Bull. Environ. Contam. Toxicol. (1993) 50:348–355

Environmental
Contamination
and Toxicology

Organic Compounds Found in Dokai Bay, Japan

Akiko Terashi, Yoshifumi Hanada, Azuma Kido, and Seiichi Ishikawa

The Kitakyushu Municipal Institute of Environmental Health Sciences, Shin-ike 1-2-1, Tobata-ku, Kitakyushu-shi, 804, Japan

Dokai Bay is a 13km-long narrow bay with average depth of 7m, which is located in north Kyushu in Japan. There had been serious environmental pollution caused by untreated domestic and industrial wastewater in this area. The pollution was accelerated by the characteristics of this enclosed shape with little water exchange to open sea. Nowadays, the water quality of this bay has improved as a result of the regulation of wastewater, the spreading of sewage treatment system and the dredging of polluted sediment. But there are still many factories producing chemicals, iron and steel around the bay. And large residence area stretches upstream. Therefore, many kinds of chemical substances may accumulate in this area than in an open area.

Today computer-assisted gas chromatography / mass spectrometry has improved to be very useful for analysis of environmental pollutant(Sheldon et.al.1978; Coleman et.al.1980; Elder et.al. 1981). In this study, we analyzed organic chemicals remaining in the sediment by this method to understand the pollution state of Dokai Bay and to obtain information of chemicals that persist in the environment.

MATERIALS AND METHODS

Sediment sample was collected on 28 September 1990 and stored at 5℃ until analysis. Sample was treated with following three methods in order to get additional information in mass spectral identification.

A fifty gram sample was extracted twice with 100ml of acetone-hexane(1:1). The extract was washed with water and concentrated to 10ml. Acidic and basic components were then extracted with 10ml of 1N-NaOH and 1N-HCl, respectively. The organic layer was concentrated again and applied to a silica-gel column and eluted with 5% ether in hexane. The eluate was concentrated to 1ml. The extracts with 1N-NaOH and 1N-HCl were adjusted to pH<2 and pH>12

Send reprint requests to A.Terashi at the above address.

respectively, then extracted with hexane and concentrated to 1ml. For the acidic fraction, a derivatization reaction was performed with bis(trimethylsilyl)trifluoroacetoamide at 80℃ for 30min (Knapp 1979)(method A).

A sample was extracted with 1N-HCl and re-extracted with dichloromethane after pH adjustment. The organic layer was concentrated and trimethylsilyl(TMS) derivatization was done at the same condition as mentioned above (method B).

A sample was extracted with 1N-NaOH and re-extracted with dichloromethane after pH adjustment, and concentrated to 1ml (method C).

A JEOL DX303 mass spectrometer coupled to a Hewlett-Packard 5890A gas chromatograph was used under the following conditions: column, Ultra 2 fused silica capillary column (0.32mm i.d. x 25m x 0.52μm); column temperature, 60℃(1min) increased at 10℃/min to 300℃, held for 10min; injection temperature, 300℃; carrier gas, helium; GC/MS interface temperature, 280℃; ionization voltage, 70eV; scan time, 1sec/c; scan range, 50-500 mass units. Electron impact mass spectral data were acquired and processed with a JEOL DA5100 data system.

The identifications presented here are based on coincidence of gas chromatographic retention times and equivalence of sample spectra with those of authentic compounds or published spectra (Mclafferty et.al.1989). Some compounds were inferred to be present by comparison of mass spectra with those of similar compounds.

RESULTS AND DISCUSSION

The organic compounds found in Dokay Bay sediment are listed in Table 1. Most of them originated from industrial uses.

Alkanes were not prominent, but their presence was confirmed by a reconstructed ion chromatogram at m/z 85. It sugests that their origins were artificial sources, since there was no predominance of odd carbon numbers(matsumoto et.al. 1981). Triterpanes such as hopane, methylhopane, trisnorhopane and cholestane, which were well known to exist in petroleum(Eganhouse 1982), were also identified.

Aliphatic acids were a major component in the acidic fraction. C12-C26 Acids were identified from the acidic fraction of method A, and C5-C18 acids were identified from the extract of method B. The difference of carbon number of the two methods is attributed to increasing hydrophobidity with carbon chain length. C12-C18 Aliphatic acids with even carbon numbers were predominant. These are commonly distributed in all living organisms and known to be

Table 1 Organic Compounds Identified in Dokai Bay

No.	Compounds	Elemental Formula	M.W.	Ext.[a]	Ident.[b]	Relative Size
	Polycyclic Aromatic Hydrocarbons					
1	naphthalene	$C_{10}H_8$	128	A	RT+MS	+++
2	C1-naphthalene	$C_{11}H_{10}$	142	A	MS	++
3	C2-naphthalene	$C_{12}H_{12}$	156	A	MS	+
4	C3-naphthalene	$C_{13}H_{14}$	170	A	MS	+
5	C4-naphthalene	$C_{14}H_{16}$	184	A	MS	+
6	C5-naphthalene	$C_{15}H_{18}$	198	A	MS	++
7	C3-tetrahydronaphthalene	$C_{13}H_{18}$	174	A	MS	+
8	C2-propenylnaphthalene	$C_{15}H_{16}$	196	A	MS	+
9	C3-Propenylnaphthalene	$C_{16}H_{18}$	210	A	MS	++
10	biphenyl	$C_{12}H_{10}$	154	A	RT+MS	+
11	C1-biphenyl	$C_{13}H_{12}$	168	A	MS	+
12	acenaphthylene	$C_{12}H_8$	152	A	RT+MS	++
13	acenaphthene	$C_{12}H_{10}$	154	A	RT+MS	+
14	fluorene	$C_{13}H_{10}$	166	A	RT+MS	++
15	phenanthrene	$C_{14}H_{10}$	178	A	RT+MS	+++
16	anthracene	$C_{14}H_{10}$	178	A	RT+MS	++
17	C2-anthracene[c]	$C_{16}H_{14}$	206	A	MS	+
18	C4-anthracene[c]	$C_{18}H_{18}$	234	A	MS	++
19	benzo(def)fluorene	$C_{15}H_{10}$	190	A	MS	++
20	fluoranthene	$C_{16}H_{10}$	202	A	RT+MS	+++
21	pyrene	$C_{16}H_{10}$	202	A	RT+MS	+++
22	C1-pyrene[c]	$C_{17}H_{12}$	216	A	MS	++
23	C2-pyrene[c]	$C_{18}H_{14}$	230	A	MS	+
24	phenylnaphthalene	$C_{16}H_{12}$	204	A	MS	++
25	benzo(ghi)fluoranthene	$C_{18}H_{10}$	226	A	MS	++
26	benz(a)anthracene	$C_{18}H_{12}$	228	A	RT+MS	+++
27	chrysene / triphenylene	$C_{18}H_{12}$	228	A	RT+MS	+++
28	naphthacene	$C_{18}H_{12}$	228	A	MS	++
29	C1-chrysene[c]	$C_{19}H_{14}$	242	A	MS	++
30	C2-crycene[c]	$C_{20}H_{16}$	256	A	MS	+
31	dihydrobenz(a)anthracene[c]	$C_{18}H_{14}$	230	A	MS	+/-
32	o-terphenyl	$C_{18}H_{14}$	230	A	RT+MS	+/-
33	m-terphenyl	$C_{18}H_{14}$	230	A	RT+MS	+
34	p-terphenyl	$C_{18}H_{14}$	230	A	RT+MS	+
35	methylenechrysene[c]	$C_{19}H_{12}$	240	A	MS	++
36	benzo(b)fluoranthene	$C_{20}H_{12}$	252	A	RT+MS	+++
37	benzo(k)fluoranthene / benzo(j)fluoranthene	$C_{20}H_{12}$	252	A	RT+MS	+++
38	benzo(e)pyrene	$C_{20}H_{12}$	252	A	RT+MS	+++
39	benzo(a)pyrene	$C_{20}H_{12}$	252	A	RT+MS	+++
40	perylene	$C_{20}H_{12}$	252	A	RT+MS	+++
41	C1-benzo(a)pyrene[c]	$C_{21}H_{14}$	266	A	MS	++
42	C2-benzo(a)pyrene[c]	$C_{22}H_{16}$	280	A	MS	+
43	methyleneperylene[c]	$C_{21}H_{12}$	264	A	MS	+
44	indeno(1,2,3,-cd)pyrene	$C_{22}H_{12}$	276	A	RT+MS	+++
45	benzo(ghi)perylene	$C_{22}H_{12}$	276	A	RT+MS	+++
46	C1-indeno(1,2,3-cd)pyrene[c]	$C_{23}H_{14}$	290	A	MS	+
47	benzo(b)chrysene[c]	$C_{22}H_{14}$	278	A	MS	+++
48	C1-benzo(b)chrysene[c]	$C_{23}H_{16}$	292	A	MS	+
49	dibenzo(a,e)pyrene[c]	$C_{24}H_{14}$	302	A	MS	++
50	triphenylbenzene	$C_{24}H_{28}$	306	A	MS	+

Table 1 (Continued)

No.	Compounds	Elemental Formula	M.W.	Ext.[a]	Ident.[b]	Relative Size
	Azaarenes					
51	C2-quinoline	$C_{11}H_{11}N$	157	A	RT+MS	+
52	C3-quinoline	$C_{12}H_{13}N$	171	A	MS'	+
53	azafluorene	$C_{12}H_{9}N$	167	A	RT+MS	+/-
54	acridine	$C_{13}H_{9}N$	179	A	RT+MS	+
55	phenanthridine	$C_{13}H_{9}N$	179	A	RT+MS	+
56	C1-acridine[c]	$C_{14}H_{11}N$	193	A	MS'	+/-
57	azapyrene[c]	$C_{15}H_{9}N$	203	A	MS'	++
58	C1-azapyrene[c]	$C_{16}H_{11}N$	217	A	MS'	+
59	C2-azapyrene[c]	$C_{17}H_{13}N$	231	A	MS'	+/-
60	azachrysene[c]	$C_{17}H_{11}N$	229	A	MS'	++
61	C1-azachrysene[c]	$C_{18}H_{13}N$	243	A	MS'	+
62	azabenzo(a)pyrene[c]	$C_{19}H_{11}N$	253	A	MS'	++
63	C1-azabenzo(a)pyrene[c]	$C_{20}H_{13}N$	267	A	MS'	+/-
64	azabenzo(ghi)perylene[c]	$C_{21}H_{11}N$	277	A	MS'	+
65	azabenzo(b)chrysene[c]	$C_{21}H_{13}N$	279	A	MS'	+
66	azadibenzo(a,e)pyrene[c]	$C_{23}H_{13}N$	303	A	MS'	+/-
	Aliphatic Hydrocarbons					
67	heptadecane	$C_{17}H_{36}$	240	A	RT+MS	++
68	docosane	$C_{22}H_{46}$	310	A	RT+MS	+
69	tricosane	$C_{23}H_{48}$	324	A	RT+MS	+
70	tetracosane	$C_{24}H_{50}$	338	A	RT+MS	+
71	cholestane	$C_{27}H_{48}$	372	A	MS	+
72	trisnorhopane	$C_{27}H_{46}$	370	A	MS	++
73	norhopane	$C_{29}H_{50}$	398	A	MS	++
74	hopane	$C_{30}H_{52}$	412	A	MS	++
75	methylhopane	$C_{31}H_{54}$	426	A	MS	++
76	dimethylhopane	$C_{32}H_{56}$	440	A	MS'	+
	Chlorobenzenes					
77	o-dichlorobenzene	$C_6H_4Cl_2$	146	A	RT+MS	+
78	m-dichlorobenzene	$C_6H_4Cl_2$	146	A	RT+MS	+
79	p-dichlorobenzene	$C_6H_4Cl_2$	146	A	RT+MS	+
80	trichlorobenzene	$C_6H_3Cl_3$	180	A	MS	+
	Aliphatic Acid - as trimethylsilyl derivatives					
81	pentenoic acid -TMS	$C_8H_{16}O_2Si$	172	B	MS	++
82	hexanoic acid -TMS	$C_9H_{20}O_2Si$	188	B	MS	+++
83	heptanoic acid -TMS	$C_{10}H_{22}O_2Si$	202	B	RT+MS	++
84	octanoic acid -TMS	$C_{11}H_{24}O_2Si$	216	B	RT+MS	+++
85	nonanoic acid -TMS	$C_{12}H_{26}O_2Si$	230	B	RT+MS	+++
86	decanoic acid -TMS	$C_{13}H_{28}O_2Si$	244	B	RT+MS	++
87	decenoic acid -TMS	$C_{13}H_{26}O_2Si$	242	B	MS'	+
88	undecanoic acid -TMS	$C_{14}H_{30}O_2Si$	258	B	RT+MS	++
89	undecenoic acid -TMS	$C_{14}H_{28}O_2Si$	256	B	MS'	++
90	dodecanoic acid -TMS	$C_{15}H_{32}O_2Si$	272	A,B	RT+MS	++
91	dodecenoic acid -TMS	$C_{15}H_{30}O_2Si$	270	B	MS	++
92	tridecanoic acid -TMS	$C_{16}H_{34}O_2Si$	286	B	RT+MS	+
93	tetradecanoic acid -TMS	$C_{17}H_{36}O_2Si$	300	A,B	MS'	++
94	tetradecenoic acid -TMS	$C_{17}H_{34}O_2Si$	298	B	MS'	+++
95	pentadecanoic acid -TMS	$C_{18}H_{38}O_2Si$	314	A,B	RT+MS	+++
96	hexadecanoic acid -TMS	$C_{19}H_{40}O_2Si$	328	A,B	RT+MS	+++

Table 1 (Continued)

No.	Compounds	Elemental Formula	M.W.	Ext.[a]	Ident.[b]	Relative Size
97	hexadecenoic acid -TMS	$C_{19}H_{38}O_2Si$	326	B	MS	++
98	heptadecanoic acid -TMS	$C_{20}H_{42}O_2Si$	342	A	RT+MS	++
99	heptadecenoic acid -TMS	$C_{20}H_{40}O_2Si$	340	A	MS'	+
100	octadecanoic acid -TMS	$C_{21}H_{44}O_2Si$	356	A,B	RT+MS	+++
101	octadecenoic acid -TMS	$C_{21}H_{42}O_2Si$	354	A	MS	++
102	eicosanoic acid -TMS	$C_{23}H_{48}O_2Si$	384	A	RT+MS	+
103	heneicosanoic acid -TMS	$C_{24}H_{50}O_2Si$	398	A	RT+MS	+/-
104	docosanoic acid -TMS	$C_{25}H_{52}O_2Si$	412	A	RT+MS	+
105	tricosanoic acid -TMS	$C_{26}H_{54}O_2Si$	426	A	RT+MS	+
106	tetracosanoic acid -TMS	$C_{27}H_{56}O_2Si$	440	A	MS	++
107	pentacosanoic acid -TMS	$C_{28}H_{58}O_2Si$	454	A	MS'	+/-
108	hexacosanoic acid -TMS	$C_{29}H_{60}O_2Si$	468	A	MS'	+
109	oxalic acid -2TMS	$C_8H_{18}O_4Si_2$	234	B	MS	++
110	nonanedioic acid -2TMS	$C_{15}H_{32}O_4Si_2$	332	B	RT+MS	++
111	pentadecanedioic acid -2TMS	$C_{21}H_{44}O_4Si_2$	416	B	RT+MS	+
112	hexadecanedioic acid -2TMS	$C_{22}H_{46}O_4Si_2$	430	B	RT+MS	+
113	octadecenedioic acid -2TMS	$C_{24}H_{48}O_4Si_2$	456	B	MS'	+/-
114	tetradecanetrioic acid -3TMS	$C_{23}H_{48}O_6Si_3$	504	B	MS'	+/-
	Hydroxylated Polycycic Aromatic Hydrocarbons - as trimethylsilyl derivatives					
115	hydroxyanthracene -TMS[c]	$C_{17}H_{18}OSi$	266	A	MS'	+
116	C1-hydroxyanthracene -TMS[c]	$C_{18}H_{20}OSi$	280	A	MS'	+
117	hydroxypyrene -TMS[c]	$C_{19}H_{18}OSi$	290	A	MS'	+
118	C1-hydroxypyrene -TMS[c]	$C_{20}H_{20}OSi$	304	A	MS'	+
119	hydroxychrysene -TMS[c]	$C_{21}H_{20}OSi$	316	A	MS'	+
120	hydroxybenzo(a)pyrene-TMS[c]	$C_{23}H_{20}OSi$	340	A	MS'	+/-
	Phenols - as trimethylsilyl derivatives					
121	iso-propylphenol-TMS	$C_{12}H_{20}OSi$	208	A	MS	+
122	hydroxybenzaldehyde-TMS	$C_{10}H_{14}O_2Si$	194	B	MS	++
123	hydroxymethoxybenzaldehyde-TMS	$C_{11}H_{16}O_3Si$	224	B	MS	++
124	bromophenol-TMS	$C_9H_{13}BrOSi$	244	A,B	MS	++
	Aromatic Acids - as trimethylsilyl derivatives					
125	benzoic acid -TMS	$C_{10}H_{14}O_2Si$	194	B	MS	+++
126	phenylacetic acid -TMS	$C_{11}H_{16}O_2Si$	208	B	MS	+
127	dichlorobenzoic acid -TMS	$C_{10}H_{13}Cl_2O_2Si$	262	B	RT+MS	+/-
128	dibromobenzoic acid -TMS	$C_{10}H_{12}Br_2O_2Si$	350	B	MS'	+
129	dibromohydroxybenzoic acid -2TMS	$C_{13}H_{20}Br_2O_3Si_2$	438	B	MS'	+/-
	Miscellaneous					
130	dibenzofuran	$C_{12}H_8O$	168	A	MS	++
131	benzonaphthofuran	$C_{16}H_{10}O$	218	A	MS	+
132	benzonaphthothiofen	$C_{16}H_{10}S$	234	A	MS	++
133	anthraquinone	$C_{14}H_8O_2$	208	A	MS	+
134	dibutyl phthalate	$C_{16}H_{22}O_4$	278	A	MS	+
135	dioctyl phthalate	$C_{24}H_{38}O_4$	390	A	MS	++
136	methoxybenzenediamine	$C_7H_{10}N_2O$	138	C	MS	+
137	tributylmethyltin	$C_{13}H_{30}Sn$	306	A	MS	+/-

[a] Extraction method by which the compound was identified. [b] Compound identification methods were as follows: RT+MS, comparing the retention time and mass spectrum with authentic reference; MS, comparing the mass spectrum with published data; MS', comparing the mass spectrum with similar compounds. [c] Or other isomers.

used in soap and synthetic detergent manufacture.

The most predominant compounds in thid study were polycyclic aromatic hydrocarbons(PAHs). Around Dokai Bay, there are many factories which use petroleum and coal as raw materials and for fuel. Hanada et.al.(1990) reported much of PAHs in Dokai Bay originated from industrial effluents. Hydroxylated PAHs identified suggest oxidative transformation occurring in the environment.

Benzoic acids, phenylacetic acid, hydroxybenzaldehyde and hydroxy methoxybenzaldehyde could be of natural origin. However, they are widely used in various industries and also may be formed by oxidative degradation of other chemical substances. Further study is necessary to know the exact origins of these substances.

Concerning Basic compounds, some azaarenes and methoxybenzenediamine were identified. Many kind of basic compounds in the sediment could probably not be extracted with 1N-HCl or acetone-hexane, since basic compounds may be strongly adsorbed to sediment. It is probable that azaarenes have the same origin as PAHs. Methoxybenzenediamine itself is not in industrial use. It may have occurred as a result of reductive transformation and/or biological methylation in the environment.

Figure 1 shows mass spectrum of tributylmethyltin(Bu_3MeSn). Although the sample mass spectrum overlapped with dimethylnaphthalene(mw=156), the isotopic pattern of tin was obvious. The possible origin is impurity of tributyltin compounds or transformation products of them in the environment.Maguire(1984; 1986) detected Bu_3MeSn in coastal sediment with concentration of zero to 8% of that of Bu_3Sn^+. They also found natural occurring of methylation of Bu_3Sn^+ by experiment(1985). Considering the concentration of Bu_3Sn^+ in Dokai Bay(0.02-0.05mg/kg)(Japan Environment Agency 1989), Bu_3MeSn should have been at very small amount and could hardly have given the mass spectrum if it had been present as less than 10% of Bu_3Sn^+ in concentration. Therefore, the concentration of Bu_3MeSn in Dokai Bay is thought to be unusually high. It may suggest that the transformation from Bu_3Sn^+ to Bu_3MeSn could be a significant pathway in a certain condition. However, Bu_2Me_2Sn, $BuMe_3Sn$ and Me_4Sn were not detected in this study.

Dichlorobenzene and trichlorobenzene were found reflecting industrial activities. Bromophenol, dibromobenzoic acid and dibromohydroxybenzoic acid were identified as TMS derivatives(See Figure 2). Since dibromobenzoic acid and dibromohydroxybenzoic acid were not commercially available, they were identified by comparing with the mass spectral pattern of corresponding

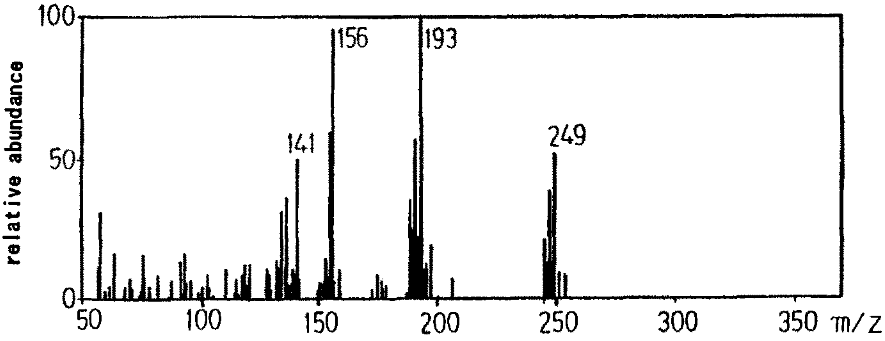

Figure 1. Mass spectrum of tributylmethyltin found in the sample

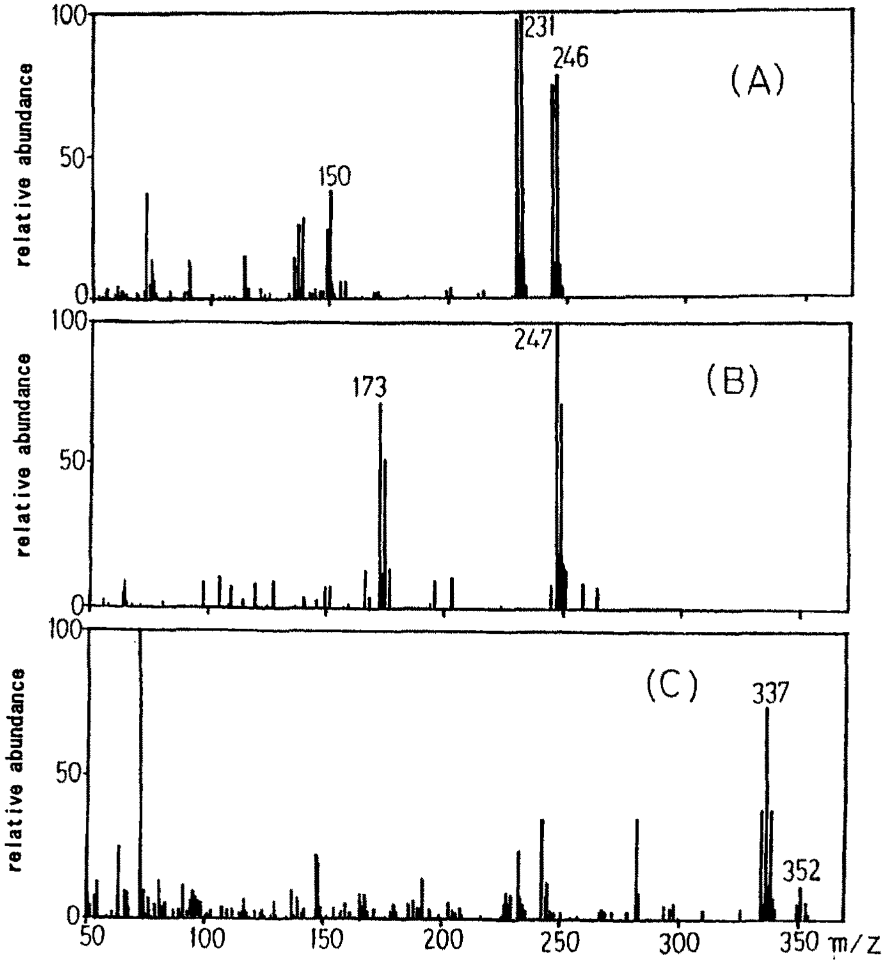

Figure 2. Mass spectra of (A)bromophenol, (B)dichlorobenzoic acid and (C)dibromobenzoic acid found in the sample (as TMS derivatives)

chlorinated compounds. The brominated compounds found here are not in industrial use. Chlorophenol and dichlorobenzoic acid are useful raw material in chemical industry. But chlorophenol could occur by hydroxylation of dichlorobenzenes which are abundant in this area. Furtheremore, benzoic acid and relative compounds could be formed by oxdative degradation of other chemical substances such as PAHs and pesticides(Tahori 1971; Haque 1974) in the environment.

REFEREFCES

Coleman WE, Melton RG, Kopfler FC, Barone KA, Aurand TA, Jellison MG (1980) Identification of organic compounds in a mutagenic extract of a surface drinking water by a computerized gas chromatography/mass spectrometry. Environ Sci Technol 14:576-588

Eganhouse RP, Kaplan IR (1982) Extractable organic matter in municipal wastewaters, 2 hydrocarbons : molecular characterization. Environ Sci Technol 16: 541-551

Elder VA, Proctor BL, Hites RA (1981) Organic compounds found near dump sites in Niagara Falls, New York. Environ Sci Technol 15: 1237-1243

Hanada Y, Mizuguchi M, Sueta S, Takeuchi R, Kido K (1990) The state of pollution causing fossil fuel in enclosed coastal sea. J Environ Lab Assoc 15: 149-155

Haque R, Fread VH (1974) Environmental dynamics of pesticides. Plenum Press, New York

Japan Environment Agency (1989) Chemicals in the environment. Tokyo

Knapp DR (1979) Handbook of analytical derivatization reaction. John Wiley & Sons, New York

Maguire RJ (1984) Butyltin compounds and inorganic tin in Ontario. Environ Sci Technol 18: 291-294

Maguire RJ, Tkacz RJ (1985) Degradation of the tri-n-butyltin species in water and sediment from Toronto Harbor. J Agr F Chem 33: 947-953

Maguire RJ, Tkacz RJ, Chau YK, Bengert GA, Wong PTS (1986) Occurrence of organotin compounds in water and sediment in Canada. Chemosphere 15: 253-274

Matsumoto G, Hanya T (1981) Comparative study on organic constituents in polluted and unpolluted waters. Water Res 15: 217-224

Mclafferty FW, Stauffer DB (1989) The Wiley/NBS registry of mass spectral data. Wiley-Interscience, New York

Sheldon LS, Hites RA (1978) Organic compounds in the Delaware river. Environ Sci Technol 12: 1188-1194

Tahori AS (1971) Pesticide terminal residues. Butterworths, London

Received May 28, 1992; accepted October 10, 1992.