Amino acids in the sediments and interstitial waters from ODP Holes 677B and 678B in the Panama basin

Hodaka KAWAHATA [a] and Toshio ISHIZUKA [b]

[a] Geological Survey of Japan, Tsukuba, Ibaraki, 305, Japan.

[b] Ocean Research Institute, University of Tokyo, Nakano, Tokyo, 164, Japan.

ABSTRACT

Biogenic siliceous and calcareous sediments were drilled at sites 677 and 678 on the flank of Costa Rica Rift in the Panama basin. We analysed DCAA (dissolved combined amino acids) and DFAA (dissolved free amino acids) in the interstitial waters as well as THAA (total hydrolyzable amino acids) in the sediments in order to evaluate the relationship between THAA, DCAA and DFAA.

Comparison of the THAA in the sediments and in marine settling particulate matter (Ittekkot *et al.*, 1984 *b*) showed that the concentration of non-protein amino acids (*e.g.*, ß-alanine, γ-aminobutyric acid, and ornithine) was much higher in the sediments and that the neutral amino acid fraction was depleted in the sediments. These results, together with the organic carbon profile, indicated that the production of non-protein amino acids occurred mainly at shallow depth but still continued through the sedimentary column, at a slow rate and that the neutral amino acid fraction was more liable to decomposition than the acidic and basic amino acid fractions.

The positive correlation between total DCAA and the DFAA and the weak correlation of the total amino acid concentrations between the interstitial waters and sediments suggested that biological and/or chemical reaction rates between the DCAA and the DFAA were much higher than the digestion and/or transformation rates of the THAA in the sediments. The higher ratio of neutral to acidic amino acid fractions in the DFAA than in the DCAA was attributed mainly to reaction or adsorption with carbonates and partly to the contribution of decomposed products of the neutral fraction in the THAA of the sediments.

Oceanologica Acta, 1993. **16**, 4, 373-379.

RÉSUMÉ

Acides aminés dans les sédiments et les eaux interstitielles des sites ODP 677 B et 678 B du bassin de Panama

Les sédiments biogéniques siliceux et calcaires ont été forés sur les sites 677 et 678 sur le flanc du rift de Costa Rica, dans le bassin de Panama. Les acides aminés combinés dissous (DCAA) et libres dissous (DFAA) des eaux interstitielles et les acides aminés hydrolysables totaux (THAA) des sédiments ont été analysés pour évaluer les rapports entre eux.

La comparaison de THAA dans les sédiments et dans les particules en suspension (Ittekot *et al.*, 1984 *b*) montre que la concentration des acides aminés non-protéiques (par exemple β-alanine, acide g-aminobutyrique, et ornithine) est très

supérieure dans les sédiments, et que la fraction d'acide aminé neutre est épuisée dans les sédiments. Ces résultats, ainsi que le profil de carbone organique, indiquent que la production des acides aminés non-protéiques intervient surtout à de faibles profondeurs, mais se poursuit faiblement à travers la colonne sédimentaire, et que la fraction d'acide aminé neutre est plus sujette à décomposition que les fractions d'acides aminés acide et basique.

La corrélation positive entre DCAA total et DFAA et la faible corrélation des concentrations en acides aminés totaux entre les eaux interstitielles et les sédiments suggèrent que les vitesses des réactions biologiques et/ou chimiques entre DCAA et DFAA sont très supérieures aux vitesses de digestion et/ou transformation de THAA dans les sédiments. Le rapport des fractions d'acides aminés neutre et acide prend une valeur plus grande dans DFAA que dans DCAA, ce qui est surtout attribué à la réaction ou à l'adsorption avec les carbonates et, en partie, à la contribution des produits de décomposition de la fraction neutre dans THAA des sédiments.

INTRODUCTION

Amino acids are common components of all organisms and constitute a major fraction of nitrogenous compounds in sediments and interstitial waters (Degens, 1970; Henrichs and Farrington, 1979). They undergo decomposition and transformation during early diagenesis. Amino acid compounds dissolved in interstitial waters should be very sensitive indicators of these processes. Analyses of total dissolved amino acids show much higher concentrations in interstitial waters than in the overlying sea water, indicating that some processes in sediments must be supplying soluble compounds. Several studies of amino acids in interstitial waters have been carried out but they concerned only dissolved free amino acids (Lee and Bada, 1975; Henrichs and Farrington, 1979; Bada *et al.*, 1982; Michaelis *et al.*, 1982). However, in studies by Ishizuka *et al.* (1988), Kawahata and Ishizuka (1989), and Kawahata *et al.* (1990), both dissolved combined and free amino acids in interstitial waters were analysed.

Holes 677A and 678B are of interest in studying the distribution of amino acids through marine long cores since inorganic and mineralogical studies indicate a fairly constant composition of the sediments through the holes (Leg 111 Shipboard Scientific Party, 1988; Kawahata *et al.*, 1991). Furthermore, high biogenic productivity results in a rapid sedimentation of organic matter (Leg 111 Shipboard Scientific Party, 1988).

The purposes of this study are to present the characteristics of the amino acid composition of sediments and interstitial waters and to evaluate their relations during early diagenesis.

SUMMARY OF GEOLOGICAL FRAMEWORK AND CORE DESCRIPTION

Two holes were drilled at Site 677 (1°12.14', 83°44.22'W) at a water depth of 3,461 m. Site 678 was drilled at 1°13.01'N, 83°43.39'N at a water depth of 3,435 m (Fig. 1).

The former site was located in a lower (166 mW/m^2) and the latter in a higher (250 mW/m^2) heat flow zone (Langseth *et al.*, 1988; Leg 111 Scientific Drilling Party, 1987; Leg 111 Shipboard Scientific Party, 1987; 1988).

Hole 677B was cored to 97.1 m subbottom depth. The sediments were composed of alternating clayey biogenic calcareous and siliceous oozes and clayey biogenic siliceous and calcareous oozes, ranging in age from Pleistocene to late Pliocene. The sedimentation rate, established on microfossils, averaged about 48 mm/ky with little variation for the last 5.95 Ma (Leg 111 Shipboard Scientific Party, 1988).

Hole 678B was drilled down to 170.1 m subbottom depth. Three sedimentary units and one basaltic unit were recognized. Unit 1 consisted of clayey calcareous and siliceous ooze (0.0-27.7 m). Unit 2 consisted of clayey diatom nannofossil chalk (95.5-111.8 m). Unit 3 was composed of
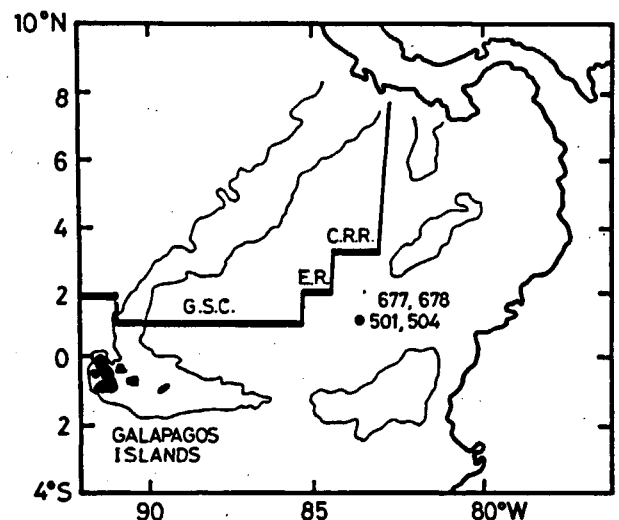

Figure 1

Location of ODP Sites 677 and 678 in the Panama Basin of the Costa Rica Rift. (G.S.C.= Galapagos Spreading Center; E.R.=Ecuador Rift; C.R.R.=Costa Rica Rift).

limestone (111.8-169.8 m) and Unit 4 consisted of basalt fragments and conglomerates (169.8-170.1 m).

Porosity values ranged from 84.0 % at the seafloor to 76.9 % at the bottom of Hole 677B, and from 88.0 to 71.2 % at the base of Hole 678B. Generally, bulk density decreased *versus* subbottom depth (Leg 111 Shipboard Scientific Party, 1988). Downhole temperature was measured in the upper 100 m of Hole 677A. Its linear extrapolation yielded a basement temperature of the order of 60-70°C.

Smear slide results for Holes 677B and 678B showed that biogenic carbonate and silica comprise about 46 and 34 % of the sediments, respectively. Within the calcareous fraction, nannofossils were the dominant constituents (average 38 %). Within the siliceous fossil fraction the percentage of diatoms (average 16 %) often exceeded that of radiolarians (average 12 %), but each of these fossil groups formed a significantly larger portion than sponge spicules (average 5 %) and silicoflagellates (average 0.7 %). Clay minerals were a third dominant fraction in the sediments, ranging from 3 to 40 %, averaging 16 %. Smectites dominated chlorite, illite and kaolinite and accounted for about 70 % of clay minerals. Detrital materials including quartz and feldspar made up less than 5 %. Framboidal pyrite originating in the reduction of sulfate by bacteria constituted on average only 1 % of the sediments (Kawahata *et al.*, 1991).

METHODS

Sampling and storage

Interstitial waters from Holes 677B and 678B were extracted on R/V *ODP Joides Resolution* by hydraulic squeezing. All interstitial water samples were filtered through 0.22-mm Millipore cellulose acetate filters in the shipboard laboratory. For amino acid analysis in interstitial waters, a 5 ml aliquot of the water sample was taken in a precombusted glass ampoule, which was sealed after flushing with helium, stored in a refrigerator for one month and returned to the land-based laboratory. After sampling interstitial waters, squeezed sediments were stored at - 20°C before the analysis. We analysed the total of thirty samples. Fourteen samples were taken from Hole 677B at regular intervals. Also, a few samples were selected from each lithologic unit of Hole 678B.

Analysis of amino acids procedures

Dissolved free amino acids (DFAA) in interstitial water were analysed after the addition of 7 ml of distilled 2M HCl with 0.7 ml of each interstitial water sample. Each amino acid concentration was determined by direct injection into an automated liquid chromatograph (Hitachi Model 835). The reagent blank was suitably corrected.

For analyses of total hydrolyzable amino acids (THAA), samples of 1 ml of the interstitial water were added to distilled concentrated HCl to a final concentration of 6M solution and hydrolyzed at 110°C for 22 hours in precombusted glass ampoules under an argon atmosphere. The hydrolyzed
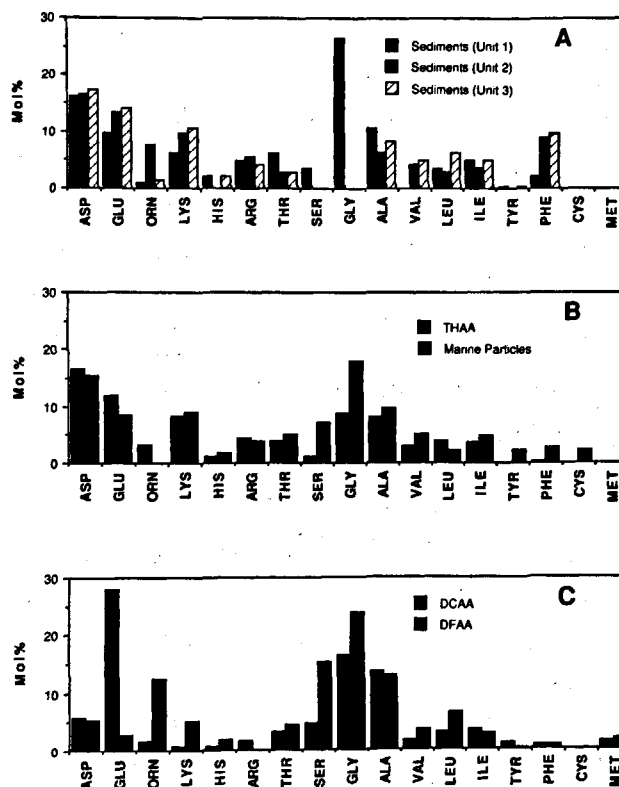

Figure 2

A) Relative abundances of THAA (total hydrolyzable amino acids) of the sediments from Holes 678B; B) Average relative abundances of THAA of the sediments from Holes 678B and of marine particulate matter (Ittekkot et al., 1984 b); C) Average relative abundances of DCAA (dissolved combined amino acids) and DFAA (dissolved free amino acids) in interstitial waters from Holes 677B and 678B.

samples were gently evaporated using a rotary evaporator at a temperature below 42°C and the residue redissolved in 1ml of 0.01 M HCl. The aliquots were then injected into the amino acid analyzer. The standard deviation for amino acid analysis based on replicated measurements of a standard solution (about 1mmol/l for amino acids) was less than 10 % and the detection limit of our method was about 0.03 mmol/l. Low values in free and hydrolyzable amino acids near the detection limit may not be dependable. Nevertheless, we retain these values because they confirm that the amino acid concentration is low (Ishizuka *et al.*, 1988).

For the total hydrolyzable amino acids (THAA) of a sediment, 0.1 g of the sediments, together with distilled concentrated HCl, to make 6N solution, were placed in a precombusted glass ampoules and hydrolyzed at 110°C for 22 hours under an argon atmosphere. After hydrolization, the samples were filtered through a 0.2 mm filter. Other procedures were the same as those followed in the analysis of THAA in interstitial waters.

RESULTS

THAA in the sediments

Total THAA concentrations were 2.09 mmol/g at 0.5 m subbottom depth and 0.183 mmol/g at 18.7 and 97.0 m.

Table

Amino acid compositions of the sediments, interstitial waters, and marine particulate matter.

|	Sediment THAA (a) (%)	Interstitial water DCAA (a) (%)	Interstitial water DFAA (a) (%)	Marine particulate matter (b) (%)
Acidic (sum)	28.7	33.8	8.1	24.0
Aspartic acid	16.6	5.6	5.4	15.4
Glutamic acid	12.1	28.0	2.7	8.6
Basic (sum)	17.2	4.8	19.4	14.9
Ornithine	3.3	1.6	12.4	0.1
Lysine	8.3	0.8	6.1	9.0
Histidine	1.2	0.8	1.8	1.8
Arginine	4.4	1.6	0.1	4.0
Neutral (sum)	32.7	45.8	68.8	52.3
hydroxy				
Threonine	3.9	3.3	4.4	6.1
Serine	1.2	4.6	15.2	7.2
straight				
Glycine	8.8	16.4	23.7	18.0
Alanine	8.0	13.5	12.9	9.7
branch				
Valine	3.0	1.7	3.4	5.1
Iso-leucine	4.0	2.9	6.4	2.4
Leucine	3.8	3.4	2.8	4.6
Aromatic (sum)	0.2	1.8	0.9	5.1
Tyrosine	0.0	1.1	0.3	2.3
Phenylalanine	0.2	0.7	0.6	2.8
Sulfur (sum)	0.0	1.3	1.9	2.2
Cysteine	0.0	0.0	0.0	2.2
Methionine	0.0	1.3	1.9	-
The rest (sum)	21.2	11.7	0.9	1.5
Taurine	1.5	0.5	0.0	-
Citrulline	0.0	8.4	0.0	-
Cystathionine	0.0	0.1	0.7	-
β-alanine	7.0	1.2	0.1	0.9
α-aminobutyric acid	0.0	1.5	0.1	-
β-amino iso-butyric acid	0.0	0.0	0.0	-
γ-aminobutyric acid	10.8	0.0	0.0	0.6
α-aminoadipic acid	1.9	0.0	0.0	-
Total	100.0	99.2	100.0	100.0

Note: -: not reported.
(a) this study,
(b) Ittekkot et al., 1984b (excluding an unusual coccolithophorid bloom)

Neutral amino acids were the most abundant THAA fraction and accounted for 32.7 mol % of total THAA in the sediments from Hole 678B (Fig. 2A and B, Tab.). The second abundant fraction was acidic amino acids, which constituted 28.7 mol %, of the total THAA. Basic and aromatic amino acid fractions made up 17.2 mol % and 0.2 mol %, respectively. The remaining amino acids constituted 21.2 mol %; γ-aminobutyric acid, ß-alanine; α-aminoadipic acids accounted for 10.8 mol %, 7.0 mol %, and 1.9 mol %, respectively.
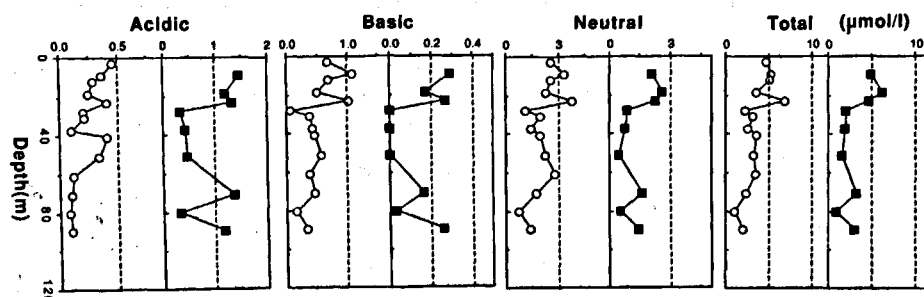
DCAA in the interstitial waters

DCAA concentration was calculated by subtracting the dissolved free amino acid (DFAA) concentration from the total hydrolyzable amino acid concentration in the interstitial waters. Average DCAA concentration in the interstitial waters is given in the Table and presented in Figure 2C. DCAA downcore distribution is plotted in Figure 3 and Figure 4. Total concentration of DCAA ranged from 0.92 mmol/l to 9.23 mmol/l, averaging 4.53 mmol/l. The total concentrations decreased with depth in Hole 677B whereas they remained nearly uniform in Hole 678B. But the relative high values were found in the deeper part.

Total DCAA showed fairly similar spectral compositions. The relative abundances of the acidic, basic, neutral, aromatic and sulfur-containing amino acid fractions averaged 33.8 mol %, 4.8 mol %, 45.8 mol %, 1.8 mol %, and 1.3 mol %, respectively (Tab., Fig. 5).

DFAA in the interstitial waters

Average DFAA concentration in the interstitial waters is given in the Table and presented in Figure 2C. DFAA concentrations are plotted *versus* subbottom depth in Figure 3 and Figure 4. The total concentration of DFAA ranged from 0.87 mmol/l to 6.92 mmol/l, averaging 3.09 mmol/l. The profiles are similar to those of DCAA. The relative abundances of the acidic, basic, neutral, aromatic, and sulfur-containing fractions from Holes 677B and 678B were on average 8.1 mol %, 19.4 mol %, 68.8 mol %, 0.9 mol %, and 1.9 mol %, respectively (Tab., Fig. 6).

DISCUSSION

Comparison of sediment THAA with settling organic particles

Marine particulate matter settling on the sea floor constitutes the most important amino acid contributor to sediments. The average amino acid composition of settling particles collected at 3,560 m by time-series sediment traps in the Panama Basin (5°22'N, 85°35'W) from December 1979 through May 1980 is presented in the Table and Figure 2B (Ittekkot et al., 1984 b). The main sources of the amino acids were calcareous and siliceous biogenic debris.

Comparison of the amino acid composition of Hole 677B sediments and settling particulate matter provides informa-
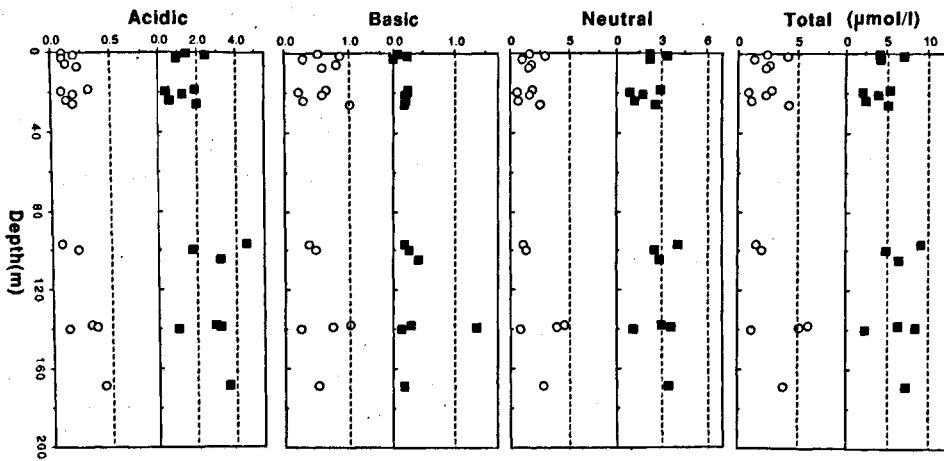
Figure 3

Vertical distribution of the concentrations of acidic, basic, neutral and total amino acids of DFAA (dissolved free amino acid) (open circle) and DCAA (dissolved combined amino acid) (solid square) in interstitial waters from Hole 677B.

Figure 4

Vertical distribution of the concentrations of acidic, basic, neutral and total amino acids of DFAA (dissolved free amino acid) (open circle) and DCAA (dissolved combined amino acid) (solid square) in interstitial waters from Hole 678B.

tion on the degree of compositional modification during early diagenesis. Especially significant in this regard is the presence of appreciable concentrations of ß-alanine and γ-aminobutyric acid in the sediments. These are absent from ultracleaned foraminifera tests (Schroeder, 1975) and are generally considered to be enzymatic decomposition products of aspartic and glutamic acids, respectively. Such decomposition can take place either on particles (Lee and Cronin, 1982) or in the guts of organisms (Ittekkot et al., 1984 a). The rapid transport to the deep sea of freshly produced particulate matter is so immediate that the relative abundance of non-protein amino acids collected in sediment traps is generally fairly low [< 3 mol % (Ittekkot et al., 1984 a, b)]. The concentration of non-protein amino acids as well as the total THAA concentrations were high in surface sediments and low at the 18.7 and 97.0 m levels of Hole 678B. The total organic carbon profile showed gradual decrease through the hole, indicating slight but continuous

degradation of organic matter through the hole. These results suggest that the production of non-protein amino acids occurred mainly in surface sediments but nevertheless continued through the sedimentary column at slow rate.

The THAA spectra might indicate a trend of amino acid degradation in the sediments. The neutral and aromatic amino acid fractions were more depleted in the sediments than in the settling particulate matter, while the acidic and basic amino acid fractions were more enriched. If we take into consideration the fact that ß-alanine and γ-aminobutyric acid were produced from aspartic and glutamic acids (Ittekkot et al., 1984 b), the total abundance of these four amino acids accounts for as much as 50 % of the total THAA. On the other hand, the neutral fraction was more liable to alteration during early diagenesis and might produce a dissolved free neutral fraction as the decomposed product, an event which may be responsible for the higher

Figure 5

A) Acidic amino acids versus neutral amino acids of DCAA in interstitial waters from Holes 677B (open square) and 678B (open triangles); B) Basic amino acids versus neutral amino acids of DCAA from Hole 677B and 678B.
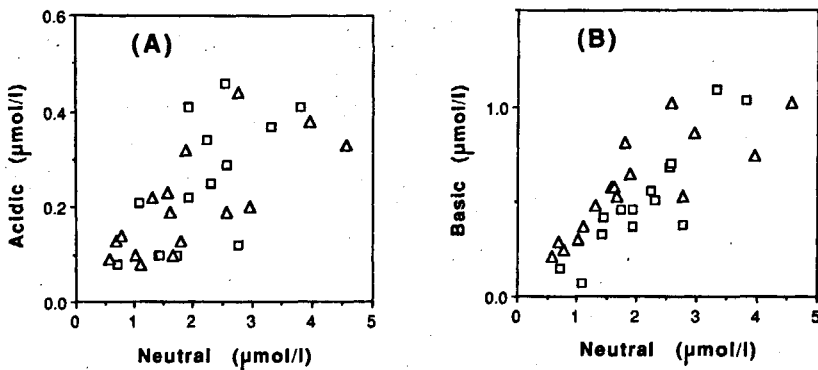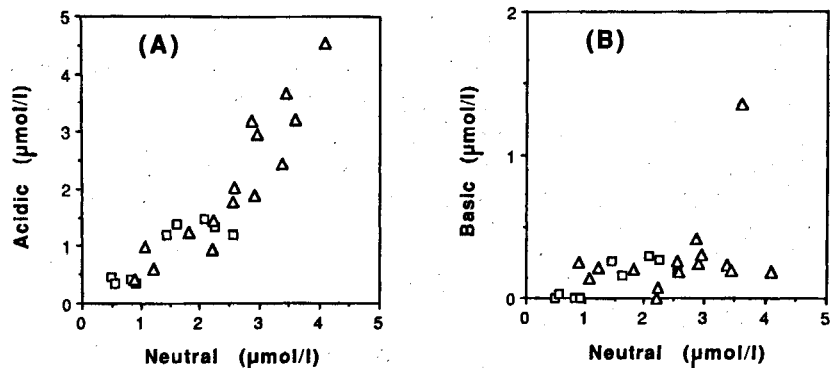

Figure 6

A) Acidic amino acids versus neutral amino acids of DFAA in interstitial waters from Holes 677B (open square) and 678B (open triangles); B) Basic amino acids versus neutral amino acids of DFAA from Hole 677B and 678B.

ratio of neutral to acidic amino acid fractions in the DFAA than in the THAA of the sediments. Within the basic fractions, ornithine was the major amino acid in the sediments. This is also considered to be a decomposition product of arginine. The relative abundances of the other basic fractions in the sediments are comparable to those in the marine settling particulate matter. Since basic amino acid is positively charged, the basic fractions may have escaped decomposition more than other fractions in their reaction with clay minerals.

Amino acids in the interstitial waters

The major characteristics of DCAA and DFAA in the interstitial waters were: 1) positive correlation between total DCAA and total DFAA; 2) higher values of the total DCAA and DFAA at shallow depth and above the basaltic basement; 3) higher ratios of neutral to acidic amino acid fractions in the DFAA than in the DCAA (Fig. 5 and 6); and 4) lower concentrations of non-protein amino acids in the DFAA than in the DCAA.

Amino acids in sea water and interstitial waters are present in the combined and free states. As also observed by Lee and Bada (1975), there was a concentration some ten times greater of combined than of free amino acids in sea water with DCAA enriched in alanine and aspartic and glutamic acids. However, in interstitial water from ODP Hole 695A in the Weddel Sea of the Antarctic Ocean, where diatom oozes and silty mud were predominant (Kawahata et al., 1990), DFAA were more predominant than DCAA. The DCAA concentrations in the interstitial waters from Hole 677B and 678B were only about 1.5 times greater than those of DFAA. Microbiological productivities and stability of combined and free amino acids in the solution would have affected their relative abundance.

The relation between THAA in the sediments and total dissolved amino acids (DCAA + DFAA) in the interstitial water is of interest. Concentrations of THAA, DCAA and DFAA were 2.09 mmol/g, 4.17 mmol/l and 2.30 mmol/l at 0.5 m subbottom depth, 0.183 mmol/g, 5.48 mmol/l, and 2.85 mmol/l at 18.7 m, and 0.183 mmol/g, 9.23 mmol/l and 1.55 mmol/l at 97.0 m, respectively. The sediment at 0.5 m contained about ten times the amount of THAA at 18.7 and 97.0 m. But up to a twofold change of total DCAA and DFAA was found although total DCAA positively correlated with total DFAA through the holes. These results suggest that the digestion and/or transformation rates of THAA in the sediments were much slower than biological and/or chemical reaction rates between DCAA and DFAA in the interstitial waters.

Although the organic carbon content of the sediments generally decreased with depth in Hole 678B, higher values were found in the lower part of the hole. Since Hole 678B is located in the higher heat flow zone, a small amount of warm water migrates upward from the basaltic basement (e.g., Leg 111 Shipboard Scientific party, 1988). The present temperature of the lower part was estimated to be 30-60°C. As the sediments were buried so deep, they were heated up. In general, amino acid constitutes one of most unstable biochemical compounds under the influence of thermal stress. It might raise the total DCAA and DFAA during the decomposition and/or transformation of organic matter in the sediments.

As discussed above, the far higher ratio of neutral to acidic amino acid fractions in the DFAA than in the DCAA resulted partly from the contribution of decomposed products of neutral amino acid fraction of the THAA. Another plausible explanation is the reaction of acidic fraction with sedimentary particles.

Glutamic acid is often one of the major constituents in interstitial waters (Garder and Hanson, 1979; Jørgensen et al., 1981; Burdige and Martens, 1984; Jørgensen, 1984; Henrichs et al., 1984; Henrichs and Farrington, 1987). Garder and Hanson (1979) suggest that bacteria may produce relative high levels of glutamic acid and alanine in DFAA in interstitial waters. Henrichs et al. (1984) also suggest a bacterial source for most of the DFAA in sediments where sulfate reduction occurs. Also acidic amino acids are more stable than basic amino acid in the free state (Abelson, 1959). However, aspartic and glutamic acids are much more depleted in the DFAA than in the DCAA (Tab.; Fig. 2C, 3 and 4).

Acidic amino acids react with calcite, which can be best explained by a specific episodic effect in which aspartic or glutamic acid forms a protective overgrowth on the carbonate surface (Jackson and Bischoff, 1971). Carter (1978) and Carter and Mitterer (1978) indicate that the carbonate surface appears to selectively adsorb aspartic acid-enriched organic matter while the non-carbonate fraction in our sediments make up 46 % of the total sediments. The low concentration of acidic amino acids of DFAA was partly due to the reaction or adsorption with carbonates.

Acknowledgements

REFERENCES

Abelson P.H. (1959) Geochemistry of organic substances. in: *Research in geochemistry*, P.A. Abelson editor, John Wiley, New York, 79-103.

Bada J.L., E. Hoopes and M.-S. Ho (1982). Combined amino acids in Pacific Ocean waters. *Earth planet. Sci. Letts*, **58**, 276-284.

Burdige D. and C.D. Martens (1984). Amino acid cycling in an organic-rich marine sediments. *Eos*, **65**, 960.

Carter P.W. (1978). Adsorption of amino acid-containing organic matter by calcite and quartz. *Geochim. cosmochim. Acta*, **42**, 1239-1242.

Carter P.W. and P.M. Mittere (1978). Amino acid composition of organic matter associated with carbonate and non-carbonate sediments. *Geochim. cosmochim. Acta*, **42**, 1231-1238.

Degens E.T. (1970). Molecular nature of nitrogenous compounds in seawater and recent marine sediments. in: *Symposium on Organic Matter in natural waters*, D.W. Hood, editor. Institute of Marine Science, Alaska, Occasional Publication, 1, 77-106.

Garder W.S. and R.B. Hanson (1979). Dissolved free amino acids in interstitial water of Georgia salt marsh soils. *Estuaries*, **2**, 113-118.

Henrichs S.M. and J.W. Farrington (1979). Amino acids in interstitial waters of marine sediments. *Nature*, **279**, 319-322.

Henrichs S.M. and J.W. Farrington (1987). Early diagenesis of amino acids and organic matter in two coastal marine sediments. *Geochim. cosmochim. Acta*, **51**, 1-15.

Henrichs S.M., J.W. Farrington and C. Lee (1984). Peru upwelling region sediments near 15°S. 2: Dissolved free and total hydrolyzable amino acid. *Limnol. Oceanogr.*, **29**, 20-34.

Ishizuka T., Y. Nozaki and K. Shimooka (1988). Amino acids in the interstitial waters of ESOPE long cores from two North Atlantic abyssal plains. *Geochem. J.*, **22**, 1-8.

Ittekkot V., W.G. Deuser and E.T. Degens (1984 a). Seasonality in the fluxes of sugars, amino acids, and amino sugars to the deep ocean: Sargasso Sea. *Deep-Sea Res.*, **31**, 1057-1069.

Ittekkot V., E.T. Degens and S. Honjo (1984 b). Seasonality in the fluxes of sugars, amino acids, and amino sugars to the deep ocean: Panama Basin. *Deep-Sea Res.*, **31**, 1071-1083.

Jackson T.A. and J.L. Bischoff (1971). The influence of amino acids on the kinetics of the recrystallization of aragonite to calcite. *J. Geol.*, **79**, 493-497.

Jørgensen N.O.G. (1984). Microbial activity in the water-sediment interface: assimilation and production of dissolved free amino acids. *Oceanis*, **10**, 437-365.

Jørgensen N.O.G., P. Lindroth and K. Mopper (1981). Extraction and distribution of free amino acids and ammonium in sediments and overlying sea waters from the Limfjord, Denmark. *Oceanologica Acta*, **4**, 465-474.

Kawahata H. and T. Ishizuka (1989). Organic property of sediments and amino acids in interstitial waters from the flank of Costa Rica rift, Galapagos Spreading Center (ODP Site 677 and 678). in: *Proceedings of the ODP Scientific Results*, **111**, K. Becker, H. Sakai *et al.*, editors. College Station, TX (Ocean Drilling Program), 215-225.

Kawahata H., T. Ishizuka and T. Nagao (1990). Amino acids in interstitial waters from the ODP Site 695 in the Weddell Sea, Antarctic Ocean. in: *Proceedings of the ODP Scientific Results*, **113**, P.F. Barker, J.P. Kennett *et al.*, editors. College Station, TX (Ocean Drilling Program), 179-187.

Kawahata H., S. Aoki, S.D. Scott and T. Ishizuka (1991). Geochemistry of sediments from the flank province of Costa Rica rift in the Panama basin. *Bull. geol. Surv. Japan*, **42**, 199-220.

Langseth M.G., M.J. Mottl, M.A. Hobart and A. Fisher (1988). The distribution of geothermal and goechemical gradients near Site 501/504: implications for hydrothermal circulation in the oceanic crust. in: *Proceedings of the ODP Scientific Results*, **111**, K. Becker, H. Sakai *et al.*, editors. College Station, TX (Ocean Drilling Program), 23-32.

Lee C. and J.L. Bada (1975) Amino acids in equatorial Pacific Ocean water. *Earth planet. Sci. Letts*, **26**, 61-68.

Lee C. and C. Cronin (1982). The vertical flux of particulate organic nitrogen in the sea: decomposition of amino acids in the Peru upwelling area and the equatorial Atlantic. *J. mar. Res.*, **40**, 227-251.

Leg 111 Scientific Drilling Party (1987). Costa Rica rift hole deepened and logged. *Geotimes*, **8**, 14-16.

Leg 111 Shipboard Scientific Party (1987). New from a deepening hole. *Nature*, **325**, 484-485.

Leg 111 Shipboard Scientific Party (1988). Site 677 and 678. in: *Proceedings of the ODP. Initial Reports*, **111**, K. Becker, H. Sakai *et al.*, editors. Washington, US Government Printing Office, 253-348.

Michaelis W., B. Mycke, J. Vogt, G. Schuetze and E.T. Degens (1982). Organic geochemistry of interstitial waters, sites 474 and 479, Leg 64. in: *Initial Reports. DSDP*, **64**, J.R. Curray, D.G. Moore *et al.*, editors. Washington, US Government Printing Office, 933-937.

Shroeder R.A. (1975). Absence of β-alanine and γ-aminobutyric acid in cleaned foraminiferal shells: implications for use as a chemical criterion to indicate removal of non-indigenous amino acid contaminants. *Earth planet. Sci. Letts*, **25**, 272-278.