

THE PERSEUS EXOBIOLOGY MISSION ON MIR: BEHAVIOUR OF AMINO ACIDS AND PEPTIDES IN EARTH ORBIT

F. BOILLOT¹, A. CHABIN¹, C. BURÉ¹, M. VENET², A. BELSKY³,
M. BERTRAND-URBANIAK¹, A. DELMAS¹, A. BRACK¹ and B. BARBIER^{1*}

¹ Centre de Biophysique Moléculaire, Rue Charles Sadron, 45071 Orléans Cedex 2, France;
² CNES, DSO ED US, BP 22–21, 18 Avenue E. Belin, Toulouse, France; ³ Centre Lasers Intenses et Applications, 351, Cours de la Libération, 33405 Talence Cedex, France
(* author for correspondence, e-mail: barbier@cnrs-orleans.fr; fax: +33 238 631517)

(Received 2 March 2002; accepted in revised form 22 May 2002)

Abstract. Leucine, α -methyl leucine and two peptides were exposed to space conditions on board the MIR station during the Perseus-Exobiology mission. This long duration space mission was aimed at testing the delivery of prebiotic building blocks. During this mission, two amino acids (leucine and α -methyl leucine) and two peptides (leucine-diketopiperazine and trileucine thioethylester) were exposed in Earth orbit for three months. Basalt, clay and meteorite powder were also mixed with the samples in order to simulate the effects of potential meteorite protection. Analysis of the material after the flight did not reveal any racemization or polymerisation but did provide information regarding photochemical pathways for the degradation of leucine and of the tripeptide. Amino acids appeared to be more sensitive to UV radiation than peptides, the cyclic dipeptide being found to be as particularly resistant. Meteorite powder which exhibits the highest absorption in Vacuum UltraViolet (VUV) afforded the best protection to the organic molecules whereas montmorillonite clay, almost transparent in VUV, was the least efficient. By varying the thickness of the meteorite, we found that the threshold for efficient protection against radiation was about 5 μ m. The possible exogenous origin of biological building blocks is discussed with respect to the stability to the molecules and the nature of the associated minerals.

Keywords: amino acids, Earth orbit, exposure in space meteorites, origin of life, photochemistry

1. Introduction

Since the pioneer statement of Oro (1960), the search for primitive building blocks of life has progressively led scientists to consider space as a possible source. About 70 organic molecules have been identified in the interstellar medium by radioastronomy, including many chemical precursors of the biological building blocks such as H₂O, HCN, NH₃, CH₄, CO₂, HCHO or CH₃CN (Oro, 1996). Recently, interstellar glycolaldehyde, a precursor of sugars, was detected in the Milky Way (Hollis *et al.*, 2000). These molecules are probably embedded in the icy mantle of interstellar grains. Greenberg modeled such grains and found a good correlation with the measured spectral signature of interstellar grains (Greenberg, 1984; Greenberg *et al.*, 1995). Under interstellar radiation, some quantum organic chemical reactions (i.e. via tunneling effect) can take place either within the icy film surrounding the



grains or on sublimated gaseous molecules around the grains. Some proton and cation transfer could occur to produce more complex organic molecules that would be subsequently trapped back into ice (Charnley, 1997).

Roughly 40 000 tons of extraterrestrial organic matter impact the Earth upper atmosphere each year (Love and Brownlee, 1993), of which interstellar dust particles (IDP) represent the main fraction. The flux of carbon about 4 billion years ago, during the period of heavy bombardment, has been estimated at about 10^8 kg yr⁻¹ (Chyba and Sagan, 1992). A large collection of micrometeorites extracted from Antarctic old blue ice was studied by Maurette (Maurette, 1998). The size fraction ranging from 50 to 400 μ m can strike the Earth without damage and delivers an organic fraction which is about 100 000 times more important than that delivered by meteorites (>1 cm). Larger particles burn during the atmospheric entry while the smaller ones, of a few micrometers in size, are probably sensitive to UV radiation which destroys the organic fraction.

Cometary grains are small icy particles released by comets. Ground-based analysis indicates that comets have an organic-rich composition. Artificial cometary grains exposed in Earth orbit onboard the EURECA facility showed a spectral signature close to that reported for the interstellar medium (Greenberg *et al.*, 1995). The Giotto and Vega mass spectrometer *in situ* analysis of the comet Halley showed that organic matter could represent up to 25% of the total composition. Formaldehyde and methane could represent 4 and 5%, respectively, of the cometary organic mass and hydrogen cyanide has been estimated up to 7% (Chyba and Sagan, 1992; Oro, 1996).

Meteorites are relatively large bodies. Among the meteorites, carbonaceous chondrites contain up to 5% of carbon in mass. The Murchison meteorite, a CM2-type chondrite which fell in 1969 in Australia, revealed about 500 organic compounds including nucleic bases (Stoks and Schwartz, 1981) sugar-related organic compounds (Cooper *et al.*, 2001) and about 80 amino acids, 8 of them being proteinaceous (Cronin and Moore, 1971; Jungclaus *et al.*, 1976; Kvenvolden *et al.*, 1970, 1971; Oro *et al.*, 1971). Thus, building blocks of life can reach the Earth without damage when carried by meteorites. Excesses of L-enantiomers up to 9.2% of several C α -disubstituted amino acids (NH₂-CRR'-COOH) were recently found in the Murchison and Murray meteorites (Pizzarello and Cronin, 2000). These amino acids are not proteinaceous and cannot, therefore, be the result of terrestrial contamination. On the other hand, the corresponding C α -monosubstituted amino acids (NH₂-CHR-COOH) are present as racemic mixtures. C α -monosubstituted amino acids probably racemized via C α -hydrogen atom abstraction whereas the chemical structure of disubstituted amino acids prevents them from such a reaction.

We exposed amino acids in Earth orbit to test the hypothesis of a safe delivery of amino acids to the Earth. During the orbital missions Biopan 1 and Biopan 2, amino acids and short peptides were exposed to solar radiation for about 10 days. Preferential degradation of acidic amino acids occurred. Some of the activated amino acids polymerized under radiation but no racemization was observed (Barbier *et*

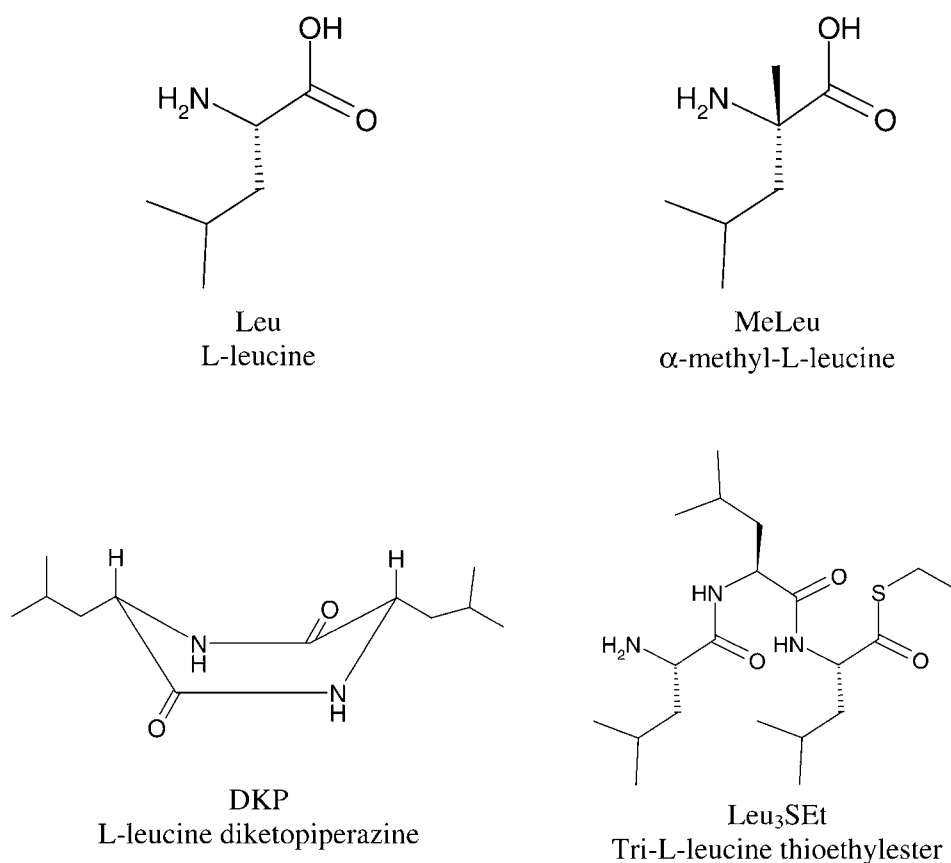


Figure 1. Chemical structure of the samples exposed during the Perseus mission.

al., 1998; Hénin, 1998). The opportunity for further investigation during a 99 day duration flight was offered in 1999 onboard the Russian MIR Space station.

The Perseus mission was launched on 20 February 1999 with a Soyuz TM-29 from Baikonour (Kazakhstan) with three astronauts onboard. The scientific material was carried to MIR with a Progress cargo on 2 April. The Exobiology experimental hardware (COMAT, Toulouse) was installed outside the station on the Kvant 2 module on 16 April 1999.

The Perseus-Exobiology experiment was run from 16 April to 23 July 1999. For 97 days, two amino acids (L-leucine, Leu, and α -methyl L-leucine, MeLeu), one cyclic dipeptide (L-leucine diketopiperazine, DKP) and one activated tripeptide (tri-L-leucine thioethylester, Leu₃SEt), were exposed to space conditions outside MIR (Figure 1).

The aim of the experiment was to study the stability and the resistance to racemization using the two amino acids Leu et MeLeu, the chemical stability of the peptide bond using the two peptides DKP and Leu₃SEt and a possible

photochemically-induced polymerization using the activated tripeptide Leu₃SEt. L-leucine, an amino acid present in the Murchison meteorite, was selected for the long duration Perseus Exobiology mission because it exhibited good UV stability during the Biopan flights. α -methyl-L-leucine (MeLeu), a C _{α} -disubstituted amino acid, was exposed to compare its behavior with that of L-Leu, the monosubstituted analogue. The stability of the peptide bond was checked with the two peptides. The activated tripeptide tri-L-leucine thioethylester (Leu₃SEt) was chosen to study possible oligomerization. A tripeptide was chosen in order to limit intramolecular cyclisation and sublimation. The thiol ester was selected with regard to the key role that it might have played in early metabolism (DeDuve, 1998; Huber and Wächtershauser, 1997, 1998). The cyclic dipeptide L-leucine diketopiperazine (DKP) is considered as a possible storage form of the linear peptide. DKP, which is generally considered as an unproductive by-product in amino acid polymerization, can, nevertheless, be opened into its linear form when reacting on mineral surfaces (Bujdak and Rode, 1999).

The samples were exposed in two modes: unprotected (free form) and associated with different ground mineral compounds to mimic micrometeorites, i.e. montmorillonite clay, powdered basalt and Allende meteorite. Typically, 5 μ m thick mineral films were used. In the case of Leu, DKP and Leu₃SEt, different thicknesses of meteoritic deposits were used to determine the protection threshold.

Clays, and particularly montmorillonites have already shown their ability to catalyze the synthesis and the polymerization of amino acids (Bujdak *et al.*, 1995; Bujdak and Rode, 1997, 1999; Ferris *et al.*, 1996; Paecht-Horowitz, 1977; Paecht-Horowitz and Lahav, 1977). This mineral was expected to protect the samples and to induce the oligomerization of amino acids and peptides.

Basalt is a effusive magmatic rock with a microlithic structure, consisting of microscopic crystals of plagioclase, pyroxene and olivine. The later two minerals can be found in meteorites. To some extent, basalt can chemically mimic meteorites (Jarosewich, 1990; Jurewicz *et al.*, 1991). The 4.6 billion years old Allende meteorite is a CV3 carbonaceous chondrite which fell in 1969 in the Chihuahua region, Mexico. It is composed of silicates, iron (II) oxide and magnesium (II) oxide. The organic fraction of Allende has been analyzed and the amount of amino acids detected does not exceed 1% of the amount measured in CM2 (Cronin and Moore, 1971; Cronin and Pizzarello, 1983). It was selected to mimic the mineral fraction of a meteorite.

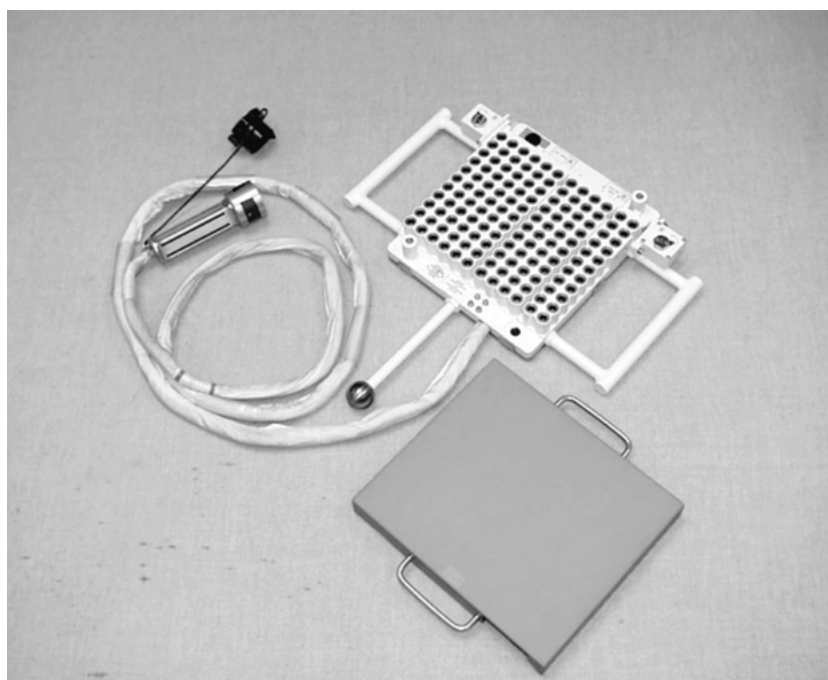


Figure 2. Experimental exposure device (plate: 620 × 490 × 110 mm).

2. Material and Methods

2.1. EXPERIMENTAL SETUP

The experimental setup consisted of a large piece of anodized aluminum (Figure 2).

The two opposite faces were been hollowed out into 66 symmetrical cells. A system of ventilation enabled each row of 11 cells to be evacuated or filled with an inert gas through 0.22 μm filters to prevent cross contamination. To each cell a sample holder was attached with a magnesium fluoride (MgF_2) window glued on top (Epotek E501, G-Brot, Paris). MgF_2 permits a maximum irradiation (120–7000 nm) of the samples deposited as solid films on the cavity-facing side of the window. Moreover, MgF_2 is a resistant material with a dilation coefficient identical to that of aluminum.

The outer surface of the hardware was covered with white silicon, light-reflecting paint. Temperature sensors installed inside two opposing cavities measured the temperature in both exposed and light protected cells. The radiation flux was recorded outside the cavities by two UV sensors.

2.2. EXPOSED SAMPLES

The two amino acids, L-leucine and L-methyl leucine, were purchased from Sigma, St Louis and Novabiochem, respectively. The amino acids were desalted in order to obtain the zwitterionic forms, known to be less sensitive to sublimation and photolysis. The two peptides, DKP and Leu₃SEt, were synthesized in our laboratory.

All the compounds were exposed as 0.5 μm solid films. The films were prepared by dissolving about 0.2×10^{-6} mole of each compound in water or in a 3/7 (v:v) mixture of water/methanol. 100 μL of the solutions were dropped into the cavity facing side of each window and dried under vacuum over phosphorus pentoxide. A first outgassing was achieved under 80 mm Hg for one hour before increasing the vacuum by steps of 20 mm Hg. After complete evaporation, the samples were stored under a dry nitrogen atmosphere before being fixed onto the hardware at the very last moment. The thickness of the films was calculated from the molecular volume of each species loaded. No physical measurement of the thickness was made. The samples were exposed in triplicate.

The montmorillonite used was a generous gift from James Ferris (RPI, Troy, U.S.A.). The basalt sample was collected in the Massif Central, France. After being crushed, the powder was washed with HPLC grade water to remove all UV absorbing compounds and filtered. The finest size fraction <20 μm was selected by settling a suspension in water and dried by lyophilization. The same protocol was used for a fraction of the Allende meteorite which was purchased from 'The Meteorite Market' (<http://www.alaska.net/~meteor/>).

2.3. FLIGHT PARAMETERS

During solar irradiation in Earth orbit, the compounds were exposed to three major parameters: vacuum, temperature and radiation.

The interplanetary vacuum has been estimated at 10^{-14} Pa. In the neighborhood of a satellite, a natural outgassing of the vessel creates a residual atmosphere that decreases the vacuum quality. The residual pressure measured in the vicinity of the shuttle was found to be in the range of 10^{-4} to 10^{-6} Pa (Horneck and Brack, 1992; Horneck *et al.*, 1996). During the Perseus mission, the vacuum was not measured.

The temperature of a body in Earth orbit depends on its surface, size and mass. It absorbs and emits energy. Different energy sources operate in Earth orbit: solar radiation (1371 W m^{-2}), Earth's albedo (480 W m^{-2}) and terrestrial radiation (230 W m^{-2}). Thus, a body can experience extreme temperatures depending on its position. The temperature was recorded every 5 min during the mission. The temperature inside the cavities varied between $-14 \text{ }^\circ\text{C}$ and $+44 \text{ }^\circ\text{C}$. Temperature fluctuations followed the orbiting of the station. As the MIR station orbits in 90 min, it is exposed to the sun during 45 min and remains in the dark for the remaining 45 min. Thus, alternating high and low temperature peaks were reached

every 45 min ($-10\text{ }^{\circ}\text{C}$, $+44\text{ }^{\circ}\text{C}$) with a $5\text{ }^{\circ}\text{C}$ difference between the exposed and dark cavities.

Earth orbit is subjected to intense radiation of both solar and galactic in origin. Cosmic radiation entering the solar system is composed of protons, electrons, α -particles and heavy ions. Solar particle radiation is emitted as solar wind during chromospheric eruptions. It is composed of up to 95% of protons, α -particles and heavy ions. Protons and electrons do not reach Earth vicinity, being trapped by the geomagnetic field.

The solar electromagnetic radiation above the Earth atmosphere is $1371 \pm 10\text{ W m}^{-2}$ (Anderson and Smith, 1994). This value is defined as one solar constant (1 SC). The radiation is composed of 45% infrared radiation, 48% visible light and only 7% UV light. Most of the biological compounds absorb in the VUV domain, below 190 nm, where photons are the most energetic and are, therefore, able to induce photochemical damages.

Two UV sensors were mounted onboard the Mir station (silicon photodiode 190–1000 nm HAMAMATSU S5226-8BQ). The UV dose received by the samples was recorded in two range of wavelengths. A 260 nm filter (UV-MAZ 8, Schott) and a 265–385 nm filter (8max 333.6 nm, DUG 11X, Schott) were used. Nevertheless, the sensitivity of the detectors and the broad band of the filters were not sufficient to record the total amount of radiation received during exposure. This was due to the low transmission of the filters and to the high UV dose received which saturated the filters and truncated the measurements. The UV dose was estimated by considering the solar spectrum recorded during previous missions such as Spacelab, Eureca (Heroux and Hinteregger, 1978; Mecherikunnel, 1996; Nicolet, 1989) or UARS-SUSIM (Brueckner *et al.*, 1993; Reber *et al.*, 1993). The full solar exposure time was deduced by summing up all the periods of high temperature. The total irradiance received for a 1045 day exposure was estimated at $5.16 \times 10^9\text{ J m}^{-2}$. The UV flux deduced is $2.9 \times 10^{21}\text{ photons cm}^{-2}$. It has been estimated that this photon dose corresponds to the dose received during a 0.93 million-year trip in diffuse interstellar clouds (Greenberg, 1995).

2.4. GROUND BASED SIMULATION

The temperature and vacuum conditions as recorded during the flight were simulated in the laboratory. $0.5\text{ }\mu\text{m}$ thick films of each sample were loaded onto MgF_2 window as for the flight. They were subjected for 10 days to high vacuum (10^{-4} mbar) and to the highest temperature conditions experienced by the in-flight samples during 10 days, i.e. 2.5 days at $34\text{ }^{\circ}\text{C}$, 2.5 days at $35\text{ }^{\circ}\text{C}$, 2 days at $36\text{ }^{\circ}\text{C}$, etc. (Table I).

2.5. SPECTRAL REFERENCES

Amino acids and peptides absorb light in the VUV region below 200 nm. Absorption band spectra were already available for some amino acids and oligopeptides

TABLE I

Cumulated time at a given temperature, above 34 °C, experienced on board MIR for 10.5 days

Temperatures (°C)	34	35	36	37	38	39	40
Cumulated time (days)	2.5	2.5	2	1.5	1	0.5	0.5

in the solid state but not for the samples exposed in the Perseus Exobiology experiment (Inagaki, 1973; McMillin *et al.*, 1973). Spectra of 0.5 μm films of Leu, MeLeu, DKP and Leu₃SEt were recorded using the synchrotron radiation of the SuperAco storage ring at LURE – Orsay, France. The SA61 beamline was used with a 3 m long Balzers-type monochromator and a 300 lines mm^{-1} network. The spectral range was between 30 and 1200 nm and the resolution of this network was 0.1 nm. Samples on MgF₂ windows were fixed perpendicularly to the beam. The transmitted radiation was measured by a photomultiplier counting photon impacts. The beam has been defocused such that a 3 mm^2 rectangular beam could pass through the sample. Thus, potential photolysis phenomena were avoided and the recorded spectra were more representative of a surface than of a single point. Several spectra were obtained from three different parts of the sample by moving the sample in front of the incident radiation beam. The averaged spectra obtained were considered as representative of the surface state of the samples. The spectra are represented on Figures 3 and 4.

The absorption of Leu, MeLeu, DKP and Leu₃SEt is mainly due to the -CO-X group where X can be O, N or S. For amino acids, three bands were identified at 208, 184 and 163 nm, corresponding to the $n-\pi^*$, $\pi_1-\pi^*$ and $\pi_2-\pi^*$ electronic transitions, respectively (Inagaki, 1973; McMillin *et al.*, 1973). The increase in absorption between 160 and 120 nm is due to the excitation of σ electrons of C–C and C–H bonds and to the photoionization process (Dodonova, 1993). Unfortunately, the 208 nm band is not visible on a 0.5 μm film of Leu or MeLeu. For peptides, two maxima at 140 and 190 nm were attributed to the absorption of the amid groups by comparison with acetamide spectrum (Dodonova, 1993; McMillin *et al.*, 1973). For Leu₃SEt, an additional absorption band at 234 nm was attributed to the absorption of the thioester group of Leu₃SEt.

The VUV spectroscopic studies of the compounds exposed during the Perseus Exobiology experiment demonstrated that the 0.5 μm sample deposits were transparent in the 120–250 nm range, thus indicating that the solar radiation traversed the entire thickness of each exposed unprotected sample. The spectroscopic studies also confirmed that the -CO-X absorbing group was the main photochemical target in the space irradiation experiments.

Spectroscopic studies were also made on mineral films. The shielding effect of minerals was studied on 5 μm thick films of montmorillonite clay, basalt powder

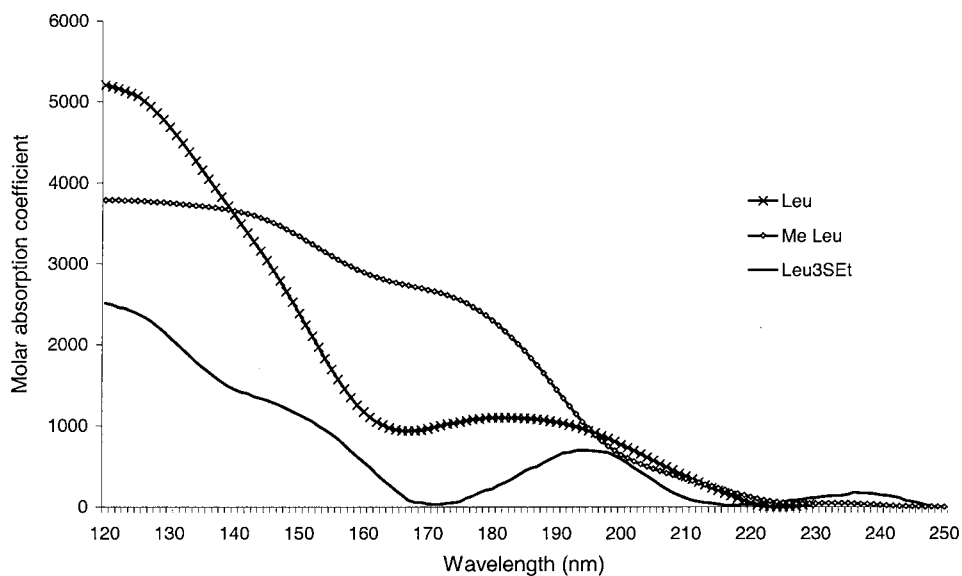


Figure 3. VUV-UV spectra of leucine, α -methyl leucine and trileucine thioethylester in the solid state ($0.5 \mu\text{m}$ thick film on MgF_2 window).

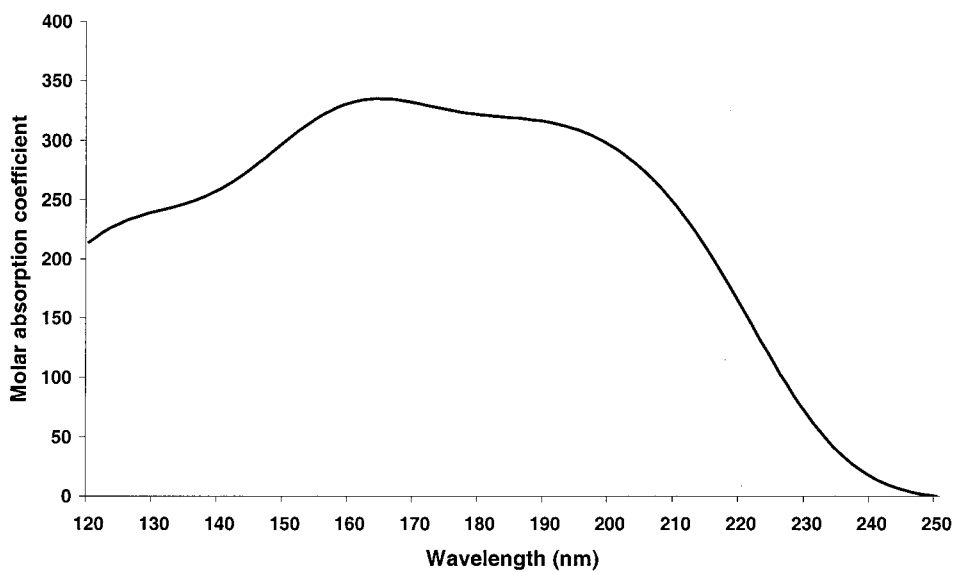


Figure 4. VUV-UV spectra diketopiperazine of leucine, in the solid state ($0.5 \mu\text{m}$ thick film on MgF_2 window).

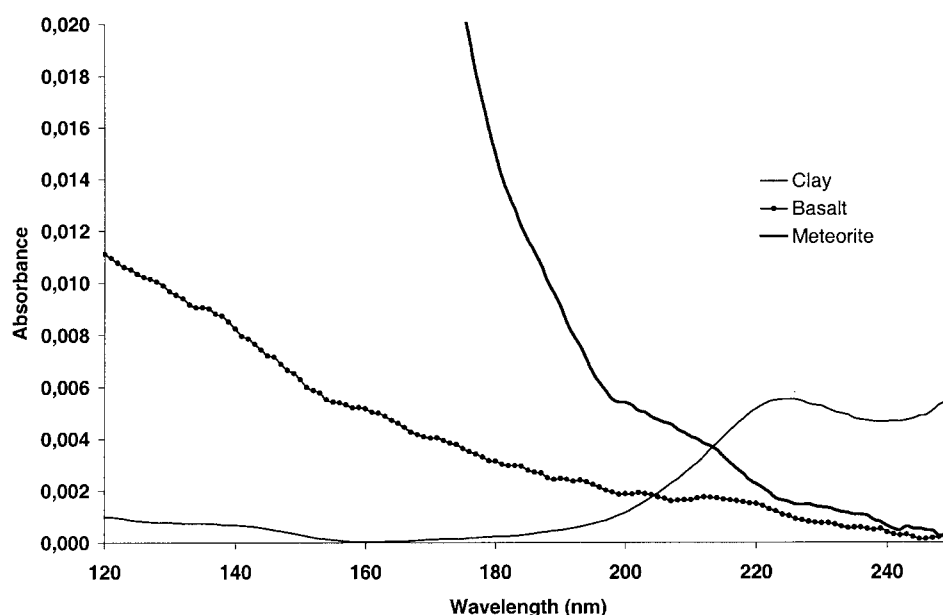


Figure 5. Absorption spectra of 5 μm thick films of minerals in the VUV – UV region.

and powered Allende meteorite. The powders were studied with synchrotron radiation in the range of 120–250 nm. The energy of the beam was adjusted to measure 10 000 photon impacts at 170 nm for each sample. The spectra were recorded with respect of significant transmission over the whole spectrum. The spectra were plotted as described for organic films and the average transmission has been considered to be representative of an homogenous 5 μm thick film. The results are presented in Figure 5.

The meteoritic powder exhibits the most important absorption in the 120–250 nm range, whereas basalt powder shows a similar but lower absorption suggesting a lower protective potential compared to the meteorite. The absorption spectrum of the clay revealed different spectral properties since it absorbed in UVC domain and was quite transparent in VUV. Photons corresponding to the shorter wavelengths are more energetic and therefore more destructive for organic compounds, thus, montmorillonite was considered as the least protective mineral. These could be explained by structural differences since the clay forms organized layers whereas basalt and meteoritic powder are just aggregates of grains, better able to scatter incident light. Regarding the spectral properties, meteoritic powder is expected to be the best shield against UV light.

2.6. SAMPLE RECOVERY AND ANALYTICAL TECHNIQUES

The infrared spectra of the solid samples were obtained from the MgF_2 windows, immediately before and after space exposure, with a Spectrum 1000 FT-IR ap-

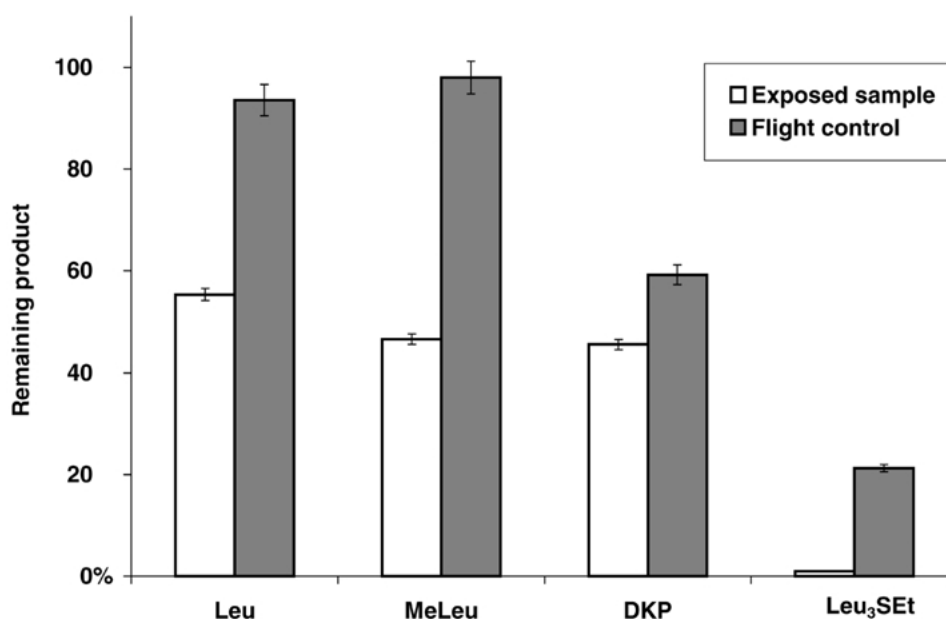


Figure 6. Remaining product after Earth orbit exposure.

paratus from Perkin Elmer. Infrared spectroscopy reveals which groups of the molecule have been damaged during solar exposure.

Samples were recovered by several washings of the windows with water and with a methanol/water mixture (7/3). The washing solutions were put into Eppendorf tubes and evaporated under reduced pressure and over P_2O_5 . They were stored at $-20\text{ }^\circ\text{C}$ until analysis. Prior to analysis, the samples were dissolved in water and a small volume was subsampled. Analysis of the remaining organic compounds associated with minerals was performed as for free samples. The mineral fraction was not extracted and water was added to dissolve samples prior to centrifugation. HF treatment (Stevenson and Cheng, 1970) did not extract remnant molecules eventually retained by clays.

Each sample was analyzed by capillary electrophoresis. Each analysis was repeated three times and was calibrated with standard solutions. The amounts of samples recovered after the flight in the exposed and the dark side cavities are reported in Figure 6.

Two capillary electrophoresis methods were optimized to separate amino acids and their oligomers, as well as their esterified derivatives. The amino acids, as well as the products of polymerization and degradation of the amino acids and peptides, were detected by UV at 200 nm. The sensitivity of capillary electrophoresis is of the order of the picomole. This technique allows the use of very small volumes of samples ($2\text{ }\mu\text{L}$) and, thus, numerous analyses for a given sample.

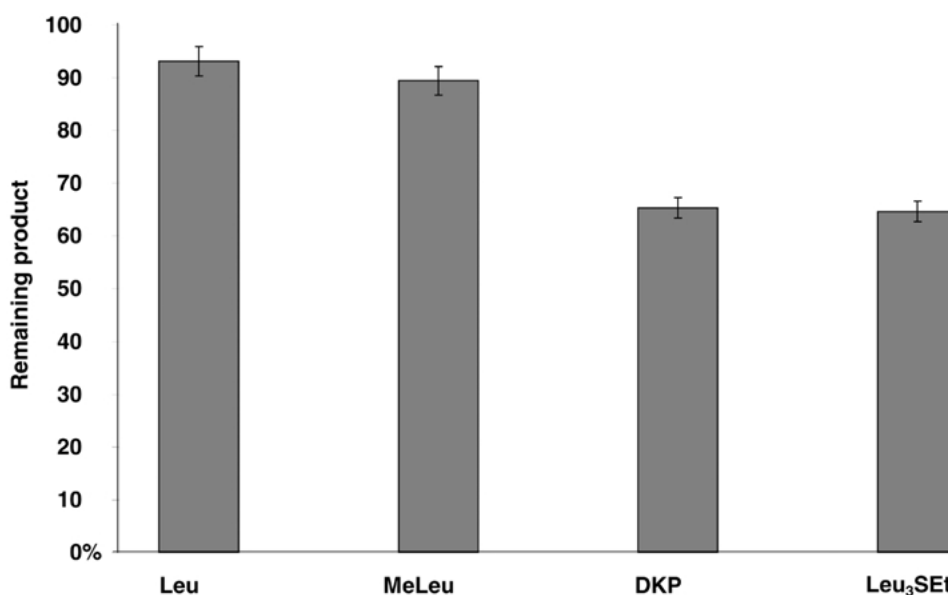


Figure 7. Simulation experiment. Remaining products after 10.5 days as the highest temperatures recorded during Perseus – Exobiology experiment.

Electrospray mass spectrometry (VG Quattro II mass spectrometer, Fisons Instruments) was used to characterize the remaining compounds and their by-products. This apparatus allows the detection of positive and negative ions in a mass range of 1 to 4000 m/z . The concentration of the sample was up to 100 pmol μL for an injection of 3 μL . The accuracy of the mass measurement was 0.03%.

3. Results

3.1. SURVIVAL OF UNPROTECTED SAMPLES

After 97 days of exposure in Earth orbit, the sample films were recovered without significant damage or broken windows. All exposed samples turned yellow after the flight. The loss of material was measured in both the dark and exposed cavities.

As reported on Figure 6, the unprotected amino acids, Leu and MeLeu, were fully recovered in the dark cavities whereas a loss of about 50% was measured in the exposed ones indicating that solar radiation was the main degradation factor. Capillary electrophoresis analysis did not reveal any new products. For the peptides, a deficit was observed in the dark cavities and was attributed to some sublimation of the molecules under vacuum. The deficit reached about 40% for DKP and 80% for Leu₃SEt. For DKP, the losses observed in space and those measured in the ground based experiments are in good agreement (Figures 6 and 7).

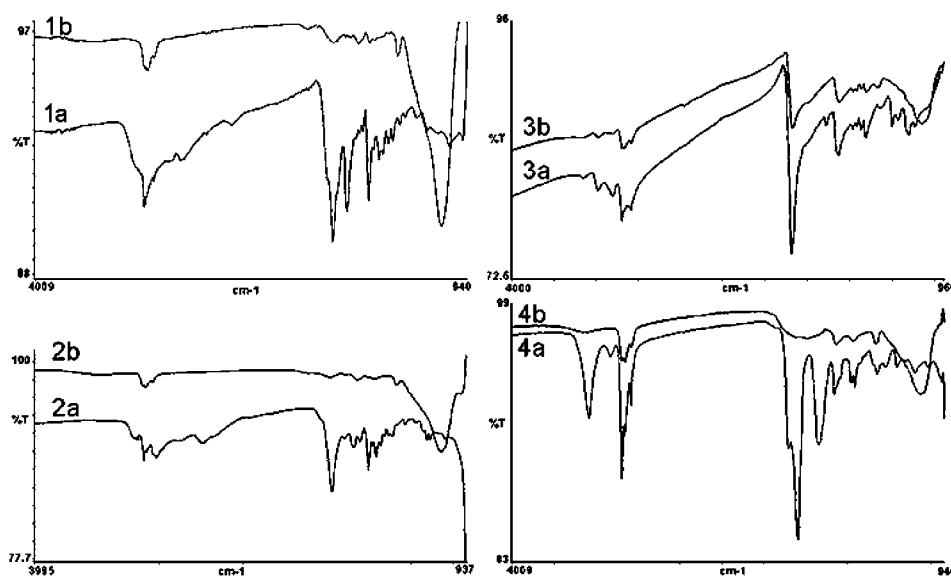


Figure 8. Comparison of IR spectra of Leu (1), MeLeu (2), DKP (3) and Leu₃SEt (4) before (a) and after (b) solar exposure in Earth orbit.

For Leu₃SEt, such an agreement was not observed, suggesting that the ground based simulations did not perfectly match those experienced in space. The difference between exposed and dark cavities was attributed to solar radiation. We found that solar radiation photolyzed 13.6% of DKP, whereas the photodecomposition of Leu₃SEt was calculated to be about 20%.

Peptides are known to sublime under vacuum. Ground simulations have confirmed this sensitivity since a loss of about 35% was measured for DKP and Leu₃SEt. However, when comparing the losses due solely to UV photolysis, peptides appear to be more stable towards solar radiation than amino acids (Figure 6).

3.2. PHOTOCHEMISTRY OF COMPOUNDS

IR spectra between 950 and 4000 cm⁻¹ were directly measured on the films deposited on the windows before and after the flight, prior to any treatment (Figure 8).

They indicate that carbonyl and hydroxyl groups were affected by solar radiation, thus confirming that decarboxylation is the main photochemical effect (Collins and Grant, 1969; Johns *et al.*, 1967; Johns and Seuret, 1972). MeLeu was more photolyzed than Leu by about 13%, probably because of the presence of the methyl group on the C_α-carbon which generates a mesomeric electron-donor effect that weakens the C-COOH bond.

After exposure, the samples exhibit similar general infrared absorption profiles that are very different from those of the starting material. The profile for DKP shows the least before-and-after-flight differences, thus confirming its good sta-

TABLE II
Attribution of the IR absorption bands exhibited by the samples after the flight

Frequency (cm ⁻¹)	Functional group
1060	C-N and/or n-propyl
1300	>C=CH-
1700 (weak)	NH ₃ ⁺
2900	CH
2929	CH ₂
2950	CH ₃
2960	NH ₃ ⁺

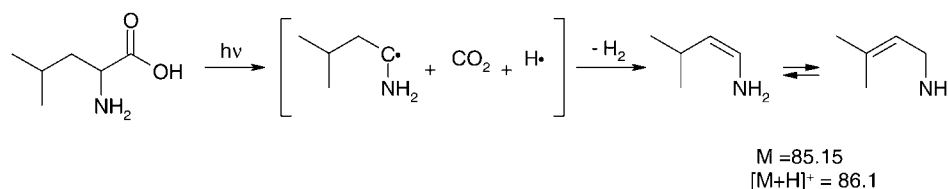


Figure 9. Photochemistry of leucine in solid state.

bility in solar radiation, already shown in Figure 6. The spectra of Leu, MeLeu and Leu₃SEt have been more affected by light. A broad absorption band appeared around 1060 cm⁻¹ on the spectrum of Leu₃SEt. This band is characteristic of hydrocarbons. On the other hand, the carbonyl bands of amino acids and peptides, as well as the hydroxyl band of amino acids, were drastically decreased (Figure 8). Table II refers to the main absorption bands and their attributed functional group detected after solar exposure. This attribution is in good agreement with a decarboxylation reaction. The presence of photochemical by-products revealed by IR studies was not confirmed by capillary electrophoresis analyses. These photo-products might have been produced in quantities that were too small to be measured or not be revealed by UV detection.

Electrospray mass spectrometry was used in order to identify additional compounds. Leucine decarboxylation was first confirmed by the detection of a mass at [M+H]⁺ = 86, corresponding to the unsaturated amine (Figure 9).

It has been established that a radical formed by decarboxylation can further follow different pathways, such as recombination with another radical, or formation of a saturated compound by the capture or loss of a hydrogen radical to form an unsaturated compound. For MeLeu, neither the corresponding derivative nor the saturated corresponding by-product could be detected.

Mass spectrometry measurements made on the light exposed and in dark cavities of Leu₃SEt (Figure 10) show a great difference of composition.

Some starting material was recovered in dark cavities ($[M+H]^+ = 402.7$), but none in the exposed ones. A great number of new masses were obtained and some of them identified as leucine, dileucine, trileucine and diketoperazine of leucine, all present in small quantities. Attribution of molecular structures to the detected masses was done by reference to solid state photochemistry of similar compounds.

Decarboxylation was considered as the major reaction in solid state amino acid photochemistry and decarbonylation, i.e. the rupture of the CO-NH bond, followed by the loss of the CO group, as the main photochemical reaction for peptides. From the more abundant compounds identified (Table III), a degradation pathway of Leu₃SEt is proposed in Figure 11.

The more likely structures were attributed to the detected masses based on radical mechanisms. Formation of unsaturated compounds by radical rearrangement was foreseen since the transformation of a primary radical to a more stable secondary one, leads to an unsaturated molecule. Such a pathway was confirmed by the IR analyses. The spectrum of Leu₃SEt exhibits a C=C double bond absorption band occurring at 1300 cm^{-1} , whereas no absorption band was detected around 1450 cm^{-1} . This supports the presence of a $R_1R_2C=CHR_3$ bond rather than a $R_1CH=CHR_2$ double bond.

The main photochemical process highlighted during Leu₃SEt photolysis is a decarbonylation of the starting product with possible emission of carbon monoxide and formation of an unsaturated compound. The smallest mass detected in irradiated Leu₃SEt samples was $[M+H]^+ = 86$. This compound was already detected with Leu and attributed to the 2-methyl but-2-ene amine. The loss of carbon dioxide led to the formation of the unsaturated amine and emission of dihydrogen.

As already mentioned, DKP was unstable under the flight conditions. Mass spectrometry analyses were made to characterize both photochemical and possible thermal degradation of the molecule. However, only one compound was detected at $[M+H]^+ = 87$ and its structure remains unknown. This seems to confirm the observation that the loss of DKP was mainly due to sublimation. The absence of the carboxyl group in DKP and in Leu₃SEt was expected to reduce their photodecomposition since no decarboxylation can occur. Photochemistry of such molecules is based on the reactivity of the peptide bond CO-NH. Such a bond needs higher energies in order to be broken and to produce CO (Collins and Grant, 1969; Johns *et al.*, 1968; Johns and Seuret, 1970, 1972). Considering the solar spectrum, photons sufficiently energetic to induce such reactions are present in the VUV domain but are not abundant, thus limiting the photoreaction.

3.3. RACEMIZATION

Photochemical processes can lead to racemization by proton abstraction on the asymmetric carbon. The formation of a trivalent carbon radical can induce an in-

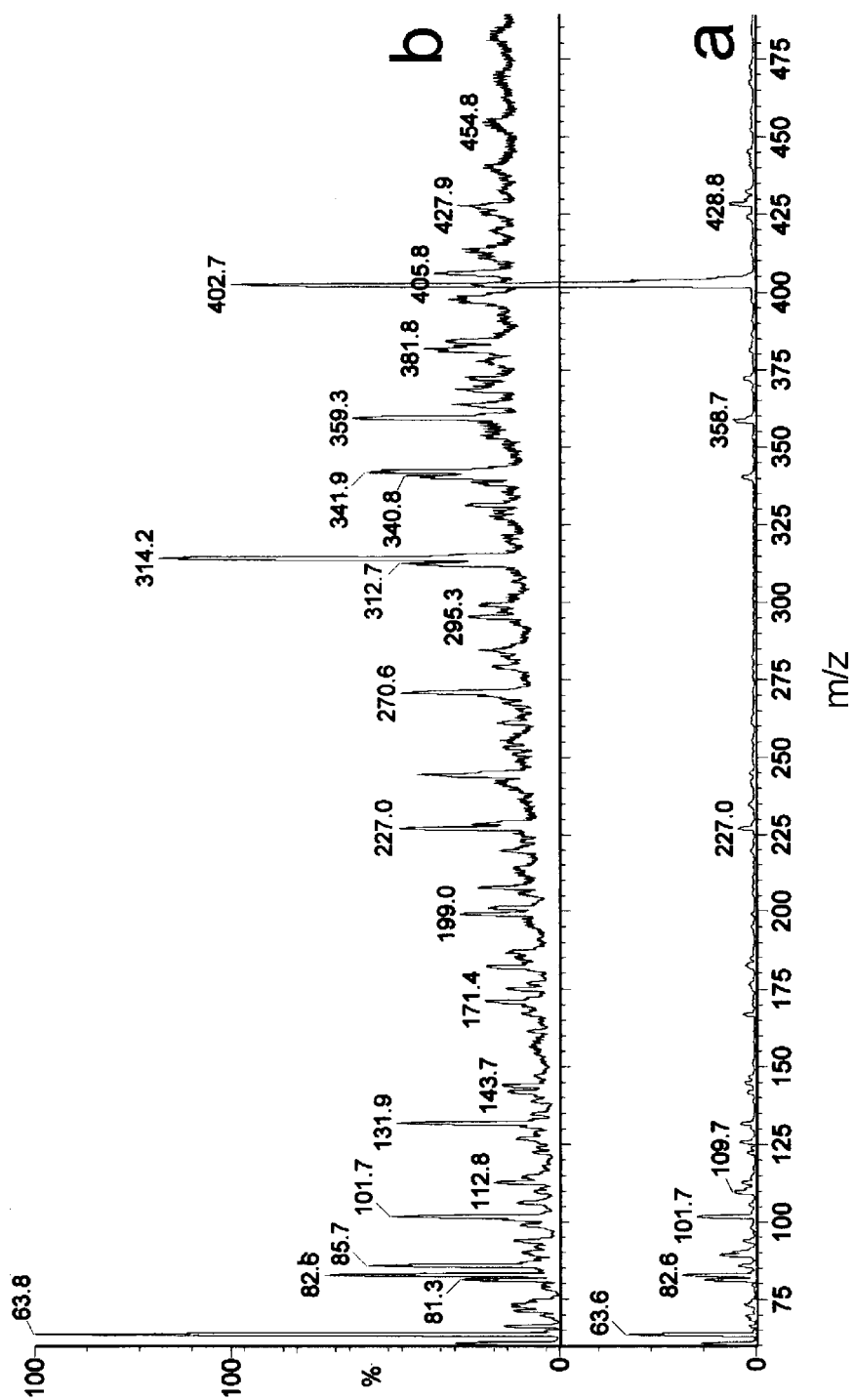
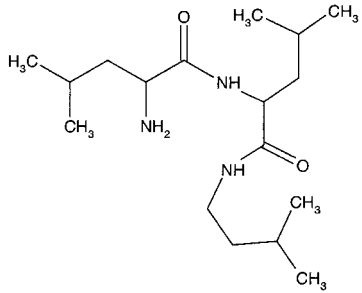
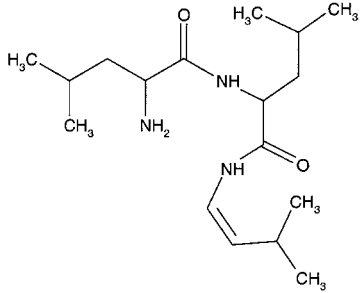
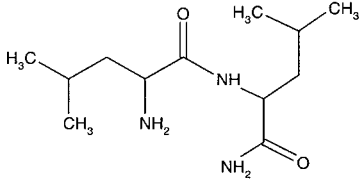
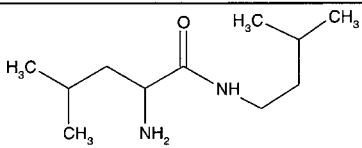
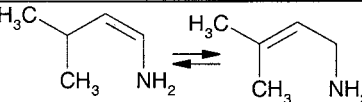


Figure 10. Comparison of electrospray ionization mass spectra of flight control (a) and solar exposed (b) Leu₃SEt sample.

TABLE III

Molecular structures of detected masses in Leu₃SEt after solar exposure

Atomic mass units	Compounds
$[M+H]^+ = 402.7$ M = 401	H-Leu ₃ SEt
$[M+H]^+ = 358.7$ M = 357	H-Leu ₃ -OH
$[M+H]^+ = 341.9$ M = 341	H-Leu ₃ -H
$[M+H]^+ = 314.2$ M = 313	
$[M+H]^+ = 312.7$ M = 311	
$[M+H]^+ = 244.5$ M = 243	
$[M+H]^+ = 227.0$ M = 226	DKP
$[M+H]^+ = 201$ M = 200	
$[M+H]^+ = 131.9$ M = 131	H-Leu-OH
$[M+H]^+ = 85.7$ M = 85	

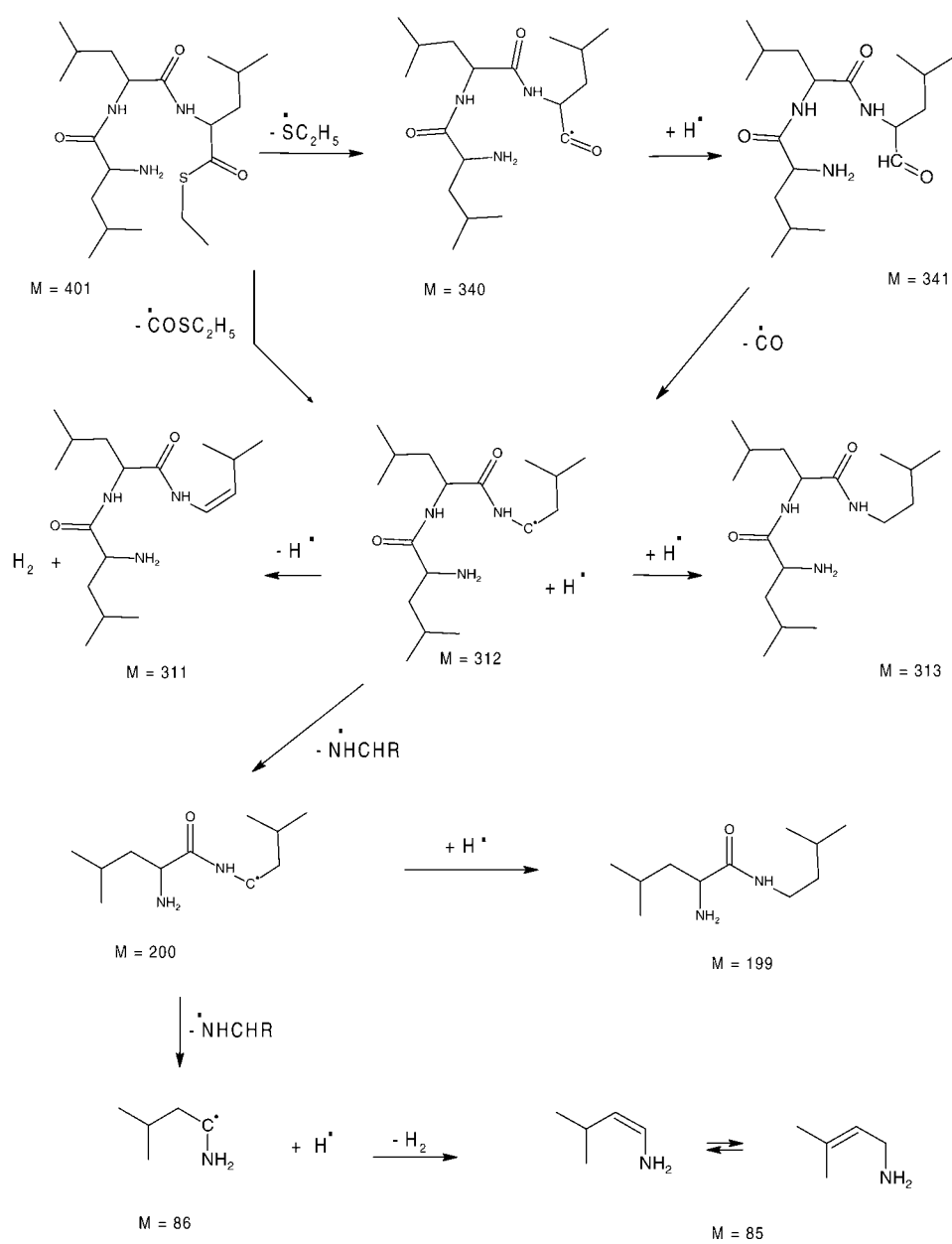


Figure 11. Proposed photodecomposition pathway for Leu₃SEt in the solid state.

TABLE IV
Loss of exposed samples embedded in minerals

	Unprotected	Clay	Basalt	Meteorite
Leu	38%	49%	21%	17%
MeLeu	51%	52%	33%	19%
DKP	14%	3%	0%	12%
Leu ₃ SEt	20%	41%	8%	14%

version of configuration of the initial tetrahedron asymmetric carbon. The energy of a C–H bond is weak enough to be broken in a radical process by UV radiation (Berkowitz *et al.*, 1994) and may lead to racemization.

L-Leu et L-MeLeu were chosen to study possible racemization with the presence of a methyl group on the asymmetric carbon atom. MeLeu should be less sensitive to racemization than Leu, the energy of a C–C bond being higher than the energy of a C–H bond. Investigation for possible racemization was made using both HPLC and capillary electrophoresis, after derivatization with the chiral reactant (+)-FLEC, which forms diastereoisomers separable by HPLC (Einarsson *et al.*, 1987). No D-amino acid was detected within the limit of 0.1%. Either racemization does not occur to a detectable extent under the conditions used, or it may be masked by the photolysis of the intermediary trivalent carbon radical. This result shows that the remaining amino acids after solar radiation photolysis are not racemized. However, in the interstellar medium, where the UV dose is several magnitudes weaker and is of longer duration, racemization could perhaps compete with photodegradation.

3.4. PROTECTING EFFECT OF MINERALS

When the samples were embedded in 5 μm -thick mineral films, the protecting effect is limited and depends on the nature of the mineral (Table IV).

Except for DKP, montmorillonite enhanced the photolysis of the compounds. The meteoritic powder ensured the best protection, with a loss never exceeding 10 to 20%. The better protecting effect of the meteorite is in good agreement with the mineral spectroscopic data, since Allende meteorite exhibits higher absorption in the VUV domain (Figure 5). Montmorillonite is nearly transparent in the VUV domain while absorbing in UVC (Figure 5). Additionally, some water molecules are known to remain trapped between the clay sheets. This remnant water might have increased the photo-destruction of the embedded compounds. Water molecules are photo-ionized at 184.9 nm (Nikogosyan and Görner, 1992, 1995) and produce aqueous electrons which probably reacted with the organic compounds to form free radicals and subsequent by-products. Among the exposed compounds, DKP

TABLE V
Volumetric concentration of organic compounds embedded in mineral fraction

	0.5 μm	2 μm	5 μm	7 μm	10 μm	25 μm
DKP (mmol cm^{-3})	6.75	1.35	0.61	0.45	0.32	0.13
Leu ₃ Set (mmol cm^{-3})	3.84	0.77	0.35	0.26		

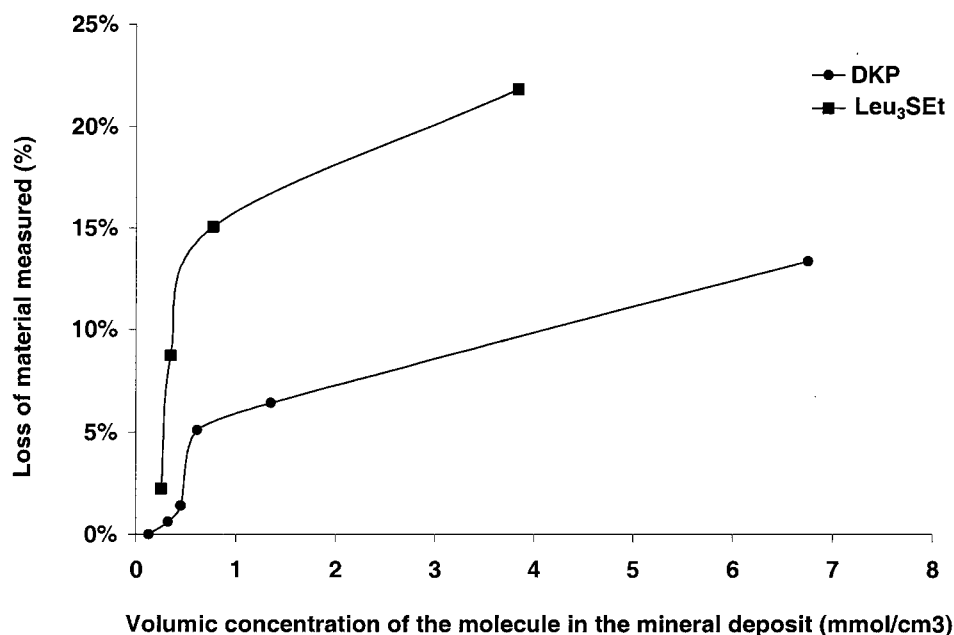


Figure 12. Determination of the shielding effect of the Allende meteorite.

is the less sensitive to UV because it lacks reacting groups. In a general feature, the presence of clay minerals does not modify the scale of UV sensitivity of the unprotected exposed compounds, as shown in Table IV.

In order to quantify the protective effect of minerals on associated organic molecules, the required amounts of DKP and Leu₃SET needed to make 0.5 μm unprotected films were embedded into powered meteorite films ranging in thickness from 0.5 to 25 μm for DKP and from 0.5 to 7 μm for Leu₃SET. As expected, the protecting effect increased with the thickness of the mineral film. Since a constant amount of peptide was used to make the films, its volumetric concentration decreased with increasing thickness of the films (Table V).

The loss of organics versus the volumetric concentration of the compound is reported in Figure 12.

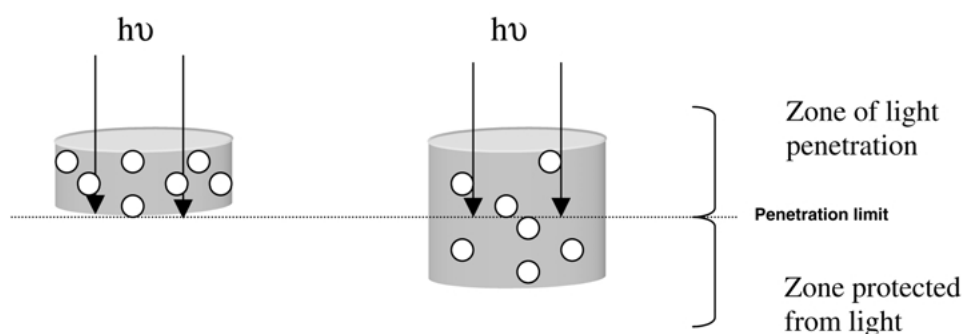


Figure 13. Schematic representation of the shielding effect.

For the two compounds, a curve in two steps is obtained. For the thicker films, i.e. at low volumetric concentrations, the peptides are quite unaffected by radiation since most of the organic material is not affected by UV light as exemplified in Figure 13.

When the thickness decreases, more molecules are accessible to light and thus the losses are more important. The meteorite protection threshold, deduced from Figure 12, is estimated to be about $4.8 \mu\text{m}$ and $4.0 \mu\text{m}$ for DKP and Leu₃ Set films, respectively. These values are considered as the protection threshold for micrometeorite-associated organics.

3.5. QUANTUM YIELDS

The photochemical degradation of exposed molecules during the Perseus Exobiology experiment is mediated by the C=O groups, either carboxylic or peptide amide, because of their high absorption in the VUV and in UVC (Figures 3 and 4). The absorption bands of the C=O groups, $\pi_2-\pi^*$ at 140 nm, $\pi-\pi^*$ at 185 nm and $n-\pi^*$ at 208 nm, have been identified by deconvolution of the whole spectrum and their intensity calculated for a $0.5 \mu\text{m}$ thick film. The flux of photons available in the absorption window of the samples was determined from a previous record of the solar irradiance (Nicolet, 1989). The photon flux necessary for damaging the molecules was deduced by taking into account the absorption of each compound. Quantum yields M , defined as the number of molecules transformed per photon absorbed, are given in Table VI.

Quantum yields of peptides are smaller than those of amino acids, probably because they absorb light in a larger range of wavelengths.

According to Greenberg's calculations, the solar flux received in 1045 hr in Earth orbit between 120 and 275 nm is equivalent to a 0.93 million year voyage in the interstellar medium (Greenberg *et al.*, 1995). From this estimation and from our data, we estimated the times necessary for the full degradation of the sample in the interstellar medium and in the vicinity of the Sun (Table VII).

TABLE VI
Quantum yield of the photodegradation of the samples

Compounds	Molecules transformed	Photons absorbed	Quantum yield Φ
Leu	$7.62 \times 10^{+16}$	$4.55 \times 10^{+19}$	1.7×10^{-3}
MeLeu	$8.98 \times 10^{+16}$	$2.60 \times 10^{+19}$	3.5×10^{-3}
DKP	$1.65 \times 10^{+16}$	$3.92 \times 10^{+20}$	4.0×10^{-5}
Leu ₃ SEt	$1.33 \times 10^{+16}$	$2.12 \times 10^{+20}$	6.2×10^{-5}

TABLE VII

Estimated time necessary for complete degradation of amino acids and peptides in interstellar medium (in million of years) and in the solar system (in years)

Compounds (molecules)	Unprotected		Clay		Basalt		Meteorite	
	0.5 μm		5 μm		5 μm		5 μm	
	ISM	Solar system	ISM	Solar system	ISM	Solar system	ISM	Solar system
Leu ($1.99 \cdot 10^{17}$)	2.43	114	1.88	88	4.37	204	5.60	262
MeLeu ($1.75 \cdot 10^{17}$)	1.81	85	1.78	83	2.82	132	4.84	227
DKP ($1.15 \cdot 10^{17}$)	6.84	320	26.57	1244	465	21771	7.95	373
Leu ₃ SEt ($6.55 \cdot 10^{16}$)	4.58	216	2.24	105	11.62	544	6.69	313

4. Discussion

The main phenomenon observed during the ‘Perseus-Exobiology’ mission was the photolysis of compounds exposed to sunlight. Photo-degradation of the peptides did not lead to the formation of amino acids since the main photolytic process determined by mass spectrometry analyses was shown to be decarbonylation. This result is in good agreement with previous laboratory simulations performed in the solid state, as reported above. The predominance of peptide bond decomposition processes probably impeded the oligomerization of Leu₃SEt. Similarly, the absence of observable racemization could be due to a real resistance to the racemization process or to the photodegradation of the intermediary compounds. Laboratory experiments have shown that racemization occurred after irradiative destruction of 99% of the starting material. Since 60% of the starting material was recovered for L-leucine in our experiment, photolysis was probably not sufficient to generate significant racemization.

When comparing the stability of Leu and MeLeu, it appeared that the C_d-disubstituted MeLeu exhibited lower stability towards irradiation. It is likely that

the extra methyl group linked to the C α -carbon atom weakens the C–COOH bond via a mesomeric electron donor effect.

Cyclic peptides remain stable under irradiation. The difference of stability between linear and cyclic peptides can be explained by photochemical processes leading to the decarboxylation of the functional groups. In cyclic peptides, the functional groups sensitive to UV radiation are engaged in peptide bonds and decarboxylation is impeded. After being delivered to Earth, cyclic peptides could produce linear peptides by reacting with water on mineral surfaces, as proposed by Bujdak *et al.* (1995) and Bujdak and Rode (1996, 1997, 1999). However, cyclic peptides never exist as zwitterions and are, therefore, more sensitive to sublimation.

The use of magnesium fluoride windows in the exposure device stopped all photons below 120 nm. The effect of the hydrogen Lyman α -emission was, therefore, not taken into account. The quantum yields were difficult to determine because of the continuous decomposition of the sample and of the heterogeneity of the sample films. Film heterogeneity resulting from the method of preparation, generated up to 10% discrepancies for some samples exposed in triplicate. For further missions, a new loading technique based on the sedimentation of samples will be developed.

The choice of thioester activation was justified by its prebiotic relevance but its preservation at temperatures up to 40 °C, the highest temperature recorded during the flight, was rather poor and a spontaneous decomposition of the activating ester probably occurred in the solid state during the flight.

The amino acid films exposed during the Perseus Exobiology experiment were obtained by evaporation of an aqueous solution of the compounds. The procedure probably led to zwitterions forming microcrystal structures (Clark *et al.*, 1976; Keniry *et al.*, 1984; Wang *et al.*, 2001). This zwitterionic form may have provided good stability in space conditions i.e. vacuum and temperature. For peptides, the longer distance between the charges increased the propensity to sublimation.

Some space exposure parameters cannot be easily anticipated, thus ground simulations before the mission are difficult to perform. Moreover, some difficulties can appear during the flight. For instance UV acquisition during the mission partially failed due to the saturation of the UV sensors. Nevertheless, the reconstitution of the energy profile received by the samples was accomplished by different methods and gave satisfactory results. A prediction of the thermal conditions based on previous Biopan flight parameters was used for pre-flight simulations. Unfortunately, the temperature experienced by the mission was higher than expected, and sublimation phenomena and thermal degradation occurred, as confirmed by post-flight ground controls. Ground simulations were helpful in determining the chemical modifications relating only to photochemical processes.

The newly designed hardware with individual removable exposure cells was easy to handle and facilitating the loading and recovery operations. In addition, individual windows allowed direct IR spectroscopy of the samples before and after

the mission, prior to any extraction or treatment. This was a major improvement because some side-products could not be analyzed by UV spectroscopy.

5. Conclusion

The 97 day Perseus Exobiology experiment represents the first long duration space exposure experiment with amino acids and peptides, whereas the previous Biopan flights did not exceed 2 weeks. This exposure experiment was successful in terms of scientific return, and also in terms of hardware with all parameters, technical and chemical, being optimized. This led us to propose plausible photochemical degradation processes involved during solar exposure of the studied compounds and to understand the differences in chemical behavior between these molecules. The experiment utilizing mineral surfaces to mimic micrometeorites indicated that the threshold for efficient protection against radiation was about 5 μm . This supports the hypothesis that the 50–100 μm micrometeorite fraction represents a protected environment for the importation of organic compounds.

Acknowledgements

This work was been supported by CNES and the Région Centre, France. The authors wish to thank all persons involved in the success of the orbital mission, CNES staff, astronauts Viktor Afanassiev, Jean-Pierre Haignere and Ivan Bella. Special thanks to Delphine FAYE-LECAT and her team (CNES-Toulouse) for performing the simulation experiments.

References

- Anderson, J. and Smith, R. E.: 1994, 'Natural Orbital Environmental Guidelines for use in Aerospace Vehicle Development', *NASA Technical Memorandum* (4527).
- Barbier, B., Chabin, A., Chaput, D. and Brack, A.: 1998, 'Photochemical processing of amino acids in Earth orbit', *Planet. Space Sci.* **46**(4), 391–398.
- Berkowitz, J., Ellison, G. B. and Gutman, D.: 1994, 'Three methods to measure RH bond energies', *J. Phys. Chem.* **98**, 2744–2765.
- Brueckner, G. E., Edlow, K. L., Floyd, L. E., Lean, J. L. and E. V. M.: 1993, 'The Solar Ultraviolet Spectral Irradiance Monitor (SUSIM) experiment on board the Upper Atmosphere Research Satellite (UARS)', *J. Geophys. Res.* **98**(D6), 10,695–10,711.
- Bujdak, J., Faybikova, K., Eder, A., Yonyai, Y. and Rode, B. M.: 1995, 'Peptide chain elongation: A possible role of montmorillonite in prebiotic synthesis of protein precursors', *Orig. Life Evol. Biosphere* **25**, 431–441.
- Bujdak, J. and Rode, B. M.: 1996, 'The effect of smectite composition on the catalysis of peptide bond formation', *J. Mol. Evol.* **43**, 326–333.
- Bujdak, J. and Rode, B. M.: 1997, Silica, Alumina, and clay-catalyzed alanine peptide bond formation. *J. Mol. Evol.* **45**, 457–466.

- Bujdak, J. and Rode, B. M.: 1999, 'The effect of clay structure on peptide bond formation catalysis', *J. Mol. Catal. A: Chem.* **144**, 129–136.
- Charnley, S. B.: 1997, 'On the Nature of Interstellar Organic Chemistry', in C. B. Cosmovici, S. Bowyer and D. Werthimer (eds), *Astronomical and Biochemical Origins and the Search for Life in the Universe*, pp. 89–96.
- Chyba, C. and Sagan, C.: 1992, 'Endogenous production, exogenous delivery and impact-shock synthesis of organic molecules: An inventory for the origins of life', *Nature* **355**, 125–132.
- Clark, D. T., Peeling, J. and Colling, L.: 1976, 'An experimental and theoretical investigation of the core level spectra of a series of amino acids, dipeptides and polypeptides', *Biochim. Biophys. Acta* **453**(2), 533–545.
- Collins, M. A. and Grant, R. A.: 1969, 'Ultraviolet light induced free radicals in glycine peptides in the solid state', *Photochem. Photobiol.* **9**, 369–375.
- Cooper, G., Kimmich, N., Belisle, W., Sarinana, J., Brabham, K. and Garrel L.: 2001, 'Carbonaceous meteorites as a source of sugar-related organic compounds for the early Earth', *Nature* **414**(6866), 879–883.
- Cronin, J. R. and Moore, C. B.: 1971, 'Amino acid analyses of the Murchison, Murray, and Allende carbonaceous chondrites', *Science* **172**, 1327–1329.
- Cronin, J. R. and Pizzarello, S.: 1983, 'Amino acids in meteorites', *Adv. Space Res.* **3**(9), 5–18.
- DeDuve, C.: 1998, 'Possible Starts for Primitive Life. Clues from Present-day Biologie: The Thioester World', in A. Brack (ed.), *The Molecular Origins of Life: Assembling Pieces of the Puzzle*, Cambridge University Press, pp. 219–236.
- Dodonova, N. Y.: 1993, 'Vacuum UV – Photophysics and photochemistry of biomolecules', *J. Photochem. Photobiol. B: Biol.* **18**, 111–121.
- Einarsson, S., Josefsson, B., Möller, P. and Sanchez, D.: 1987, 'Separation of amino acids enantiomers and chiral amines using precolumn derivatization with (+)-1-(9-fluorenyl)ethylchloroformate and reversed-phase liquid chromatography', *Anal. Chem.* **59**, 1191–1195.
- Ferris, J. P., Hill Jr., A. R., Liu, R. and Orgel, L. E.: 1996, 'Synthesis of long prebiotic oligomers on mineral surfaces (see comments)', *Nature* **381**(6577), 59–61.
- Greenberg, J. M.: 1984, 'Chemical evolution in space', *Orig. Life Evol. Biosph.* **14**, 25.
- Greenberg, J. M.: 1995, 'Chirality in Interstellar Dust and in Comets: Life from Dead Stars', *Proceedings from the Symposium in Santa Monica, 'Physical Origin of Homochirality in Life'*, AIP Press.
- Greenberg, J. M., Li, C., Mendoza-Gomez, C. X., Schutte, W. A., Gerakines, P. A. and De Groot, M.: 1995, 'Approaching the interstellar grain organic refractory component', *ApJ* **455**, L177–L180.
- Hénin, O.: 1998, 'Des acides aminés prébiotiques aux peptides catalytiques: Recherche d'une activité catalytique', *Mémoire de Thèse*, Université d'Orléans.
- Heroux, L. and Hinteregger, H. E.: 1978, 'Aeronomical reference spectrum for solar UV below 2000 Å', *J. Geophys. Res.* **83**(A11), 5305–5308.
- Hollis, J. M., Lovas, F. J. and Jewell, P. R.: 2000, 'Interstellar Glycolaldehyde: The first sugar', *Ap. J.L.* **540**, L107–L110.
- Horneck, G. and Brack, A.: 1992, 'Study of the origin, evolution and distribution of life with emphasis on exobiology experiments in Earth orbit', *Adv. Space Biol. Med.* **2**, 229–262.
- Horneck, G., Rettberg, P., Rabbow, E., Strauch, W., Seckmeyer, G., Facius, R., Reitz, G., Strauch, K. and Schott, J.-U.: 1996, 'Biological dosimetry of solar radiation for different simulated ozone column thicknesses', *J. Photochem. Photobiol. B: Biol.* **32**, 189–196.
- Huber, C. and Wächtershauser, G.: 1997, 'Activated acetic acid by carbon fixation on (Fe, Ni)S under primordial conditions', *Science* **276**, 245–247.
- Huber, C. and Wächtershauser, G.: 1998, 'Peptides by activation of amino acids with CO on (Ni, Fe)S surfaces: Implications for the origin of Life', *Science* **281**, 670–672.

- Inagaki, I.: 1973, 'Optical absorptions of aliphatic amino acids in the far ultraviolet', *Biopolymers* **12**, 1353–1362.
- Jarosewich, E.: 1990, 'Chemical analyses of meteorites: A compilation of stony and iron meteorite analyses', *Meteoritics* **25**, 323–337.
- Johns, R. B., Looney, F. D. and Whelan, D. J.: 1967, 'Photochemistry of biological molecules – I. Photolysis of amino acids in the solid state', *Biochim. Biophys. Acta* **147**, 369–380.
- Johns, R. B., Looney, F. D. and Whelan, D. J.: 1968, 'Photochemistry of biological molecules – II. Photolysis of dipeptides in the solid state', *Photochem. Photobiol.* **7**, 65–72.
- Johns, R. B. and Seuret, M. G.: 1970, 'Photochemistry of biological molecules – III. Mechanism of photodamage of alanine peptides in the solid state', *Photochem. Photobiol.* **12**, 405–417.
- Johns, R. B. and Seuret, M. G.: 1972, 'Photochemistry of biological molecules - IV. Gaseous products from the photolysis of alanine peptides in the solid state', *Photochem. Photobiol.* **16**, 413–424.
- Jungclauss, G., Cronin, J. R., Moore, C. B. and Yuen, G. U.: 1976, 'Aliphatic amines in the Murchison meteorite', *Nature* **261**, 126–128.
- Jurewicz, A. J. G., Mittlefehldt, D. W. and Jones, J. H.: 1991, 'Partial melting of the Allende (CV3) meteorite: Implications for the origins of basaltic meteorites', *Science* **252**, 695–698.
- Keniry, M. A., Kintanar, A., Smith, R. L., Gutowsky, H. S. and Oldfield, E.: 1984, 'Nuclear magnetic resonance studies of amino acids and proteins. Deuterium nuclear magnetic resonance relaxation of deuteriomethyl-labeled amino acids in crystals and in Halobacterium halobium and Escherichia coli cell membranes', *Biochemistry* **23**(2), 288–98.
- Kvenvolden, K., Lawless, J., Pering, K., Peterson, E., Flores, J., Ponnampuruma, C., Kaplan, I. R. and Moore, C.: 1970, 'Evidence for extraterrestrial amino-acids and hydrocarbons in the Murchison meteorite', *Nature* **228**, 923–926.
- Kvenvolden, K. A., Lawless, J. G. and Ponnampuruma, C.: 1971, 'Nonprotein amino acids in the Murchison meteorite', *Proc. Nat. Acad. Sci. U.S.A.* **68**(2), 486–490.
- Love, S. G. and Brownlee, D.: 1993, 'A direct measurement of the terrestrial mass accretion rate of cosmic dust', *Science* **262**, 550–553.
- Maurette, M.: 1998, 'Carbonaceous micrometeorites and the origin of life', *Orig. Life Evol. Biosph.* **28**(4–6), 385–412.
- McMillin C. R., Rippon W. B. and Walton A. G.: 1973, 'Vacuum ultraviolet spectroscopy of poly- α -amino acids', *Biopolymers* **12**, 589–597.
- Mecherikunnel, A. T.: 1996, 'Solar total irradiance observations from spacecraft: 1992–1993', *J. Geophys. Res.* **101**(A8), 17,073–17,079.
- Nicolet, M.: 1989, 'Solar spectral irradiances with their diversity between 120 and 900 nm', *Planet. Space Sci.* **37**(10), 1249–1289.
- Nikogosyan, D. N. and Görner, H.: 1992, 'Photolysis of aromatic amino acids in aqueous solution by nanosecond 248 and 193 nm laser light', *J. Photochem. Photobiol. B: Biol.* **13**, 219–234.
- Nikogosyan, D. N. and Görner, H.: 1995, 'Photolysis (193 nm) of aliphatic amino acids in aqueous solution', *J. Photochem. Photobiol. B: Biol.* **30**, 189–193.
- Oro, J.: 1996, 'Comets and Life on the Primitive Earth. Astronomical and Biochemical Origins and the Search for Life in the Universe', C. B. Cosmovici, S. Bowyer and D. Werthimer (eds), *Proceedings of the 5th International Conference on Bioastronomy IAU Colloquium No. 161*, pp. 97–120.
- Oro, J., Gibert, J., Lichtenstein, H., Wikstrom, S. and Flory, D. A.: 1971, 'Amino-acids, aliphatic and aromatic hydrocarbons in the Murchison meteorite', *Nature* **230**, 105–106.
- Paecht-Horowitz, M.: 1977, 'The mechanism of clay catalyzed polymerization of amino acid adenylates', *Biosystems* **9**, 93–98.
- Paecht-Horowitz, M. and Lahav, N.: 1977, 'Polymerization of alanine in the presence of a non-swelling montmorillonite', *J. Mol. Evol.* **10**, 73–76.
- Pizzarello, S. and Cronin, J. R.: 2000, 'Non-racemic amino-acids in the Murray and Murchison meteorites', *Geochim. Cosmochim. Acta* **64**(2), 329–338.

- Reber, C. A., McNeal, C. A., Trevathan, C. E. and Luther, M. R.: 1993, 'The Upper Atmosphere Research Satellite (UARS) mission', *J. Geophys. Res.* **98**(D6), 10,643–10,647.
- Stevenson, F. J. and Cheng, C.-N.: 1970, 'Amino acids in sediments. Recovery by acid hydrolysis and quantitative estimation by a colorimetric procedure', *Geochim. Cosmochim. Acta* **34**, 77–88.
- Stoks, P. and Schwartz, A. W.: 1981, 'Nitrogen-heterocyclic compounds in meteorites: significance and mechanisms of formation', *Geochim. Cosmochim. Acta* **45**, 563–569.
- Wang, R., Liu, H., Carducci, M. D., Jin, T., Zheng, C. and Zheng, Z.: 2001, 'Lanthanide coordination with alpha-amino acids under near physiological pH conditions: Polymetallic complexes containing the cubane-like $[\text{Ln}_4(\mu_3\text{-OH})_4]^{8+}$ cluster core', *Inorg. Chem.* **40**(12), 2743–2750.