ORGANIC MATTER AND LIGANDS

4. Carbon and Nitrogen Biogeochemical Cycle at Earth's Surface

The abundance of carbon at the earth's surface potentially exchangeable with atmospheric carbon dioxide is approximately 43×10^{18} gC. The ocean holds about 90% of this "active" carbon, in either inorganic or organic form. Carbon and nitrogen are incorporated into the biogeochemical cycle in the ocean and buffer the level of atmospheric carbon dioxide.

Improved knowledge of the carbon and nitrogen biogeochemical cycle is of critical importance to understanding and responding effectively to issues on regional and global climatic change. Our research focused on clarifying the dynamics of chemical compounds in the ocean and their relationship to the global carbon cycle.

4.1 Detection of dissolved protein molecules in oceanic waters and bacterial membranes: Possible source of a major dissolved protein in seawater

Tanoue (1995)

Tanoue, Nishiyama, Kamo and Tsugita (1995)

Dissolved organic matter (DOM) in seawater is one of the three reactive reservoirs of organic matter on the planet; the other two are living plants and organic matter in soil on land. Although evidence indicates that the majority of DOM originated in marine environments, only less than 30% of the component molecules, such as combined amino acids, carbohydrates, and solvent-extractable lipids, have been identified so far. To date, there is a gap in our understanding of organic constituents between those of marine organisms (sources of organic matter) and those of the inanimate organic (DOM) pool in both classes and quality.

Tanoue (1995) and Tanoue *et al.*(1995) developed a new method for extraction and detection of dissolved protein molecules in oceanic waters and studied bacterial membranes as a possible source of a major dissolved protein in seawater. They measured dissolved protein at a variety of depths at three stations in the Pacific, ranging from the tropics to the subarctic. Most dissolved protein is distributed over a wide range of molecular masses, but consists of fewer than 30 individual proteins. One, with an apparent molecular mass of 48 kDa, is a major constituent at all stations (Fig. 95-14). Its N-terminal amino acid sequence was found to be a homologue of porin P, a trans-outer-membrane channel protein of gram-negative bacteria.

The correspondence of N-terminal amino acid sequences and apparent molecular masses between this dissolved protein and porin P indicates that almost the complete homologue of porin P, from the N-terminus to (probably) the C-terminus, survives without modification in the water column (Table 95-5). Persistence of appreciable amounts of an identifiable protein suggests a pathway for the production of dissolved organic matter whereby enzyme-resistant biopolymers survive and accumulate in the sea. The accumulation of appreciable amounts of a relatively limited number of proteins leads to the hypothesis that particulate proteins that make up the majority of dissolved protein components in seawater are derived from specific sources and contribute quantitatively to the oceanic organic nitrogen pool.

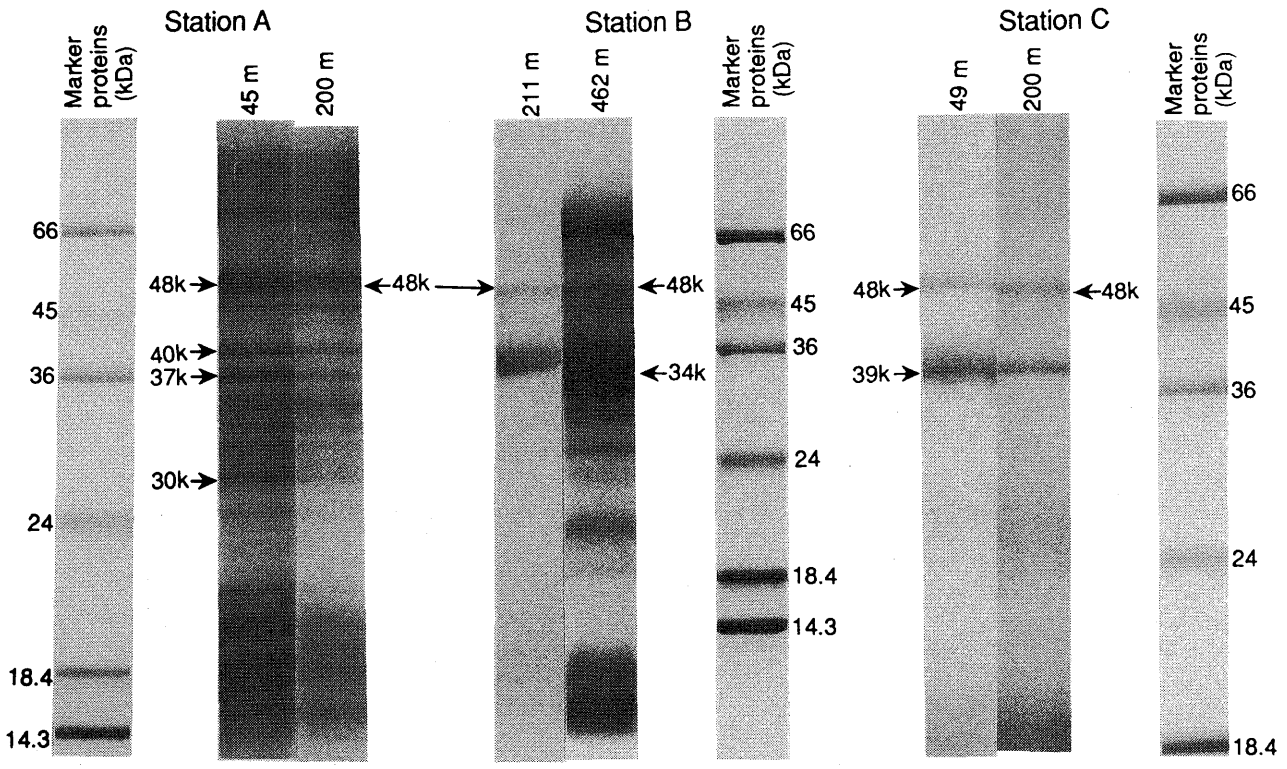

Fig. 95-14 Electrophoresis of dissolved proteins extracted from surface and intermediate waters at three stations: subarctic Station A (45°10.3′N, 165°34.4′E); subtropical Station B (24°35.0′N, 170°00.1′E); and tropical Station C (22°47.1′N, 158°04.6′W). Standard marker proteins are shown for all stations. Proteins in certain bands (arrows) on gels were subjected to N-terminal amino acid sequencing (Table 95-5). Amounts of samples loaded on gels were equivalent to 1 liter of original seawater at Stations A and C, and 0.25 liters of original seawater at Station B. Each marker protein was loaded at 1 μg for Stations A and C, and at 2 μg for Station B. Reprinted from *Geochim. et Cosmochim. Acta*, **59**, Tanoue *et al.*, Bacterial membranes: Possible source of a major dissolved protein in seawater, 2643-2648, Copyright (1995), with permission from Elsevier Science.

Table 95-5 N-terminal amino acid sequences of the proteins in the dissolved phase. "Xaa" indicates that an identifiable phenylhydantoin derivative was not recovered.

Cycle	mol.mass of protein	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Station A (45°10.3′N, 165°34.4′E; depth, 5,934 m)																
45 m	48 kDa	Gly Gln*	Thr	Val	Thr	Thr	Asp	Gly	Ala	Asp	Ile	Val	Ile	Lys	Thr	
	40 kDa	Thr Gly*	Val	Thr	Val	Thr	Pro	Leu	Met	Leu	Gly	Tyr	Thr	Phe	Gln	Leu
	37 kDa	Ala	Asp	Val	Lys	Ile	Tyr	Gly	Arg	Ala	His	Val	Ser	Leu	Asp	Tyr
	30 kDa	Thr	Gln	Ala	Glu	Val	Gly	Ala	Ser	Ala	Gly	Leu	Ile	Asp	Pro	Asp
200 m	48 kDa	Gly Ala*	Thr	Val	Thr	Thr	Asp	Gly	Thr**	Asp	Ile	Val	Ile	Lys	Thr	Lys
Station B (24°35.0′N, 170°0.1′E; depth, 5,966 m)																
211 m	48 kDa	Gly	Thr	Val	Thr	Thr	Asp	Gly	Ala	Asp	Leu**	Val	Ile	Lys	Thr	Lys
462 m	48 kDa#	Gly	Thr	Val	Thr	Thr	Asp	Gly	Ala	Asp	Xaa	Val	Ile	Lys	Thr	Lys
		Met	Lys	Asp	Gly	Leu	Val	Glu	Arg	Thr	Gln	Gly	Ser	Glu	Val	Asp
	34 kDa	Val	Thr	Gly	Gly	Tyr	Ala	Arg	Leu	Pro	Val	Glu	Leu	Tyr	Lys	
Station C (22°47.1′N, 158°4.6′W; depth, 4744 m)																
49 m	48 kDa	Gly	Thr	Val	Thr	Thr	Asp	Gly	Xaa	Asp	Ile	Val	Ile	Lys	Thr	Lys
	39 kDa	Ala	Val	Val	Gly	Gly	Gly	Ala	Thr	Leu	Pro	Gln	Asn	Leu	Tyr	Asn
200 m	48 kDa	Gly	Thr	Val	Thr	Thr	Asp	Gly	Xaa	Asp						
	Porin P##	Gly	Thr	Val	Thr	Thr	Asp	Gly	Ala	Asp	Ile	Val	Ile	Lys	Thr	Lys

* This amino acid was also detected. ** The amino acid was detected but not at a significant level. # The protein in this sample with a molecular mass of 48 kDa was a mixture of two proteins, and two amino acids were detected at every step. One protein was identified as a homologue of porin P and no sequence resembling that of the other protein was found in the PIR database. ## After Worobec et al. (1988) and Siehnel et al. (1990). Reprinted from *Geochim. et Cosmochim. Acta*, **59**, Tanoue *et al.*, Bacterial membranes: Possible source of a major dissolved protein in seawater, 2643-2648, Copyright (1995), with permission from Elsevier Science.

4.2 Detection, characterization, and dynamics of dissolved organic ligands in oceanic waters

Tanoue and Midorikawa (1995)

In the last two decades, increased attention has been paid to the chemistry of metalorganic compounds. The ability of dissolved organic matter to form complexes with metal ions in natural water is of interest because of associated biological implications, such as the bioavailability and toxicity of metals to living organisms, and because of its relevance to efforts to understand geochemical cycles of metals in the environments.

Tanoue and Midorikawa (1995) studied the detection, characterization and dynamics of dissolved organic ligands in oceanic waters, focusing on interaction with copper. Three classes of organic ligands – L_1, L_2 and L_N – concentrated by repeated lyophilization and dialysis, were distinguished by differences in their copper complexing. L_1 and L_2 appear to belong to the group of weak ligands in the literature (Table 95-6). The conditional stability constant of ligand L_N for Cu(II) was extremely high and comparable to that of EDTA (Table 95-7).

Two types of ligand similar to weak ligands L_1 and L_2 were extracted directly from seawater using immobilized metal ion affinity chromatography (IMAC). IMAC gave new insights in showing that the weak ligands were a mixture of at least two types of different organochemical ligands and that their dynamics may be active in the water column.

Table 95-6 Conditional stability constants (K'_{ML}) and concentrations (C_L) of natural ligands in samples of seawater at pH8.15 (EPPS), at an ionic strength of 0.7 M (KNO_3), and at a temperature of 25.0℃. The concentration of ligand has been converted to that in seawater. The volume of the original sample of seawater used was about 51, except in the case of site S-1, 0 m (+: 1.71, #: 1.01)

		Cu(II)								Cd(II)	
Site[a]	Depth	C_{L1} (nM)		log K'_{ML1}		C_{L2} (nM)		log K'_{ML2}		C_L (nM)	log K'_{ML}
	(m)	B	A	B	A	B	A	B	A	A	A
S-1	0+	9.4	8.6	8.34	8.61	62	19	6.38	7.25	-	-
	0#	7.9	7.0	8.48	8.68	45	17	6.57	7.30	-	-
P	0	-	2.2	-	8.89	-	7.3	-	7.09	2.7	6.81
	191	5.1	3.9	8.26	8.41	20	5.5	6.55	7.75	3.8	6.74
J	0	1.9	1.2	9.36	9.60	7.6	6.5	7.50	7.57	1.2	7.21
	523	1.3	1.1	9.20	9.44	5.4	4.0	7.59	7.94	1.4	6.75
	1071	*	3.5	*	9.05	14	11	7.15	7.77	4.5	6.82

[a]Site S-1: at 34°56′ N, 138°41′ E on Apr. 26, 1988. Site P: at 41°32′ N, 147°00′ E on Aug. 13, 1987. Site J: at 44°15′ N, 130°58′ E on Aug. 26, 1987.

Key: *, not detected; -, not determined; B, before demetallization; A, after demetallization.

Table 95-7 Estimated lower limits for the concentration of the undemetallizable ligand, L_N, and the conditional stability constant for copper in various samples of seawater at an ionic strength below 10^{-5} M, at pH5.71 and at 4℃.

Site	Depth (m)	Lower limit C_{L_N}[a] (nM)	Lower limit $\log K'_{CuL_N}$
S-1[b]	0	0.79 ± 0.04	13.9 ± 0.2
	0	0.77 ± 0.04	14.0 ± 0.2
	0	0.80 ± 0.06	14.2 ± 0.2
	0	0.74 ± 0.15	14.0 ± 0.3
	ave.	0.78 ± 0.04	14.1 ± 0.1
S-2[b]	0	1.08 ± 0.20	14.2 ± 0.3
	0	1.04 ± 0.18	13.5 ± 0.3
	ave.	1.06 ± 0.13	14.0 ± 0.2
A	0	0.31 ± 0.03	13.8 ± 0.3
P	0	0.200 ± 0.004	14.3 ± 0.3
	191	0.071 ± 0.004	14.0 ± 0.3
J	0	<0.01	*
	523	0.050 ± 0.002	13.9 ± 0.3
	1071	0.170 ± 0.004	14.0 ± 0.3

*Not estimated.

[a]Concentration of ligand is that in seawater. The values of $[Cu]_{ex}$ was taken as the lower limit for C_{LN} (MIDORIKAWA and TANOUE, 1994a).

[b]Values estimated from multiple subsamples of seawater from Suruga Bay are averaged.

4.3 Relationship between particulate uranium and thorium-complexing capacity of oceanic particulate matter

Hirose (1995b)

Chemical characterization of particulate organic matter (POM) surfaces in their metal complexing is of importance to understanding oceanographic roles of particulate matter (PM). Hirose (1995b) studied the relationship between particulate uranium and thorium-complexing capacity of oceanic particulate matter. Thorium-complexing capacity (ThCC), defined as the amount of thorium adsorption onto PM in 0.1 mol l^{-1} HCl by complexing, has been introduced as a new oceanographic parameter. ThCC implies the concentration of a strong organic ligand in PM.

To specify chemically strong ligands in PM, Hirose compared ThCC in PM with that of particulate uranium, which exists as an organic complex in sea water. ThCC in PM correlated with particulate uranium, and this relationship enables the conditional stability constant of the organic uranium complex in PM to be calculated based on mass action law. The estimated conditional stability constant of the uranium complex in seawater ($10^{14.5}$ l mol^{-1}) is greater than that determined for organic copper complexes, whose order of magnitude coincides with the result of the metal adsorption on microorganisms (Table 95-8). These findings suggest that the strong ligand corresponding to ThCC in PM, which is directly related to the complexation of metals in PM, originates in marine organisms.

Table 95-8a The thorium-complexing capacity in particulate matter and particulate uranium in seawater: surface waters

Sampling date	Location	ThCC (nmol l^{-1})	Particulate U (μBq l^{-1})
The western North Pacific			
Apr. 1991	32°00'N 140°15'E	3.41 ± 0.19	2.90 ± 0.42
Apr. 1991	30°00'N 140°07'E	2.30 ± 0.13	2.13 ± 0.38
May 1991	28°00'N 137°00'E	5.84 ± 0.32	4.96 ± 0.35
May 1991	18°00'N 137°00'E	3.37 ± 0.16	2.79 ± 0.22
May 1991	10°00'N 137°00'E	7.31 ± 0.42	6.07 ± 0.36
May 1991	3°00'N 144°00'E	9.47 ± 0.42	8.24 ± 0.58
May 1991	7°00'N 144°00'E	3.22 ± 0.16	2.17 ± 0.15
May 1991	22°00'N 144°00'E	4.63 ± 0.21	3.29 ± 0.23
The Japan Sea			
May 1993	38°09'N 134°27'E	13.5 ± 0.5	11.3 ± 0.6
May 1993	38°36'N 143°03'E	9.92 ± 0.48	

Table 95-8b The thorium-complexing capacity in particulate matter and particulate uranium in seawater: vertical distribution (location: 39°25'N 133°25'E)

Depth(m)	Temp. (°C)	Salinity (‰)	ThCC	Particulate U
0	13.06	34.51	11.4 ± 0.6	10.5 ± 0.7
10	12.99	34.50	9.67 ± 0.48	
25	10.92	34.44	6.89 ± 0.48	
50	9.05	34.13	4.49 ± 0.31	
100	6.08	34.04	3.05 ± 0.21	2.83 ± 0.40
200	1.63	34.05	1.62 ± 0.21	
300	0.78	34.06	1.42 ± 0.18	
500	0.38	34.07	1.28 ± 0.16	1.22 ± 0.22
750	0.29	34.07	1.23 ± 0.16	1.25 ± 0.22
1000	0.23	34.07	1.06 ± 0.14	1.16 ± 0.21

ACID DEPOSITION

5. Acid Deposition at Summit of Mt. Fuji

Dokiya, Tsuboi, Sekino, Hosomi, Igarashi, and Tanaka (1995)

The summit of Mt. Fuji, the highest mountain in Japan, is a solitary 3,776m peak considered to be in the free atmosphere and presumably free from local pollution. The JMA has run a weather station at the summit since 1932. Because of severe meteorological conditions, no data is available on the amount of precipitation and only a few studies have been made on the chemical species in precipitation at the summit.

Dokiya *et al.*(1995) have measured chemical species in precipitation since August 1990 to evaluate the site as a background air quality station and to obtain information on the long-range transport of chemical species. They conducted intensive observations of chemical species in aerosols, gases, and other samples at the summit of Mt. Fuji and at Tarobo (1,300m up on the mountain's southern slope) during summer 1993 and 1994 to evaluate the sources of chemical species in precipitation.

From July 26 to August 3, 1993, concentrations of gaseous HCl and SO_2 were low and comparable to those at remote sites. The concentration of NH_3 during this period was higher with diurnal variation, however,