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A Detailed Study of the Amino Acids Produced from the Vacuum UV Irradiation of Interstellar Ice Analogs

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Abstract We present a detailed analysis of the variety, quantity and distribution of the amino acids detected in organic residues after acid hydrolysis. Such organic residues are produced in the laboratory after the vacuum ultraviolet (VUV) irradiation of several astrophysically relevant ice mixtures containing H_2O, CO, CO_2, CH_3OH, CH_4 and NH_3 at low temperature (10–80 K), and subsequent warm-up to room temperature. We explore five experimental parameters: the irradiation time, the temperature, the ice mixture composition, the photon dose per molecule and the substrate for the ice deposition. The amino acids were detected and identified by *ex-situ* liquid chromatography analysis of the organic residues formed after warming the photolysed ices up to room temperature. This study shows that in all experiments amino acids are formed. Their total quantities and distribution depend slightly on the experimental parameters explored in the present work, the important requirement to form such molecules being that the starting ice mixtures must contain the four elements C, H, O and N. We also discuss the effects of the chemical treatment needed to detect and identify the amino acids in the organic residues. Finally, these results are compared with meteoritic amino acid data from the carbonaceous chondrite Murchison, and the formation processes of such compounds under astrophysical conditions are discussed.

Keywords Amino acids · Interstellar medium (ISM) · Laboratory simulations · Molecular processes · UV irradiation

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Introduction

The production of complex organic molecules such as amino acids from the irradiation of interstellar ice analogs with UV photons at low temperature in the laboratory is nowadays a clearly known and established result (Bernstein et al. 1995, 2002; Muñoz Caro et al. 2002; Nuevo et al. 2006, 2007).

In these laboratory simulations of irradiation of ices, amino acids are detected in the organic residues formed from the vacuum ultraviolet (VUV) photolysis at 10–80 K of the starting icy material, subsequently warmed up to room temperature after irradiation. After sublimation of the ices, a refractory thin layer of organic compounds remains on the surface of the substrate. This refractory material, called *organic residue* or simply *residue* thereafter, and whose compounds are stable at room temperature, can be analysed by *ex-situ* chemical techniques to study its composition.

Among the great variety of organic molecules that could constitute such residues, amino acids were searched for because they are among the smallest organic molecules – although large from an astronomical point of view – and because the methods to detect and identify them are well known (Pfeifer and Hill 1983; Abe et al. 1996). Since only 20 of these amino acids are the building blocks of proteins, present in all living organisms on Earth, they are also of great interest for prebiotic chemistry, and thus likely to be connected with the origins of life.

A large variety of amino acids were also detected in extraterrestrial material, in particular meteorites such as Murchison (Engel and Macko 1997; Cronin and Pizzarello 1997, 1999; Pizzarello et al. 2003) and Murray (Cronin and Pizzarello 1999), where more than 70 of them were found (see Shock and Schulte 1990, and references therein). However, the link between these meteoritic amino acids and (1) those that may be present in the interstellar medium (ISM) (Kuan et al. 2003), (2) the organics including amino acids that may be present in asteroids, comets, interplanetary dust particles (IDPs) (Irvine 1998) and micrometeorites (Brinton et al. 1998; Matrajt et al. 2004), and (3) the prebiotic molecules from which life started 3.8 billion years ago on the primitive Earth, has not been established yet. A possible scenario, first proposed by Oró (1961), favours a delivery of the prebiotic material by asteroids, comets and IDPs during the period of heavy bombardment that our planet experienced in its first hundreds million years of existence.

In this paper, we present a detailed study of the absolute quantities and distribution of the amino acids found after the acid hydrolysis of the organic residues formed by the VUV irradiation of interstellar ice analogs in the laboratory and their subsequent warm-up to room temperature, focusing on the detection of proteinaceous amino acids. We explore five experimental parameters (irradiation time, temperature, ice mixture composition, photon dose per molecule and substrate for deposition), and discuss the effects of the chemical treatment (extraction solvent, acid hydrolysis) required to detect and identify such compounds. Finally, we compare these results with meteoritic data, and discuss the formation mechanisms of such molecules in astrophysical environments.

Experimental

UV Irradiation of the Ices

The protocol for UV irradiation is explained in detail elsewhere (Nuevo et al. 2007). Briefly, in a vacuum chamber evacuated with a turbo-molecular pump down to ~10^{-7}mbar

and cooled down to about 10 or 80 K with liquid helium or nitrogen, respectively, gas mixtures are deposited onto a cold substrate (MgF_2 window or Cu disc, see Results section). When they reach the substrate, the ices condense to form a thin film. The ices are then irradiated by the UV photons (mainly 121.6 and ~160 nm photons) emitted by a H_2 discharge lamp. In this study, the ices were deposited and irradiated simultaneously, in order to obtain a thick sample (a few μm) photolysed over its whole thickness. A Fourier-transform infrared (FTIR) spectrometer connected to the chamber allowed us to control the evolution of the ices deposited on the MgF_2 substrate during the whole experiments.

The components of the ice mixtures were H_2O, CO, CO_2, CH_3OH, CH_4 and NH_3, chosen to be relevant to the solid compounds observed in the ISM (e.g. Dartois et al. 2002; Gibb et al. 2004 and references therein). H_2O was purified to a 18.2-MΩ cm resistivity (Millipore Direct-Q 5), the gases were purchased from Messer (CO, >99.997% purity; CO_2, 99.998% purity; CH_4, 99.995% purity; NH_3, 99.98% purity) and CH_3OH from Aldrich (liquid, 99.9% purity). Different mixtures were prepared for the irradiations, by mixing two to five compounds at the same time, and changing the relative proportions between the components of a mixture (see column 2 of Table 1 for the composition of these mixtures). The relative proportions between the components were controlled by their partial pressures in the handling gas system.

After irradiation, we let the samples warm up slowly under dynamic vacuum to room temperature (at about 10 K h^{-1}), to avoid their loss by explosive sublimation of the ices due to radical exothermic recombination (d'Hendecourt et al. 1982). At room temperature, the refractory organic residues remaining on the substrate were kept under static vacuum (~10^{-5}mbar) before being analysed with chromatographic techniques (see Ex-situ Analysis of the Residues section).

Ex-situ Analysis of the Residues

To determine the chemical composition of the so-formed organic residues, infrared (IR) spectroscopy is not enough. Indeed, the IR spectroscopy monitoring only allows us to observe the depletion of the starting components' IR features during the UV irradiation at low temperature, the apparition of IR features assigned to the newly-formed photo-products (Bernstein et al. 2004; Nuevo et al. 2006), as well as their evolution during the warm-up to room temperature (Bernstein et al. 1995; Nuevo et al. 2006). However, these IR identifications are limited to families of organic molecules (alcohols, carboxylic acids, carbonyls, alkyl chains, etc.), and cannot allow the identification of any given organic compound. Therefore, *ex-situ* chemical techniques are required to determine the composition of the residues. In the present work, we chose to search for amino acids.

Extraction and Injection Protocol

The organic residues were first extracted from their substrate with 4 × 100 μL of water, and then with 4 × 100 μL of acetonitrile CH_3CN (thereafter named ACN). Water was demineralised by percolation through type SG30 resin (Aquadem), then distilled in an Aquatron model A4S glass apparatus (Bibby Sterilin, Ltd.), and ACN was of analytical grade (Merck). This double extraction was performed to be sure that the whole residues were dissolved whatever the polarity of their components. The extraction solvent was evaporated in a desiccator. The obtained extracts were then submitted to acid hydrolysis (6 M HCl, 105°C, 24 h), and the hydrolysates were dried under vacuum on P_2O_5 and

Table 1 Experimental parameters and total amino acid quantities (in nmol) in the hydrolysed residues determined for each irradiation experiment

Exp. #	Ice mixture H_2O:CO:CO_2:CH_3OH:CH_4:NH_3	T (K)	Irradiation time (h)	Photon dose per molecule	Substrate	Total amino acids (nmol)[a]	Abundances (nmol)			
E0 (blank)	0:0:0:0:0:0	81	47.5	–	MgF_2	2.8	Ser (0.5)	Gly (0.4)	His?(0.3)	Glu (0.2)
E1	2:1:0:1:1:1	11	18.1	16.03	MgF_2	45.1	Ala(20.3)	Gly (15.8)	Ser (2.2)	Tyr?(2.0)
E2	1:2:0:0:1:1	11.5	26.7	0.05	MgF_2	59.0	Gly(21.8)	Ala (17.4)	Glu (3.6)	Ser (2.9)
E3[b]	2:1:0:1:1:1	9.5	21.3	0.29	MgF_2	4.2	Tyr?(2.1)	His?(1.1)	Gly (0.5)	Ser (0.3)
E4[c]	2:1:0:1:1:1	cyclic 10–60	21.3	0.15	MgF_2	24.1	Gly (9.9)	Ala (6.5)	Ser (2.9)	Lys?(1.3)
E5[c]	1:2:0:0:1:1	cyclic 10–60	22.0	0.06	MgF_2	12.2	Gly (5.8)	Ala (1.3)	Ser (1.2)	Glu (1.2)
E6[c]	2:1:0:1:1:1	19	43.2	0.06	drilled Cu[d] disc	23.4	Gly(14.7)	Ala (3.8)	Ser (1.1)	Asp (0.9)
					MgF_2	2.0	Gly (0.6)	Ser (0.4)	Glu (0.3)	Asp (0.3)
E7	6:0:0:2:0:1	81	23.3	0.04	MgF_2	7.6	Gly (4.1)	Glu (1.1)	Ala (1.0)	Ser (0.5)
E8	0:0:0:1:0:1	81	49.0	0.04	drilled MgF_2	57.0	Gly(28.0)	Ala (18.5)	Lys?(4.7)	Ser (1.9)
E9[e]	0:0:0:1:0:1	81	49.0	0.04	MgF_2	31.9	Gly(18.2)	Ala (8.9)	Ser (1.7)	Glu (0.6)
E10	6:0:0:2:0:1	82	49.7	0.05	drilled MgF_2	7.6	Gly (4.8)	Ser (0.6)	Ala (0.6)	Glu (0.3)
E11	1:0:3:1:0:1	83.5	50.4	0.06	MgF_2	19.4	Gly(11.3)	Ala (5.6)	Ser (1.1)	Leu (0.4)
E12	1:0:1:1:0:1	81.5	48.1	0.06	MgF_2	44.3	Gly(25.9)	Ala (15.1)	Ser (1.3)	Glu (0.5)
E13	3:0:1:1:0:1	81	48.3	0.06	MgF_2	16.7	Gly (8.6)	Ala (5.0)	Ser (1.3)	Glu (0.3)

In the last column are given the four most abundant amino acids with their individual quantities (in nmol). The compounds followed by a question mark are amino acids with complex structures (Lys, Tyr, His) which are not expected to be formed in our residues with such high quantities (see Remarks on the Detection of Some Amino Acids section).

[a] The quantities reported in columns 7 and 8 correspond to the sum of quantities measured for both hydrolysed water and ACN extracts. [b] In this experiment the extracted residue was not hydrolysed. [c] For these samples, the water- and ACN-extracted residues were divided into two fractions: one was hydrolysed, the other was not. Only the quantities measured after hydrolysis are reported here. [d] See text about the nature of the substrate for details. [e] The chromatogram and detailed identification of the amino acids for this sample are given in Fig. 2 and Table 2, respectively.

NaOH before being buffered in a 67-mmol L^{-1} trisodium citrate–HCl solution (pH 2.2) and injected into the amino acid analyser.

Amino Acid Analyses

The hydrolysates were analysed with a Hitachi L-8800 Amino Acid Analyzer, namely a liquid chromatography system equipped with a Hitachi Packed Column #2620MSC-PS (inner diameter 4.6 mm, length 80 mm) filled with an Na^+-form sulfonic resin (cation-exchange chromatography). The separation of the compounds is performed with a discontinuous gradient of ionic strength, the concentration of Na^+ ranging from 0.10 to 1.26 mol L^{-1}. After each injection, the column is regenerated by percolation of 0.2-mol L^{-1} NaOH. The detection system is a post-column ninhydrin-reaction device. Ninhydrin reacts with primary amino acids to yield: (1) aldehydes derived from the starting amino acids, (2) carbon dioxide, (3) hydrindantin and (4) ammonia. The reaction of hydrindantin and ammonia leads to the formation of Ruhemann's purple (Greenstein and Winitz 1961), absorbing at 570 nm and thereby allowing the detection of the amino acids.

This technology was set up 50 years ago for the separation and quantification of amino acids in protein hydrolysates (Spackman et al. 1958). It has been improved ever since, and dedicated instruments such as the one used in this study now allow the automatic analysis of numerous samples with reliability, accuracy and reproducibility (Le Boucher et al. 1997). Consequently, only the proteinaceous amino acids are identified and quantified in our samples. Moreover, similar amino acid analyses were performed on similar samples produced under similar experimental conditions using gas chromatography coupled with mass spectrometry (GC-MS) (Muñoz Caro et al. 2002; Nuevo et al. 2006, 2007). These results showed similar distributions for the amino acids detected with both techniques, indirectly supporting the present study.

The standard used for the identification and the quantification of the amino acids (ATC – Ajinomoto-Takara Corporation) is a mixture of ammonia (NH_3) and 17 proteinaceous amino acids: aspartic acid (Asp), threonine (Thr), serine (Ser), glutamic acid (Glu), proline[1] (Pro), glycine (Gly), alanine (Ala), cystine (Cys), valine (Val), methionine (Met), isoleucine (Ile), leucine (Leu), tyrosine (Tyr), phenylalanine (Phe), lysine (Lys), histidine (His) and arginine (Arg). Since the starting mixtures did not contain sulphur, cystine and methionine, which are sulphur-bearing amino acids, were not present in our residues. The standard mixture is provided by the manufacturer in 0.1 M HCl at a concentration of 2.500 ± 0.075 μmol mL^{-1} for each component. A dilution to 0.1 nmol μL^{-1} in the pH 2.2 buffer was made in the laboratory, and 20 μL (2 nmol) of this diluted solution were injected before each series of analyses.

In order to estimate the risk of contamination between samples injected successively and the relative uncertainties of the measurements in each injection, we performed injections of three diluted standards to obtain amounts of 0.1, 1 and 10 nmol (i.e., ranging over two orders of magnitudes) for each amino acid. Each diluted solution was injected three times into the amino acid analyser. The results show that for amounts of the order of 0.1 nmol, the relative uncertainty is 1.1%. This uncertainty drops down to 0.6% for amounts ranging from 1 to 10 nmol. In other words, the absolute uncertainties for individual amino acids do not exceed 0.01, 0.1 and 0.2 nmol for measured quantities smaller than 1, 16 and 33 nmol, respectively. Thus, since the quantities measured after two successive injections for a given

[1] Proline is a secondary amino acid. Its reaction with ninhydrin yields a compound absorbing at another wavelength. In the present work, the presence of proline has not been searched for.

amino acid are very close and smaller than 28.0 nmol (maximum measured for glycine in the E8 sample, see column 8 of Table 1 and Results section), the uncertainties are smaller than 0.2 nmol, so that the contamination between samples appears to be negligible.

Results

In this work, we prepared and analysed 14 samples, numbered from E0 to E13. E0 was a blank where an MgF_2 window without any ice deposition was irradiated at 80 K and analysed at room temperature under exactly the same conditions as for the E1~E13 experiments. The effects of five experimental parameters were studied:

1. *Irradiation time*: we performed experiments with irradiation times ranging from about 18 to 50 h.
2. *Temperature in the cryostat*: the experiments can be divided into three groups: a first group where irradiations were performed at about 10 K (E1~E3 and E6 experiments), a second one where the temperature was around 80 K (E7~E13 and blank E0), and a last one where we made the temperature vary cyclically between 10 and 60 K (E4 and E5). The idea for this last group of experiments was to condense the ices around 10 K and then to slowly increase the temperature to ~60 K, so the radicals formed from the photolysis of the ices might become more mobile and react with higher efficiency to form complex molecules. Then, the chamber was cooled down to ~10 K again to re-condense the ices, then warmed up to 60 K again, and so on cyclically, with a period of about 1 h.
3. *Starting ice mixture composition*: we used six different starting compounds (see UV Irradiation of the Ices section), the most complex mixtures containing five of these compounds. Different mixture compositions were chosen, as well as different relative proportions between the components. The important requirement for the formation of complex organic molecules such as amino acids was that all starting mixtures had to contain the four elements C, H, O and N.
4. *Photon dose per molecule*: for each experiment, the number of photons per deposited molecule, averaged over the whole irradiation time, was calculated. All numbers of photons per molecule were found to be smaller than 1, except for the E1 experiment.
5. *Nature of the substrate*: two kinds of MgF_2 windows were tested in our experiments: regular windows (E1~E5, E7, E9 and E11~E13 experiments, and the blank E0), and windows where tiny cavities (1-mm large, 0.5-mm deep) were drilled on the surface (E8 and E10), to increase the effective interaction surface between the deposited ices and the photons. A thin 1-mm-thick copper (Cu) disc, whose surface was also drilled with tiny cavities (1-mm large, 0.5-mm deep) was used for the E6 experiment (Fig. 1). This Cu disc, placed over an MgF_2 window, allowed us to check if metallic surfaces have any catalytic effect on the production of amino acids. A small hole in its middle (3-mm large) allowed the beam of the FTIR to pass through the MgF_2 window in order to control the evolution of the ices deposited on the available MgF_2 (small) surface (see UV Irradiation of the Ices section). For each residue obtained, the extraction protocol and the analysis were the same.

As observed in previous laboratory simulations where interstellar ice analogs were UV-irradiated (Bernstein et al. 2002; Muñoz Caro et al. 2002; Nuevo et al. 2006, 2007), the extracted and hydrolysed organic residues produced led to the formation of a large variety of amino acids with measurable quantities in all our experiments. For each irradiation

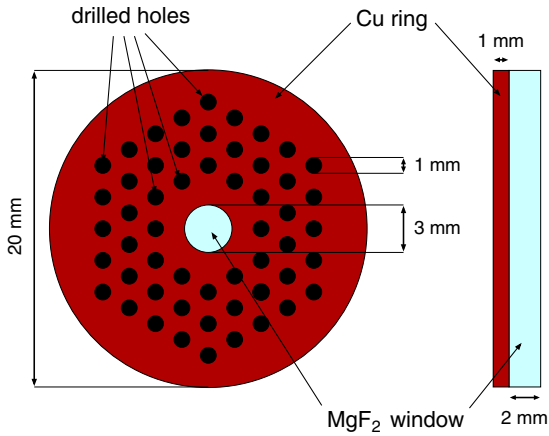

Fig. 1 Schematic diagram of the copper disc used as a substrate for the E6 experiment, seen from the face (*left*) and profile (*right*) views. The hole in the middle of the disc allows the infrared beam to pass through the MgF_2 window

experiment E0~E13 performed in the present work, the experimental conditions, the total amino acid quantity (in nmol), as well as the individual quantities measured for the four most abundant amino acids (in nmol) are given in Table 1. The reported total amino acid quantities (column 7) correspond to the sum of the quantities measured in both hydrolysed water and ACN extracts.

In a general way, the amino acid quantities measured vary significantly from one sample to another, ranging from 7.6 (E7 and E10) to 59.0 nmol (E2) for the hydrolysates. These amounts are low in absolute values, but detectable with chromatographic techniques, and comparable with those which can be deduced from previous experiments (Muñoz Caro et al. 2002), as well as those measured in 1 g of the Murchison meteorite, where they range from 2.5 to 24.5 nmol (Engel and Macko 1997). The highest total amino acid quantities were obtained (after hydrolysis) for the E2 (mixture: H_2O:CO:CH_4:NH_3 = 1:2:1:1, T = 11.5 K, irradiation time: 26.7 h, substrate: regular MgF_2 window) and E8 (CH_3OH:NH_3 = 1:1, T = 81 K, 49.0 h, drilled MgF_2 window) experiments, with 59.0 and 57.0 nmol, respectively.

Figure 2 shows the chromatograms obtained for the water-extracted hydrolysates of the E9 sample (CH_3OH:NH_3 = 1:1, T = 81 K, 49.0 h, MgF_2) and of the blank E0 (T = 81 K, 47.5 h, MgF_2), prepared under the same conditions. We clearly see that the chromatogram for E9 (trace a) displays several peaks of appreciable areas, unlike the one for E0 (trace b). By comparison with the ATC standard (see Ex-situ Analysis of the Residues section), most of the peaks were assigned to amino acids and their quantities were calculated (Table 2). As already mentioned (Amino Acid Analyses section), the identification of the amino acids which were also routinely detected by an independent technique (GC-MS) in similar residues (Muñoz Caro et al. 2002; Nuevo et al. 2006, 2007) can be considered as significant. The other amino acids will be referred as tentatively identified and marked with an asterisk in Table 2.

Among the amino acids identified in this water-extracted hydrolysate, the most abundant ones were glycine (Gly, 17.9 nmol) and alanine (Ala, 8.8 nmol). The peaks ascribed to tyrosine and histidine (Fig. 2 and Table 2) may actually correspond to smaller compounds eluting from the column with the same retention times. This point will be discussed in Remarks on the Detection of Some Amino Acids section. The total amino acid amount in

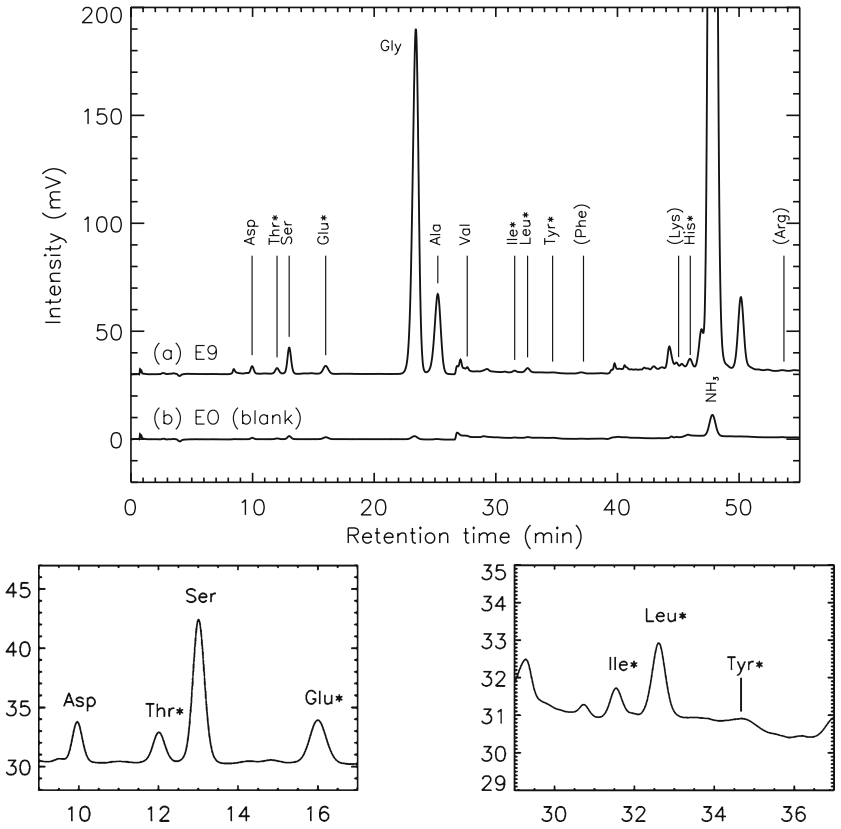

Fig. 2 Chromatograms of **a** the E9 (CH_3OH:NH_3=1:1, T=81 K, 49 h, MgF_2) and **b** the corresponding blank E0 water-extracted hydrolysates (volume injected onto the column: 75% of the total volume). The peak identification is given on the chromatogram. The amino acids whose names are given between parentheses were not detected in the E9 sample. The amino acids marked with an asterisk (*) were only tentatively identified (see Results section and Table 2). The two traces below the main chromatogram are details of the 9–17 min (*left*) and 29–37 min ranges (*right*) for E9. The amino acid quantities, calculated for the whole water-extracted hydrolysate of E9, are listed in Table 2. The highest peak, eluted at 48 min, corresponds to residual NH_3 (see Ex-situ Analysis of the Residues section)

the blank (E0) water-extracted hydrolysate (2.8 nmol) is small compared with that of E9 (31.9 nmol). This shows that contamination during the extraction and hydrolysis steps is negligible.

Irradiation Time

This parameter seems to have only a small effect on the production of amino acids. Indeed, if we consider the total quantities of amino acids obtained for all experiments, it seems that from ~18 h of irradiation (E1 experiment), the production of organic molecules is efficient enough for amino acids to be detected. The photo-process efficiency does not seem to increase significantly with the irradiation time (see column 7 of Table 1), certainly because of a competition between the formation of new species from the starting ices and the degradation of the already formed photo-products. However, although the measured

Table 2 Identification of the peaks from the chromatogram of the water extract of E9 (Fig. 2), with the chemical formulas, the retention times R_t, the quantities (in nmol) and corresponding masses (in μg) deduced for each detected amino acid

Amino acid	Formula	R_t (min)	Quantity[a] (nmol)	Mass[a] (μg)
Aspartic acid (Asp)	$C_4H_7NO_4$	9.96	0.3	0.04
Threonine* (Thr)	$C_4H_9NO_3$	12.01	0.3	0.03
Serine (Ser)	$C_3H_7NO_3$	13.01	1.3	0.13
Glutamic acid* (Glu)	$C_5H_9NO_4$	16.01	0.5	0.08
Glycine (Gly)	$C_2H_5NO_2$	23.43	17.9	1.35
Alanine (Ala)	$C_3H_7NO_2$	25.23	8.8	0.78
Valine (Val)	$C_5H_{11}NO_2$	27.65	0.1	0.01
Isoleucine*(Ile)	$C_6H_{13}NO_2$	31.55	0.1	0.01
Leucine* (Leu)	$C_6H_{13}NO_2$	32.61	0.2	0.03
Tyrosine* (Tyr)[b]	$C_9H_{11}NO_3$	34.67	<0.1	<0.01
Histidine* (His)[b]	$C_6H_9N_3O_2$	45.96	0.2	0.04

[a] The quantities and masses given here are for the whole water extracts. [b] See discussion about the identification of these amino acids (Remarks on the Detection of Some Amino Acids section).

The amino acids marked with an asterisk (*) were tentatively identified, since they were only detected by the present liquid chromatography technique. Their identification needs to be confirmed by other independent techniques.

absolute amino acid quantity may increase with the irradiation time since more material is deposited on the substrate, the results reported in Table 1 clearly show that other factors have more significant effect on the final amino acid quantities, comparing for instance E1 vs. E4, and E7 vs. E10.

Temperature

When we consider the results reported in Table 1 for the series of experiments at ~10 (E1~E3 and E6) and ~80 K (E7~E13), we clearly see that the temperature has also only a small effect on the production of amino acids. Indeed, for the ~10 K series, the amino acid quantities after hydrolysis range from 25.4 (E6, total amount on both Cu disc and MgF_2 window) to 59.0 nmol (E2), and for the ~80 K series the measured quantities are of the same order of magnitude, ranging from 7.6 (E7 and E10) to 57.0 nmol (E8).

This confirms the results obtained previously by Muñoz Caro and Schutte (2003), who already noticed that the temperature has little effect on the production of organic residues, the residues formed at ~10 and ~80 K displaying similar visual aspects, infrared spectra and amino acid quantities after water extraction and acid hydrolysis. From a kinetic point of view and assuming that no equilibrium is reached within the irradiation time, more compounds should be formed at 80 K than at 10 K. All these results suggest that organic molecules are formed at a temperature higher than 80 K, probably during the warm-up, by radical–radical recombination.

For the E4 and E5 experiments, we irradiated the ices while the temperature was varying cyclically from 10 to ~60 K in the cryostat, with a period of about 1 h. As shown in Table 1, this cyclic change of temperature has no significant enhancement effect on the production of the amino acids recovered in our samples. It may even have led to the

production of less organic material, comparing E1 vs. E4 and E2 vs. E5, where the same starting mixtures led to amino acid quantities from two to five times smaller in the cases of a cyclic temperature. This result also supports a formation of complex organic molecules during the warm-up to room temperature.

Starting Ice Mixture Composition

According to the results reported in Table 1, it seems that the starting ice mixtures leading to the highest amino acid quantities in the hydrolysed residues are those containing less water ice in relative proportions (see E2, E8, E9 and E12 experiments). Only the E1 experiment, where the proportion of water is twice as high as those of the other components, displays high quantities of amino acids.

These observations can be surprising, since water ice matrices are expected to protect the formed organic molecules from their destruction by the UV photons. However, experiments carried out in our laboratory showed that when water ice is the main component of the ices, the UV photons emitted by the H_2 lamp do not penetrate deeper than 0.1 μm inside the ice matrix (unpublished results). This shield will thus prevent the production of complex organic molecules from the moment the deposited layer of ice becomes optically thick for the UV photons (see also Photon Dose Per Molecule section). The mixtures leading to the highest amino acid quantities under our experimental conditions are therefore those containing less water and where the other components have relative proportions close to 1 (E2, E4 and E12 experiments), as well as those which do not contain water at all (E8 and E9).

Moreover, since the total pressure in the bottle where the gases are mixed strongly depends on the water vapour pressure at room temperature (about 18 mbar), the less water we put into the mixture, the higher the carbon quantity and thus carbon reservoir is. Consequently, the *absolute* quantity of organic molecules will increase with respect to experiments where the mixtures contain more water. However, Muñoz Caro and Schutte (2003) showed that the relative proportions between the quantities of organic molecules formed and those of carbonaceous compounds in the starting ices are of the same order of magnitude whatever the proportion of water. In some cases, they even showed that the production efficiency for organic molecules is higher when the proportion of water is high. Obviously, water-rich mixtures are more relevant to astrophysical observations, since H_2O is by far the most abundant component of interstellar ices. Therefore, other experiments have to be performed to study the role of water ice in the formation of organic molecules and understand the discrepancies between our experiments and those of Muñoz Caro and Schutte (2003).

Photon Dose Per Molecule

After the calculation of the number of photons per deposited molecule for the E1 experiment, found to be high (16.03), the deposition of the gas mixtures was controlled in order to increase the deposition flux of the molecules on the substrate and thus decrease the average photon dose, and to keep a roughly constant number of photons per molecule of 0.04–0.06 for the next experiments (E2, E5~E13). Only E3 and E4 experiments have slightly higher photon doses of 0.29 and 0.15 photons per molecule, respectively.

Regarding the E1~E6 experiments, since the photon dose is different for each experiment, it is difficult to find a correlation between the photon dose and the total quantity of amino acids formed, or with their distribution. However, for the E7~E13

experiments, where the photon dose is roughly the same, it appears that the total amino acid quantity measured in the extracted and hydrolysed residues is correlated with the relative proportion of H_2O with respect to the other components in the starting mixture. Indeed, the higher this relative proportion of water ice, the smaller the total amino acid quantity: for the E7 and E10 experiments, where the starting mixture is H_2O:CH_3OH:NH_3 = 6:2:1, the total amino acid quantities were measured to be 7.6 nmol, i.e., much less than for the E8, E9 and E11~E13 experiments where the relative proportion of H_2O in the starting mixture is smaller. This result suggests that water ice has a screening effect on the UV photons irradiating the ice matrices, leading to smaller yields of photo-products, and therefore to smaller quantities of organic molecules formed (see also Starting Ice Mixture Composition section).

Finally, the absolute photon dose may have consequences for the (photo)chemical processes leading to the formation of amino acids or their precursors, and hence on their final distribution. Thus glycine, alanine, serine and glutamic acid always appear to be the four most abundant compounds in most of the extracted and hydrolysed residues of the E7~E13 experiments, where the photon dose is roughly the same. As a last remark, it can be pointed out that in contrast with all other samples where glycine was found to be the most abundant amino acid, alanine was the most abundant compound detected in the E1 sample, where the photon dose was much higher (16.03) than for the other experiments and greater than 1. This again shows a correlation between the photon dose and the final distribution of the amino acids found in the samples.

Substrate

As for the irradiation time and the temperature, the substrate also seems to have only little effect on the production of amino acids. The experiments where regular MgF_2 windows were replaced by drilled windows, thought to increase the effective interaction surface between the ices and the UV photons, do not allow any definitive conclusion. If we compare the E8 and E9 experiments, performed under similar conditions and with identical starting mixtures, we see that the drilled MgF_2 window (E8) increases the amino acid production by more than 50% with respect to the regular MgF_2 window (E9). However, the amino acid production efficiency appears to be the same for E7 (regular MgF_2 window) and E10 (drilled MgF_2 window) samples, which have a common starting mixture composition, different from that of E8 and E9.

For the copper disc used in the E6 experiment, we did not observe any measurable catalytic effect on the production of amino acids. Indeed, if we compare the E1 and E6 experiments (same starting ice mixtures, $T < 20$ K), the total quantity measured for the Cu-disc experiment (E6) is smaller than the quantity obtained with the regular MgF_2 window (E1). The deposition thickness and the absence of catalytic effect from the substrate surface imply that only bulk effects are important: the photo- and thermochemical reactions take place in the icy bulk, including during the warm-up, and the ice/metal interface does not play any important role in the studied processes.

Discussion

The results reported in Table 1 and the study of the five experimental parameters (Results section) allow us to draw the conclusion that whatever the irradiation time, the temperature in the cryostat, the composition of the starting mixture, the photon dose and the substrate

used for the ice deposition, amino acids are always formed and detected in our organic residues. However, these results depend on the extraction solvent (water then ACN), and the amino acid quantities are very different for hydrolysed and non-hydrolysed fractions of the residues. In this section we analyse different parameters of the chemical analysis of the extracted residues, and make important remarks on the results obtained.

Extraction Solvent

All residues including the blank E0 were extracted first with water and then with acetonitrile (ACN), in order to be sure that all compounds in the residues, in particular the most hydrophobic ones, were dissolved. Although most of the molecules present in the organic residues are polar and thus soluble in water, amino acids such as glycine and serine were also found to be present in the ACN extracts. Table 3 gives the quantities of amino acids found in the water and ACN extracts, before and after hydrolysis, as well as the ratios between the quantities in both extracts.

The quantities measured in the ACN extracts are significantly smaller than those measured for the water extracts. Of course, this result is mostly due to the fact that the extractions with ACN were performed after those with water. In a general way, the ratios between the quantities measured for water and ACN extracts for the residues range from 1.2 to 41.7, showing that in some cases the amounts of amino acids contained in the ACN extract is negligible. When taking a deeper look, one can notice that these ratios are in average higher for E8~E13 experiments, irradiated at temperatures around 80 K, compared with those irradiated around 10 K (E2~E6). Only the blank sample E0 has a ratio smaller than 1, showing that most of the few contaminants are not polar. This tends to suggest that

Table 3 Amino acid quantities (in nmol) obtained in the water and ACN extracts, before and after acid hydrolysis, and ratios between the quantities in both extracts

Experiment	Water fraction (nmol)		ACN fraction (nmol)		Ratio
	Non-hydrolysed	Hydrolysed	Non-hydrolysed	Hydrolysed	Water/ACN[a]
E0	–	0.8	–	2.0	0.4
E1	–	41.5	–	3.5	11.8
E2	–	43.3	–	15.8	2.7
E3[b]	3.0	–	1.2	–	2.4
E4	2.1	18.5	0.2	5.6	3.6
E5	0.8	6.6	0.5	5.6	1.2
E6[c]	1.9	21.0	0.6	4.4	4.6
E7	–	5.3	–	2.3	2.3
E8	–	50.7	–	6.3	8.1
E9	–	30.0	–	2.0	15.4
E10	–	7.0	–	0.5	13.4
E11	–	18.3	–	1.2	15.5
E12	–	42.9	–	1.5	29.5
E13	–	16.3	–	0.4	41.7

[a] For E4~E6 samples, the ratios were calculated by summing up the quantities obtained before and after hydrolysis for each extract. [b] This sample was extracted with water then ACN, but was not hydrolysed. [c] For this sample, the reported quantities were obtained by summing up the quantities for the amino acids detected both on the Cu disc and the MgF_2 window.

Only E4~E6 samples were divided into hydrolysed and non-hydrolysed fractions.

the molecules present in the residues produced from ices irradiated at 80 K are more polar than those formed at 10 K, although the infrared spectra of the residues produced after irradiation of the ices at 10 and 80 K appear to be similar (Muñoz Caro and Schutte 2003).

Effects of the Acid Hydrolysis

The quantities reported in Table 1 were measured after acid hydrolysis of the extracted residues (except for E3 which was not hydrolysed), as it was also the case in the previous irradiation experiments (Bernstein et al. 2002; Muñoz Caro et al. 2002; Nuevo et al. 2006, 2007), and in the analysis of meteorites such as Murchison and Murray (Engel and Macko 1997; Cronin and Pizzarello 1997, 1999; Pizzarello et al. 2003; Meierhenrich et al. 2004). But some amino acids can also be detected before hydrolysis, as shown by the results obtained for E3~E6 samples (Table 3) and in some analyses of Murchison (see Shock and Schulte 1990, and references therein). The presence of these amino acids before hydrolysis shows that at least some of their formation processes occur during the photolysis in the ices at low temperature and/or during the warm-up, and/or during the extraction of the samples with the solvents.

Table 4 gives the total amino acid quantities measured before and after hydrolysis in the E4~E6 samples, as well as the ratios between these two quantities. For these three samples, the residues extracted with the solvents (water then ACN) were divided into two fractions, one to be hydrolysed with HCl as described in the experimental protocol (Ex-situ Analyses of the Residues section), the other fraction to be directly injected into the amino acid analyser without hydrolysis. The total amino acid quantity obtained for the E3 sample, extracted with water and ACN, and injected directly onto the column without hydrolysis, is also given for comparison. Thus, we could determine the order of magnitude of the proportion of amino acids formed/released during the hydrolysis step. Table 4 shows clearly that the non-hydrolysed fractions contain much less amino acids than the hydrolysed ones by a factor of 10, roughly constant for the E4~E6 samples, confirming that they are mainly formed during the hydrolysis step, and that the processes are similar for all samples.

In all cases, these results indicate that *most of the molecules present in the extracted residues before hydrolysis are different from the amino acids finally detected*. These compounds may be amino acid oligomers (linear peptides, cyclic peptides or diketopiperazines). They may also be amino acid derivatives (amino acid esters or amides, *N*-formyl or acetyl amino acids, etc.) or precursors, becoming amino acids after hydrolysis. Among all organic molecules, many candidates could play the role of amino acid precursors, in particular α-aminonitriles (involved in the Strecker synthesis of α-amino acids) and hydantoins.

Table 4 Total amino acid quantities (water plus ACN extracts, in nmol) obtained before and after acid hydrolysis

Experiment	Amino acids Before hydrolysis	Amino acids After hydrolysis	Hydrolysed/ Non-hydrolysed
E3	4.2	–	–
E4	2.3	24.1	10.7
E5	1.3	12.2	9.2
E6*	2.5	25.4	10.3

*Total amino acids found on both the Cu disc and the MgF_2 window.

This type of analysis was performed for the three samples E4~E6. The total amino acid quantity for the E3 sample, not hydrolysed, is also given for comparison.

These results can be compared with proteinaceous amino acid data from the Murchison carbonaceous chondrite. Table 5 gives some quantities measured for the three most abundant amino acids in the E4~E6 samples, namely glycine (Gly), alanine (Ala) and serine (Ser), before and after acid hydrolysis in the water extracts, as well as the quantities for the same compounds in 1 g of the Murchison meteorite (see Shock and Schulte 1990, and references therein).

The first result that can be drawn from Table 5 is that before hydrolysis, these three amino acids are produced with roughly the same abundances in our residues. This is not the case in Murchison, where glycine appears to be the most abundant amino acid before alanine and then serine. This same distribution is also displayed by these three amino acids after hydrolysis in Murchison and in our residues, where their abundance decreases with the increasing number of atoms in the lateral chain (see Distribution of the Amino Acids section for a detailed analysis of the amino acid distribution in our samples).

As mentioned in the Effects of the Acid Hydrolysis section, another important result is that most of the detected amino acids are formed/released after the acid hydrolysis of the residues, the amino acid quantities measured after hydrolysis being one order of magnitude higher than before hydrolysis (Table 4). Identically, more amino acids were detected after hydrolysis in the Murchison samples, but with smaller enhancement factors, from 2 to 5 (Table 5). In particular, meteoritic glycine was found to be only twice as abundant after hydrolysis as before.

These results suggest that the formation mechanisms of amino acids in our experiments – and therefore probably in the ISM – may be different from the processes that occurred in the Solar Nebula and/or in Solar System cold bodies such as asteroids and comets, i.e., the parent bodies of meteorites. One possible explanation is that organic molecules in these bodies are believed to experience contact with liquid water for periods of time that are short from an astrophysical point of view, but long enough for these compounds to be chemically processed before they reach the Earth. In this case, the organic fraction of extraterrestrial material collected on Earth may not be directly connected to the observed interstellar matter.

Remarks on the Detection of Some Amino Acids

We can make five important remarks on the identification of some amino acids in our samples:

1. To the best of our knowledge, this is the first time that threonine (Thr), glutamic acid (Glu), isoleucine (Ile) and leucine (Leu) have been detected in residues formed by the UV irradiation of interstellar ice analogs, taking all chromatographic techniques for analysis (liquid and gas) into account. However, their identification has to be confirmed by independent techniques such as GC-MS.
2. On the chromatograms, the peaks tentatively ascribed to tyrosine (Tyr) and histidine (His) are in fact probably due, at least in part, to other compounds eluting from the column with the same retention times (phenomenon of *coelution*). Indeed, if we look at the tyrosine and histidine molecular structures, which contain a phenyl ring with a hydroxyl group and a heterocycle with two nitrogen atoms, respectively, we can doubt that these complex amino acids were formed in such high amounts. An argument supporting our doubts is the absence of phenylalanine (Phe), an amino acid with a structure similar to tyrosine, but without the hydroxyl group on the phenyl ring, among the most abundant amino acids. Only trace amounts of phenylalanine were measured in eight of our residues. Concerning the nature of these coeluting compounds, the only

Table 5 Quantities (in nmol) measured before and after hydrolysis in the water extracts for the three most abundant amino acids (glycine, alanine and serine) in our residues, and comparison with the data obtained for different samples of Murchison (for 1 g of meteorite) (see Shock and Schulte 1990, and references therein)

	Before hydrolysis (nmol)		After hydrolysis (nmol)	
	Murchison[a]	Residues[b]	Murchison[a]	Residues[b]
Gly	25.5–45.3	0.5–0.7	13.3–102.7	3.7–27.5
Ala	15.9–23.6	0–0.1	12.3–90.7	0.5–20.3
Ser	0–1.9	0.1–0.7	0.9–9.0	0.3–2.0

[a] The data for Murchison are the minima and maxima reported in Shock and Schulte (1990). [b] The data for our experiments are the quantities measured for E4~E6 samples. For the E6 sample, the quantities are the sum of the quantities measured for both the Cu disc and the MgF_2 window.

certainty is that they are amino-bearing molecules since they react with ninhydrin (Amino Acid Analyses section). Nevertheless, since their retention time on the ion-exchange column is related to their isoelectric point, we can make further assumptions about their chemical nature. The compound coeluting with tyrosine (i.e., in the range of the neutral amino acids) probably bears an acidic function to compensate for the positive charge of the amine. The compound coeluting with histidine (i.e., in the range of the basic amino acids) might either be a monoamine or a diaminoacid (see the case of lysine/ethanolamine in remark 3). For all these reasons, the (small) contributions of the tyrosine and histidine peaks to the amino acid total quantities were taken into account in Table 1.

3. The peak tentatively ascribed to lysine (Lys) by the analyser is certainly also due, at least in part, to a coeluting compound. In that precise case, the molecule is probably ethanolamine ($NH_2CH_2CH_2OH$), an amino-alcohol derived from ethanol, likely to be formed easily in our experiments. The injection of a commercial sample of ethanolamine into the amino acid analyser showed that it does coelute with lysine. If its identification is confirmed, this could be the second detection of ethanolamine in organic residues formed by irradiation of interstellar ice analogs, after that reported by Bernstein et al. (2002). It should be noted, however, that the presence of lysine in our residues cannot be totally ruled out since Muñoz Caro et al. (2002) found in their residue two diaminohexanoic acids, which are constitutional isomers of lysine, with relatively high abundances.
4. Among all proteinaceous amino acids, when considering those containing a short lateral carbon chain, it can be surprising to find only small amounts of threonine (Thr). Indeed, threonine is a hydroxylated amino acid whose molecular structure is close to that of serine (having only one carbon more). Whereas serine was present in all the samples, threonine was detected in only a few samples, and never among the four most abundant compounds.
5. Finally, it is important to keep in mind that this study could only allow the detection and identification of 15 proteinaceous amino acids, among all similar compounds existing in nature, since the amino acid analyser used in the present work is optimized for such compounds (see Amino Acid Analyses section). It is thus probable that other simple non-proteinaceous amino acids, such as sarcosine, isovaline or 2,3-diaminopropanoic acid are also present in our residues. The actual complete distribution of the amino acids is therefore certainly different, and the comparison with the analyses of Murchison is in that sense limited to proteinaceous compounds. To detect and identify non-proteinaceous amino acids, other standards and other chromatographic techniques are needed.

Distribution of the Amino Acids

An important result we can draw from Table 1 is that whatever the experiment the most abundant amino acids present in the hydrolysates are roughly the same. In particular, there are only a few samples where glycine (Gly), alanine (Ala) and serine (Ser) are not among the most abundant amino acids. As far as the other compounds are concerned, if we take the restrictions mentioned about the presence of tyrosine, histidine and lysine into account (see Remarks on the Detection of Some Amino Acids section), the only amino acids which appear regularly are aspartic (Asp) and glutamic (Glu) acids, i.e., amino dicarboxylic acids. *This amino acid distribution only depends a priori on the molecular complexity of the compounds: the most abundant amino acid (glycine) is also the simplest one, and the abundances of the others decrease as their molecular weight and molecular complexity increase*. Alanine and serine, which contain only one carbon atom more than glycine, appear to be the most abundant amino acids after glycine. The experimental blank E0 contains minute amounts of amino acids, whose distribution is different from that displayed in the other samples. When comparing the total amino acid quantities measured in the blank E0 with those measured in the other residues, we can consider that contamination in the samples is negligible.

However, the four most abundant amino acids have random relative abundances, and it seems that there is no correlation between the abundance of one amino acid with respect to the others, or with respect to any of the five explored experimental parameters. If we look for instance at the glycine to alanine ratios (the two most abundant amino acids) measured after hydrolysis for all experiments (not including E3 which was not hydrolysed, see Table 6), we see that for all residues except E1, these ratios are higher than 1 and range from 1.2 to 8.6. Half of them range between 1.2 and 2.0, with no apparent correlation with any experimental parameter. Indeed, E2 and E4 samples, which have different relative proportions between the components of their starting ice mixtures, have close [Gly]/[Ala] ratios. But E5 sample, which was produced from the same mixture as E2 and using cyclic temperature for irradiation as E4, displays a [Gly]/[Ala] ratio three times higher. The E8 sample displays a ratio of 1.5, close to

Table 6 Ratio between the total quantities (water plus ACN extracts) measured for glycine and alanine for each residue after hydrolysis (E3, whose extracted residue was not hydrolysed, is not reported here)

Experiment	Irradiated mixture H_2O:CO:CO_2:CH_3OH:CH_4:NH_3	*T* (K)	Substrate	[Gly]/[Ala]
E0	0:0:0:0:0:0	81	MgF_2	2.3
E1	2:1:0:1:1:1	11	MgF_2	0.8
E2	1:2:0:0:1:1	11.5	MgF_2	1.2
E4	2:1:0:1:1:1	10–60	MgF_2	1.5
E5	1:2:0:0:1:1	10–60	MgF_2	4.5
E6	2:1:0:1:1:1	19	drilled Cu	3.8
			MgF_2	2.9
E7	6:0:0:2:0:1	81	MgF_2	4.1
E8	0:0:0:1:0:1	81	drilled MgF_2	1.5
E9	0:0:0:1:0:1	81	MgF_2	2.0
E10	6:0:0:2:0:1	82	drilled MgF_2	8.6
E11	1:0:3:1:0:1	83.5	MgF_2	2.0
E12	1:0:1:1:0:1	81.5	MgF_2	1.7
E13	3:0:1:1:0:1	81	MgF_2	1.7

those calculated for E2 and E4, but with a completely different starting mixture (it contains only one source of carbon – CH_3OH – and no water), irradiated at a temperature of 81 K, and using a drilled MgF_2 window as a substrate. Finally, alanine appears to be the most abundant amino acid in E1 ([Gly]/[Ala] = 0.8). However, the comparison with the E4 and E6 samples shows that this result is not related to the starting ice mixture composition.

Another indication that the substrate for the ice deposition has no effect on the amino acid distribution is that both E8 and E10 samples, where drilled MgF_2 windows were used, display very different [Gly]/[Ala] ratios of 1.5 and 8.6, respectively. One can also notice that the E6 sample, where the Cu disc was used as a substrate, displays a [Gly]/[Ala] ratio of 3.8, different from most of the ratios calculated for experiments where a regular MgF_2 window was used, except for E7 (residue ratio of 4.1). This shows again the absence of clear correlation between [Gly]/[Ala] ratios and the substrate.

Therefore, *there is no significant link between the relative abundances of the formed amino acids, the starting ice mixture composition, the temperature of irradiation and the substrate used for deposition*.

Finally, the amino acid distribution found for the E3 sample, whose residue was not hydrolysed before injection into the amino acid analyser, was found to be very different from the others. The presence of tyrosine and histidine, or more probably other simple compounds eluting from the column with the same retention times (see Remarks on the Detection of Some Amino Acids section), among the four most abundant amino acids, as well as their small quantities (even for glycine and serine) compared with other samples, show again that most of the amino acids detected here are formed and/or released during the hydrolysis step.

The distribution of the amino acids detected in our experiments, in particular that of the most abundant compounds, can be compared with those obtained for previous irradiations of ices where liquid chromatographic techniques were used for the chemical analysis of the samples. The results obtained by Bernstein et al. (2002) show a lower variety of amino acids formed from the UV irradiation of a H_2O:CH_3OH:NH_3:HCN = 20:2:1:1 ice mixture. However, the three amino acids detected in their hydrolysed residue are the three most abundant produced in our samples, namely glycine, alanine and serine.

The analysis of organic residues with gas chromatographic techniques, such as GC-MS, does not allow the detection of large quantities of serine, as already shown by Muñoz Caro et al. (2002). Moreover, we mentioned in Remarks on the Detection of Some Amino Acids section that the peak ascribed to lysine in our chromatograms is probably due to ethanolamine, which may be present in our samples. In the chromatogram published by Bernstein et al. (2002), the fourth identified compound (besides glycine, alanine and serine) is precisely ethanolamine, a molecule that was only detected with liquid chromatography.

These results tend to show that the correlation between the distribution of the detected amino acids and the experimental conditions of irradiation is not straightforward. They also show that the distribution depends to a certain extent on the chromatographic technique (liquid or gas phase) used for the chemical analyses of the residues. In particular, compounds containing an alcohol function (OH) such as serine, threonine or ethanolamine seem to be more easily and efficiently detected in the liquid phase than in the gas phase, certainly because of a weak volatility of the derivatives of these compounds necessary for the analysis with gas chromatography.

Astrochemical Considerations

With regard to the results reported in this work, the production of complex organic molecules is an efficient process if the ice mixture contains the four elements essential to

build such molecules, namely C, H, O and N. In the ISM, such dirty ices, which are largely observed towards protostars, will indeed undergo UV photon irradiation as it is known that molecular clouds are continuously permeated by such UV radiation originating from the interaction of cosmic rays with H_2 molecules (Prasad and Tarafdar 1983).

To what extent UV irradiations in astrophysical media are comparable with these laboratory simulations is unknown. For a given molecular mixture, the production of organic species will depend essentially on the UV photon flux. In our experiments, we saw that photon doses per deposited molecule of about 0.05 are enough to produce amino acids and/or their precursors in our samples (see Photon Dose Per Molecule section), and certainly many other organic species which were not searched for in the present work.

Mathis et al. (1983) showed that 60 h of irradiation in the laboratory using a H_2 discharge lamp similar to the one used in the present work are equivalent, in terms of photon dose in the energy range of photochemistry, to 10^5 years of irradiation in the diffuse medium and from 10^8 to 10^9 years in the dense medium, assuming a UV photon flux for the diffuse ISM of 8×10^7 photons cm^{-2} s^{-1}, for $E_{photons} \geq 6$ eV. In our experiments, the irradiation times ranged from 18 to 50 h, i.e., the integrated UV photon doses were of the same order of magnitude as during the lifetime of a molecular cloud. However, these numbers are certainly lower limits since matter can recycle closer to new-born stars and undergo further UV photon irradiation.

The formation of complex organic molecules, in particular amino acids and/or their precursors, at the surface of cold grains may therefore be an efficient process in interstellar clouds and protostars. These organics could then be incorporated into small bodies such as asteroids and comets in the Solar System, and collected on Earth after being delivered by meteorites, IDPs and/or micrometeorites (Maurette 1998). However, since (micro) meteorites are not pristine bodies, the nature of the organics identified in such extraterrestrial material is different from what is observed in the ISM and laboratory simulations. Cometary ices are pristine material and should therefore be more relevant for the study of the origins of life. In this connection, the European ROSETTA mission, which will analyse the organic composition of cometary ices *in-situ* with an enantioselective gas chromatography coupled with mass spectrometry (GC-MS) technique (Thiemann and Meierhenrich 2001), will be of great interest.

Amino acids are the organic molecules which have been most searched for in meteorites such as Murchison (see Shock and Schulte 1990, and references therein), but their meteoritic distribution is different from what is observed in laboratory simulations (Bernstein et al. 2002; Muñoz Caro et al. 2002; Nuevo et al. 2006, 2007; Effects of the Acid Hydrolysis and Distribution of the Amino Acids sections). In addition, meteoritic chiral amino acids were found to display measurable L enantiomeric excesses (Cronin and Pizzarello 1997, 1999; Engel and Macko 1997, 2001; Pizzarello et al. 2003), whereas only biological processes were previously known to produce enantiomeric excesses. Several scenarios were proposed to explain how such excesses could be produced in space (Jorissen and Cerf 2002). Circularly polarized UV light (UV CPL) appears to be a good candidate (Rubenstein et al. 1983), and laboratory experiments using such a circularly polarized UV light as a chiral inductor were performed (Flores et al. 1977; Bonner and Bean 2000; Meierhenrich et al. 2005; Nuevo et al. 2006). These experiments could not lead to conclusive results about the role of UV CPL on the production of non-racemic molecules under interstellar conditions. Therefore, further studies are required to understand the link between the organic matter from an interstellar origin, and the origin of an enantiomeric excess that is required for an evolution towards the homochirality observed in biological processes on Earth.

Conclusion

The chemical analyses of several organic residues formed by the VUV irradiation of interstellar ice analogs under different experimental conditions and subsequent warm-up to room temperature clearly indicate that amino acids, chosen as significant complex organic molecule tracers, *are always formed* after the acid hydrolysis of the residues. It was shown that the temperature, the irradiation time and the substrate have no significant effect on the distribution of the amino acids detected after chemical analysis of the residues. However, the relative proportions of the ice mixture components may affect the absolute total amino acid quantities in the residues, and the photon dose may have an effect on their final distribution.

According to the present work, the production of complex organics is an efficient process when the ice mixture contains C, H, O and N. Moreover, the irradiation times in our experiments are compatible in terms of photon dose to what is expected for a molecular cloud during its lifetime, where the irradiation of interstellar grains can initiate the photochemical reactions leading to complex interstellar molecules in such clouds and protostars.

The other important result of this study is that most of the analysed residues display similar amino acid distributions. The most abundant compounds are also the simplest ones, and their abundances decrease with molecular weight. Such an amino acid distribution is expected for organic compounds formed via abiotic processes from the starting ices. Finally, among the amino acids identified in this work, threonine, glutamic acid, isoleucine and leucine were tentatively detected for the first time in such organic residues formed from the VUV photo-processing of ices in the laboratory, but their identification has to be confirmed by independent chromatography techniques.

Nevertheless, the distribution of the amino acids in our experiments is not in agreement with the one displayed in meteorites such as Murchison, indicating either that their formation mechanisms are different, or that organic molecules in the Solar System underwent thermal and/or chemical alteration, leading to a different final distribution.

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