobases in addition to the four canonical nucleobases (adenine, uracil, cytosine, and guanine). In prebiotic times, non-canonical nucleobases may have provided additional stability or catalytic functionality not available with the standard nucleobase library. Furthermore, alternative extraterrestrial nucleobases such as purine, 2,6-diaminopurine, 6,8-diaminopurine, xanthine, and hypoxanthine could have served as precursors for synthesis of an assortment of nucleobases, either through abiotic reactions or biosynthesis by the first life on Earth.

The discoveries of amino acids, nucleobases and other molecules used in modern life provide evidence that meteorites could have jump-started life on Earth.

### Future directions for research in meteoritic amino acids and nucleobases organics research

The last forty years have seen many advances in the analysis of organic compounds in meteorites, as well as in our understanding of how meteoritic delivery of amino acids and nucleobases may have contributed to the origin of life. The future of organic meteoritic research will most likely focus on three areas of growth: (i) advanced technologies for improved separation and structural and isotopic characterization of both soluble and insoluble organic compounds; (ii) unraveling the association between minerals and complex organic mixtures at microscales; and (iii) linking meteorites to their parent bodies *via in situ* analysis and sample return missions to asteroids and comets.

### Improvements in analytical technology

The first growth area driving the current renaissance of meteorite research is the development of increasingly selective and sensitive analytical tools. One direction of increased selectivity is the ability to perform position-specific compound-specific isotopic analysis. This would allow, for example, the isotopic ratio of each carbon in an amino acid to be determined, providing invaluable information in deducing the mechanism of formation of the species. Current developments in this area are promising,<sup>57</sup> but still lack the sensitivity and general applicability to amino acids required for meteorite analyses.

Just as the growth of discoveries in the putative role of RNA in origin of life has been fueled by advancements in biotechnology, increasingly sophisticated organic analyses are driven by innovations in the pharmaceutical analysis industry. Nevertheless, improvements in inorganic analyses of meteorites in the last few years have eclipsed the compound-specific organic analyses possible today. For example, NanoSIMS (nanoscale secondary ion mass spectrometry) provides researchers the ability to map isotopes in fine scales. These methods were driven by the need for high-precision surface analyses in the semiconductor industry; innovations were also driven by NASA's Stardust comet sample return mission. In addition, high-resolution spectroscopy techniques (*e.g.*, deep UV, Raman and XANES) are now being used to map the organic composition of meteorites with high spatial resolution and minimal destruction of the sample.<sup>58</sup> While these are impressive and valuable tools for understanding the mineralogy and organic distribution by compound class, in most cases these techniques do not provide the information necessary to identify specific organic molecules, let alone quantitate enantiomeric ratios. The microprobe two-step laser mass spectrometry ( $\mu$ L2MS) technique approaches the scales and information needed for spatially-mapped, compound-specific measurements;<sup>59</sup> however, this technique focuses exclusively on UV-absorbing species, such as polycyclic aromatic hydrocarbons, lacks the ability to separate isomers or enantiomers, and relies on comparatively low resolution mass spectrometry such that unambiguous molecular formulae cannot be assigned.



Hence, further improvements that allow for the quantitation of organic isomers in fine scales are needed.

### Connecting meteorite organic chemistry and mineralogy

As discussed previously, carbonaceous chondrite class and alteration history are linked to amino acid and nucleobase abundances and distributions. This link is only beginning to be understood, as sufficient meteorite samples of various petrographic types are catalogued and made available through ANSMET and other meteorite recovery efforts. By analyzing macroscopic samples of the Murchison meteorite, a correlation between L-isovaline excesses and hydrated silicates was observed.<sup>60</sup> Similarly, analysis of different samples of the Tagish Lake meteorite saw some correlation between aqueous alteration and amino acid abundances.<sup>61</sup> Obtaining a more complete understanding of the relationship between aqueous alteration and thermal metamorphism to amino acid abundances and distributions will require integrated studies of both organic content and mineralogy at fine scales.

### Miniaturizing instruments for *in situ* extraterrestrial analysis

Organic analysis of meteorites is necessarily limited to those samples which are recovered on Earth. This means that a targeted understanding of the chemical nature of specific planetary bodies is difficult. *In situ* analysis via analytical instrumentation on planetary missions addresses this limitation. In all cases of instrumentation developed for *in situ* measurements on planetary surfaces, the precision, sensitivity, and selectivity of laboratory instrumentation are sacrificed for more efficient use of power and reductions in weight and volume.

The first GC-MS for a planetary mission landed on Mars in 1976 as part of the Viking Mission; the instrument failed to detect organics for a multitude of reasons, including experimental design and possible destruction of organics by soil oxidants during heating as part of the sample analysis. In 2012, the next instrument with gas chromatographic capabilities will reach Mars on the Mars Science Laboratory (MSL). Sample Analysis at Mars (SAM) on MSL is an integrated evolved-gas mass spectrometer, gas chromatograph, and tunable laser spectrometer that will inventory the chemical and isotopic composition of organic and inorganic volatiles for both atmospheric and pyrolyzed solid samples on Mars.<sup>62</sup> SAM features a “one-pot” solvent extraction and chemical derivatization experiment targeted at detection of amino acids and other organics in Martian regolith. The ability to detect organic acids using the SAM wet chemistry experiment will, however, be more limited than current state of the art laboratory approaches, and enantiomeric separation of amino acids will not be possible using the derivatization reagents on SAM. A similar chemical derivatization and GC-MS approach<sup>63</sup> has also been incorporated into the Cometary Sampling and Composition (COSAC) evolved-gas experiment on ESA’s Rosetta Lander and will provide amino acid detection on the surface of comet 67P/Churyumov-Gerasimenko in 2014. The Mars Organic Molecule Analyzer (MOMA), an ion trap mass spectrometer in development for the joint European/Russian ExoMARS 2018 mission, will also target biologically

important molecules including amino acids and nucleobases. Another analytical approach currently in development for future planetary missions is microchip capillary electrophoresis ( $\mu$ CE), which can be miniaturized to lab-on-a-chip systems with microfluidics for sample delivery and routing. “Urey Instrument” is a  $\mu$ CE device with laser-induced fluorescence (LIF) detection. Recently, a fully automated end-to-end  $\mu$ CE analysis of amino acids was demonstrated using a four-layer microchip electrophoresis device with LIF detection.<sup>64</sup> These microfluidic devices are particularly attractive because they have very low mass, volume, and power requirements compared to current separation and detection of organics; however, this technology has not yet been demonstrated in space.

### Sample return missions

Sample return missions of organic-rich targets significantly contribute to the analysis of extraterrestrial amino acids, nucleobases, and other organic compounds. These missions can reduce the terrestrial contamination of a sample by controlling and documenting the chain of exposure to potential contaminants in a way virtually impossible with either meteorites or *in situ* missions. Sample return missions also allow researchers to utilize analytical instruments to examine the returned sample without regard to the limitations in mass, power, volume, data rate, complexity, and sample preparation that limit flight instrumentation. Since the parent body of the returned sample is known and can be studied by *in situ* instrumentation, a sample return mission provides the opportunity to compare the chemistry (organic and inorganic) with the geology and astronomy of the parent body. Two recent examples of such missions are Japan’s Hayabusa mission and NASA’s OSIRIS-REx. Hayabusa visited Asteroid 25143 Itokawa and returned  $\sim$ 1500 grains. Initial analyses of asteroidal particles for amino acids and other molecules did not find any indigenous organic compounds above the part-per-million level, although this was perhaps unsurprising given that Itokawa’s composition appears to match ordinary, and not carbonaceous, chondrites.<sup>65</sup>

In 2011, NASA selected OSIRIS-REx to characterize near-Earth asteroid 101955 (1999 RQ36) and return at least 60 g of regolith to the global scientific community to advance our understanding of the generation, evolution, and maturation of regolith on small bodies. This B-type Apollo Near Earth Asteroid is a primitive body that may be related to CI or CM meteorites based on reflectance spectra.<sup>66</sup> However, the relationship between this asteroid and the meteorites in curators’ vaults, as well as its amino acid, nucleobase, and organic content, will be unknown until the sample is returned in 2023 and is analyzed by instruments and techniques, many of which are likely not yet invented.

### Conclusions

Carbonaceous chondrites contain a wide range of organic compounds, including amino acids and nucleobases. Delivery of these compounds to the early Earth through meteoritic bombardment may have contributed to the origin of life. The structural distributions and abundances of amino acids vary between chondritic classes, reflecting differences in

parent body chemistry. Stable isotopic compositions of individual amino acids reveal differences in formation pathways, and chemical reactions leading to a variety of amino acid structural types have been identified. Certain meteoritic amino acids display enrichments of the L-enantiomer, an intriguing observation because life on Earth uses L-amino acids almost exclusively. A diverse suite of nucleobases has also been observed in meteorites. Differences in distributions of these molecules among meteorites suggest that there were multiple formation pathways leading to nucleobases in different parent bodies. The study of meteorites over the last 50 years has yielded a wealth of information about prebiotic chemistry in our Solar system. Nevertheless, considerable work, aided by new technological developments, remains to improve our understanding of the origin, evolution, and contribution to the origins of life of meteoritic organic compounds.

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