AMINO ACIDS IN THE YAMATO-74662 METEORITE, AN ANTARCTIC CARBONACEOUS CHONDRITE

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Abstract: The Yamato-74662.23 meteorite, a carbonaceous chondrite (C2), was examined for amino acids. The meteorite sample was separated into an exterior and an interior portion. Each fraction was pulverized and extracted with water. Unhydrolyzed and acid-hydrolyzed water extracts were analyzed for amino acids, qualitatively and quantitatively. Fifteen amino acids were detected. Of these, nine amino acids are proteinaceous and six amino acids are non-proteinaceous. Glycine is the most abundant amino acid in the unhydrolyzed portion (14 nm/g in the exterior and 13 nm/g in the interior) and the acid-hydrolyzed portion (34 nm/g in both the exterior and the interior). The D- and L-isomers of alanine, aspartic acid, and glutamic acid are nearly equal in abundance in the unhydrolyzed and hydrolyzed portions of the exterior and interior. These results clearly indicate that the amino acids detected were of meteoritic origin and few had terrestrial contaminants.

1. Introduction

Carbonaceous chondrites are the only known source of extraterrestrial organic compounds available for analysis on Earth. The first conclusive evidence was presented with amino acids indigenous to the Murchison meteorite by KVENVOLDEN *et al.* (1970) who showed the presence of an equal abundance of the D- and L-optical isomers. This study was extended to non-protein amino acids of the same meteorite in which the abundance of the D- and L- forms found were nearly equal (KVENVOLDEN *et al.*, 1971). This strategy was also applied to the examination of amino acids in the Murray meteorite (LAWLESS *et al.*, 1971) and the Orgueil meteorite (LAWLESS *et al.*, 1972). Other studies of amino acids in carbonaceous chondrites were reported by ORÓ *et al.* (1971a, b), CRONIN and MOORE (1971), and BUHL (1975).

The studies of the organic compounds of meteorites provide useful information on the abiotic synthesis of primordial organic compounds in the early

solar nebula (ANDERS *et al.*, 1973) and also on the origins of prebiotic organic compounds on the primitive Earth that lead to the appearance of life (LEMMON, 1970; PONNAMPERUMA, 1972; MILLER and ORGEL, 1974; PONNAMPERUMA, 1978). However the total number of studies of organic compounds in the meteorites is small.

Recent discoveries of a large number of meteorites in Antarctica (YOSHIDA *et al.*, 1971; CASSIDY *et al.*, 1977; YANAI, 1978) provide us with new opportunities to examine organic compounds in meteorites. The meteorites found in Antarctica are not only large in number, but also are uniquely preserved in the ice since their fall and later brought out onto the surface of the bare ice. This preservation may provide us with meteorites with the least possible terrestrial contamination. If this is true, the studies of the Antarctic meteorites will lead us to more accurate information and to a better understanding of the organic compounds of extraterrestrial origin.

Of the several classes of organic compounds studied, amino acids alone provide definitive clues to the indigenous nature of the compounds and help us to estimate the degree of terrestrial contamination. Therefore, we initiated an investigation of amino acids for the study of the organic compounds in the Antarctic meteorite. The meteorite we examined was the Yamato-74662, collected on the surface of the bare ice at the Motoi Nunatak in the Meteorite Ice Field, southern Yamato Mountains, East Antarctica, on December 30, 1974. This meteorite was the first meteorite identified as a carbonaceous chondrite, type II, from the nearly 1000 specimens collected by the 15th Japanese Antarctic Research Expedition (YANAI and HARAMURA, 1978).

2. Experimental

The sample examined was Yamato-74662.23 which was a piece separated in the laboratory from the Yamato-74662. The sample was separated into an exterior, middle, and interior portion in a class 100 clean room. Each portion was pulverized and the exterior (1.05 g) and interior (1.10 g) were subjected to the following sample preparation procedures. Each powdered sample was placed in a glass tube with a teflon-lined cap and refluxed with water (10 g) for 20 hours. After the refluxing, the aqueous solution was centrifuged, and the supernatant was recovered in a beaker. The residue was rinsed twice with water (5 g) and combined with the supernatant. The combined solution (approximately 30 g) was reduced under an infra-red lamp, transferred to a 1 m*l* glass vial, and dried completely. Then, 600 μl of water was added to the vial, sonicated and divided into two equal portions (300 μl).

2.1. Unhydrolyzed samples

One of the 300 μl solutions was further split into two 150 μl solutions which were dried completely under the lamp. 100 μl of water was added to one of the dried portions. This solution was sonicated to redissolve amino acids and was analyzed quantitatively by an automated Durrum-500 amino acid analyzer. To the other dried portion was added 1 m*l* isopropanol-1.5 N HCl. This alcohol solution was heated to 105°C for 2 hours and then evaporated to dryness under the lamp. After this drying, 1 m*l* of TFAA-CH_2Cl_2 (1 to 2 by volume) was added to the residue, and heated to 105°C for 5 min. Finally, the excess TFAA-CH_2Cl_2 was evaporated and the N-TFA-isopropyl esters of amino acids, thus prepared, were redissolved with CH_2Cl_2. The methylene chloride solution was injected by a solids injector into a Perkin-Elmer 900 gas chromatograph equipped with a Chirasil Val glass capillary column (25 m) and a nitrogen detector. The use of this optically active column was for the determination of enantiomeric abundance of amino acids.

2.2. Acid-hydrolyzed samples

The other 300 μl of the solution was dried under the lamp. The residue was hydrolyzed with 1 m*l* of 6 N HCl at 105°C for 20 hours. The hydrolyzed solution was divided into two equal portions and the solutions were evaporated to dryness. These two dried hydrolysates in 1 m*l* glass vials were subjected to the same sample preparation procedures as were unhydrolyzed samples for the analyses by the amino acid analyzer and the gas chromatograph.

The exterior and interior portions were processed in parallel for sample preparation along with a procedural blank with 2 g oven-baked sand. All glassware used was cleaned with chromic acid solution, rinsed with water, and oven-baked at 450°C overnight. The water used was deionized and then twice distilled. 6 N HCl was prepared with concentrated HCl and water, and was double-distilled. Methylene chloride was distilled once because of its small use. A commercial grade of TFAA (trifluoroacetic anhydride) was used without any purification. The class 100 clean room was maintained at about 20°C under a constant low humidity.

3. Results

3.1. Amino acid abundance

The ion exchange chromatogram shows that several amino acids were detected in the procedural blank in minor quantities (Fig. 1). In the unhydrolyzed portion these amino acids were 0.4 nm aspartic acid, 0.1 nm threonine, 0.6 nm serine, 0.4 nm glutamic acid, 0.3 nm glycine, and 0.3 nm alanine. Amino acids found in the acid-hydrolyzed portion were 0.8 nm aspartic acid, 0.3 nm threonine,

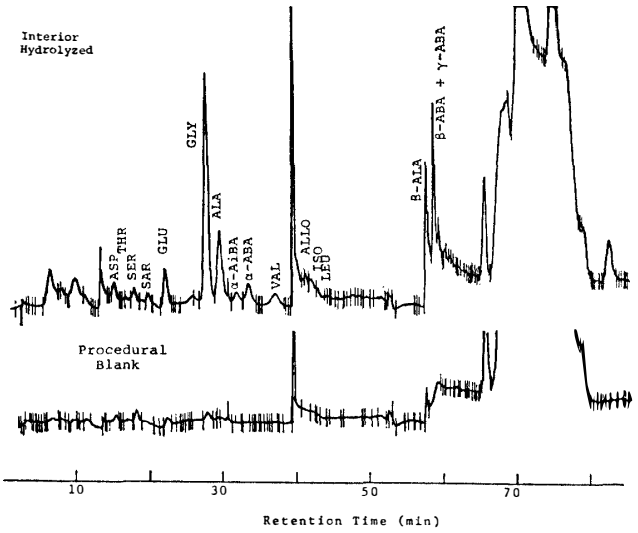

Fig. 1. Amino acid chromatograms of the hydrolyzed portion of the interior specimen.

Table 1. Amino acids in the Yamato-74662 meteorite (nm/g meteorite).

	Exterior		Interior	
	Unhydrolyzed	Hydrolyzed*	Unhydrolyzed	Hydrolyzed*
Aspartic acid	0.2	1.1	0.2	1.2
Threonine	0.2	0.1	0.1	0.2
Serine	0.1	0.0	0.1	0.2
Sarcosine	3.1	4.6	2.3	5.4
Glutamic acid	0.1	4.9	0.1	4.0
Glycine	14	34	13	34
Alanine	6.1	13	5.7	13
α-amino-i-butyric acid	2.8	3.9	2.0	3.7
α-amino-n-butyric acid	3.3	6.7	2.6	6.7
Valine	2.7	4.3	1.8	3.4
Alloisoleucine	1.2	2.7	0.6	2.7
Isoleucine	1.2	2.4	0.6	2.2
Leucine	0.6	1.6	0.4	1.3
β-alanine	4.4	16	4.5	14
β-aminobutyric acid + γ-aminobutyric acid	3.0**	18**	2.5**	17**

* Values of hydrolyzed portion contain values of unhydrolyzed portion.
** Gas chromatographic study indicates the value is mostly of γ-aminobutyric acid.

1.6 nm serine, 1.5 nm glutamic acid, 1.2 nm glycine and 0.4 nm alanine. A similar result was obtained as amino acid contamination per gram of water used when we studied lunar soils for amino acids by a similar analytical scheme in the same clean room (GEHRKE *et al.*, 1975). These amino acids are common in biological material and probably introduced from the water used during analysis. However, the concentration level of these contaminants was insignificant, except serine, when compared to amino acids recovered from the Yamato meteorite samples.

A total of 15 amino acids were identified in the Yamato meteorite based on their retention times on the ion exchange chromatograms (Fig. 1). The quantities of these amino acids are listed in Table 1. These values were obtained after subtracting the values of amino acid contaminants. Of 15 amino acids, nine amino acids are commonly found in proteins, and the other six amino acids are non-proteinaceous. After acid hydrolysis the yields increased approximately two to three times that found in the unhydrolyzed portions. This increase is striking with aspartic acid and glutamic acid, both dicarboxylic amino acids.

The amounts and kinds of amino acids recovered from the exterior and interior portions are almost identical. A minor difference in quantity is probably caused by analytical error.

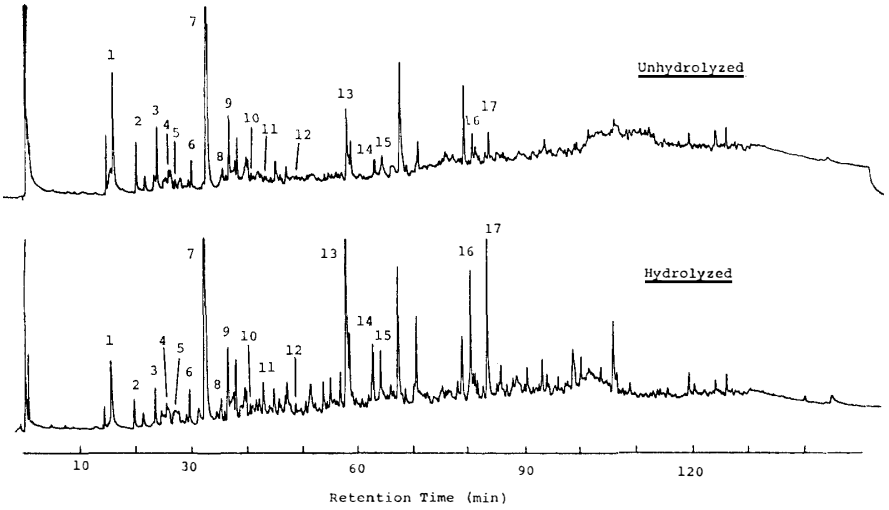

Fig. 2. Gas chromatograms of N-TFA-isopropyl esters of amino acids of the interior specimen. Peaks:
1. sarcosine, 2. D-alanine, 3. L-alanine, 4. D-α-amino-n-butyric acid, 5. D-valine, 6. L-α-amino-n-butyric acid plus L-valine, 7. glycine, 8. D-norvaline, 9. β-alanine, 10. L-norvaline, 11. D-leucine, 12. L-leucine, 13. γ-aminobutyric acid, 14. D-aspartic acid, 15. L-aspartic acid, 16. D-glutamic acid, 17. L-glutamic acid.

3.2. Optical isomers of amino acids

The gas chromatographic analysis by the optically active column and the nitrogen detector shows more than 40 recognizable peaks on one chromatogram. The chromatograms of the unhydrolyzed and hydrolyzed portion of the interior portion are shown in Fig. 2. The corresponding chromatograms of the exterior portion show very similar patterns to those of the unhydrolyzed and hydrolyzed portion of the interior of the meteorite.

Many of the peaks lack good baseline separation from other peaks. However, D- and L-alanine show clearly equal abundance in both the unhydrolyzed and hydrolyzed portion. Other enantiomers which are in nearly equal abundance are D- and L-aspartic acid and D- and L-glutamic acid, when other peaks overlapping near the baseline level were subtracted. Although the chromatograms are very complicated, the nearly equal abundance of D- and L-enantiomers of such protein amino acids as alanine, aspartic acid, and glutamic acid suggests the presence of equal abundance of D- and L-isomers of other protein and non-protein amino acids.

In order to verify that the equal abundance of the D- and L-isomers were present in the meteorite and not by artifact, L-isomers of alanine, isoleucine, leucine, phenylalanine, and glutamic acid were spiked in clean sand and were subjected to the same procedures as for the hydrolyzed samples. No D-isomers were detected on the chromatogram. This control experiment clearly indicates that the observed equal abundance of the enantiomers were of meteoritic origin.

4. Discussion

4.1. Amino acids of the Yamato-74662 meteorite

The finding of both protein and non-protein amino acids and a nearly equal abundance of the D- and L-isomers of these amino acids clearly indicated that they were extraterrestrial in origin and products of abiotic synthesis. These amino acids found are from the simplest amino acid, glycine (C_2), to C_6 amino acids. Some are α-amino acids and others are β- and γ-amino acids. Normal and iso-structures are present. The N-methyl derivative, sarcosine, was also found. Aspartic acid and glutamic acid are dicarboxylic amino acids. It seems that a variety of amino acids might be present in the meteorite, although diamino acids were not detected.

The amount of amino acids found was in the same order of magnitude as in the Murchison and Murray (both C2 chondrites). For example, glycine content in the Murchison meteorite was about 80 nm/g (KVENVOLDEN *et al.*, 1970; ORÓ *et al.*, 1971; CRONIN and MOORE, 1971). Amino acids found in the unhydrolyzed portion were present as free molecules in the meteorite. However, the chemical nature of amino acids yielded by the acid-hydrolysis is not known clearly. They

might be hydrolyzed from simpler molecules (*e.g.*, nitriles) and/or more complex molecules (*e.g.*, peptides). Other possibilities are salt forms with inorganic ions and bound forms on organic and inorganic particulates. A study of the Murchison meteorite indicated only a small fraction accounted for amino acids by the hydrolysis of peptide material (CRONIN, 1976b). The current study found that amino acids whose yields increase markedly by the hydrolysis are dicarboxylic amino acids, aspartic acid and glutamic acid, and the non-α-amino acids, β-alanine and β-aminobutyric acid plus γ-aminobutyric acid as seen in Table 1. Aspartic acid and glutamic acid yields were also markedly increased after the acid-hydrolysis of water extract of the Murchison meteorite (CRONIN and MOORE, 1971). More studies of carbonaceous meteorites are necessary to clarify the nature of acid-hydrolyzable amino acids because, unlike those in geologic samples, organic com-

Table 2. Molar ratios of amino acids found in carbonaceous chondrites.

C♯	Amino acids	Yamato-74662		Murchison*	Murray**
		Exterior	Interior		
C_2	Glycine	100	100	100	100
C_3	Alanine	38	38	45	37
	β-alanine	47	41	7	34
	Serine	0	1	8	
	Sarcosine	14	16	20	
		99	96	80	71
C_4	Aspartic acid	3	4	14	30
	Threonine	0	1	7	
	α-amino-n-butyric acid	20	20	12	
	α-amino-i-butyric acid	11	11	43	277
	β-amino-i-butyric acid			7	7
	β-aminobutyric acid + γ-aminobutyric acid	53	50		
		87	86	83	326
C_5	Valine	13	10	18	19
	Glutamic acid	14	12	32	27
	Proline			14	9
		27	22	64	55
C_6	Leucine	5	4	8	
	Isoleucine	7	6	8	
	Alloisoleucine	8	8		
		20	18	16	

* Data from CRONIN (1976a).
** Data from CRONIN and MOORE (1971).

pounds in the meteorites are non-biological in origin.

The relative abundance of amino acids found in the Yamato-74662, the Murchison, and the Murray meteorites is shown in Table 2. It is reasonable to note that glycine is the most abundant amino acid of all. This is because glycine is the simplest amino acid and, thereby, the easiest amino acid to be synthesized abiotically.

However, the Yamato sample shows a nearly equal abundance of the total C_3 amino acids to glycine. The total abundance of C_4 amino acids are 87 and 86, which might become approximately 100 if other C_4 amino acids (*i.e.*, diaminobutyric acids, N-ethylglycine and N-methylalanine) are identified and quantitized. This observation suggests that the total amino acid quantity does not necessarily decrease with the increase of carbon numbers. The abundances of individual amino acids synthesized abiotically should decrease with the increase of carbon number because the number of structurally possible amino acids with a given carbon number increases markedly. More quantitative studies of amino acids in carbonaceous chondrites are necessary for a better understanding of primordial organic synthesis during condensation of the meteorite and of their subsequent history.

4.2. Amino acids of meteorite origin

The Yamato-74662 meteorite shows nearly equal abundance of amino acids both in the exterior and in the interior portion. If the meteorite was contaminated by terrestrial amino acids or biological material, the exterior portion should have shown more abundance of amino acids, specifically protein amino acids. This is the case for the Orgueil, the Murray, and even the Murchison meteorite. The study of these meteorites was always conducted with the interior portions. The exterior of the meteorite was not generally used for analysis.

The finding of non-protein amino acids in significant quantity in the Yamato meteorite indicates the presence of amino acids indigenous to the meteorite. However, this evidence alone cannot be used as a criterion for the extraterrestrial origin of the amino acid assemblage identified, as the assemblage might be a composite of meteoritic and terrestrial origin.

A close examination of the D- and L-isomer abundance for alanine, aspartic acid, and glutamic acid, all protein amino acids, indicates nearly equal abundances both in the exterior and interior portion of the Yamato meteorite. This evidence reveals the terrestrial amino acid contamination was negligible for the meteorite. The examination of D/L ratios of the Mighei meteorite, which fell in the Soviet Union in 1889, showed a pronounced L-isomer predominance of protein amino acids in the exterior portion. This predominance decreases toward the middle and interior portion. However, non-protein amino acids show the equal abundance

throughout the exterior to interior portion (BUHL, 1975). Apparently the amino acid assemblage of each portion of the Mighei meteorite was a mixture of the meteorite and terrestrial origin. A similar result was obtained by a recent examination of the exterior and interior portion of the Murchison meteorite (KOTRA *et al.*, 1979).

Considering the special environment for preservation in Antarctica, observation of the recovered body of the meteorite suggested that it was not altered and contaminated after its fall (YANAI and HARAMURA, 1978). The current study of amino acids supports this observation and emphasizes the importance of studies of the Antarctic meteorites.

Acknowledgments

We are grateful to Dr. P. E. HARE, Carnegie Institution of Washington, for his help with the amino acid analysis. We thank Prof. T. NAGATA, Director of the National Institute of Polar Research of Japan, for the gift of a sample of the Yamato meteorite for analysis. This research was supported by NASA Grant NGR-21-00-317.

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(*Received May 2, 1979*)