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Chlorinated Benzenes in Herring Gull and Double-Crested Cormorant Eggs from Three Locations in the Maritime Provinces

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CHLORINATED BENZENES IN HERRING GULL AND DOUBLE-CRESTED CORMORANT EGGS FROM THREE LOCATIONS IN THE MARITIME PROVINCES

by

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ABSTRACT

Herring gull (Larus argentatus) and double-crested cormorant (Phalacrocorax auritus) eggs were collected from colonies at Boot Island, Nova Scotia; Manawagonish Island, New Brunswick and Cherry Island, Prince Edward Island and analyzed for chlorinated benzene content. Measurement by combined gas chromatography/mass spectrometry showed all sample extracts contained chlorinated benzenes with concentrations dependent primarily on the particular isomer and sample and to a lesser extent on site and species. No significant statistical variations were determined for average isomeric content among locations although the Boot Island eggs appeared somewhat more contaminated. Values for the three Maritime locations were approximately one order of magnitude lower than measurements reported for a similar study in Lake Ontario.

RÉSUMÉ

Des oeufs de goélands (Larus argentatus) et de cormorans (Phalacrocorax auritus) ont été recueillis de colonies situées à Boot Island en Nouvelle-Ecosse, Manawagonish Island au Nouveau-Brunswick et Cherry Island à l'Ile-du-Prince-Edouard. Les oeufs ont été examinés dans le but d'analyser leur teneur en benzène chloré. Les analyses ont été effectuées en utilisant à la fois la chromatographie au gaz et un spectographe de masse. Cette évaluation a permis de déceler la présence de benzène chloré dans tous les échantillons et a révélé que le degré de concentration provenait particulièrement de l'isomère de benzène chloré même et de l'échantillon. L'espèce et le site de cueillette des oeufs n'influençaient que moindrement les concentrations de benzène chloré. Bien que les oeufs provenant de Boot Island semblaient relativement un peu plus contaminés que ceux des autres sites, il n'a pas été possible de déterminer dans les divers sites des variations statistiques significatives dans le contenu isomérique moyen. Les valeurs établies pour les trois sites de la région des Maritimes étaient d'un rang de grandeur inférieur aux mesures rapportées dans une étude semblable faite dans la région du lac Ontario.

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1 INTRODUCTION

Chlorinated benzenes are included on the DOE/NHW List of Priority Chemicals. This list is intended to focus upon those substances for which regulations are being developed under the Environmental Contaminants Act (ECA) and upon those substances about which further information must be obtained to determine whether regulation is necessary. Materials are evaluated on the basis of toxic effects, persistence, and quantity and use criteria. Chlorinated benzenes appear in Category III of the list, indicating that the federal government considers they may pose a significant danger to human health or the environment and that further detailed information is required. Many chlorinated benzenes have been identified in the tissue of fish and fish-eating birds from the Great Lakes, indicating not only their presence in the environment, but also their persistence and accumulation.

A survey of selected industrial effluents in the Atlantic Region (Travers, 1978) indicated a number of industries were discharging chlorinated benzenes in their effluents; however, in most cases quantities were not considered significant. Sediments, collected in the

immediate vicinity of effluent outfalls found to have the highest concentrations, were contaminated in all cases(MacLaren Marex, 1979).

To complement this data, information relevant to an appropriate biotic monitoring species was desirable. Eggs of the herring gull (Larus argentatus) and double-crested cormorant (Phalacrocorax auritus) were selected for study because of the availability of comparable data on hexachlorobenzene from the Great Lakes. The Canadian Wildlife Service, as part of their Toxic Chemicals Program, regularly sample several gull colonies in the Atlantic Provinces. As part of a cooperative effort with the Environmental Protection Service, they organized and conducted the sampling portion of this survey.

2 CHLORINATED BENZENES IN THE ENVIRONMENT

2.1 Production, Use and Occurrence of Chlorinated Benzenes

Chlorinated benzenes are synthetic organic compounds which, on the basis of their physical and chemical properties, have widespread industrial and

agricultural application. These compounds are insoluble in water, highly soluble in organic solvents (e.g. carbon tetrachloride, ether), have low vapour pressures and high octanol-water partitioning coefficients (EPA, 1978).

All chlorinated benzene isomers are highly stable in environmental matrices and degradation via photolytic, physico-chemical and biological mechanisms is restricted (Gilbertson, 1978). The stability and lipophilic qualities of these compounds accentuate their bioaccumulation potential and subsequent magnification up the food chain.

Chlorinated benzenes are ubiquitous, having been detected in air, water, sediments and biota in North America, Europe and Japan. Their widespread occurrence is attributed to varied agricultural, industrial and commercial production and usage. Figures for the United States indicate that in 1972, 15.6 million pounds of 1, 2,4-trichlorobenzene were manufactured; while in 1976, over 392 million pounds of monochlorobenzene, 48 million pounds of o-dichlorobenzene and 37 million pounds of p-dichlorobenzene were produced (EPA, 1979, 1978).

Chlorinated benzene applications include

organic intermediates in the dye industry (1,2,3-trichlorobenzene); heat transfer media (monochlorobenzene); fumigants (m-dichlorobenzene); spatial odorants (o- and p-dichlorobenzene); transformer lubricants (1,2,4-trichlorobenzene); and pesticides (penta- and hexachlorobenzene) (EPA, 1978). Chlorinated benzenes are also produced as by-products or impurities in the manufacture of chlorinated hydrocarbons such as polychlorinated biphenyls and carbon tetrachloride (Isensee, et. al., 1976).

The broad application of these compounds results in both their deliberate and accidental release to and subsequent accumulation in the environment. For example, levels of trichlorobenzenes in two river systems in the southern United States ranged between 5-460 $\mu g/l$ (EPA, 1979). Aerial concentrations of p-dichlorobenzene in Japan were found to vary from 1.5 $\mu g/m^3$ in suburban Tokyo to 4.2 $\mu g/m^3$ in the central area of the city (Morita and Ohi, 1975).

In the Maritime Provinces a 1978 survey indicated the presence of several chlorinated benzenes in the discharge of local industries. Chlor-alkali and wood preserving plants were found to be discharging penta- and

hexachlorobenzene while textile mills contained di-, tri-, tetra-, and pentachlorobenzenes in their effluents (Travers, 1978).

An incident of human poisoning by hexachlorobenzene-treated seed wheat in south-east Turkey prompted the establishment of criteria for human foodstuffs by W.H.O. in 1969 (Beall, 1976). Recently, the U.S. Environmental Protection Agency has published a guideline for hexachlorobenzene content in foodstuffs; poultry meat, fat, shellfree eggs and dairy products other than cheese are restricted to .1 ppm, while cheese is set at .3 ppm (Tuttle, 1979).

Criteria have also been established by this agency to protect fresh and salt water life. For fresh water the 24 hour average concentration for the following chlorinated isomers are: o-dichlorobenzene - 44 µg/l; m-dichlorobenzene - 310 µg/l; p-dichlorobenzene - 190 µg/l. For salt water, these criteria are respectively 15 µg/l, 22 µg/l, and 15 µg/l (EPA, 1978). Criteria for other chlorobenzene isomers must await the results of further investigation.

2.2 Metabolism of Chlorinated Benzenes

Studies on the metabolism of chlorinated benzenes have concentrated on laboratory investigations involving rabbits, rats, chickens, and Japanese quail (Coturnix coturnix Japonica). These studies have shown that chlorobenzene contamination can occur by normal respiration and dietary uptake.

The toxic effects observed in mammals and birds include liver and kidney damage, necrosis, central nervous system depression, hyperexcitability, restlessness and hepatic porphyria (EPA, 1978, 1979), an affliction which interferes with the normal synthesis of heme, hemoglobin, myoglobin, cytochromes and various enzymes (NRC, 1975).

Chlorinated benzenes, as mentioned previously, are highly lipophilic and bioaccumulation into fatty tissues from the digestive tract of animals has been reported (Kohli, et. al., 1976; EPA, 1976b). The assimilation efficiency is directly related to the chlorination of the aromatic ring (EPA, 1979).

Hexachlorobenzene (HCB) has been studied more

rigorously than any other chlorobenzene isomer due to its widespread use as an insecticide and its detection in many environmental surveys assessing organohalogens. HCB has a low acute but high chronic toxicity due to tissue accumulation effects (NRC, 1975). Chickens fed HCB-treated seed for six months had body fat levels 21-31 times their feeding levels (Avrahami and Steele, 1972a and b). Quail, fed 80 μg/g HCB per day, exhibited reduced egg production after three weeks and reduced egg fertility and hatchability after six weeks.

The metabolism of chlorinated benzenes occurs very slowly in all organisms studied and is believed to occur via hydroxylation through arene oxide intermediates (Kohli, et. al., 1976; EPA, 1977) with subsequent reduction to phenolic metabolites. These metabolities are excreted in the urine as glucuronides, sulfate and mercapturic acid conjugates (EPA, 1978a).

Depuration studies with rabbits have shown that the greater proportion of mono- and dichlorobenzenes are metabolized and expelled from the body in a 1-5 day period following dosage (Azouz, et. al., 1955; Parke and Williams, 1955). Six days after dosing, 48% of the least readily metabolized tetrachlorobenzene (the 1,2,4,5-isomer)

remained in the tissues of rabbits (Jondorf, et. al., 1958; EPA, 1979), while more than seven days were required for the removal of 50% of accumulated pentachlorobenzene (Parke and Williams, 1960; EPA, 1979). HCB had the longest half-life of the chlorobenzenes, ranging from 24-27 days in the body fat of chickens (Hansen, et. al., 1978) to 4-5 months in rats (Koss, et. al., 1978).

Because of such long term retention, second generation effects have been documented in the progeny of chickens (Avrahami and Steele, 1972a and b), rats (Vos, et. al., 1979), rabbits (Agatha Corp, 1976), and quail (Leoni and D'Arca, 1976). Effects vary with the species and the parental dose applied. Chickens, transferring HCB from body fat to egg yolk, produced young showing no detrimental effects when fed HCB at levels below 100 µg/g (Avrahami and Steele, 1972a and b), while the young of pregnant rats fed 150 µg/g HCB in feed exhibited reduced cell immunity and increased liver weights (Vos, et. al., 1979).

2.3 Chlorinated Benzenes in the Aquatic Environment

The intricate food chain of the aquatic

environment is particularly sensitive to major accumulations of chlorinated benzenes. They enter the environment via air or water-borne effluents, are readily retained in sediments (Isensee, et. al., 1976; Beall, 1976; Niimi, 1979) and are distributed throughout the primary consumers (e.g. algae, bacteria), thus assuring contamination up the food chain. Accumulation rates are dependent on the bioaccumulation potential of the particular chlorobenzenes, the organism's resistance to the cumulative effects, species habitat, feeding habits and behaviour; as well as individual age, maturity and body weight (Hickey, et. al., 1976; Zitko, et. al., 1974; Blus, et. al., 1979; Szaro, et. al., 1979). High bioaccumulation rates have been reported for some species. For instance, juvenile largemouth bass (Micropterus salmoides) exposed to 2 µg/l HCB for 15 days exhibited a biomagnification of 44,000 and damage at the cellular and organ level was detected (EPA, 1976b). Adult catfish, which are benthic feeders, concentrated HCB by a factor of 15,000 with no apparent toxic effect (EPA, 1976a). Lake Huron trout accumulated p-dichlorobenzene to a factor of 215 in muscle tissue and the more chlorinated HCB to 7880.

2.4 Chlorinated Benzenes in Bird Eggs

The potential hazard associated with concentrations of chlorinated hydrocarbons measured in bird eggs is species dependent. Levels of HCB (.6 μg/g); dieldrin (.091 μg/g); DDT (.24 μg/g) and PCB's (20 μg/g) measured in the great skua (Catharacta skua) have no apparent effect but are 2-3 times higher than values impairing reproductive success in the brown pelican (Pelecanus occidentalis) (Furness and Hutton, 1979).

High levels of chlorinated hydrocarbons have been linked to reduced reproductive success of fish-eating birds in the Great Lakes (Peakall, 1970; Gilbertson, 1974; Gilbertson and Fox, 1977; King, et. al., 1978; Blus, et. al., 1979; Szaro, et. al., 1979) although a recent study (Gilman, et. al., 1978) found no conclusive proof that organochlorines had any such effect on herring gulls.

The population effects of chlorinated benzenes on fish-eating birds are not well documented and only comparative studies exist in the literature. A "slightly polluted" environment for the common tern (Sterna hirundo) in Bathurst, New Brunswick resulted in average egg levels

of .028 µg/g HCB, while an HCB value of 2.3 µg/g (Holden, 1973) was determined for eggs from the "polluted" environment of Hamilton, Ontario.

Chlorinated benzene levels in herring gull eggs collected in 1977 from Lake Ontario colonies (Table 1) have a mean HCB level of .45 µg/g fresh weight (Hallett, et. al., 1978). This is considerably higher than the trace levels reported for double-crested cormorants, herring gulls, and black ducks (Anas rubripes) in Passamaquoddy Bay and the Bay of Fundy (Zitko and Choi, 1972; Zitko, 1976). Presumably levels are representative of the relative contamination of the surrounding environment. Herring gulls have been used successfully as an indicator species for chlorinated hydrocarbons because of their habitat and feeding characteristics as well as their potential for bioconcentrating even trace level contaminants (Peakall, et. al., 1978).

TABLE 1 CHLORINATED BENZENES IN LAKE ONTARIO HERRING GULL EGGS (From Hallett, et. al., 1978)

(ng/g fresh weight)

	Tetrachlorobenzenes	Pentachlorobenzene	Hexachlorobenzene
	24	37	464
Lake Ontario	26	44	494
Bird Colonies	36	47	466
	23	30	370
	13	25	355
	29	34	449
	29	39	521
	14	31	392
	19	46	463
	22	43	574
	26	37	433
	39	51	425
	32	39	454
m ± c*	26 ± 4.6	39 ±4.5	450± 36
S**	7.7	7.5	60

* m ± c = mean ±95% confidence interval

** S = standard deviation

3 METHODS AND MATERIALS

3.1 Apparatus

1. Gas chromatography, Hewlett-Packard 5713A equipped with a Ni-63 electron capture detector and a 1.0 mv single pen recorder.

2. Gas chromatograph-mass spectrometer system (GC/MS), Hewlett-Packard 5980A mass spectrometer interfaced with an HP 5933A data system. The quadrupole mass spectrometer was coupled to the gas chromatograph via a glass-lined jet separator held at 270@ and was operated in the selected ion mode through data system control.

3. Gas chromatographic columns (electron capture system); 180 cm long x 3 mm I.D.

4. Column packings (electron capture system); (a) 3% Dexil 300 GC on 100/120 Supelcoport, (b) 1.5% SP-2250/1.95% SP-2401 on 100/120 Supelcoport.

5. Gas chromatographic column (GC/MS system); 90 cm long x 2 mm I.D.

6. Column packing (GC/MS system); 3% SP-2100 on 100/120 Supelcoport.

7. Polytron homogenizer.

3.2 Reagents

All solvents were glass distilled.

1. Benzene
2. Hexanes
3. Acetone
4. Acetonitrile
5. Silica gel, 100/120 mesh - activated at 250° for 8 hr., deactivated with 5% water (by weight), shaken for 2 hr., stored in a dessicator.
6. Sodium sulfate - anhydrous, reagent grade, heated for 8 hr. at 600° and stored in a closed glass container. Washed with benzene prior to use.
7. Chlorinated benzene standards - Aldrich Chemical Company

o-Dichlorobenzene

m-Dichlorobenzene

p-Dichlorobenzene

1,2,3-Trichlorobenzene

1,2,4-Trichlorobenzene

1,3,5-Trichlorobenzene

1,2,3,4-Tetrachlorobenzene

1,2,3,5-Tetrachlorobenzene

Pentachlorobenzene

Hexachlorobenzene

3.3 Sample Collection

Herring gull and double-crested cormorant eggs were collected from colonies at three different locations in the Maritime Provinces: Manawagonish Island, Cherry Island and Boot Island. These locations are graphically depicted in Figure 1. The number of egg samples collected at each site ranged between 7-18. Contents of individual eggs were stored in glass containers which had been pre-rinsed with benzene. Sample caps were lined with aluminium foil to prevent contamination. Samples were stored frozen until analyzed.

3.4 Sample Preparation

After thawing, the contents of each egg were homogenized in the collection vessel using a blender. A 15 g aliquot of the homogenate was added to a 200 ml Berzelius beaker containing 75 ml of 20% acetone/80% acetonitrile and extracted using a polytron homogenizer. The supernatant obtained after centrifuging was transferred to a 2000 ml separatory funnel containing 100 ml hexane and 750 ml of a 5% sodium sulfate solution. The fat residue was re-extracted with a second 75 ml

portion of acetone/acetonitrile which was combined with the original extract.

After vigorous shaking and subsequent settling, the hexane layer was decanted and the aqueous layer extracted with an additional 100 ml portion of hexane. The combined hexane extracts were washed twice with distilled water, dried with anhydrous sodium sulfate, concentrated to approximately 5 ml on a vacuum rotary evaporator and finally reduced in volume to 1 ml under a stream of dry nitrogen.

The concentrated extract was fractionated on a silica gel column (10 x 100 mm) (a small amount of anhydrous sodium sulfate was added to the top of the column to remove any extraneous water). The initial 15 ml of eluate were collected, transferred to a 15 ml centrifuge tube and concentrated to 2 ml. One ml of concentrated sulfuric acid was slowly added to the concentrate and the tube carefully shaken using a vibrating mixer. After the tube was cooled in an ice bath, the hexane layer was withdrawn by pipette (centrifuging may be necessary if an emulsion forms). The sulfuric acid was then re-extracted with a second 2 ml portion of hexane. The combined extract was treated

with 3 ml of 10% sodium bicarbonate, washed with 2 ml of deionized- distilled water, dried using anhydrous sodium sulfate and concentrated to 1 ml.

The concentrate was eluted through a micro silica gel column and the initial 10 ml collected. The volume was adjusted for analysis under a stream of dry nitrogen.

3.5 Sample Analysis

3.5.1 Gas Chromatographic Analyses

Sample volumes were adjusted to either 1 ml or 0.5 ml for analysis by electron capture gas chromatography. In either case 2 µl portions were injected and all extracts were analyzed on both of the columns described previously. The operating conditions were as follows:

Column temperature	-	115^{o}
Inlet temperature	-	200^{o}
Detector temperature	-	300^{o}
Carrier gas	-	Argon/Methane (95/5)
Carrier gas flow rate	-	60 ml/min

Chlorinated benzenes present in the egg extracts were identified by comparing retention times of unknowns with those of reference materials. Peak areas were determined manually and quantitative evaluation was based on a comparison with areas obtained for known amounts of standard substances.

3.5.2 Gas Chromatographic/Mass Spectrometric Analyses

The four ions of greatest relative abundance in the mass spectra of di-, tri-, tetra-, penta- and hexachlorobenzene(s) were selected for monitoring. Initiation of spectrometer monitoring for each of these groups of four ions was based on retention time data obtained using reference materials. The instrumental parameters and conditions used in the study are listed below:

Gas Chromatograph:

Initial temperature	-	85^{o} (4 min hold)
Final temperature	-	150^{o} (hold)
Program rate	-	8^{o}/min
Injection port	-	250^{o}
Carrier gas	-	helium
Carrier gas flow rate	-	22 ml/min
Sample size	-	2 µl

Mass Spectrometer:

Ion source	-	240^{o}
Analyzer manifold	-	110^{o}
Glass-lined jet separator	-	250^{o}
Operating pressure	-	5×10^{-6} torr
Electron energy	-	70 ev

4 RESULTS AND DISCUSSION

4.1 Analytical Measurement

All sample extracts were analyzed by combined gas chromatography/mass spectrometry (GC/MS) as well as by electron capture gas chromatography. The recorded values are those obtained using the former system, although similar results were determined with the electron capture instrument. With its capability of monitoring particular ions in the mass fragmentation pattern, the GC/MS served as a specific detector for tri-, tetra-, penta- and hexa-substituted benzenes. Identification of particular tri- and tetra-substituted isomers was based on retention times.

Although both analytical systems allow the measurement of mono- and dichlorobenzenes, no values have been reported. Reagent and sample blanks analyzed concurrently with the eggs showed high background levels of these materials which prevented reliable quantitation. Chemical pretreatment of reagents was only partially successful in removing these extraneous interferences. Consequently, little confidence could be attached to values for these compounds measured in environmental samples and they have not been included for discussion.

With this one exception, no problems were encountered with the analytical measurements. Recoveries with spiked egg samples were in excess of 80% for all isomers.

4.2 Chlorinated Benzene Content of Egg Samples

Herring gull and double-crested cormorant eggs were collected from the three Maritime locations shown in Figure 1. The quantities of chlorinated benzenes measured in each sample are listed in Tables 2-6. Boot Island is on the south side of Minas Basin, Nova Scotia and the bird colonies there might possibly be influenced by contaminant discharges emanating from the Windsor and

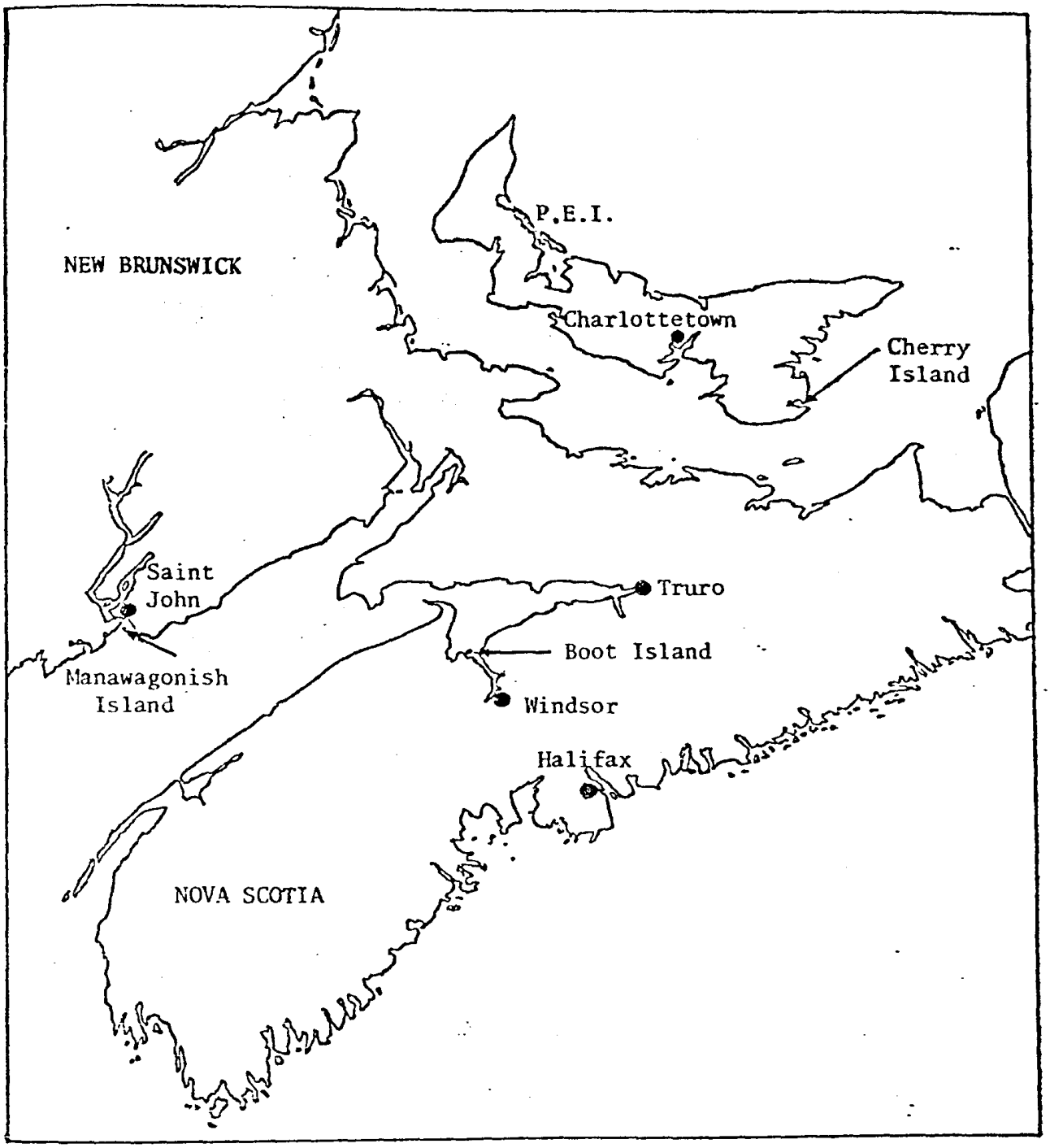

FIGURE 1 MAP OF THE MARITIME PROVINCES INDICATING THE LOCATION OF BIRD COLONIES WHERE EGGS WERE COLLECTED, AND THEIR PROXIMITY TO MAJOR PROVINCIAL INDUSTRIAL CENTRES

TABLE 2 CHLORINATED BENZENES IN HERRING GULL (Larus argentatus) EGGS FROM MANAWAGONISH ISLAND, N.B., MAY 9, 1979. (ng/g fresh weight)

Sample	Trichlorobenzene			Tetrachlorobenzene		Pentachlorobenzene	Hexachlorobenzene
	1,3,5-	1,2,4-	1,2,3-	1,2,3,5-* +1,2,4,5-	1,2,3,4-		
1	n.d.	n.d.	n.d.	1.0	0.6	0.8	5.8
2	n.d.	n.d.	n.d.	0.2	0.2	0.5	6.0
3	n.d.	n.d.	n.d.	0.5	0.2	0.6	3.2
4	n.d.	2.9	n.d.	0.4	0.2	0.8	13
5	n.d.	n.d.	n.d.	n.d.	0.1	0.3	4.2
6	n.d.	n.d.	n.d.	0.6	0.3	1.6	9.4
7	n.d.	2.8	1.0	0.4	0.5	0.2	2.0
8	n.d.	n.d.	n.d.	2.3	2.0	2.1	29
9	n.d.	n.d.	n.d.	0.3	0.4	0.6	1.8
10	n.d.	n.d.	n.d.	0.2	0.1	0.2	2.8

* quantitated as the 1,2,4,5-tetrachlorobenzene isomer

n.d. = not detected; detection limits:

1,3,5-trichlorobenzene = 0.2 ng/g

1,2,4-trichlorobenzene = 1.0 ng/g

1,2,3-trichlorobenzene = 0.5 ng/g

1,2,4,5-tetrachlorobenzene = 0.2 ng/g

1,2,3,4-tetrachlorobenzene = 0.1 ng/g

pentachlorobenzene = 0.1 ng/g

hexachlorobenzene = 0.1 ng/g

TABLE 3 CHLORINATED BENZENES IN HERRING GULL (Larus argentatus) EGGS FROM BOOT ISLAND, N.S., MAY 7, 1979.
(ng/g fresh weight)

Sample	Trichlorobenzene			Tetrachlorobenzene		Pentachlorobenzene	Hexachlorobenzene
	1,3,5-	1,2,4-	1,2,3-	1,2,3,5-* +1,2,4,5-	1,2,3,4-		
1	-	-	-	7.8	3.7	1.9	30
2	n.d.	14	2.4	7.9	3.1	4.3	19
3	n.d.	13	2.0	4.2	3.0	0.8	2.7
4	n.d.	37	3.3	11	5.5	1.9	16
5	n.d.	5.4	1.6	2.9.	2.8	1.3	11
6	n.d.	12	2.3	2.9	1.5	0.5	2.7
7	n.d.	3.4	n.d.	n.d.	8.5	4.8	3.4
8	n.d.	4.1	n.d.	1.3	1.0	0.6	4.3
9	n.d.	7.0	1.0	8.6	2.4	2.8	7.9
10	n.d.	13	2.0	5.5	2.8	0.7	1.9

* quantitated as the 1,2,4,5-tetrachlorobenzene isomer

n.d. = not detected; detection limits·

1,3,5-trichlorobenzene = 0.2 ng/g

1,2,4-trichlorobenzene = 1.0 ng/g

1,2,3-trichlorobenzene = 0.5 ng/g

1,2,4,5-tetrachlorobenzene = 0.2 ng/g

1,2,3,4-tetrachlorobenzene = 0.1 ng/g

pentachlorobenzene = 0.1 ng/g

hexachlorobenzene = 0.1 ng/g

TABLE 4 CHLORINATED BENZENES IN HERRING GULL (Larus argentatus) EGGS FROM CHERRY ISLAND, P.E.I. MAY 7, 1979.
(ng/g fresh weight)

Sample	Trichlorobenzene			Tetrachlorobenzene		Penta-chloro-benzene	Hexa-chloro-benzene
	1,3,5-	1,2,4-	1,2,3-	1,2,3,5-* +1,2,4,5-	1,2,3,4-		
1	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	1.0
2	n.d.	n.d.	n.d.	4.4	1.0	3.6	62
3	n.d.	n.d.	n.d.	0.8	0.2	0.5	11
4	n.d.	2.9	n.d.	1.1	0.5	2.0	31
5	n.d.	n.d.	n.d.	3.1.	1.1	2.9	38
6	n.d.	n.d.	n.d.	0.6	0.8	1.0	16
7	n.d.	2.8	1.0	0.9	0.3	0.8	12

* quantitated as the 1,2,4,5-tetrachlorobenzene isomer

n.d. = not detected; detection limits:

1,3,5-trichlorobenzene = 0.2 ng/g
1,2,4-trichlorobenzene = 1.0 ng/g
1,2,3-trichlorobenzene = 0.5 ng/g
1,2,4,5-tetrachlorobenzene = 0.2 ng/g
1,2,3,4-tetrachlorobenzene = 0.1 ng/g
pentachlorobenzene = 0.1 ng/g
hexachlorobenzene = 0.1 ng/g

TABLE 5 CHLORINATED BENZENES IN DOUBLE-CRESTED CORMORANT (*Phalacrocorax auritus*) EGGS FROM MANAWAGONISH ISLAND, N.B., MAY 9, 1979 (ng/g fresh weight)

Sample	Trichlorobenzene			Tetrachlorobenzene		Penta-chloro-benzene	Hexa-chloro-benzene
	1,3,5-	1,2,4-	1,2,3-	1,2,3,5-* +1,2,4,5-	1,2,3,4-		
1	n.d.	n.d.	n.d.	0.2	0.4	0.4	3.5
2	n.d.	1.5	n.d.	0.8	0.7	1.4	11
3	n.d.	2.6	1.0	6.4	0.2	1.9	18
4	n.d.	2.9	n.d.	0.3	0.2	0.6	4.4
5	n.d.	4.0	0.8	5.0	2.6	5.5	41
6	n.d.	n.d.	n.d.	0.2	0.2	0.7	6.3
7	n.d.	2.8	1.0	n.d.	0.2	0.8	13
8	n.d.	n.d.	n.d.	0.4	0.4	0.8	7.5
9	n.d.	n.d.	n.d.	0.5	0.6	1.2	11
10	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	0.7

* quantitated as the 1,2,4,5-tetrachlorobenzene isomer

n.d. = not detected; detection limits:

1,3,5-trichlorobenzene = 0.2 ng/g

1,2,4-trichlorobenzene = 1.0 ng/g

1,2,3-trichlorobenzene = 0.5 ng/g

1,2,4,5-tetrachlorobenzene = 0.2 ng/g

1,2,3,4-tetrachlorobenzene = 0.1 ng/g

pentachlorobenzene = 0.1 ng/g

hexachlorobenzene = 0.1 ng/g

TABLE 6 CHLORINATED BENZENES IN DOUBLE-CRESTED CORMORANT (Phalacrocorax auritus) EGGS FROM BOOT ISLAND, N.S., JUNE 5, 1979. (ng/g fresh weight)

Sample	Trichlorobenzene			Tetrachlorobenzene		Pentachlorobenzene	Hexachlorobenzene
	1,3,5-	1,2,4-	1,2,3-	1,2,3,5-* +1,2,4,5-	1,2,3,4-		
1	n.d.	n.d.	n.d.	0.8	0.6	1.3	9.9
2	n.d.	1.5	n.d.	1.2	1.1	3.0	22
3	n.d.	2.6	1.0	0.7	0.8	2.5	46
4	n.d.	2.9	n.d.	0.3	0.4	1.3	15.4
5	n.d.	4.0	0.8	n.d.	0.7	0.7	6.9
6	n.d.	n.d.	n.d.	0.4	0.7	1.1	7.9
7	n.d.	2.8	1.0	0.5.	0.6	1.0	11
8	n.d.	n.d.	n.d.	0.9	0.7	1.2	11

* quantified as the 1,2,4,5-tetrachlorobenzene isomer

n.d. = not detected; detection limits:

1,3,5-trichlorobenzene = 0.2 ng/g

1,2,4-trichlorobenzene = 1.0 ng/g

1,2,3-trichlorobenzene = 0.5 ng/g

1,2,4,5-tetrachlorobenzene = 0.2 ng/g

1,2,3,4-tetrachlorobenzene = 0.1 ng/g

pentachlorobenzene = 0.1 ng/g

hexachlorobenzene = 0.1 ng/g

Truro areas. Manawagonish Island is near the port of Saint John, New Brunswick, a comparatively heavily industrialized area. Cherry Island is on the Prince Edward Island coast near Murray River, some distance removed from potential sources of industrial pollution. All samples analyzed contained at least some of the chlorinated benzene isomers, although concentrations and isomeric distribution varied somewhat with species and collection site. In fact, substantial differences were sometimes determined among egg samples of the same species and colony. Of the particular chlorinated benzenes monitored, only the 1,3,5-isomer was undetected in any sample.

Average values for each of the colonies sampled are listed in Table 7. With the exception of hexachlorobenzene (HCB), levels for eggs from Boot Island are somewhat higher than those for the other two locations. Herring gull eggs from Cherry Island have an average HCB content of 26 ng/g, highest of the three sampling sites. A rationale for this anomaly is not readily discernible, although it may suggest a localized use in a commercial or agricultural operation. HCB is the predominant isomer in all cases regardless of species and location. This may be attributable to a number of

TABLE 7 ASSESSMENT OF CHLORINATED BENZENE ISOMERS IN THE EGGS OF HERRING GULL (Larus argentatus) AND DOUBLE-CRESTED CORMORANT (Phalacrocorax auritus) COLONIES FROM THREE LOCATIONS IN THE MARITIME PROVINCES

Chlorinated Benzenes	Herring Gull (ng/g)			Double-Crested Cormora	
	Manawagonish Island	Boot Island	Cherry Island	Manawagonish Island	Boot Island
Hexachloro-benzene					
m ± c*	7.7±5.9	9.9±6.6	26±19	12±8	16±11
S**	8.3	9.2	20	11	13
Pentachloro-benzene					
m ± c*	.8±.4	2.0±1.1	1.8±1.3	2±1	1.5±0.7
S**	.6	1.6	1.2	2	0.8
1,2,3,4-Tetrachloro-benzene					
m ± c*	.5±.4	3.4±1.5	.6±.4	.6±.6	.7±.2
S**	.6	2.2	.4	.8	.2
1,2,4,5-(1,2,3,5-)*** Tetrachloro-benzene					
m ± c*	.7±.5	5.8±2.5	1.8±1.[illegible]	[illegible]	.[illegible]±.3
S**	.7	3.2	1.6	[illegible]	[illegible]
1,2,3-Trichloro-benzene					
m ± c*	n.d.	2.1±.66	n.d.	n.d.	n.d.
S**		.71			
1,2,4-Trichloro-benzene					
m ± c*	n.d.	12±8	n.d.	n.d.	n.d.
S**					
1,3,5-Trichloro-benzene					
m ± c*	n.d.	n.d.	n.d.	n.d.	n.d.
S**					

* m ± c = mean ±95% confidence interval
** S = standard deviation
*** quantitated as the 1,2,4,5-Tetrachlorobenzene isomer
n.d. = not detected; detection limits:
1,2,3-trichlorobenzene = 0.5 ng/g
1,2,4-trichlorobenzene = 1.0 ng/g
1,3,5-trichlorobenzene = 0.2 ng/g

factors such as its ubiquitous nature, a slow rate of biodegradation in the environment and/or the fact that its bioaccumulation potential is the highest of any of the chlorinated benzene isomers.

Other trends are not readily identifiable and no statistical differences could be determined among chlorinated benzene concentrations from the three locations. It is, however, interesting to note that values for eggs collected at Manawagonish Island, the colony of closest physical proximity to an industrialized area, are similar to those for Cherry Island samples and less than those from Boot Island. Comparatively high concentrations of 1,2,3- and 1,2,4-trichlorobenzenes were detected only in herring gull eggs from Boot Island. In an earlier industrial effluent survey (Travers, 1978), these isomers were associated with textile maufacturers and, coincidentally, Nova Scotia Textiles is located at Windsor, Nova Scotia.

Less variability was observed in the average chlorinated benzene concentrations of double-crested cormorants between sampling sites than was the case with herring gulls. The dynamics of the metabolic action of chlorinated benzenes in gulls is unclear (Hallett, 1980).

Some of the variation noted in this experiment may be the result of gulls with dissimilar feeding habits sharing the same nesting site.

The Lake Ontario environment is considerably more contaminated than the Maritime areas sampled in this program; a fact emphasized by comparison of chlorinated benzene values in Tables 1 and 7. Chlorinated benzene levels are approximately one order of magnitude higher in the case of gull eggs collected in Lake Ontario, [illegible] indicating higher contamination of [illegible].

5 CONCLUSIONS AND RECOMMENDATIONS

Concentrations of hexachlorobenzene in extracts from eggs of herring gulls and double-crested cormorants were considerably higher than those measured for any of the other chlorinated benzene isomers. Pentachloro-; 1,2,3,4-tetrachloro- and 1,2,3,5-tetrachlorobenzene were found in most of the samples while 1,2,4-trichloro- and 1,2,3-trichlorobenzene were detected less frequently. Comparable relative distributions of these isomers are assumed to occur in the environments surrounding the colonies, particularly in the immediate food sources.

Samples analyzed in this investigation showed considerably less chlorinated benzene content than similar samples from Lake Ontario, no doubt reflecting relative contamination of the two areas. These results complement the two earlier effluent monitoring and sediment sampling studies and taken together the three surveys suggest that chlorobenzenes do not represent a serious environmental problem in the Atlantic Region.

While there seems little point in repeating this exercise on a yearly basis, a similar study in 3-5 years would be extremely useful from a trend monitoring standpoint; increases or decreases in chlorinated benzene concentrations in such egg samples would be of use in assessing the environmental status of these compounds.

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