Volatile Halogenated Hydrocarbons in Foods

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Volatile halogenated organic compounds were determined in foods. Statistical treatment of the data for 13 foods sampled from 20 families living in suburban Tokyo (Saitama prefecture) indicated that the foods were contaminated by water pollution and/or substances introduced by the process of food production. Butter and margarine were contaminated by chlorinated ethylene, ethane, and related compounds released by dry cleaning and other operations. Soybean sprouts and tofu (soybean curd) contained chloroform and related trihalomethanes absorbed during the production process.

Keywords: *Trihalomethanes; halogenated organic solvents; purge–trap; GC-MS; daily intake; principal component analysis; cluster analysis*

INTRODUCTION

As part of our studies of contaminants in food (Miyahara et al., 1991, 1992a,b; Miyahara and Saito, 1993) we have expanded our work to include volatile halogenated hydrocarbons (VHC). These substances are toxic and suspected carcinogens (Symons et al., 1981; Eriksson et al., 1993); therefore, it is important to check the levels in foods and to identify the sources to reduce the daily intake. Limited studies on halogenated compounds in food have been monitored and the sources were tentatively identified (Clower, 1980). The most likely sources are the water used for production, wrapping materials, polluted air, retained solvents used for extraction of natural components (pigments, caffeine, oil, etc.) from plants, and so on. The halogenated compounds are also used as dry cleaning solvents, pesticides for fumigation, and washing solvents for metals and semiconductors. Trihalomethanes are formed by bleaching crops or organic materials and by chlorinating water in a disinfection process (Rook, 1977). Thus, there are many possible sources of food contaminants. Food may be contaminated by VHC's from many sources. To reduce contamination of foods with volatile halogenated carbons, more precise sources are desirable.

Many analytical methods are available for a field study on contamination with halogenated carbons. There are established official methods for the determination of residual fumigants in grains (AOAC, 1990), vegetables (MacLoad, 1986), food additives in spices (FDA, 1983), and trihalomethane in water (APHA, 1992; EPA-Japan, 1992). Few studies of halogenated organic compounds in variety of foods are known (Heikes and Hopper, 1986). A combination of purge–trap sampling and gas chromatography with a mass spectrometric detector or head-space sampling with gas chromatography equipped with electron capture detector was used to study VHC's in food.

The data obtained in our study were treated statistically. Pattern recognition analysis and chemometric analysis had previously been applied to the data for coffees, flavors, honeys, and beans to determine significant effects of the raised places, or to evaluate the quality (Crecente and Latorre, 1993; Bicchi, 1993; Aijima, 1992). Those studies have established the usefulness of the statistical analysis to categorize data and to find unobservable and subtle factors in the data set. Preliminary cluster analysis and principal component analysis (PCA) are very commonly studied in the field. However, few reports have appeared using the original data and statistical methods to determine main sources of pollutants in foods.

This paper describes the results of monitoring volatile halogenated hydrocarbons (Figure 1) in foods and a trial of estimation of the pollutants sources using statistical analysis of the data. The analysis of the data gave significant results for the estimation of the sources for the pollutants.

EXPERIMENTAL PROCEDURES

Apparatus. (*a*) *GC-MS.* A Hewlett-Packard Model 5890A gas chromatograph with VG Model Masslab TRIO-1 mass spectrometric detector and a NEUTRABOND-1 capillary column (30 m × 0.25 mm i.d., GL Science, Tokyo) with a film thickness of 0.15 μm were used with helium carrier gas at 1 mL/min and a helium septum purge at 1 mL/min, and a splitter was used. Injector temperature was 240 °C. The column oven temperature was maintained at 40 °C for 1 min and increased to 135 °C at 5 °C/min. Transfer tube temperature was 200 °C. Injection with a purge–trap injector was utilized. The monitor ions for the determinations were as follows: *m/z* 96 for 1,1-dichloroethylene and *cis*-1,2-dichloroethylene) with *m/z* 100 for internal standard (1,1-dichloroethylene-d_2); *m/z* 62, 64 for 1,2-dichloroethane and 1,2-dichloropropane with *m/z* 65, 67 internal standard (1,2-dichloropropane-d_6); *m/z* 75, 77 for 1,3-dichloropropene and *m/z* 83, 85, 97 for 1,1,2-trichloroethane with *m/z* 99, 101, 103 internal standard (1,1,2-trichloroethane-$^{13}C_2$).

(*b*) *ECD-1.* A Shimadzu Model GC14A gas chromatograph with electron-capture detector (^{63}Ni) and a splitter were used. A DB-624 capillary column (30 m × 0.53 mm i.d., J&W Scientific, Folsom, CA) with a film thickness of 3 μm was used with helium carrier gas at 5 mL/min. Injector and detector temperatures were 300 °C. The column oven temperature was programmed as follows: initially maintained at 40 °C for 5 min and then increased to 50 °C at 2 °C/min and then increased at 3 °C/min to 80 °C. A 3 μL injection in the splitless mode followed by a waiting time of 2 min was utilized. Make-up gas consisted of nitrogen at 60 mL/min.

(*c*) *ECD-2.* A Hewlett-Packard Model 5890A gas chromatograph with electron-capture detector (^{63}Ni) and a splitter were used. A DB-624 capillary column (30 m × 0.53 mm i.d.) with

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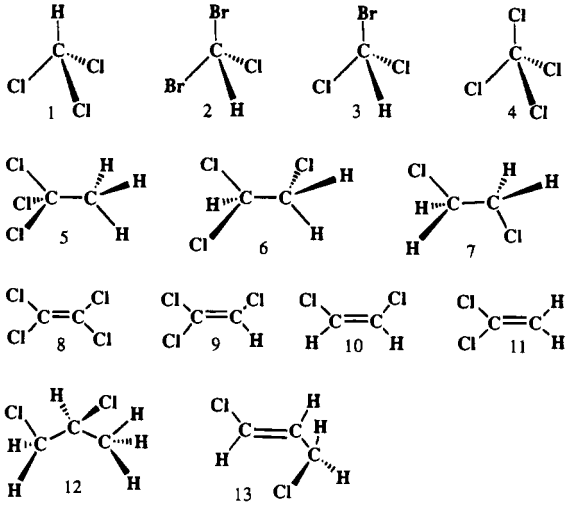

Figure 1. Structures of volatile halogenated carbons: **1**, chloroform; **2**, dibromochloromethane; **3**, bromodichloromethane; **4**, carbon tetrachloride; **5**, 1,1,1-trichloroethane; **6**, 1,1,2-trichloroethane; **7**, 1,2-dichloroethane; **8**, tetrachloroethylene; **9**, trichloroethylene; **10**, *cis*-1,2-dichloroethylene; **11**, 1,1-dichloroethylene; **12**, 1,2-dichloropropane; **13**, 1,3-dichloropropene.

a film thickness of 3 μm was used with helium carrier gas at 5 mL/min. Injector and detector temperatures were 200 and 300 °C, respectively. The column oven temperature was programmed as follows: maintained at 45 °C for 5 min and increased to 135 °C at 6 °C/min. A 3 μL injection in the splitless mode followed by a waiting time of 2 min was utilized. Make-up gas consisted of nitrogen at 60 mL/min.

(*d*) *Purge–Trap System.* A Chrompack purge and trap system was used. The desorber was operated at 250 °C for 5 min. The sample was trapped in a silica wide bore column (0.53 mm i.d.) cooled at −130 °C. A condensed sample was purged at 240 °C for 5 min. The purge gas consisted of helium at 20 mL/min.

(*e*) *Computers.* An Apple Computer Model Macintosh Quadra 700 and NEC (Nippon Electrical Co.) Model PC9801LV were used for data elaboration and statistical analysis.

Reagents. (*a*) *Standards.* The standards used were as follows: chloroform (**1**), >99%; dibromochloromethane (**2**), >98%; bromdichloromethane (**3**), >98%; carbon tetrachloride (**4**), >99%; 1,1,1-trichloroethane (**5**), >99%; 1,1,2-trichoroethane (**6**), >98%; 1,2-dichloroethane (**7**), >99%; tetrachloroethylene (**8**), >99%; trichloroethylene (**9**), >98%; *cis*-1,2-dichloroethylene (**10**), >97%; 1,1-dichloroethylene (**11**), >99%; 1,2-dichloropropane (**12**), >98%; 1,3-dichloropropene (**13**), >95%. These standards were purchased from GL Science Co, Tokyo.

Internal standards included 1,1-dichloroethylene-d_2, 1,2-dichloropropane-d_6, and 1,1,2-trichloroethane-$^{13}C_2$ that were purchased from Supelco Co., Bellefonte, PA.

Warning! These standards are suspected carcinogens and should be handled with care.

(*b*) *Grade.* All organic solvents for analysis were of pesticide residue grade.

(*c*) *Sample Bag.* A sample bag which was made with polyvinylfluoride film (Maeda Seisaku-syo Co., Tokyo) was used for solid sample storage (tofu, soybeans spores, butter, margarine, rice, and cake). The bag had a 2 L capacity. For liquid samples, 200 mL vials with screw caps and Teflon seals were used.

Samples. Volunteers purchased food in the usual manner at retail stores in Saitama prefecture. Samples were unpacked and stored as usual at the volunteers' homes for 1 week in order to simulate typical home storage conditions. The samples were then packed in glass vials or plastic bags to prevent contamination during transportation to the laboratory. The volunteers were selected at random and sampling was abstracted at random so that the samples reflected normal histories of food transportation and storage.

Analytical Procedure for Chloroform (1), Dibromochloromethane (2), Bromodichloromethane (3), Carbon Tetrachloride (4), 1,1,1-Trichloroethane (5), Tetrachloroethylene (8), and Trichloroethylene (9). The analytical procedures were already reported and briefly described (EPA-Japan, 1992).

(*a*) *Juice and Cola.* Halogenated organic carbons were extracted from the sample (30 g) with *n*-hexane (3 mL). An aliquot of the organic layer was injected into a gas chromatograph equipped with ECD.

(*b*) *Samples Other than Juice and Cola.* The sample (50–100 g) was mixed with 20 mL of water and 10 mL of *n*-hexane. Halogenated organic carbons were distilled from the sample with a Dean-Stark apparatus for 60 min (3 mL). An aliquot of the organic layer was injected into the gas chromatograph with ECD.

Analytical Procedure for 1,1,2-Trichloroethane (6), 1,2-Dichloroethane (7), 1,2-Dichloroethylene (10), 1,1-Dichloroethylene (11), 1,2-Dichloropropane (12), and 1,3-Dichloropropene (13). A sample (5–10 g) was placed in a 125 mL vial for head-space analysis. To the sample were added 30 g of sodium chloride, 80 mL of water, and internal standard. The vial was sealed with an aluminum cap and shaken for 30 min at 50 °C. The head-space gas (5 mL) was injected into the thermal desorption cold trap injector which was connected to the gas chromatograph–mass spectrometer.

Statistical Analysis. (*a*) *Statistical Values.* Basic statistical values were calculated through "Excel Analysis ToolPack" program (Microsoft Co., Delaware, 1992, Ver. 4).

(*b*) *Cluster Analysis.* Each food sample or chemical was considered as an assembly of variables represented by the chemical data or food data. Similarities between each food sample were calculated as Euclidean distances and clusters were created by the nearest-neighbor method in this work (Massarti et al., 1988a). This classification procedure provides a primary evaluation of the food or chemical group similarities. The cluster analyses were processed through a "Multivariate Statistical Analysis" program package (Kogakusya, Tokyo, 1993, Ver. 2.0).

(*c*) *Principal Component Analysis (PCA).* Each food sample was also considered as an assembly of variables (VHC levels) (Massarti et al., 1988b). This classification method was used to categorize foods into the groups trihalomethane polluted cleaning solvent polluted, and a combination of the two. The PCA was also processed through the "Multivariate Statistical Analysis" program package.

RESULTS AND DISCUSSION

Sampling. The samples were collected during the period November 20–27, 1992, in cooperation with the Saitama Institute of Public Health. The sampling locations were scattered throughout the Kanto plain (Figure 2) and did not include any industrial park. The volunteers purchased the food at neighborhood supermarkets or grocery stores. In the home, the samples were unpacked and stored in the individual volunteer's refrigerator (5–10 °C) or kitchen (room temperature) for 1 week. No effort was made to inhibit absorption of gases from surrounding atmosphere or trihalomethane generation by reactions in food. Thus, the samples were stored initially in a manner that simulated the usual consumer proclivities. After exposure, the solid samples were packed in gas impermeable plastic bags. The bags were sealed with a heat sealer to guarantee that no gas could pollute the samples in the bags. These were stored in a freezer at −20 °C until analysis. The liquid samples were bottled in 200 mL vials and were stored in a refrigerator at 4 °C until analysis. These sampling procedures are very simple and reliable.

Analytical Conditions. The extraction and purification procedures are modifications of the Japanese Environmental Protection Agency's methods (EPA-Japan, 1992.). Some official analytical methods for

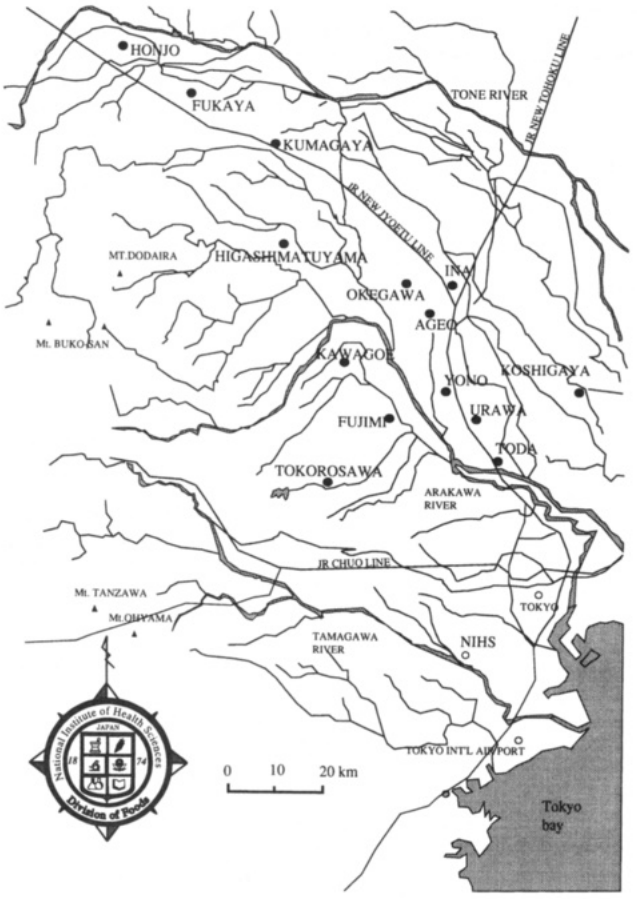

Figure 2. Sampling points. Twenty sampling points located in 14 cities of the Kanto plain, Japan.

VHC's in food include acetone extraction from grains (AOAC, 1990), a combination of acetonitrile extraction and Florisil column clean up from vegetables (McLeod, 1986), and a closed-system vacuum distillation with toluene for spices at 30 ppm (FDA, 1983). These are methods for specific samples. Few methods for water are usable (APHA, 1992; EPA-Japan, 1992) for VHCs in food. The performances of the methods have been well established for environmental samples (APHA, 1992). To apply the methods to our samples, some samples were tested. The methods performed as well for food as for the environmental samples. The results were satisfactory for the food samples. Recoveries at 0.002–0.005 ppm ranged from 91 to 95% (CV%, 1.84–3.81).

Monitoring Results. The 13 compounds in 13 kinds of foods that were monitored and the levels are shown in Table 1. Chloroform was found in all of the samples examined. The levels in moyashi were extremely higher than those in the other samples. This may result from the production process, which will be discussed later. The products (tofu, milk, lactic beverage, plain yogurt, juice, ice milk, and cola) which include water as a main component were contaminated with chloroform, bromodichloromethane, and dibromochloromethane. However, it was not clear if water was the source of the contaminants. The levels of the compounds were quite low and may not be hazardous to health. To judge the hazard, the total daily intakes were estimated from the results and shown in Table 2. These results illustrate that all of the compound levels were below the MAC (maximum acceptable concentrations) levels as specified by the Japanese government. The levels observed for chloroform were somewhat lower than those reported (daily intakes for chloroform ranged 3.38–178.38 μg and the mean was 45.4 μg) (Tamagawa et al., 1988; Toyoda et al., 1986, 1987, 1990).

Entz and Hollifield (1982) reported tetrachloroethylene was present in many foods at a level of 0.4–1050 ppb. However, our study shows a less frequent incidence than that reported by them. This may be because the samples examined were entirely different.

Classification of the Contaminants and Foods by Cluster Analysis. To identify the probable sources of the contaminants, the data were analyzed by a cluster analysis method. This statistical analysis provided the preliminary classification of the compounds according to the similarities of data sets (nonsupervised pattern recognition approach). In this case, the levels for 13 compounds in foods were analyzed as one data set and the distances of the data sets were calculated. The 13 compounds were classified into three groups as shown in Figure 3. The three groups are named chloroform, trihalomethanes (dibromochloromethane, bromedichlorothane), and cleaning solvents (the other compounds). The classifications are named after the probable sources of pollutants.

The data sets for foods were also analyzed by cluster analysis. The results are shown in Figure 4. The foods were classified into five clusters—moyashi, cola, high fat foods (margarine, butter), lactic semisolid foods (tofu, yogurt, lactic beverage, ice cream, and ice milk), and foods contaminated at low levels (rice, juice, cake, and milk). The results cannot be uniquely described to product processes or materials, so it is difficult to identify the sources of the contaminants.

Relationship between Contaminant Levels and Storage Periods. Foods may be contaminated by

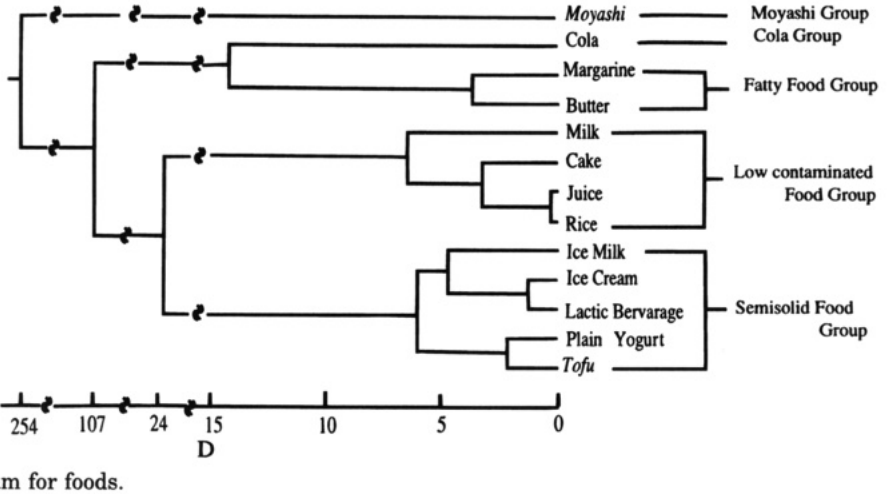

Figure 3. Dendrogram for foods.

Table 1. Monitoring Results (ppb)

	moyashi (bean sprout)			cola			butter		
	incidence	range	mean	incidence	range	mean	incidence	range	mean
chloroform	20/20	0.8–320	153.7	5/5	0.9–41	29	19/19	7.3–37	27.4
dibromochloromethane	19/20	ND-3.6	4.32	4/5	ND-5.8	3.1	19/19	ND-3.6	1.45
bromodichloromethane	1/20	ND-0.5	0.025	4/5	ND-19	12	17/19	ND-3.0	1.59
carbon teterachloride	0/20	ND		0/5	ND		6/19	ND-6.6	0.81
1,1,1-trichloroethane	7/20	ND-1.3	0.27	0/5	ND		19/19	1.0–4	1.82
1,1,2-trichloroethane	0/20	ND		0/5	ND		0/19	ND	
1,2-dichloroethane	0/20	ND		0/5	ND		16/19	ND-5.4	1.30
tetrachloroethylene	1/20	ND-0.5	0.07	0/5	ND		17/19	ND-5.7	1.23
trichloroethylene	0/20	ND		0/5	ND		0/19	ND	
cis-1,2-dichloroethylene	1/20	ND-0.8	0.04	0/5	ND		0/20	ND	
1,1-dichloroethylene	0/20	ND		0/5	ND		0/20	ND	
1,2-dichloropropane	0/20	ND		0/5	ND		0/19	ND	
1,3-dichloropropene	0/20	ND		0/5	ND		0/19	ND	
storage time (days)	N/A			5/5	34–156	76.8	19/19	31–174	66.6

	margarine			milk			cake		
	incidence	range	mean	incidence	range	mean	incidence	range	mean
chloroform	20/20	1.7–32	27.4	20/20	1.1–11	2.54	3/3	1.3–1.5	1.4
dibromochloromethane	20/20	ND-3.6	1.54	1/20	ND-4.6	4.32	0/3	ND	
bromodichloromethane	13/20	ND-6.5	1.81	1/20	ND-8.2	0.03	0/3	ND	
carbon tetrachloride	9/20	ND-24	2.12	0/20	ND		0/3	ND	
1,1,1-trichloroethane	19/20	ND-74	5.29	0/20	ND		3/3	1.2–2.1	1.6
1,1,2-trichloroethane	0/20	ND		0/20	ND		0/3	ND	
1,2-dichloroethane	13/20	ND-24	1.96	1/20	ND-0.6	0.03	1/3	ND-0.6	0.2
tetrachloroethylene	13/20	ND-4.1	0.79	0/20	ND		3/3	ND-1.8	1.2
trichloroethylene	0/20	ND		0/20	ND		2/3	ND-1.7	0.8
cis-1,2-dichloroethylene	0/20	ND		0/20	ND		0/3	ND	
1,1-dichloroethylene	0/20	ND		0/20	ND		0/3	ND	
1,2-dichloropropane	0/20	ND		0/20	ND		0/3	ND	
1,3-dichloropropene	0/20	ND		0/20	ND		0/3	ND	
storage time (days)	20/20	23–270	55.2	20/20	23–270	10.35	N/A		

	juice			rice			lactic beverage		
	incidence	range	mean	incidence	range	mean	incidence	range	mean
chloroform	7/20	ND-8.2	1.25	1/20	ND-2	0.10	16/16	ND-29	4.16
dibromochloromethane	3/20	ND-1.4	0.18	0/20	ND		5/16	ND-2.6	0.46
bromodichloromethane	3/20	ND-2.9	0.28	0/20	ND		5/16	ND-5.0	0.86
carbon tetrachloride	0/20	ND		0/20	ND		0/16	ND	
1,1,1-trichloroethane	0/20	ND		3/20	ND-0.8	0.11	1/16	ND-1.2	0.08
1,1,2-trichloroethane	0/20	ND		0/20	ND		0/16	ND	
1,2-dichloroethane	0/20	ND		0/20	ND		0/16	ND	
tetrachloroethylene	0/20	ND		1/20	ND-0.5	0.03	0/16	ND	
trichloroethylene	1/20	ND-0.6	0.03	0/20	ND		1/16	ND-0.5	0.03
cis-1,2-dichloroethylene	0/20	ND		1/20	ND-1.5	0.08	1/16	ND-2.8	0.18
1,1-dichloroethylene	0/20	ND		0/20	ND		0/16	ND	
1,2-dichloropropane	0/20	ND		0/20	ND		0/16	ND	
1,3-dichloropropene	0/20	ND		0/20	ND		0/16	ND	
storage time (days)	20/20	5–288	51.7		N/A		16/16	5–16	8.4

	ice cream			plain yogurt			tofu (soybean curd)		
	incidence	range	mean	incidence	range	mean	incidence	range	mean
chloroform	15/15	1.2–27	4.96	20/20	1.0–5.1	2.17	20/20	1.1–36	11.4
dibromochloromethane	3/15	ND-1.4	0.26	5/20	ND-1.1	0.56	20/20	2.3–7.1	4.83
bromodichloromethane	1/15	ND-4.2	0.58	14/20	ND-2.3	0.37	7/20	ND-7.1	1.57
carbon tetrachloride	1/15	ND-1.5	0.100	0/20	ND		0/20	ND	
1,1,1-trichloroethane	1/15	ND-3.2	0.88	1/20	ND-1.2	0.06	2/20	ND-1.6	0.145
1,1,2-trichloroethane	0/15	ND		0/20	ND		1/20	ND-1.4	0.07
1,2-dichloroethane	1/15	ND-1.5	0.03	1/20	ND		0/20	ND	
tetrachloroethylene	5/15	ND-0.9	0.22	0/20	ND		0/20	ND	
trichloroethylene	2/15	ND-1.3	0.16	1/20	ND-0.6	0.03	0/20	ND	
cis-1,2-dichloroethylene	0/15	ND		0/20	ND		0/20	ND	
1,1-dichloroethylene	0/15	ND		0/20	ND		1/20	ND-2.8	0.14
1,2-dichloropropane	1/15	ND-0.6	0.04	0/20	ND		0/20	ND	
1,3-dichloropropene	0/15	ND		0/20	ND		0/20	ND	
storage time (days)		N/A		20/20	5–12	8.2	20/20	4–8	6.3

	ice milk		
	incidence	range	mean
chloroform	4/4	0.6–26	8.2
dibromochloromethane	3/4	ND-2.5	1.1
bromodichloromethane	2/4	ND-3.6	1.4
carbon tetrachloride	0/4	ND	
1,1,1-trichloroethane	1/4	ND-0.9	0.2
1,1,2-trichloroethane	0/4	ND	
1,2-dichloroethane	0/4	ND	
tetrachloroethylene	1/4	ND-1.8	0.5
trichloroethylene	1/4	ND-1	0.3
cis-1,2-dichloroethylene	0/4	ND	
1,1-dichloroethylene	0/4	ND	
1,2-dichloropropane	0/4	ND	
1,3-dichloropropene	0/4	ND	
storage time (days)		N/A	

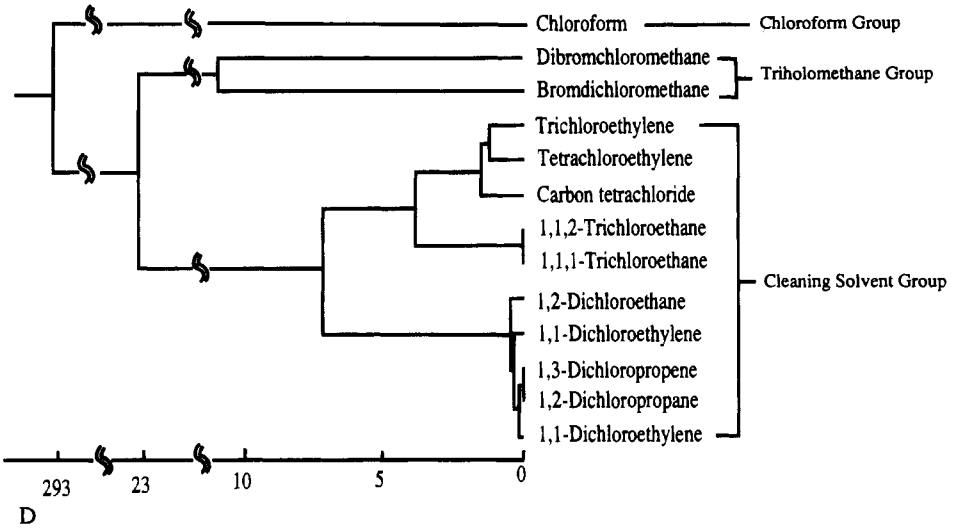

Figure 4. Dendrogram for contaminants.

Table 2. Total Daily Intake (Milligrams per Day per Person)

compound	intake through food[a]	mean intake through water[b]
chloroform (**1**)	1.030	43.7
bromodichloromethane (**2**)	0.188	13.4
dibromochloromethane (**3**)	0.228	6.7
carbon tetrachloride (**4**)	0.004	0.0
1,1,1-trichloroethane (**5**)	0.040	
1,1,2-trichloroethane (**6**)	0.003	
1,2-dichloroethane (**7**)	0.004	
tetrachloroethylene (**8**)	0.005	
trichloroethylene (**9**)	0.004	
1,2-cis-dichloroethylene (**10**)	0.020	
1,1-dichloroethylene (**11**)	0.005	
1,1,2-dichloropropane (**12**)	0.000	
1,3-dichloropropene (**13**)	0.000	

[a] Calculated from the levels and diet consumption of food in Japan. [b] Calculated based on the data reported by Symons et al. (1975).

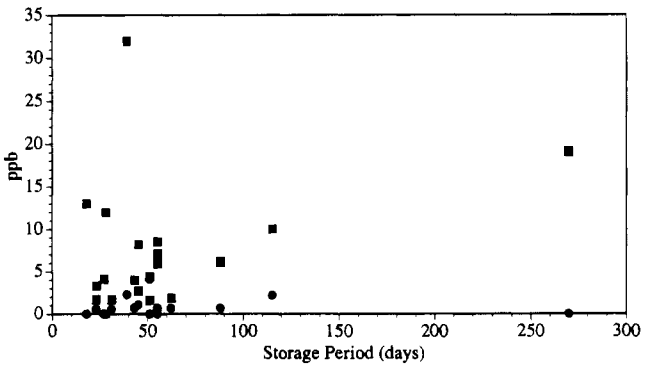

Figure 5. Storage period vs chloroform and trichloroethylene levels in margarine. (■) Chloroform; (●) trichloroethylene.

contact with contaminated materials or by production processes and/or storage environments. The contaminant levels in foods that declare production dates were correlated with length of storage (days). The results are shown in Table 3. This illustrates that contaminant levels in tofu, butter, plain yogurt, cola, lactic beverage, and milk are indifferent to the storage periods. In contrast, contaminant levels in margarine, cake, and juice are related to storage periods. The relationship between contamination and storage periods for margarine is shown in Figure 5. The results exhibit tendencies toward contaminant accumulations in these foods. The trend is not definitive because of the limited number of samples. The contamination levels depend on the individual storage circumstances. As a result, it is very hard to establish that the relationship between the storage period and the contaminants levels are due to the storage atmosphere.

PCA Results. As discussed earlier in this report, it is not clear whether the major sources of contamination are polluted storage atmospheres or circumstances of transportation. The sources of contaminants are tentatively assigned in the raw materials of the foods. This statistical method identifies the most significant component or contaminant in food. The results are shown in Tables 4 and 5.

As shown in Table 4, the main load factor of first principle is assigned to chloroform and the contribution of the first principle is over 80%. The first principle axis is considered as a trihalomethane contamination level of those foods (ice cream, ice milk, tofu, lactic beverage, moyashi, and cola) and main contaminants in the foods are trihalomethanes.

Shown in the Table 5 are the load factors of those foods (butter, margarine, and yogurt) which are calculated with the total contributions of principles (>90%). The first principles are different from each other. First and second components of yogurt are considered to be chloroform level and brominated methanes, respectively. First and second components of butter are those of trihalomethanes and cleaning solvents. Those of margarine are 1,1,1-trichloroethane and chloroform. Thus main components vary with the foods and are interpreted as main sources of pollution.

Estimation of the Sources of Contaminants in Foods. Cola is produced from water and cola extracts. The water is disinfected by chlorination with sodium hypochlorite and is dechlorinated with active carbon. The trihalogenated carbon sources may be the water which was chlorinated. The trihalogenated carbon levels depend on the factories which produced the samples. The levels of samples produced by O bottling factory ranged from 30 to 34 ppb for chloroform, from 5.8 to 1.4 ppb for dibromochloromethane, and from 7.5 to 19 ppb for bromodichloromethane. On the other hand, the levels of samples by N factory were 0.9, 0, and 0 ppb for chloroform, dibromochloromethane, and bromodichloromethane, respectively. This estimation was confirmed by the relationships among the levels of trihalomethane (correlation coefficient for the levels of chloroform vs dibromochloromethane, 0.87; chloroform vs bromodichloromethane, 0.93; dibromochloromethane

Table 3. Correlation Coefficients (r) between Compound Levels and Storing Time

compound	tofu	butter	margarine	plain	cola	cake	lactic beverage	juice	milk
chloroform	0.13	−0.02	0.70	0.01	−0.83	−0.90	−0.13	0.71	−0.06
dibromochloromethane	0.09	−0.13	−0.07	−0.15	−0.60	ND	−0.09	−0.10	N/A
bromodichloromethane	−0.20	0.11	−0.03	−0.19	−0.60	ND	−0.05	−0.16	N/A
trichloroethylene	ND	−0.23	0.62	ND	ND	0.91	ND	ND	ND
1,1,1-trichloroethane	ND	−0.04	0.02	ND	ND	0.97	ND	ND	ND
tetrachloroethylene	ND	−0.08	0.31	ND	ND	−0.37	ND	ND	ND

Table 4. Results of PCA (I) (Load Factors of First Component and Total Contribution)

food	ice cream	ice milk	tofu	lactic beverage	cola
chloroform	6.48	11.93	11.96	6.91	16.10
bromodichloromethane	1.12	1.54	0.37	1.34	7.73
dibromochloromethane	0.21	−0.22	0.31	0.32	2.25
1,1,1-trichloroethane	−0.30	−0.12	−0.12		
tetrachloroethylene	−0.11	0.25			
trichloroethylene	−0.09	−0.14			
cis-1,2-dichloroethylene				0.09	
total contribution (%)	95.5	98.1	91.6	95.8	97.49

Table 5. Results of PCA (II) (Load Factors of the Components and Total Contribution)

	moyashi			butter		margarine			yogurt	
food component	1	2	3	1	2	1	2	3	1	2
chloroform	0.866	0.029	0.252	7.81	0.00	−1.05	7.12	0.22	1.22	−0.266
bromodichloromethane	−0.514	0.436	0.252	0.35	−0.28	−0.46	1.02	0.27	0.23	0.255
dibromochloromethane	−0.139	0.875	−0.434	−0.50	0.25	−0.16	0.36	−0.07	0.57	0.466
1,1,1-trichloroethane	0.805	0.398	0.126	−0.50	0.09	16.24	0.51	0.23		
tetrachloroethylene				−0.94	−0.18	−0.02	1.19	−3.15		
trichloroethylene				0.19	0.30	−0.13	0.11	−0.50		
carbon tetrachloride				0.02	1.91	−0.76	0.39	4.72		
total contribution(%)	42.02	69.88	90.19	89.79	95.39	69.60	83.67	92.22	80.1	96.0

vs bromodichloromethane, 0.97). Thus, the high relationship among them implies that those compounds were generated simultaneously at the sources. The levels of trihalomethanes were higher than those (0.6–6.1 ppb) previously determined (Toyoda et al., 1986). The ratio of the trihalomethane in cola was almost same as that in tap water (Symons et al., 1975).

The high lipid foods group was contaminated by both trihalomethane and cleaning solvents. As discussed in the previous paragraph, the relationship between contaminant levels and storage may depend upon the circumstances. However, the sources of the trihalomethane are not obvious. Production procedures for butter and margarine involve a washing step and a salt solution addition step. The trihalomethane levels in the samples which were manufactured by two different producers were almost the same. They probably used chlorinated water to wash the materials and to make the salt solution. The sources of the trihalomethanes may be the water that was used for production.

The lactic semisolidified foods group was contaminated by relatively high level of trihalomethanes and low levels of cleaning solvents. The main ingredient of these foods is water (88–60% by weight). If tap water or water disinfected by chlorination was used, then trihalomethanes will be retained in the final products. The raw materials for these foods (butter, cream, soybeans, and so on) contain high levels of lipids. During storage these foods may become contaminated or cleaning solvents may accumulate as discussed in the previous paragraph. The water used for production is disinfected by chlorination; therefore, relative amounts of chloroform, bromodichloromethane, and dibromochloromethane are essentially same as those reported (Symons et al., 1975). However, the levels of trihalomethanes in the foods were lower than those in tap water (Symons et al., 1975). Those differences are due to different source waters with variations in the kinds and levels of trihalomethanes (Trussel and Umphres, 1978).

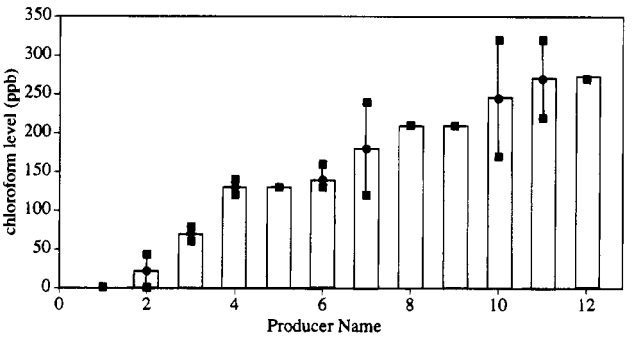

Figure 6. Chloroform level vs food producer of moyashi.

As a group the low level contaminated foods differed because the sources were different for each production process and source of raw materials. The water in juice does not require chlorination; cleaner water is used to produce juice. Therefore, when condensed juice is diluted with tap water, the levels of trihalomethanes were about the same as those found in cola. Milk is a natural product and chlorination is not required. However, the bottling process line is disinfected and washed with chlorinated sanitizers (Green et al., 1993), and the retained water may be a source of contaminants. The levels found were very low. The major part of cake is puff bread and will not contain volatile compounds because of the baking process. But the topping often consists of cream and butter. The homogenates of these mixtures were analyzed and the levels observed to be very low. Rice with hulls is stored in warehouses and polished just before it is packed for sale; therefore, the levels in rice were low.

The contaminants levels in the moyashi were very high. Especially, the levels of chloroform which were 10–100 times higher than those found in other foods. The levels depended upon the producing factories. The relationship between food processor and the chloroform level is shown in Figure 6. Moyashi is produced in high humidity and high temperature. The product must be

disinfected with sodium hypochloride before and after the process to reduce bacteria in and on the moyashi. The levels of trihalomethanes in moyashi produced by factories that do not use sodium hypochloride were very low. They may use phosphoric acid instead of sodium hypochloride.

The contaminant production mechanism is same as that for bleached flour (Heikes and Hopper, 1986). We did not determined carbon tetrachloride in moyashi samples. It has been determined in wheat and reported by several researchers (Clower, 1980).

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LITERATURE CITED

Aijima, T. *Chemometrics*; Maruzen: Tokyo, 1992 .

AOAC. Volatile Fumigants in Grain. In *AOAC Official Methods of Analysis*, 2nd ed.; Sawyer, L. D., McMahon, B. M., Parker, G. A., Eds.; Association of Official Analytical Chemists: Arlington, VA, 1990; pp 290–291.

APHA. Purge and trap packed-column gas chromatographic/mass spectrometric method I. In *Standard Methods for the Examination of Water and Wastewater*, 15th ed., Baumann, F. J., Ed.; American Public Health Association: Washington, DC, 1992; p 6210B.

Bicchi, C. P.; Binello, E. A.; Legovich, M. M.; Pellegrino, G. M.; Vanni, A. C. Characterization of Roasted Coffee by S-HSGC and HPLC-UV and Principal Component Analysis. *J. Agric. Food Chem.*, **1993**, *42*, 2324–2328.

Clower, M., Jr. Modification of the AOAC Method for Determination of Fumigants in Wheat. *J. AOAC* **1980**, *63*, 539–545.

Crecente, R. P.; Latorre, H. H. Pattern Recognition Analysis Applied to Classification of Honeys from Two Geographic Origins. *J. Agric. Food Chem.* **1993**, *41*, 560–564.

EPA-Japan. *Manual for Capillary Gas chromatography-Mass Spectrometric Determination of Environmental Pollutants*; Enviornmental Protection Agency of Japan: Tokyo, 1992.

Entz, R.; Hollifield, H. C. Head space gas chromatographic analysis of foods for volatile halocarbons. *J. Agric. Food Chem.* **1982**, *30*, 84–88.

Eriksson, L.; Johsson, J.; Berglind, R. External Validation of a QSAR for the Acute Toxicity of Halogenated Aliphatic Hydrocarbons. *Environ. Toxicol. Chem.* **1993**, *12*, 1185–1191.

FDA. Ethylene Dichloride, Trichlorethylene. In *Food Additive Analytical Manual*, 2nd ed.; Warner, C., Modderman, J., Faazio, T., Eds., Association of Official Analytical Chemists: Arlington, VA, 1983; Vol. 1, pp 176–183, 317–319.

Green, A. K.; Few, B. K.; Serafini, J. O. A Comparison of Ozonation and Chlorination for Disinfection of Stainless Steel Surfaces. *J. Dairy Sci.* **1993**, *76*, 3617–3620.

Heikes, D. H.; Hopper, M. L. Purge and Trap Methods for Determination of Fumigants in Whole Grains, Milled Grain Products, and Intermediate Grain-Based Foods. *J. AOAC* **1986**, *69*, 990–998.

Massarti, D. L.; Vandeginste, B. G. M.; Deming, S. N.; Michotte, Y.; Kaufman, L. Clustering Techniques in *Chemometrics: a textbook*; Vandeginste, B. G. M., Kaufman, L. Eds.; Data Handling in Science and Technology; Elsevier: Amsterdam, 1988a; Vol. 2, Chapter 22.

Massarti, D. L.; Vandeginste, B. G. M.; Deming, S. N.; Michotte, Y.; Kaufman, L. Principal Components and Factor Analysis. In *Chemometrics: a textbook*; Vandeginste, B. G. M., Kaufman, L. Eds.; Data Handling in Science and Technology; Elsevier: Amsterdam, 1988b; Vol. 2, Chapter 21.

McLeod, H. A. GC of Fumigant Residues From Procedure 7.5. In *Analytical Methods for Pesticide Residues in Foods*, 2nd revision ed.; McLeod, H. A., Graham, R. A., Eds.; Minister of Supply and Services Canada: Ottawa, Canada, 1986; Section 8-11, 8-89-94.

Miyahara, M.; Saito, Y. Efficiency of Refining Process Pesticides in Oil. *J. Agric. Food Chem.* **1993**, *41*, 1150–1153.

Miyahara, M.; Sasaki, K.; Suzuki, T.; Saito, Y. Expanded coagulating cleanup procedures for simultaneous gas chromatographic determination of organophosphorus pesticides in crops and fruits. *Chem. Pharm. Bull.* **1991**, *39*, 1055–1058.

Miyahara, M.; Suzuki, T.; Saito, Y. Multi-residue Methods for Some Pesticides in Lanolin by Capillary Gas Chromatography with Detection by Electron Capture, Flame Photometric, Mass Spectrometric, and Atomic Emission Techniques. *J. Agric. Food Chem.* **1992a**, *40*, 64–69.

Miyahara, M.; Suzuki, T.; Saito, Y. Capillary Gas Chromatographic Determination of Captafol in Vegetables, Fruits and Grains. *J. Agric. Food Chem.* **1992b**, *40*, 1150–1153.

Rook, J. J. Chlorination Reactions of Fulvic Acids in Natural Waters. *Environ. Sci. Technol.*, **1977**, *11*, 478–482.

Symons, J. M.; Bellar, T. A.; Carswell, J. k.; DeMarco, J.; Kropp, K. L.; Robeck, G. G.; Seeger, D. R.; Slocum, C. J.; L., S. B.; Stevens, A. A. National Organics Reconnaissance Survey for Halogenated Organics. *J. Am. Water Works Assoc.* **1975**, *67*, 634–647.

Symons, J. M.; Stevens, A. A.; Clark, R. M.; Geldreich, E. E.; Love, O. T., Jr.; DeMarco, J. *Treatment Techniques for Controlling Trihalomethanes in Drinking Water*; EPA: Ohio, 1981, Chapter 2.

Tamagawa, K.; Mishima, Y.; Seki, T.; Tunoda, A. Daily Dietary Intakes of Trihalomethanes. *J. Food Hyg. Soc. Jpn.* **1988**, *29*, 156–160.

Toyoda, M.; Ishizaka, T.; Saito, I. Determination of Minute Quantities of Chloroform in Foods. *J. Food Hyg. Soc. Jpn.* **1986**, *27*, 246–251.

Toyoda, M.; Kobayashi, K.; Mitsura, A.; Saito, Y.; Uno, K.; Sakabe, Y. Estimation of Daily Intake of Chloroform by Japanese Housewives. *J. Food Saf.* **1987**, *8*, 219–224.

Toyoda, M.; Takagi, K.; Tsurumizu, A.; Saito, Y.; Miyata, T.; Kikawa, H.; Kawamura, T.; Nagai, Y.; Sakabe, Y. Estimation of Daily Intake of Four Kind of Trihalomethane by Japanese Housewives. *J. Food Saf.* **1990**, *10*, 219–224.

Trussel, R. R.; Umphres, M. D. The formation of Trihalomethanes. *J. Am. Water Works Assoc.* **1978**, *70*, 604–612.

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