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University of Alberta

The Use of Stable Isotope Ratios to Discern Organochlorine Bioaccumulation Patterns in a Sub-alpine Rocky Mountain Lake Food Web.

by

Linda Margaret Campbell (:)



A thesis submitted to the Faculty of Graduate Studies and Research in partial fulfillment of the requirements for the degree of Master of Science

Department of Biological Sciences

Edmonton, Alberta Fall 1997



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Dr. D. W. Schindler, Supervisor

Dr. W. M. Tonn, Committee Member

Dr. M. V. H. Wilson, External Examiner

What Cilian

Date: Soft 8, 1997

Water must be thought of in terms of the chains of life it supports - from the small-as-dust green cells of the drifting plant plankton, through the minute water fleas to the fishes that strain plankton from the water and are in turn eaten by other fishes or by birds, mink, raccoons - in an endless cyclic transfer of materials from life to life. We know that the necessary minerals in the water are so passed from link to link of the food chains. Can we suppose that poisons we introduce into water will not also enter into these cycles of nature?

-Silent Spring
Rachel Carson, 1962

PREFACE

This thesis is written according to the paper format guidelines as outlined by the Faculty of Graduate Studies and Research, University of Alberta. Because Chapters II and III are written as independent manuscripts, some repetition is unavoidable. However, repetition is kept to a minimum whenever possible.

ABSTRACT

Lake trout from sub-alpine Bow Lake had higher concentrations of organochlorines compared with other fish populations nearby. The objective of this study was to determine why this was so. The original hypothesis of biomagnification via a long foodweb was refuted by the analyses of organochlorines and the stable carbon (δ^{13} C) and nitrogen (δ^{15} N) isotope ratios in lake biota. Trophic positioning, indicated by δ^{15} N, showed that there were only three trophic levels in Bow Lake. Lake trout and mountain whitefish shared the top trophic position. Carbon flow, as indicated by δ^{13} C, showed that dietary patterns were important to organochlorine bioaccumulation. Lake trout feeding on contaminated pelagic copepods had higher concentrations than littoral-feeding fish. A secondary study examined whether finray stable isotope ratios could be used to predict that of fish muscle. The results showed that δ^{13} C of finrays correlated significantly with that of fish muscle, while δ^{15} N results were not conclusive.

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Doing the work for this thesis would not have been possible without the help of many people and organizations. Friends of Banff National Park and Circumpolar Institute (C/BAR) grants aided considerably, along with a NSERC operating grant to David. W. Schindler. My work was also supported by a NSERC PG-A scholarship and the "John and Patricia Schlosser Environmental Scholarship".

David A. Donald of Environment Canada provided the impetus for this project, and generously allowed me to use some of his data in my work. Robert (Bob) Crosley and Jim Syrgiannis were very capable assistants at Bow Lake in 1994, and they also taught me how to extract organochlorines from water samples.

Derek G. C. Muir shared his knowledge and always found the time in a busy schedule to discuss any aspect of contaminants in the environment and to comment on draft manuscripts. Derek also provided laboratory space and equipment at the Freshwater Institute (Department of Fisheries and Oceans). Norbert (Bert) P. Grift, Bruno Rosenberg, Dan Savoie, and Gary Stern taught me all about organochlorine analyses. reading chromatographs, and other important chemistry skills. Ray Hesslein and Bev Ross provided stable isotope analyses in their lab at FWI. Aaron Fisk discussed science, commiserated about the vagaries of organochlorine research, and also reviewed Chapter II of this thesis. Thanks to all the people at the FWI who gave encouragement and shared good times during those Winnipeg winters.

Karen A. Kidd offered patient encouragement, assisted with initial field planning, taught me the fundamentals of organochlorine and stable isotope analyses, and in general, was a very good "hands-on" supervisor. Her reviews of Chapter II and III were very helpful. Her time and effort will always be gratefully appreciated.

David W. Schindler, my supervisor, set high standards, and expected good science. He also reviewed all chapters in this thesis. In trying to meet these expectations, I learned much about quality work, science, and myself. Brian Parker was invaluable in helping me to fill in the gaps in my field work preparations, and always was willing to answer my questions on everything from aging fish to zooplankton nets. He also contributed valuable comments on my manuscripts. Margaret Foxcroft did much behind-the-scenes work that kept things running smoothly.

Al Shostak identified the parasitic cysts on the lake trout stomachs, and explained their life cycles in mountain lakes. Peter Aku aged the fish otoliths at Glenora Fisheries Station. Michel Agbeti showed me how to identify many of the algae species found in the Bow Lake samples.

Many people volunteered as field assistants at Bow Lake. Thanks to Margaret Campbell, John Clare, Jennifer Heibert, Scott McNaught, and Frank Wilhelm. Their help ensured the success of this project. Also, Frank suggested the bright idea of using burlap sacking for collecting elusive benthic invertebrates.

Robert (Rob) J. Thacker assisted me at Bow Lake in all sorts of weather including thick snow, howling winds, heavy rain, and occasional sunshine, and on the way, discovered a deep love for the mountains. Last, but not least, Rob and my family, Margaret, John and Ian, provided moral support and constant encouragement through thick and thin.

TABLE OF CONTENTS

ABSTRACT
PREFACE
ACKNOWLEDGEMENTS
LIST OF TABLES
LIST OF FIGURES
LIST OF ABBREVIATIONS
I. GENERAL INTRODUCTION Introduction
II. FOOD WEB STRUCTURE AND ORGANOCHLORINE CONCENTRATIONS IN SUB-ALPINE BOW LAKE (BANFF NATIONAL PARK). Introduction
Methods
2. Field methods
3. Laboratory methods
4. Data analysis
Results
2. Organochlorines
Discussion
Conclusion
Literature Cited 50

III. STABLE ISOTOPE RATIOS (δ^{13} C AND δ^{15} N) IN FISH MUSCLE AND FIN RAY TISSUE: POTENTIAL FOR NON-LETHAL SAMPLING OF FISH.
Introduction
Methods
Results
Discussion
Literature Cited
IV. GENERAL DISCUSSION .86 Literature Cited .93
APPENDIX 1. BOW LAKE DATA SET (1994-95)

LIST OF TABLES

Table I.1. Log K _{ow} of different organochlorine compounds
Table I.2. Physical and chemical parameters for Bow Lake, Banff National Park26
Table II.1. Summary data for Bow Lake biota
Table II.2. Bow Lake fish stomach contents
Table II.3. Summary data for organochlorines in Bow Lake biota
Table II.4. Nonparametric Spearman's correlation coefficients (r _s) for physical parameters of Bow Lake fish
Table II.5. Spearman's correlation coefficients (r _s) for stable isotope ratios, %lipids, age, and weight against organochlorine concentrations in lake trout and mountain whitefish from Bow Lake, Banff National Park 64
Table II.6. Organochlorine concentrations in Bow Lake water and sediments 65
Table III.1. The average δ ¹³ C and δ ¹⁵ N of fin rays and muscle tissue of lake trout, mountain whitefish and lake whitefish from the Yukon Territory and Bow Lake, Banff National Park
Table III.2. Regressions for $\delta^{13}C$ of muscle and fin ray tissue
Table III 3 Regressions for δ ¹⁵ N of muscle and fin ray tissue. 83

LIST OF FIGURES

Figure I.1.	Map of Bow Lake and area	27
Figure I.2.	Thermal stratification in Bow Lake	28
Figure II.1.	Stable isotope "map" of the Bow Lake food web	56
Figure II.2.	Lipid content in Bow Lake biota vs $\delta^{15}N$	57
Figure II.3.	Lipid content in Bow Lake biota vs. δ^{13} C	58
Figure II.4.	Organochlorine concentrations in Bow Lake biota vs. % lipid content .6	59
Figure II.5.	Organochlorine concentrations in Bow Lake biota vs. $\delta^{13}C$ values 7	70
Figure II.6.	Organochlorine concentrations in Bow Lake biota vs. $\delta^{15}N$ values 7	7 1
Figure III.1	. $\delta^{13}C$ of fin ray tissue vs. muscle tissue	34
Figure III.2	. δ ¹⁵ N of fin ray tissue vs. muscle tissue	35

LIST OF ABBREVIATIONS

 Σ CHB: Total chlorinated bornanes (commonly called toxaphene).

DDT: Total dichlorodiphenyl trichloroethane.

 Σ PCB: Total polychlorinated biphenyls.

 Σ CHL: Total chlordane.

 Σ HCH: Total hexachlorocyclohexane.

OCN: Octachloronaphthalene. This is an internal standard used to monitor efficiency of organochlorine extractions.

DCM: Dichloromethane or methylene chloride. This is an organic solvent commonly used in organochlorine extractions.

δ¹⁵N: Delta nitrogen-15. The ratio of ¹⁵N to ¹⁴N in sample versus that in standard. Expressed in parts per mil (parts per thousand, %)

δ¹³C: Delta carbon-13. The ratio of ¹³C to ¹²C in sample versus that in standard. Expressed in parts per mil (parts per thousand, %)

K_{ow}: Octanol-water partition coefficient. Usually expressed as log values. Log K_{ow} values are a surrogate measure of how well an organochlorine compound dissolves in lipids versus water.

GC-ECD: Gas chromatograph with an electron capture detector.

LIST OF ABBREVIATIONS (continued)

GC-MS: Gas chromatograph with a mass spectrometer. In this thesis, this abbreviation refers to a high-resolution gas chromatography electron capture negative high resolution mass spectrometry (HRGC-ECNI-HRMS).

FWI: Freshwater Institute, Department of Fisheries and Oceans, Winnipeg, MB.

T2: An octochlorobornane toxaphene congener commonly found in biological samples

(2-exo, 3-endo, 5-exo, 6-endo, 8, 8, 10, 10-octachlorobornane).

T12: A nonachlorobornane toxaphene congener commonly found in biological samples (2-exo, 3-endo, 5-exo, 6-endo, 8, 8, 9, 10, 10-nonachlorobornane).

I. GENERAL INTRODUCTION

Atmospheric transportation of organochlorine compounds in the northern hemisphere is a widespread concern. Researchers have found organochlorine contamination in aquatic ecosystems with little direct exposure to pesticides and insecticides (Schroeder and Lane, 1988). Work in Sweden (Larsson et al., 1992), Russia (Kucklick et al., 1993), the Canadian Arctic (Lockhart et al., 1992; Kidd et al., 1995a,b), and the Great Lakes (Oliver and Niimi, 1988; Evans et al., 1991) has revealed unexpectedly high concentrations of organochlorines in fish and other aquatic biota. The Rocky Mountain National Parks in Alberta and British Columbia are not exempt from widespread organochlorine contamination. In a survey carried out in the Canadian Rocky Mountains to investigate the presence of organochlorines in lake trout and water (Donald et al., 1993), all fish sampled contained organochlorines. While lake trout from most lakes sampled had contaminant concentrations below Health Canada's guidelines for human consumption, fish from some lakes, including Bow Lake in Banff National Park, had much higher concentrations than other lakes. There was no record of any direct addition of organochlorines to Bow Lake, providing an intriguing mystery as to why organochlorine concentrations in lake trout are so high.

In an earlier study, Kidd et al. (1995a,b) found that higher than usual concentrations of organochlorines in lake trout from Lake Laberge, Yukon Territory, could be explained by very long food chains. I hypothesized that similar food chain differences could account for the high organochlorine concentrations in lake trout from Bow Lake. The purpose of this study is to investigate the role of the Bow Lake food web structure in organochlorine bioaccumulation in fish. The overall objectives are: (1) to determine food web linkages in Bow Lake by using stable carbon and nitrogen isotope ratios; (2) to assess food web structure to see if some unique feature

is leading to organochlorine bioaccumulation; and (3) investigate the use of fin ray tissue for predicting the stable isotope values of fish.

Organochlorines - what are they?

Organochlorines are chlorinated hydrocarbons which have been used as pesticides (e.g. DDT, chlordane and toxaphene) and industrial chemicals (e.g. PCBs).

Organochlorine pesticides can be acutely toxic to insects, fungi and other pests, they also lead to chronic toxicity in larger animals, including humans. Organochlorines are usually highly resistant to biological or abiotic degradation (Howard, 1991), are easily transported (Voldner and Li, 1995), and can be adsorbed on sediment and other particulate matter in water (Knezovich et al., 1987). Organochlorine compounds are highly soluble in lipids, tending to accumulate in the fatty tissues of biota. Because of these properties, organochlorine compounds can linger in the environment for long periods, and reach high concentrations in animal lipid tissue.

The tendency of different organochlorines to dissolve in fatty tissues has been found to be predictable from octanol-water coefficients (K_{ow}) of these organochlorines (Thomann, 1989), which are a surrogate measure of how well organochlorines dissolve in lipids versus water (Thomann, 1989). Log K_{ow} values are obtained by adding a specific organochlorine congener to a well-shaken flask of octanol and water, and measuring the proportions of the compound in water and octanol, although more accurate generator column methods that duplicate "shaken flask methods" exist (Hawker and Connell, 1988). Usually, the higher the log K_{ow} , the more soluble the organochlorine is in lipids, and the higher the potential for bioaccumulation in aquatic organisms (Thomann, 1989). Log K_{ow} is one of the most common measurements used in forecasting the environmental fate of contaminants.

For this study I focused on toxaphene (chlorinated bornanes), ΣDDT (total dichlorodiphenyl trichloroethane), and ΣPCB (total polychlorinated biphenyls), because these compounds are the most common organochlorines detected in all environmental compartments (i.e., fish and other organisms, water, sediment) of Bow Lake. ΣCHL (total chlordane), dieldrin and ΣHCH (total hexachlorocyclohexanes) were also investigated because these compounds are persistent and were detected at low concentrations in every sample from Bow Lake.

Toxaphene¹ (CHB), a common name for a mixture of chlorinated bornanes and camphenes, was used for a wide range of pesticide applications before its ban in 1982 in the USA and its legal restriction in 1980 in Canada (Saleh, 1991). When DDT was banned in North America in the late 1960's and early 1970's, toxaphene replaced DDT as a major agricultural pesticide, particularly for cotton crops in the southeastern USA (Saleh, 1991). Toxaphene was also used in fishery management programs as a way of getting rid of "undesirable" fish species in lakes before restocking with fish species popular with sport anglers (Miskimmin and Schindler, 1994). Fish can accumulate toxaphene to high concentrations in lipid-rich tissues (Saleh, 1991). Proportions of toxaphene congeners in biota are different from those of technical toxaphene, for a few individual congeners such as T2 and T12 are bioaccumulated to a greater extent in aquatic biota than other congeners (Stern et al., 1992).

PCBs are resistant to breakdown via oxidation and reduction, and are extremely stable. These properties are the reason PCBs have been widely used in industrial

¹ Because toxaphene was quantified by using a single response factor based on 24 peaks in the standard, and is not the sum of congeners individually quantified, I use'toxaphene' throughout this thesis instead of 'ΣCHB'.

applications such as plasticizers, fire retardants, and dielectric fluids since the 1930's. Due to growing health concerns, the use of PCBs gradually became restricted since 1970-80's (World Health Organization, 1993). There are 209 PCB congeners that vary by their chlorine content and the position of chlorine atoms in the molecule. These factors strongly affect toxicity and bioaccumulation potential of individual congeners. The toxicity of PCBs to aquatic organisms is variable, but usually the most toxic PCBs, in terms of dioxin-like effects, are the 3, 4, 3',4'-substituted (i.e. non-ortho) PCBs. Exposure to low concentrations of PCBs often leads to impaired development and reproduction in fishes (WHO, 1993). Lower chlorinated PCBs are eliminated from biota rapidly, leading to an increase in average chlorine content of PCBs with trophic level (Oliver and Niimi, 1988; Evans et al., 1991).

DDT and its dehydrochlorination breakdown products, DDD (dichlorodiphenyl dichloroethane) and DDE (dichlorodiphenyl dichloroethene) remain ubiquitous in the environment despite being banned in late 1960s and early 1970s in most countries. DDT was used as a broad-application pesticide, as it acts as a nerve toxin on invertebrates. It was once thought to be harmless to vertebrates, until sublethal reproductive failure in birds and fish was discovered to be the result of exposure to DDT (Weseloh and Temple, 1983; Jarvinen et al., 1977). *p,p'*-DDE is often the predominant congener in aquatic organisms, and can bioaccumulate to high concentrations in aquatic food webs (Evans et al., 1991). Σ DDT concentrations in aquatic organisms have decreased gradually in recent years (World Health Organization, 1989a).

Dieldrin is a single chemical compound (Madenjian et al., 1993) and is readily converted from aldrin in the environment (Dynamic Corporation, 1989). Aldrin and dieldrin were used to control soil insects in agricultural programs, and for tsetse fly

control in the tropics (World Health Organization, 1989b). These compounds act as both a contact and a stomach toxin for insects (WHO, 1989b). Most uses were banned in 1975 (Dynamic Corp., 1989). However, dieldrin is a persistent compound for it sorbs tightly to soil and leaches into water at very low rates. Dieldrin can be highly toxic to fish and aquatic insect larvae (WHO, 1989b), but the bioaccumulation pathways are usually from water, not food. Aquatic invertebrates exposed to dieldrin in the laboratory excrete it rapidly when transported to clean water (WHO, 1989b). Lake trout can excrete dieldrin at a faster rate than DDT (Madenjian, 1993). However, fish-eating birds were found to be contaminated with dieldrin in the 1970's to 1980's (WHO, 1989b), which indicates that biomagnification potential exists for this compound.

Chlordane (Σ CHL) congeners found in aquatic environments are derived from technical chlordane, which is a mixture of more than 140 compounds, including α , β , and γ -chlordane, and heptachlor (Dearth and Hites, 1991). It was used as a lawn and garden pesticide before its restriction in 1979, and after that year, it was used to control termites by applying to soil underneath buildings (Syracuse Research Corporation, 1989). Since 1988 the use of technical chlordane has been voluntarily suspended (Dearth and Hites, 1991). Chlordane sorbs to sediment, can revolatilize in water, and does not degrade rapidly (Syracuse Research Corp., 1989). Low amounts of chlordane have been found in freshwater fish across USA and in the Great Lakes area (Syracuse Research Corp., 1989), and in sub-Arctic and Arctic regions (Muir et al., 1988; Muir et al., 1995; Kidd, 1996).

Hexachlorocyclohexane (Σ HCH) has 8 isomers, including the common isomers, α , β , δ , and γ -HCHs. Technical HCH was formerly made as an insecticide, but it is no longer manufactured in North America (Clement Associates, 1989). Lindane,

purified γ -HCH, is still in use as a broad spectrum insecticide, especially in combination with fertilizer, and as a topical medication to remove ectoparasites (lice and mites) from humans and animals (World Health Organization, 1991). γ -HCH is degraded by cyanobacteria in aquatic environments (Clement Associates, 1989; WHO, 1991). The toxicities of Σ HCH and of γ -HCH to aquatic organisms are moderate, with uptake being more rapid from water than from food (World Health Organization, 1992). Chronic toxicity includes behaviour changes such as "irritability" in fishes, and reproductive effects in snails and *Daphnia* (WHO, 1992). Often aquatic organisms, including fish, will bioaccumulate Σ HCHs until a plateau is reached, after which Σ HCH usually does not increase in the tissues (WHO, 1991; 1992). As expected from its low log K_{ow} values (Table I.1), bioaccumulation of Σ HCH in biota is less pronounced than for other organochlorine compounds described above.

Bioaccumulation of organochlorines

There are three terms used to describe organochlorine movement in food webs and biota: "Bioaccumulation" is a general term used to describe the increase in contaminant concentrations from lower to higher trophic levels, and may include any pathway of contaminant intake; "Biomagnification" is the increase of chemical compounds, by trophic transfer of food through one or more steps; "Bioconcentration" is the direct transfer of compounds from water to biota, and can play an important role in organochlorine bioaccumulation (Leblanc, 1995).

Bioconcentration is most common in small organisms such as algae and zooplankton, because their large surface-to-volume ratio allows for quicker bioaccumulation of organochlorines from the surrounding water (Swackhamer and Skoglund, 1993: Leblanc, 1995).

The most important variables in organochlorine bioaccumulation in larger organisms are trophic structure, dietary composition, and lipid content of biota (Rowan and Rasmussen, 1992). Direct correlations have been found for increased bioaccumulation of organochlorines with trophic level (Rasmussen et al., 1990; Cabana and Rasmussen, 1994; Cabana et al., 1994; Kidd et al., 1995a). Dietary composition can be important. For example, Lake Ontario amphipods have disproportionately high PCB concentrations due to their feeding patterns and high lipid content, and the fish feeding on them bioaccumulate more PCBs than fish feeding on other prey (Evans et al., 1991). Different species of fish can bioaccumulate organochlorines to different concentrations, despite apparently similar exposure concentrations (Suns and Hitchin, 1992). Rasmussen et al. (1990) found that the top predators in a longer food web with more trophic levels concentrated contaminants to a higher magnitude than the same species in shorter food webs with fewer trophic levels.

Environmental and physical parameters can affect how organochlorines are bioaccumulated. For example, older lake trout that have been exposed to ambient environmental concentrations over a longer period often have higher contaminant concentrations in their tissues than younger lake trout (Gutenmann et al., 1992). However, larger mosquitofish (*Gambusia affinis*) in experimental studies may take up less organochlorines per unit weight than for smaller mosquitofish over a short exposure period (48 hours), so size may affect uptake rates (Murphy, 1971). It is apparent that age (in terms of exposure length), size and growth rates are important for organochlorine bioaccumulation in fish. The rate of organochlorine uptake in fish may be slower with decreasing temperature (Reinert et al., 1974), so fish in colder waters may bioaccumulate less organochlorines than these in warmer waters. Water chemistry parameters such as salinity (Murphy, 1970) or water hardness (Johnson et

al., 1988) can affect organochlorine availability and the uptake of contaminants in biota. Organochlorines can bind to dissolved and particulate organic matter such as humic acids, DOC and POM, so contaminants may be less available in eutrophic lakes (McCarthy and Bartell, 1988), which leads to decreased uptake in fish and other biota (Carter and Suffet, 1982; Larsson et al., 1992). Conversely, in oligotrophic lakes, organochlorine exposure may be greater.

Determining food web structure with stable isotope ratios

Stable isotopes are naturally-occurring atoms with the same atomic number (i.e., number of protons). However, isotopes can have varying atomic masses with different number of neutrons in the nucleus. Atomic charges are not affected to a large degree, but kinetic energies of different isotopes of the same element can vary. The different isotopes of an element, such as carbon, usually are found at constant ratios, but these ratios can be affected by natural processes (Ehleringer and Rundel, 1988). In biological studies, nitrogen and carbon stable isotope ratios exhibit predictable and useful patterns (described below) that provide an integrated measurement of animal's trophic level and its dietary patterns, respectively (Estep and Vigg, 1985; Peterson and Fry, 1987; Hesslein et al., 1991). These stable isotopes can be used to describe the food web structure, with stomach contents providing taxonomic detail of fish diets. However, stomach contents do not always accurately reflect the long-term diet of fish, while stable carbon and nitrogen isotope ratios in fish incorporate the isotopic values of their diets over time. In addition, there is a large degree of error in relying heavily on stomach contents because stomach contents can be digested beyond identification or stomachs can be empty.

One useful measurement is the ratio of 13 C to 12 C in environmental samples (δ^{13} C). Planktonic algae, benthic algae and detritus all have different δ^{13} C signals because

the plants process carbon differently as part of their photosynthetic pathways (O'Leary et al., 1992; Hecky and Hesslein, 1995). Carbon isotopes fractionate at low rates (Rounick and Winterbourn, 1986) and the δ^{13} C of organisms reflects that of the original plant or other original carbon food source. For example, organic carbon in lakes can be produced in situ, or originate in the terrestrial catchment, leading to distinctive variation in δ^{13} C ratios (Wissmar et al., 1977; Rau and Anderson, 1981; France, 1995). Algae preferably take in ¹²C which is freely available in the atmosphere. Pelagic algae nearer the water-air interface are usually isotopically lighter (i.e. have lower δ^{13} C values) than benthic algae below the unstirred boundary layer (Hecky and Hesslein, 1995), so it is often possible to discriminate between pelagic-feeding and littoral (or benthic)-feeding organisms and the energy pathways based on their δ^{13} C values. Zooplankton that exclusively feed on planktonic algae will incorporate the algae's δ^{13} C signature, and the δ^{13} C of their tissue will be "lighter" (more isotopically depleted) than that of benthic invertebrates feeding on littoral plant sources. Similarly, the δ^{13} C signature for fish that feed on zooplankton will be lighter than that of other fish that feed on benthic invertebrates. Thus, stable carbon isotope ratios indicate the carbon pathways within a food web, and the carbon sources of individual organisms. The source, movement and dynamics of organochlorine contaminants in the pelagic and benthic portions of a food web can be investigated using δ^{13} C measurements to discriminate between carbon sources.

The ratios of ^{15}N to ^{14}N ($\delta^{15}N$) in environmental samples are useful in other ways. Because all animals selectively excrete ^{14}N and retain ^{15}N in their tissue there is an increase of 3 to 4 ‰ per trophic level in $\delta^{15}N$ values (Peterson and Fry, 1987). Thus, $\delta^{15}N$ can be used to estimate trophic level to compare with results based on stomach contents. In fish and aquatic food webs, significant positive correlations between the biomagnification of organochlorines and trophic position as measured by $\delta^{15}N$ have

been found (Kidd et al., 1995a; 1995b; Kiriluk et al., 1996; Kucklick et al., 1996). In Yukon lakes, trophic position of fish, as measured by $\delta^{15}N$, was a more significant predictor of organochlorine bioaccumulation than lipid content for compounds with higher log K_{ow} values (Kidd et al., 1996).

Mountain aquatic environments

Animals and plants in the alpine and sub-alpine regions of Canadian Rocky

Mountains endure a harsh cold climate with a short growing period from mid-June to

September. Lakes freeze early (September - October) and the winters can last for 8-9

months. In the summer, water surface temperatures only rise above 10°C on

particularly warm summer days (personal data; D. W. Schindler, unpublished data,

Department of Biological Sciences, University of Alberta, Edmonton AB, T6G 2E9).

Only aquatic organisms adapted to cold environments and with lifecycles and

physiologies suited to long periods under ice will thrive in this environment.

The aquatic communities of mountain lakes tend to be simple, and there is a low diversity of aquatic benthic invertebrate species. For example, there have been only 17 Plecoptera species identified in 58 lakes across the Rocky Mountain National Parks area, and of these, four species were found only once (Donald and Anderson, 1980). Zooplankton are marginally more diverse with more than 50 species identified, including 20 from the family Diaptomidae (Copepoda) alone (Anderson, 1974). However, only a few zooplankton species are common throughout the mountains, including the ubiquitous calanoid copepod, *Hesperodiaptomus arcticus*, which is a keystone predator in many small fishless lakes (Paul and Schindler, 1994), but can coexist with fish in large lakes (Donald et al., 1994).

Because of the cold harsh environments and sparse prey, only hardy coldwater fish are found in the lakes of the Rocky Mountains, and very few species are endemic to the region (Donald, 1987). Stocking lakes with popular sport fish such as lake trout (Salvelinus namaycush) in the mountains has been carried out for at least eight decades up to 1988, when Parks Canada stopped fish stocking (Schindler and Pacas, 1996). In larger lakes, trout species often co-exist with other fish, such as mountain whitefish (Prosopium williamsoni), which have different feeding niches.

Organochlorines in mountain environments

Even in protected areas such as the Rocky Mountain National Parks, many organochlorine compounds can be detected in aquatic biota long after these compounds have been restricted or banned in North America. Organochlorine presence and concentrations in the mountain parks have been studied only recently (Donald et al, 1993; Chapter II, this thesis; J. Blais, unpublished data, Department of Biological Sciences, University of Alberta, Edmonton, Alberta, T6G-2E9; D. Donald, unpublished data, Environment Canada, 2365 Albert Street, Regina, SK, S4P-4K1), but it is evident that organochlorines in aquatic mountain environments are prevalent and persistent. The only possible source of these ubiquitous contaminants in such a remote area is by atmospheric long-range transport followed by deposition with rain, snow, dry fallout or gas exchange². Backtracking air masses for 5 days from above Bow Lake, Banff National Park, using an Environment Canada trajectory model, Donald et al. (*in prep.*) showed that most of the time, air masses moved from over the Pacific and Arctic Oceans (60%), and the United States (21%). It has been estimated

² Gas exchange is a two-way exchange process between the atmosphere and the surface. It can occur in snow packs, ice and water. The more volatile compounds (i.e. HCH compounds) are most affected by gas exchange (Barrie et al., 1997).

Dry fallout is the atmospheric transport of dry particles with sorbed organochlorine compounds. Less volatile compounds (i.e. CHB, CHL and DDT) that are more easily particle bound may be most affected by dry fallout.

that some Asian countries account for about 20% of total ∑DDT (including India, Russia, and China) and total ∑HCH (including India and China) world usage, while the United States accounts for at least 20% of total world toxaphene usage, particularly in the cotton growing areas (Voldner and Li, 1995). In other words, Bow Lake may be contaminated by precipitation and gas exchange from air masses passing over distant countries that use large amounts of organochlorines.

Atmospheric organochlorines can be in the form of vapour, absorbed on particulate matter, or can be "scavenged" by snow or rain (Cotham and Bidleman, 1991). Organochlorines in the vapour phase can be exchanged between surfaces (i.e. water. soil or snow) and the atmosphere, and may be the most important process (Hoff et al., 1996). Dry deposition (particulate matter), wet deposition (contaminated precipitation) and gas exchange are sources of organochlorines in the northern hemisphere (Barrie et al, 1997). Low temperatures can lead to increased deposition of organochlorines from air, and reduce the revolatilization of organochlorines. "Cold condensation" is hypothesized to occur when temperature-dependent fractionating of atmospheric organochlorine compounds with low vapour pressure leads to increased net atmospheric deposition of organochlorine compounds in cold areas such as the Arctic (Wania and Mackay, 1993). The cold condensation effect at colder high latitudes has been incorporated into a global fractionation model (Wania and Mackay, 1993; 1995), and supported by data obtained from cores and fish collected from a series of temperate to Arctic lakes in Canada (Muir et al., 1990; Muir et al., 1996). Globally, analyses of tree bark samples from around the world revealed that the more volatile organochlorine compounds, such as α - and γ -HCH, increased with latitude, while less volatile compounds such as endosulfan tended to remain near local sources (Simonich and Hites, 1995), further supporting the cold condensation hypothesis. Some Arctic and sub-arctic lake ecosystems are so highly contaminated due to

atmospheric organochlorine inputs that some fisheries have been closed (Kidd et al., 1995b). Cold condensation may also be an important factor in the cold environments of mountains, particularly on glaciers at higher altitudes (Schindler et al., 1995). Because of increased precipitation and colder temperatures at higher elevations (J. Blais and D. W. Schindler, unpublished data, Department of Biological Sciences, University of Alberta, Edmonton AB, T6G 2E9), it is likely that atmospheric deposits increase with elevation, so high-elevation alpine lakes may be receiving more organochlorines than montane lakes at lower elevations. Toxaphene concentrations in fish collected from a series of lakes from prairie to sub-alpine environments (including Bow Lake) increased with elevation (Donald et al., *in prep.*).

Bow Lake

Bow Lake is a deep single-basin sub-alpine lake in northern Banff National Park (51°45'N, 116°30'W; Figure I.1, Table I.2). It receives most of its water from Bow Glacier, a small outlet glacier of Wapta Icefields that straddles the Continental Divide Ranges. A small discharge stream enters the north shore (near the lodge) of Bow Lake, but the stream's contribution to Bow Lake is minor (Smith, 1981; Kennedy, 1975). Bow Lake drains out of a single outflow to the Bow River, which then flows south-east to the Banff National Park boundary, and then eventually to the Saskatchewan River system. The deepest part of the lake (50 m) is located near the glacial stream inflow; the lake gradually becomes shallower (10 - 20 m) towards the outflow, with a trough-like central area that is slightly deeper (20 - 30 m; Smith and Syvitski, 1982). The geological features of Bow Lake include steep talus and debris slopes on mountains surrounding the lake and a large alluvial fan at the glacial inflow of the lake (Kennedy, 1975; personal observation). The local bedrock consists of Precambrian and Cambrian argillites and quartzites on lower slopes overlain by Lower Paleozoic carbonates where most erosion occurs (Smith et al., 1982). The

inflowing glacial stream supplies mostly carbonate clastic detris, while suspended sediment entering the lake are dominantly dolomite with lesser amounts of calcite, quartz, mica, chlorite and feldspar (Smith and Syvitski, 1982. The inflow discharge is highly seasonal, with nearly all flow occurring during summer (late May to late September). On sites mesic enough to support vegetation, the plant community consists mostly of Engelmann spruce (*Picea engelmannii*) on the west shore, and shrubby willow (*Salix* spp.), and cinquefoil (*Pontentille frutescente*) on the remainder of the lake shores (personal observation). The Icefields Parkway, a major tourist thoroughfare, runs along the eastern side of Bow Lake, and other developments in the area include a lodge, parking lots and a picnic area.

The limnology and the ecology of Bow Lake have not changed much since 1931-40's (Rawson, 1942), except for secchi depth. The secchi depth has increased from 0.4-1.5 m in 1941 (Rawson, 1942) to 4 m in 1995 (personal data). The change in light transmissivity has been attributed to the decrease in sedimentation to the lake since the formation of a sediment-trapping pond at the glacier terminus in 1955 (Smith et al., 1981).). In August 1973, the average daily sedimentation rate in Bow Lake decreased from from 0.91 to 0.25 mg cm⁻² day⁻¹ (Kennedy, 1975), leading to an average of 5-10 mm varve thickness in cores after 1955 (Smith, 1981). Thermal stratification in Bow Lake is weak (Figure I.2), with midsummer surface temperatures reaching 12°C (Table I.2). Phosphorus and nitrogen concentrations in the water are low, with total dissolved nitrogen concentrations (83.9 \pm 5.2 μ g L⁻¹) an order of magnitude higher than total dissolved phosphorus (2.2 \pm 1.2 μ g L⁻¹; Table I.2). Both dissolved organic carbon (0.46 \pm 0.1 mg L⁻¹) and sediment organic carbon content (3.1%) are low in Bow Lake (Table I.2). Chlorophyll-*a* concentrations are also low (0.31 \pm 0.2 μ g L⁻¹; Table I.2).

Bow Lake is ultra-oligotrophic with sparse biota. Benthic invertebrates in the littoral zone include Gammarus lacustris, Diptera (Chronomidae and Tipulidae), Trichoptera, Ephemeroptera, Oligochaeta, and Hydracarina. Gastropoda (Lymnaeidae) are common on rocks along the east shore. Numbers of Hesperodiaptomus arcticus are moderately high (1.7 individuals per litre) with some Daphnia middendorffiana (0.03 indiv. L⁻¹)³. Rotifer species, Kellicottia longspina and Polyarthra vulgaris (0 to 0.15 indiv. L⁻¹, varying each year) are present but rare. Two species of cyclopoid copepods exist, but have been found in separate years, with Acanthocyclops vernalis in 1995 (0.01 indiv. L⁻¹) and Diacyclops thomasi in 1994 (0.02 indiv. L⁻¹). Other zooplankton species that have been found in Bow Lake include Alona gutta, Alonella excisa, Chydrous sphaericus, Eucyclops agilis, Eucylops speratus and Canthocamptus oregonenisis, all at densities less than 0.1 indiv. L⁻¹ (Anderson, 1974). Planktonic green algae (e.g., Monoraphidium sp.) are sparse. Diatoms of the genera Navicula, Eunotia, and Cyclotella are abundant. Some cyanobacteria (Cylindrospermum sp.) are present. There are no aquatic macrophytes in the littoral zone.

Originally, Bow Lake contained bull trout (Salvelinus confluentus) and mountain whitefish (Prosopium williamsoni; Donald and Alger, 1993). Cutthroat (Oncorhynchus clarki) and rainbow trout (Oncorhynchus mykiss) were introduced to Bow Lake in 1934-47, but numbers had sharply declined by 1958 (Donald and Stelfox, 1997), with only a few cutthroat trout left. In 1964-1967, Bow Lake was stocked with 24,000 lake trout (Salvelinus namaycush; Donald and Alger, 1993), and

³ Quantitative zooplankton counts (1994 and 1995) from D. Donald, Environment Canada, 2365 Albert Street, Regina, SK, S4P-4K1 (unpublished data).

by 1984, the native bull trout and the cutthroat trout were extirpated (Donald, 1987). Now, only two fish species co-exist in Bow Lake, lake trout and mountain whitefish⁴.

⁴ Personal note: A fish that appeared to be a rainbow or a rainbow hybrid (lateral pink streaks along sides) was found among captured fish from a large fish trap set on August 1994.

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Table I.1. List of log Kow (octanol-water partition coefficients) values for study organochlorines. In general, the higher the log Kow value is, the more soluble the compound is in lipid tissues, and the less soluble it is in water. There is a wide range of log Kow values for PCB congeners, so only a few common PCB congeners found in biota samples are listed here.

Organochlorine	Common	Average	
compound	name	chemical formula	Log Kow
CHB	Toxaphene	$C_{10}H_{10}Cl_8$	6.44 ^a
Σ DDT		C ₁₄ H ₈ Cl ₄	5.75 ^b
Σ DDE		C ₁₄ H ₈ Cl ₄	5.69 ^b
Σ CHL	Chlordane	$C_{10}H_6Cl_8$	6.00 ^b
Dieldrin	Dieldrin	C ₁₂ H ₈ Cl ₆ O	5.48 ^b
∑PCB	Aroclor *	(generic)	6.5 c
118		$C_{12}H_9Cl_5$	6.74 ^d
149		C ₁₂ H ₉ Cl ₆	6.67 ^d
153		C ₁₂ H ₉ Cl ₆	6.92 ^d
180		C ₁₂ H ₉ Cl ₇	7.36 ^d
∑HCH		C ₆ H ₆ Cl ₆	
α-НСН		n	3.82e
β-НСН		n	3.80e
ү-НСН	Lindane	"	3.2-3.7 ^f

^{*} Aroclor is one of many brand names for commercial PCB mixtures.

^a Saleh, 1991

b as cited in Mackay, 1982

c as cited in Mackay, 1982
c as cited in Thomann, 1989
d Hawker and Connell, 1988
e as cited in World Health Organization, 1992
f as cited in World Health Organization, 1991

<u>Table I.2</u>. Physical and chemical parameters for Bow Lake, Banff National Park.

Physical		Chemical	`````````````````````````````````````
Parameters		Parameters	
Longitude	116°31'W	DOC 1	0.46 ± 0.12 mg L ⁻¹
Latitude	51°45'N	TP ¹	$4.6 \pm 1.3 \mu g L^{-1}$
Area	2.8 km ²	TDP ¹	$2.2 \pm 1.2 \ \mu g \ L^{-1}$
Volume	0.64 km^3	TDN I	$83.9 \pm 5.2 \ \mu g \ L^{-1}$
Maximum depth	51 m	NO ₃ + NO ₂ ¹	$46.4 \pm 2.6 \mu g L^{-1}$
Average depth	22.9 m	Chl-a ¹	$0.31 \pm 0.2 \ \mu g \ L^{-1}$
Elevation	1940 m	Conductivity ¹	$161.8 \pm 3.2 \ \mu S \ cm^{-1}$
Drainage area ²	70.5 km ² (31% glacial)	pH ¹	8.1 ± 0.06
Discharge rate ³	2.6 m ³ s ⁻¹ (0.5-5.9)	Turbidity 1	2.6 NTU
Susp. Sed. ⁴	91 mg L ⁻¹ (4-1193)	DIC δ ¹³ C	-5.86 ‰
Residence time ⁵	205 days	POM + PIM δ ¹³ C	-16.44 % (average)
Productivity	ultra-oligotrophic	POM δ ¹⁵ N ⁶	0.51 ‰
MSST ⁷	10 - 12°C	Sed. OC content 8	3.1 %

¹ The chemical analyses of water samples were done by the Limnology Laboratory, Department of Biological Sciences, University of Alberta, Edmonton, AB, T6G-2E9. Average values and standard deviations of samples collected in June, July and August, 1995 are indicated.

² Drainage area: The size of the drainage basin of glacial stream feeding the lake (Smith et al., 1980). The "% glacial" is the proportion of the drainage area that is covered by glaciers.

³ Discharge rate: the rate of inflow discharge of the principal inflowing stream, Bow Outwash (Smith et al., 1980). The daily average and the daily discharge range are indicated. Both principal inflowing streams are glacial.

⁴ Suspended sediment concentration: the concentration of suspended glacial sediments near the principal inflowing stream (Smith et al., 1980). The average and the daily range are given.

Sesidence times are the estimated period of turnover of the water during the ice-free period. These estimates may not be specific, because there is density-stratification which impedes complete turnover. Inflow and outflow sources tend to concentrate in certain areas of the water column. Bow Lake residence time is roughly estimated based on available data from Smith et al. (1980) and a flow rate estimate of the non-glacial stream entering Bow Lake.

 $^{^6}$ POM δ^{15} N was collected and analyzed independently (A. Hardie, personal data, Dept. Biological Sciences, University of Alberta, Edmonton, AB, T6G-2E9).

⁷ MSST: Mid-Summer Surface Temperature. Surface temperatures were taken in July '94 and July '95.

⁸ Sediment organic carbon content (Donald et al., in prep)

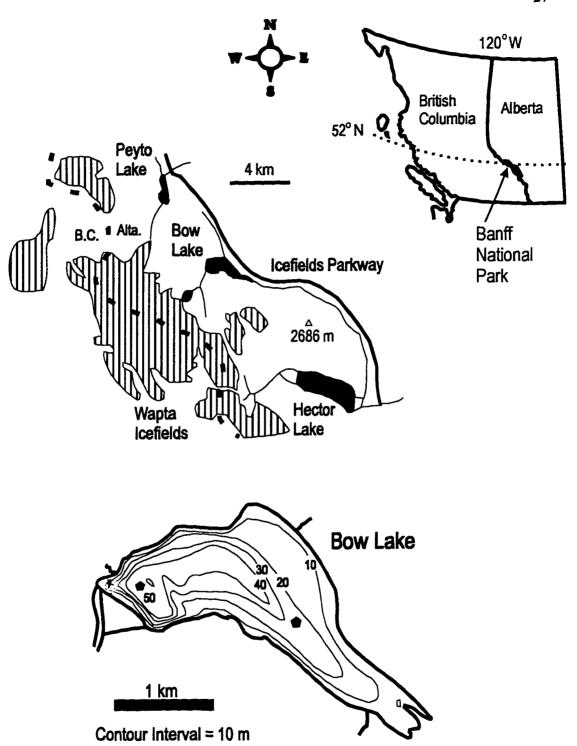


Figure I. 1. Map of Bow Lake and area. Shaded areas indicate the Wapta Icefields. The bottom map shows the contour outline of Bow Lake with marked sampling sites at the inflow and the centre of the lake. Reproduced with permission from Smith and Syvitski (1982).

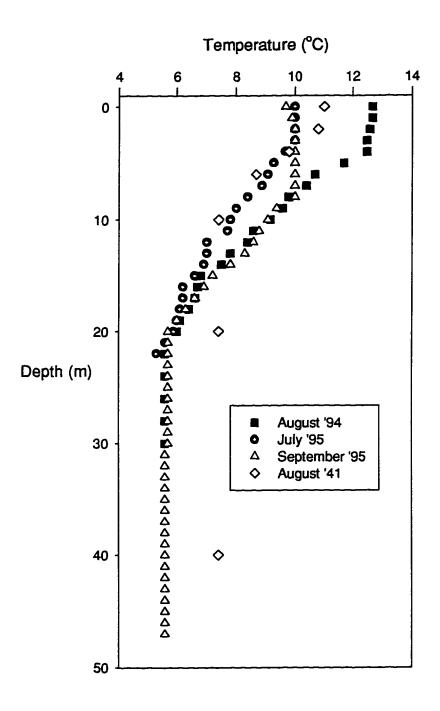


Figure I.2. The thermal stratification of Bow Lake (personal data). The 1941 values are from Rawson (1942).

II. FOOD WEB STRUCTURE AND ORGANOCHLORINE CONCENTRATIONS IN SUB-ALPINE BOW LAKE (BANFF NATIONAL PARK).

INTRODUCTION

Organochlorines in Bow Lake

In a survey done in 1991-92, organochlorine compounds (toxaphene, DDT, PCBs and other compounds) were detected in lake trout (*Salvelinus namaycush*) from 14 lakes across the Canadian Rocky Mountains (Donald et al., 1993). This caused increased concern about the presence of contaminants in the aquatic environments of the Rocky Mountain National Parks, particularly for lakes with elevated organochlorine concentrations. Bow Lake in Banff National Park is one of the more contaminated lakes, with concentrations of toxaphene (chlorobornanes, CHB) in lake trout from this lake being 10-20 times higher than in fish populations from nearby lakes (Donald et al., 1993). No direct sources could be discerned for Bow Lake (Chapter I, this thesis). Other organochlorines detected in lake trout from Bow Lake included dichlorodiphenyl trichloroethane (Σ DDT), and polychlorinated biphenyls (Σ PCB) at fairly high concentrations, and chlordane (Σ CHL), dieldrin, and hexachlorocyclohexane (Σ HCH) at low concentrations. In this study I investigated reasons for the higher organochlorine concentrations in fish from Bow Lake than in other nearby lakes.

In an earlier study of Lake Laberge, Yukon Territory, Kidd et al. (1995a,b) showed that unusually high concentrations of toxaphene and other contaminants in lake trout could be explained by longer underlying food chains. In contaminated ecosystems, there is often an increase in organochlorine concentrations with each trophic level in a food web (Oliver and Niimi, 1988; Evans et al., 1991). Top predators in Ontario food webs with longer underlying food chains have higher concentrations of PCBs than

fish from food webs with fewer trophic levels (Rasmussen et al., 1990). I hypothesized that a similar situation was happening in Bow Lake, leading to high concentrations of organochlorines in the lake trout.

Advantages of stable nitrogen and carbon isotope measurements

Characterizing trophic level and food web structure using dietary analyses of fish stomachs alone can be difficult, for contents of a stomach do not necessarily reflect long term diets. In addition, empty stomachs and highly digested contents present problems in identifying fish diets. While dietary analyses provide a taxonomic resolution of fish diets (Hecky and Hesslein, 1995), stable nitrogen (δ^{15} N) and carbon isotope ratio (δ^{13} C) measurement of biota have been invaluable in characterizing food web structure and trophic interactions (Peterson and Fry, 1987; Chapter I, this thesis). Furthermore, stable carbon and nitrogen isotope ratios integrate long-term dietary patterns, especially in cold-water fish which may have slow isotopic signature turnover rates (Hesslein et al., 1993).

Nitrogen isotopes consistently fractionate in organisms, as 14 N is selectively eliminated by organisms and 15 N incorporated in body tissues, so with each successive trophic level, δ^{15} N values in the tissue of biota increase. The average δ^{15} N difference between trophic level is 3.2 to 3.4‰ (Peterson and Fry, 1987; Wada et al., 1987; Fry, 1991; Kidd, 1996). Kidd et al. (1995a) demonstrated that contaminant uptake by fish and other organisms in Yukon aquatic food chains could be reliably predicted by using stable isotopes of nitrogen to measure trophic positioning. Similar positive significant relationships between organochlorines and stable nitrogen isotope ratios were observed in Lake Ontario (Kiriluk et al., 1996) and Lake Baikal (Kucklick et al., 1996).

In contrast, stable carbon isotopes fractionate very little in biota, with less than 1‰ enrichment in δ^{13} C per trophic level (Peterson and Fry, 1987). Because of low fractionation rates, the stable carbon isotope values of organisms reflect the average δ^{13} C of their diets. Therefore, the differences in δ^{13} C between pelagic and benthic invertebrates are passed up in the food chain, indicating the origin of organic carbon in higher trophic organisms (Hecky and Hesslein, 1995). The differences in δ^{13} C of biota can be exploited in conjunction with organochlorine measurements to determine the pathway of organochlorines into the base of the food web (Chapter 1, this thesis).

Specifically, the objectives of this study were: (1) to characterize the Bow Lake food web structure and interactions by examining trophic position as measured by $\delta^{15}N$, and carbon source as measured by $\delta^{13}C$ and (2) to examine the bioaccumulation, food chain pathways, and food web distribution of organochlorines (toxaphene, ΣDDT , ΣPCB , ΣCHL , ΣHCH , and dieldrin) in the Bow Lake food web by relating organochlorine concentrations in biota to $\delta^{15}N$, $\delta^{13}C$ and lipid concentrations. This study is the first of its kind to examine organochlorine concentrations in an aquatic food web in the Rocky Mountain National Parks using stable carbon and nitrogen isotope ratios.

METHODS

1. Study area

Bow Lake is an ultraoligotrophic sub-alpine lake located in the Bow Valley of Banff National Park (51°45'N, 116°30'W; Chapter I, this thesis). The Icefields Parkway, a major tourist thoroughfare, runs along the eastern side of Bow Lake. The majority of the inflow to Bow Lake comes from the Bow Glacier, a tongue of the Wapta Icefields that straddles the Continental Divide. The fish population consists of lake trout (Salvelinus namaycush) and mountain whitefish (Prosopium williamsoni). The

calonoid copepod *Hesperodiaptomus arcticus* is the most abundant zooplankter followed by *Daphnia middendorffiana*, and the cyclopoid copepods *Acanthocyclops vernalis* and *Diacyclops thomasi* (see Chapter I of this thesis). Rotifers (*Kellicottia longispina* and *Polyarthra vulgaris*) are present but rare (see Chapter I of this thesis). Benthic invertebrate taxa in Bow Lake include *Gammarus lacustris*, Trichoptera, Diptera (Chironomidae and Tipulidae), Oligochaeta, Gastropoda (Lymnaeidae), Plecoptera, Ephemeroptera and Hydracarina.

2. Field methods and fish stomach content analyses

Lake trout and mountain whitefish were captured using gill nets (June 1994) or fish traps (August 1994) set in the littoral zone. Otoliths were removed from fish and held in glycerin-alcohol solution. The ages of fish were later determined by counting annuli on otoliths using CSAS (Calcified Structure Analysis Software)⁵. Fish stomachs were removed upon capture and held separately. All fish and fish stomachs were wrapped in solvent-cleaned aluminum foil and stored on dry ice in the field. The fish and fish stomachs were then held in a -60°C freezer operated by Environment Canada in Calgary, Alberta, until analyzed as described in Laboratory Methods.

The fish stomachs were dissected and examined using a dissecting microscope at a later date, and the relative frequency of biota found in stomachs counted. Many stomachs contained highly digested sludge, and only organism parts were left in many cases, which precluded accurate weight estimates of the individual taxa in stomachs. Each occurrence of a taxon in a stomach was counted once, and the total number of stomachs containing that taxon was divided by the total counts of all taxa to obtain

⁵ Otoliths were analyzed by Dr. P. Aku at the Glenora Fisheries Station, Ontario.

the relative frequency of each taxon in stomachs for lake trout and mountain whitefish (Table II.2). Empty stomachs were excluded. Mountain whitefish fry, adult Ephemeroptera, and Plecoptera found in lake trout stomachs were included in stable isotope analyses because these organisms could not be collected in sufficient numbers at Bow Lake by other means. Data for lake trout stomach contents were also obtained from Environment Canada for the years of 1984, 1991 and 1993 (Table II.2; D. Donald, unpublished data, Environment Canada, 2365 Albert Street, Regina, SK, S4P-4K1).

More than 60% of lake trout captured in 1994 were found to be visibly infected with plerocercoid larval cysts of *Diphyllobothrium ditremum* on the outer wall of the stomachs⁶ (A. W. Shostak, parasite identification and personal communication, Department of Biological Sciences, University of Alberta, Edmonton, Alberta, T6G-2E9). No cysts were found in mountain whitefish. *D. ditremum* is transmitted to lake trout by feeding upon copepods or infected fish (Dick and Poole, 1984; Andersen et al., 1987). The plerocercoid cysts apparently do not harm the fish, and their cycle is completed when a fish-eating mammal or bird eats an infected fish (Anthony, 1967).

Zooplankton were collected by towing a 64 µm mesh zooplankton net of 1 metre diameter (efficiency not measured) through the entire water column, then along the surface. Zooplankton were sorted into 3 size classes using 243 µm and 947 µm screens for stable isotope and organochlorine analyses. Both sorted samples (consisting of *Hesperodiaptomus arcticus* alone) and unsorted zooplankton samples were processed for organochlorine and stable isotope analyses. Individual *Daphnia*

⁶ Personal note: I inquired with local recreational fishers and the Park Warden attending the nearby Mosquito Creek campground, about the appearance of fish stomachs in their catches from Bow Lake. They claimed to have observed these cysts in fish from Bow Lake in previous years.

middendorffiana were separated from the samples by hand for stable isotope analyses. Phytoplankton could not be collected in sufficient amounts for stable isotope or organochlorine analyses. Benthic algae were obtained by scraping rocks from the shoreline for stable isotope analyses. Benthic invertebrates were collected by sweeping the shoreline with long handled collecting nets. In addition, burlap sacks (filled with rocks, with a float) were set underwater near the shoreline to collect invertebrates. All invertebrate species were pooled by major taxa for analyses except for Hesperodiaptomus arcticus and Gammarus lacustris. Samples were stored at -60°C in pre-cleaned glass jars until analysis.

Water samples (40 L) were collected in ten 4L pre-cleaned amber glass bottles from the glacial inflow, centre and outflow of the lake over the ice-free season in 1994 and 1995. The dates of collection are given in Figure II.5. All samples were taken just below the water surface. Due to the limited numbers of bottles and transportation logistics, usually only one site was sampled on each trip. Filled bottles were kept out of direct sunlight and covered with blankets to keep them cool during transport. The filled bottles were taken to the Environment Canada laboratories in Calgary, and extracted within 24 hours (see Laboratory Methods). Organochlorine analysis of unfiltered water included both water and suspended solids.

Sediment samples were taken from the inflow area and the centre of the lake with a heavy Ekman dredge in August 1995. A core (3 cm diameter, 42.0 cm long) was also taken from the centre of the lake with a KB corer, and 5 slices were taken from the

⁷ Benthic invertebrates were attracted to the sheltering crevices and the algae that grew on the sacking material. The burlap sacking material was analyzed for organochlorines (similar to invertebrates, see Methods) but there was no indication of potential contamination of invertebrate samples. Invertebrates collected with nets and from burlap sacks were analyzed separately for organochlorines, and no significant differences were found in organochlorine concentrations.

top of the core (0.0-1.7 cm, 1.7-11.0 cm, 11.0-22.1 cm, 22.1-32.0 cm, and 32.0-42.0 cm). There are no data for ²¹⁰Pb or other means of dating the core. The sediment samples were stored at -60°C in Whirl-pac® bags and freeze-dried before laboratory analyses. The top core slice was lost during laboratory analyses, hence data are not available for this slice.

3. Laboratory methods

3.1 Stable isotope analyses

Both stable isotope and organochlorine analyses for all samples were done at the Freshwater Institute (FWI), Department of Fisheries and Oceans, Winnipeg, Manitoba. For stable isotope analyses, a small section (roughly 1.5 cm³ each) of dorsal muscle was removed from individual fish, and the skin removed. The muscle tissue was placed in individual marked plastic dishes. Trichopterans were removed from their cases. Invertebrates were pretreated with 1 N hydrochloric acid to remove surficial carbonates. Samples were not lipid-extracted prior to stable isotope analyses. All samples were oven-dried at 60°C for 1 week, ground to a fine powder. Dried samples were placed in tin capsules (8-12 mg for nitrogen analyses, and 2-5 mg for carbon analyses) and combusted in a Carlo Erba NA 1500 elemental analyzer⁸. Sample gases were introduced into a VG Optima automated mass spectrometer with helium carrier gas. Water and CO₂ were removed using magnesium perchlorate and an Ascarite® column respectively, while N2 was cryogenically trapped on a molecular sieve. The ratios of the stable isotopes were then measured against the reference standards PeeDee dolomite for δ^{13} C and air for δ^{15} N using the formulae below. The delta notation (δ) is used to indicate the parts per mil (∞ , or parts per thousand) difference in the isotopic ratio of the sample from the standard. A working

⁸ Dr. B. Ross conducted the stable isotope analyses at the FWI.

standard of Pharmamedium of known isotopic composition was run with every 10 samples. Precision over several years has been 0.4 parts per mil (2 SD; Kidd, 1996).

$$\delta^{13}$$
C or δ^{15} N = [R_{sample}/R_{standard}-1] x 1000
where R= 13 CO₂/ 12 CO₂ for δ^{13} C
R= 15 N₂/ 14 N₂ for δ^{15} N

3.2 Organochlorine extractions and analysis

Six organochlorines were quantified for this study: toxaphene (chlorobornanes), total dichlorodiphenyl trichloroethane (\(\sumeter DDT\)), total polychlorinated biphenyls (\(\sumeter PCB\)), total chlordane (\(\sumeter CHL\)), dieldrin, and total hexachlorocyclohexane (\(\sumeter HCH\)). For fish, dorsal muscle samples with skin attached were filleted, and homogenized with dry ice. After the dry ice had sublimated from the sample, 10 g of homogenate was mixed with approximately 300 g anhydrous sodium sulfate (pre-cleaned at 600°C for 16 hours). The invertebrate samples (5 - 10 g wet weight) were freeze-dried, coarsely ground and mixed with anhydrous sodium sulfate. The % water of invertebrates, as indicated by differences between wet and dry weights, was recorded. Before extraction, the organochlorine internal standards, PCB 30 and octachloronapthalene (OCN), were added. Fish and invertebrate samples were then extracted in 1:1 hexane: dichloromethane (DCM) for 4 hours using a Soxhlet apparatus. A blank consisting of 300 g pre-cleaned anhydrous sodium sulfate spiked with internal standards was included in every third run (each run consists of 6 samples) to ensure that equipment. glassware and solvents were sufficiently clean. After extraction, all samples were

⁹ See Appendix 1 for a list of all organochlorine congeners analyzed. In brief, toxaphene is quantified based on a common response factor for all peaks, including T2 (an octachlorobornane) and T12 (a nonochlorobornane). ΣDDT is the sum of all o,p'- and p,p'- DDE, DDD, and DDT congeners. ΣPCB is the sum of all PCB congeners found in samples. ΣCHL is the sum of heptachlordane, oxychlordane, trans-chlordane, cis-chlordane, trans-nonoachlor, cis-nonachlor, and other chlordane congeners. ΣHCH is the sum of α-, β-, δ-, and γ-HCH congeners (except for water, which did not include δ-HCH).

reduced in volume to 10 mL by rotary evaporation and nitrogen-stream evaporation. One-tenth of the Soxhlet extract was used to gravimetrically determine the percent extractable lipids. Invertebrate samples were pre-filtered to remove suspended particles before determining the percent extractable lipids. Lipids were removed using Gel Permeation Chromatography (GPC) by eluting though SX-3 Bio-bead columns with 1:1 hexane: DCM.

The Soxhlet extracts were separated on 1.2% deactivated Florisil (8 g) columns into 3 fractions (F1, F2, and F3). "F1" contained all PCB congeners, some DDT and toxaphene congeners, and the PCB 30 internal standard in 100% hexane. "F2" contained most of HCH, toxaphene and CHL congeners, the remaining DDT congeners, and the OCN internal standard in 85:15 hexane: DCM. "F3" contained remaining organochlorine congeners from the column including dieldrin in 1:1 hexane: DCM. Using a nitrogen stream, hexane and DCM solvents were evaporated off and gradually replaced with isooctane, and then reduced to 0.5 mL for fish samples and 0.3 mL for invertebrate samples. Samples were transferred to GC sample vials with silicone septa. Volume corrector (4.0 µL aldrin) was added to each sample to correct for possible variation in extraction volume in GC vials. Recoveries of internal standards from all samples were calculated using PCB 30 for F1 and OCN for F2. Recoveries of internal standards in all fish samples were 78 ± 10 % for F1 fractions and $77 \pm 11\%$ for F2 fractions¹⁰. No standards were used for F3. Data from chromatographs were not corrected for internal standard recoveries, but internal standards were used to monitor extraction efficiency.

¹⁰ See Appendix 1 for internal standard recoveries of all samples analysed in this study.

Each fraction was analyzed on a Varian 3600 gas chromatograph with a ⁶³Ni-electron capture detector (GC-ECD). The GC-ECD specifications included a 60 m by 0.25 mm i.d. DB-5 column (film thickness 0.25μm) with H₂ carrier gas (approximately 1 mL min⁻¹; initial flow of 2.0 mL min⁻¹; constant pressure of 22.5 psi) and N₂ makeup gas. The GC-ECD temperature started at 100°C, increased 15°C per minute to 150°C, and then increased 3°C per minute to 265°C. Temperature remained constant for the rest of the 57 minute run. External standards were used to quantify organochlorines in each sample, and were obtained from National Research Council of Canada (Halifax, NS), Ultra Scientific (Hope, RI), National Institute of Standards and Technology (Maryland), and Cambridge Isotope Laboratories (Massachusetts). toxaphene in samples was quantified from using a common single response factor that was based on 24 peaks in the standard. Detection limits are 0.01 - 0.02 ng/g. Nondetected compounds were not included in data analyses.

Water samples were extracted in approximately 1.5 L DCM using a Goulden Large-Volume Extractor (Anthony, 1985) at the Environment Canada laboratories in Calgary. Three blanks, consisting of laboratory distilled water, were also extracted, and were used to determine that the equipment and glassware were sufficiently clean. A spike, consisting of 100 ng L⁻¹ each 1,2,4,5 tetrabromobenzene, 1,3,5 tribromobenzene and δ-HCH, was added to each sample before extraction. Once the entire 40 L sample had been processed (4-6 hours), the DCM solution containing organochlorines was drawn off into a pre-cleaned amber 500 mL jar, and stored in the dark until laboratory analyses. At the Freshwater Institute, the DCM solution was spiked with PCB 30 and OCN internal standards, and poured through DCM-rinsed pre-cleaned anhydrous sodium sulfate to remove any remaining water. The DCM solution was then eluted through 1.2% deactivated Florisil columns into 3 fractions using similar procedures as for biota samples. PCB 30 and OCN values was used to

monitor efficiency of anhydrous sodium sulfate filtration and fractionation on Florisil columns. Bromobenzene standards (20 ng L^{-1} in isooctane) for the spike were used to calculate % recovery from sample, but δ -HCH was disregarded. For water, δ -HCH was not included in Σ HCH values 11 . All sediment samples were freeze-dried, and approximately 15 g of each sample was extracted with DCM on an ASE 200 Accelerated Soxhlet Extractor (ASE) under high pressure (2000 psi) and high temperatures (100°C) using N_2 gas (ultra-high purity quality). The sediment samples were run on the ASE with a static time of 10 minutes, 90% flush volume, and a purge time of 60 seconds. Internal standards (PCB 30 and OCN) were added before extraction. A 15 g blank consisting of pre-cleaned anhydrous sodium sulfate was included in the run. Sediment extracts were processed using the same procedures as for water and biota samples. Sediment and water values were not corrected for internal standard recoveries. All water and sediment extracts were treated with reduced copper (treated with 8% nitric acid and rinsed with acetone and distilled water) to remove sulfur after Florisil fractionation.

Analyzing toxaphene concentrations in abiotic samples such as water and sediment on the GC-ECD are prone to misinterpretations due to differences in chromatographic peaks between environmental samples and technical standards (Muir and de Boer, 1995). The confirmation of GC-ECD results was done by analyzing 2 water and 3 sediment samples (Table II.6) with high resolution gas chromatography electron capture negative high resolution mass spectrometry (GC-MS)¹² which measures specific ions of hexa- to nonachlorobornanes (G. Stern, *pers. comm.*, Department of Fisheries and Oceans, Freshwater Institute, 501 University Crescent, Winnipeg, MB,

¹¹ δ -HCH was not commonly found in biota or sediment samples from Bow Lake, unlike α - and γ -HCH (Appendix 1).

¹² "HRGC-ECNI-HRMS" is the proper abbreviation, but for brevity, "GC-MS" will be used in this chapter. Analyses were done by Dr. G. Stern, FWI, Winnipeg, MB.

R3T-2N6). GC separations of samples were performed on a Hewlett Packard model 5890 Series II gas chromatograph using a 60 x 0.25 mm i.d. DB-5 ms fused silica column (film thickness 0.24 µm). Helium was used as the carrier gas. Samples were run on the gas chromatograph using splitless injection with the injector temperature set at 260°C with a CTC A2000SE autosampler under data system control. Initial column temperature was 80°C; at 2 minutes, the oven was ramped at 20°C min⁻¹ to 200°C, then at 2°C min⁻¹ to 230°C, then at 10°C min⁻¹ to a final temperature of 300°C which was held for 8 minutes. Electronic pressure programming was used to increase the pressure during the injection cycle and then to maintain a constant flow of 1 mL min⁻¹ during the remainder of the run. Separated samples were then analyzed in the selected ion mode on a Kratos Concept high resolution mass spectrometer (EBE geometry) controlled using a Mach 3 data system. Selected ion electron capture negative ion mass spectrometry was performed at the resolving power of M/ΔM ~1200. The moderating gas was methane and the mass calibrant was perflurokerosene. Optimum sensitivity was obtained at a gas pressure of ~2 x 10⁻⁴ torr, as measured by the source ion gauge. The electron energy was adjusted for maximum sensitivity (180 eV), the accelerating voltage was 5.3 kV and the ion source temperature was 120°C. The characteristic ions of toxaphene congeners were monitored from the (M-Cl) isotopic cluster of the hexa- to nonachlorobornane homolog groups.

4. Data analyses

Calculations were made with SAS software (SAS Institute Inc., 1993, Version 6.10). For fish, weight was chosen as the size variable. Fish fork length and weight variables were highly correlated¹³. Non-paired Student's t-tests were used to

^{13 (}Weight)= $1.75 * 8.6 \times 10^4 (\text{Length})^3$; Parametric ANOVA F-statistic = 2403.66, $r^2 = 0.99$, p < 0.001

compare the means of stable nitrogen and carbon isotope values between lake trout and mountain whitefish and between zooplankton and benthic invertebrates. Non-parametric statistics were used for organochlorine data analyses because of the small sample sizes (Zar, 1984). Nonparametric Spearman's rank-correlation coefficients (r_s) were used to measure correlations between organochlorine concentrations and % lipids, $\delta^{15}N$ and $\delta^{13}C$ in biota, and to measure correlations between organochlorines, age, size, lipids, $\delta^{15}N$ and $\delta^{13}C$ within each fish species. Significance of correlation coefficients was determined at α <0.05 (Zar, 1984) for all correlations.

RESULTS

1. The food web of Bow Lake.

1.1 Characterization by stable nitrogen and carbon isotope ratios.

All adult fish of both species were found to feed at a similar trophic level. Lake trout (5.8‰ to 7.6‰) and adult mountain whitefish (6.1‰ to 7.8‰) had similar $\delta^{15}N$ values ranging over 2‰ (Student's t-test t= 1.43, p= 0.175; Figure II.1). Mountain whitefish fry had a $\delta^{15}N$ value (4.3‰) falling between the $\delta^{15}N$ values for adult mountain whitefish and invertebrates. $\delta^{15}N$ values for most zooplankton and benthic invertebrates were similar (1.3‰ to 2.9‰; Student's t-test t= 0.853, p= 0.216), indicating that most invertebrates occupy a similar trophic level (Figure II.1; Table II.1). Exceptions included *Daphnia middendorffiana*, with a $\delta^{15}N$ value (0.68‰) that was lower than that for any other invertebrate, and the 1994 Plecoptera and *H. arcticus* samples which had $\delta^{15}N$ values similar to these of mountain whitefish and lake trout, indicating that they are feeding on other invertebrates. Nitrogen signatures of benthic algae were divergent with one sample having a very low $\delta^{15}N$ (-1.34‰) and another sample having a nitrogen value (1.27‰) comparable to that of benthic invertebrates (Figure II.1; Table II.1). The average difference in the $\delta^{15}N$ values of Bow Lake invertebrates (excluding the exceptions outlined above) and fish was about

4‰ (Figure II.1). The range of δ^{15} N values between of algae, invertebrates and fish suggests that, including primary producers, there are only 3 trophic levels in Bow Lake.

There were at least two distinctive sources of carbon at the base of the food web, with enriched benthic and depleted pelagic signatures (Figure II.1; Table II.1). As expected, benthic invertebrates such as *Gammarus lacustris* had significantly higher δ^{13} C values (-25.8‰ to -24.2‰) than pelagic zooplankton (-33.9‰ to -31.1‰; Student's t-test t= 11.2, p<0.001). One Ephemeroptera nymph had similar δ^{13} C values to zooplankton (-30.5‰). Both benthic algae samples had different δ^{13} C values (-24.05 and -17.98‰). Mountain whitefish and mountain whitefish fry had a carbon signature similar to benthic invertebrates (-25.6‰ to -20.3‰). Lake trout δ^{13} C values (-30.7‰ to -24.8‰) fell between pelagic and benthic invertebrate δ^{13} C values. Lake trout δ^{13} C values were significantly depleted compared to mountain whitefish δ^{13} C values (Student's t-test t= 5.61, p<0.001). Taken together, the two fish species form a continuum of δ^{13} C values ranging over 11‰. From stable carbon analyses, it appeared that mountain whitefish exclusively feed on benthic invertebrates, while lake trout diets include both benthic and pelagic invertebrates.

1.2. Lipids in the food web.

Hesperodiaptomus arcticus had the highest lipid proportions (14.9 \pm 8.6%) of all Bow Lake biota, including lake trout (6.7 \pm 3.1%), while Gammarus lacustris had higher proportions (7.5 \pm 3.4%) than mountain whitefish (4.3 \pm 2.2%; Table II.1). Lipid content in biota did not increase with trophic level, as indicated by a lack of correlation with $\delta^{15}N$ (r_s = -0.357; Figure II.2). However, lipid content in biota significantly increased with more depleted $\delta^{13}C$ values (r_s = 0.547; Figure II.3).

1.3. Fish stomach contents.

Lake trout and mountain whitefish stomach contents differed. Lake trout stomach contents reflected an opportunistic feeding pattern, while mountain whitefish stomach contents consisted exclusively of benthic organisms. The average lake trout stomach content varied from year to year, with benthic invertebrates making up most of observations (50-90% of observations; Table II.2). Gammarus lacustris (average ≈23% of observations) and Chironomidae (≈28%) were most frequently found in lake trout stomachs (Table II.2). Adult Ephemeroptera in 1994 (27.3%) and unidentified aerial insects in 1991 (11.6%) made up a substantial portion of lake trout diets in those years. Small fish were found occasionally (≈8%) in lake trout stomachs (except in 1984), but did not appear to form the mainstay of lake trout diet. Zooplankton were infrequently observed in lake trout stomachs (≈7%), and were missing from sampled fish in 1991. This suggested that the mainstay of lake trout diet is benthic invertebrates, with zooplankton and smaller fish being eaten occasionally. Mountain whitefish stomach contents collected in 1994 consisted of benthic invertebrates only. Trichoptera (46%) and Lymnaeidae (30%) occurred frequently in mountain whitefish stomachs, which were found infrequently in lake trout stomachs (Table II.2). Mountain whitefish stomachs contained Plecoptera, adult Ephemeroptera and G. lacustris (each ≈8%), but in lower quantities than seen in lake trout stomach contents.

2. Organochlorines

2.1 Organochlorines in biota.

The general order of organochlorines from highest to lowest concentrations in fish was toxaphene>> ΣDDT > $\Sigma PCB \ge \Sigma CHL \ge dieldrin> \Sigma HCH$ (Table II.3). Toxaphene was highly concentrated in biota relative to other organochlorine compounds (i.e. 118 ± 46.6 ng g⁻¹ for *Hesperodiaptomus arcticus* to 1.7 ng g⁻¹ for Lymnaeidae). ΣDDT was usually found at concentrations less than half of toxaphene

in biota, while Σ PCB was found at 1/10 or less of concentrations of toxaphene. Σ CHL were usually at similar concentrations to Σ PCB in biota. Σ HCH and dieldrin were found at very low concentrations in all biota, except for zooplankton.

2.2 Organochlorine patterns in the food web.

Of all biota analyzed, *Hesperodiaptomus arcticus* had the highest organochlorine concentrations (Table II.3) and lipid content (Table II.1). Of note are the particularly high organochlorine concentrations in H. arcticus, compared to other biota, for toxaphene (118 \pm 46.6 ng g⁻¹), Σ PCB (12.9 \pm 13.4 ng g⁻¹) and dieldrin (10.5 \pm 12.8 ng g⁻¹). While lake trout also had high organochlorine concentrations, their concentrations were at least 25% lower than H. arcticus, except for Σ DDT concentrations, which were similar to that of H. arcticus. Mountain whitefish had low organochlorine concentrations, approximately one-third those of lake trout. $Gammarus\ lacustris\ usually\ had\ organochlorine\ concentrations\ less\ than\ one-quarter of <math>H$. $arcticus\ concentrations$, but higher than those of mountain whitefish. Only Σ DDT and Σ PCB were similar for both mountain whitefish and G. lacustris. There were higher organochlorine concentrations in pelagic zooplankton compared to benthic invertebrates. Among benthic invertebrates, G. $lacustris\ had\ higher\ concentrations\ than\ Tipulidae\ and\ Lymnaeidae.$

Source of carbon and lipid fraction in Bow Lake biota were important for organochlorine bioaccumulation while trophic position was less important. Biota with higher lipid content (% lipids) had significantly higher organochlorine concentrations for all compounds ($r_s = 0.577$ to 0.872; Figure II.4). The consistent negative significant correlations of δ^{13} C with all organochlorine concentrations in the biota of Bow Lake food web ($r_s = -0.477$ to -0.718; Figure II.5) indicated that organochlorine bioaccumulation is greater in organisms with more depleted carbon

signatures. Correlations between contaminants and $\delta^{15}N$ through the food web were non-significant (r_s = -0.216 to 0.186; Figure II.6).

2.3. Lake trout and mountain whitefish.

Larger lake trout had higher lipid content in muscle tissue (r_s = 0.630; Table II.4a), were older (r_s =0.540) and had more depleted δ^{13} C values (r_s =-0.750; Table II.4a). Lipid content in lake trout was not significantly correlated with age (r_s = 0.267), but was correlated with δ^{13} C (r_s = 0.754) and δ^{15} N (r_s =-0.552; Table II.4a). δ^{15} N and δ^{13} C were significantly correlated in lake trout (r_s =0.788). Larger mountain whitefish were also older (r_s = 0.986) and had more depleted δ^{13} C values (r_s = 0.529; Table II.4b). Older mountain whitefish also had more depleted δ^{13} C values (r_s = 0.554). Unlike lake trout, however, size of mountain whitefish was only weakly correlated with lipid content (r_s = 0.405).

Organochlorine concentrations, with the exception of Σ CHL, were higher in lake trout with more depleted δ^{13} C values (r_s = -0.509 to -0.726) and higher lipid content (r_s =0.549 to 0.726; Table II.5a). Σ CHL in lake trout was not correlated to δ^{13} C (r_s = -0.329), but showed a correlation to % lipids (r_s = 0.591). Lake trout with depleted δ^{15} N values had higher concentrations of Σ DDT (r_s =-0.579), Σ HCH (r_s = -0.568), and dieldrin (r_s =-0.568; Table II.5b). Older and larger lake trout had higher concentrations of toxaphene (r_s = 0.534 and 0.565 respectively), Σ PCB (r_s =0.549 and 0.656 respectively), and Σ CHL (r_s =0.456 and 0.570 respectively; Table II.5b). In mountain whitefish, higher organochlorine concentrations, except for toxaphene, were correlated with higher lipid content (r_s =0.425 to 0.749; Table II.5b) but organochlorine concentrations were not correlated with δ^{13} C values (r_s =-0.017 to -0.333). Toxaphene in mountain whitefish was an exception, showing a lack of

correlation with % lipids (r_s =0.070), but a significant correlation with δ^{13} C (r_s =-0.548; Table II.5b).

2.4. Organochlorine patterns in water and sediment.

Organochlorine concentrations in water were consistent among sampling sites (Table II.6), with more variability occurring for the more hydrophobic compounds. In all water samples, Σ PCB was the dominant compound with concentrations at least two times higher than any other organochlorine compound. Σ HCH was also highly concentrated in water in contrast with relatively low concentrations of Σ HCH in sediments and biota. Toxaphene concentrations were approximately one-quarter that of Σ PCB and half that of Σ HCH. Σ CHL concentrations were four-fold less concentrated than Σ HCH, while Σ DDT and dieldrin concentrations were an order of magnitude lower than any other compound. The GC-ECD results for toxaphene were similar to these obtained from the GC-MS (Table II.6).

Concentrations of all organochlorines in sediments collected near the inflow were similar to these from the centre of the lake (Table II.6). In terms of relative concentrations, Σ PCB was present at higher concentrations, while toxaphene and Σ DDT were an order of magnitude lower than Σ PCB in surface sediments. In sediment cores, organochlorine concentrations (except for Σ PCB) decreased by approximately 0.1 ng g⁻¹ per slice (each slice \approx 10 cm). Σ PCB concentrations appeared to remain similar in each slice, with a large increase in core slice 4 (Table II.6). Σ HCH, Σ CHL and dieldrin were much less prevalent in sediments, both surface and core, than Σ PCB, toxaphene or Σ DDT. Toxaphene concentrations in sediment were confirmed by the similarity of results from the GC-MS.

DISCUSSION

The stable carbon and nitrogen isotope measurements described a food web in Bow Lake that has only three trophic levels. The short food chain of Bow Lake did not support the original hypothesis that a long food chain was responsible for the high organochlorine concentrations in the lake trout in Bow Lake. In Bow Lake, $\delta^{15}N$ was useful for characterizing trophic levels but was not correlated with organochlorine concentrations. This finding is unusual, because in other aquatic studies, $\delta^{15}N$ measurements have been successfully used to describe the biomagnification of organochlorines with trophic level. For example, ΣDDT , ΣHCH , and toxaphene bioaccumulation in Lake Laberge was strongly correlated with $\delta^{15}N$ (Kidd, 1995a), and the slopes of the regressions were used to describe the biomagnification potential of these organochlorine compounds. Similar results were reported for p, p'- DDE, mirex and Σ PCB in the Lake Ontario pelagic food web (Kiriluk et al., 1996), and for Σ PCB in Lake Baikal (Kucklick et al., 1996). However, in most bioaccumulation studies, the aquatic food chains are longer than in Bow Lake. Usually, several species of forage fish and invertebrates provide intermediate steps between primary producers and predatory fish, leading to a wide range of $\delta^{15}N$ values within the food web. For example, in the food web of Lake Laberge, Yukon Territories, the top predators, burbot and lake trout, had $\delta^{15}N$ values about 8-9% above benthic invertebrates and 6-7‰ above zooplankton, and there were four forage fish species with intermediate δ^{15} N values (Kidd, 1996). In the pelagic food web of Lake Ontario, the δ^{15} N values of lake trout were 6-8% higher than zooplankton or amphipods, while sculpin and alewife were 3-5% above the invertebrates (Kiriluk et al., 1996). The pelagicdominated food web of Lake Baikal (Kucklick et al., 1996) had three sculpin species 6‰ higher than zooplankton, 4‰ higher than pelagic amphipods, and 2‰ higher than benthic amphipods. In the abbreviated food web of Bow Lake, $\delta^{15}N$ values of fish

were only an average of 4‰ higher than invertebrates, a range too limited to predict organochlorine bioaccumulation due to trophic position.

 $\delta^{13}C$ was correlated with organochlorine concentrations in the Bow Lake food web, with organisms having lighter $\delta^{13}C$ signatures also having higher organochlorine concentrations. Significant correlations between $\delta^{13}C$ and organochlorines were also found within lake trout. Mountain whitefish exhibited no correlation between $\delta^{13}C$ and organochlorine concentrations except for toxaphene. In Bow Lake, lake trout had a wider dietary range than mountain whitefish, as indicated by their wider range of $\delta^{13}C$ values. This affected the magnitude of organochlorines bioaccumulated in fish, because of the higher concentrations of organochlorines in pelagic organisms than benthic organisms. The relatively low complexity of the sub-alpine food web allowed $\delta^{13}C$ values to identify distinctive pelagic and benthic carbon sources. $\delta^{13}C$ measurements demonstrated that the organochlorine bioaccumulation in Bow Lake was predictable from the carbon pathways in the lake.

This is the first time significant correlations between $\delta^{13}C$ and organochlorines within a freshwater food web have been found. Only a few published studies, mostly marine, were found that attempted to correlate $\delta^{13}C$ with contaminants in biota. Spies et al. (1989) demonstrated a positive correlation (no statistics given) between average ΣDDT and $\delta^{13}C$ in two offshore marine food webs (Southern California) with grouped invertebrates (invertebrate predators and detritivores), and three species of fish. However, this relationship was attributed to changes in $\delta^{13}C$ values resulting from shifts towards more depleted isotopic values in contaminated feeding areas with high sewage input. A limitation of the study by Spies et al. (1989) is that different species of invertebrates were grouped, and average fish liver concentrations of ΣDDT were used. Muir et al. (1995), in another marine study, found a lack of correlation

between δ^{13} C and organochlorines (toxaphene, Σ PCB, Σ DDT, and Σ CHL) in Hudson Bay walrus, but they did not report any correlations (or lack of correlations) between δ^{13} C values and organochlorine concentrations for the entire food web. In another study, dietary items of the Gidra people in Papua New Guinea (Yoshinaga et al., 1992), were analyzed for mercury, elemental minerals and stable isotopes. Significant relationships were found between δ^{15} N and mercury (total, organic and inorganic; r_s >0.700, p<0.001), but only a weak correlation was found for inorganic mercury and δ^{13} C values (r_s =-0.355, p<0.05). However, the human dietary items came from disparate ecosystems that have little, if any, interrelationship (i.e. Cassowary birds, deer, ants, freshwater and marine fish and clams), so the lack of δ^{13} C correlations with mercury is not surprising. Except for Bow Lake, no published study was found that demonstrated the correlation of organochlorines in a complete freshwater food web with δ^{13} C values of individual biota.

The examination of fish stomach contents did not always fully support stable carbon isotope results. Carbon signatures of mountain whitefish are supported by their stomach contents, but this was not the case for lake trout. Lake trout stomachs contained mainly benthic invertebrates with a low frequency of zooplankton and fish. In contrast, the nearly pelagic δ^{13} C values of some lake trout suggested that those fish feed largely on pelagic invertebrates. Also, most of the lake trout collected in 1994 had plerocercoid larval parasitic infections providing indirect evidence that pelagic copepods are included in lake trout diets. The plerocercoid larval parasites in lake trout are only transmitted when the fish is preying on copepods or other infected fish (Anthony, 1967). The number of *Hesperodiaptomus arcticus* containing parasites in Bow Lake and the actual rate of infection are unknown. Because none of the mountain whitefish were found with those cysts, and young and small lake trout do not generally feed pelagically because of the threat of predation by larger fish, lake

trout probably accumulated the parasitic cysts by pelagic feeding¹⁴. In all years, the fish were usually caught in early summer soon after ice-off period, which may lead to limited observations of year-round dietary patterns. *H. arcticus* populations exist year-around, including during winter under the ice (Schindler et al., unpublished data, Department of Biological Sciences, University of Alberta, Edmonton, Alberta, T6G-2E9). Lake trout may prey on *H. arcticus* during winter when other prey are scarce, and since stable isotope signature turnover may be slow in coldwater fish (Hesslein et al., 1993), lake trout would still have pelagic carbon signatures soon after ice-off period, even if they commence to feed heavily on benthic invertebrates. A year-around sampling program, including winter, under the ice, is needed to confirm the role of *H. arcticus* and other pelagic invertebrates in lake trout diets.

Lipid content of Bow Lake biota was not related to trophic position. For example, Hesperodiaptomus arcticus had higher lipid content than both lake trout and mountain whitefish, while Gammarus lacustris had higher lipid content than most mountain whitefish. In other studies, lipid content was correlated with trophic position, with top predator fish having the highest lipid content (Kidd et al., 1996; Leblanc, 1995; Kiriluk et al., 1996; Vander Zanden et al., 1996). Lipid content was also usually correlated to the length of the food web with top predators in longer food webs having higher lipid content than these in shorter food webs (Rasmussen et al.,

¹⁴ Additional circumstantial evidence for pelagic feeding patterns in some lake trout can be observed in physical differences between lake trout with different δ^{13} C values. As *H. arcticus* were high in lipids, lake trout preying more heavily on zooplankton may be high in lipids as well, and can benefit in ways such as increased growth rates. For example, within the lake trout collected for this study, a subsample of 5 lake trout with the lightest (-29.5 ± 0.8‰) and 5 lake trout with the heaviest (-25.2 ± 0.4‰) δ^{13} C values indicated that the lake trout with more pelagic carbon signatures tended to have higher lipid content (9.2 ± 2.1%) and faster growth rates (average weight 1129 ± 151 g at 10 years) than lake trout with heavier carbon signatures (3.9 ± 2.1% lipids, average weight 700 ± 441 g at 10 years). In addition, lake trout with pelagic carbon signatures had higher organochlorine concentrations (i.e. ΣCHB = 134 ± 59.7ng g⁻¹) than lake trout with more benthic carbon values (i.e. ΣCHB = 68.2 ± 44.4ng g⁻¹).

1990; Rowan and Rasmussen, 1992; Cabana et al., 1994; Bentzen et al., 1996; Kidd, 1996). It is not known why lipid content is so variable in the food web of Bow Lake, but it may be due to increased storage of lipids in invertebrates in response to a harsh environment. For example, in less extreme environments, copepods have high caloric values due to their elevated lipid content during winter, but carry less lipids during warm summer months (Comita and Schindler, 1963). At Bow Lake, organisms must contend with short ice-free periods, constant cold temperatures, and unproductive surroundings (Chapter I, this thesis). High lipid content in invertebrates and fish is vital for energy required for survival in this environment, for overwintering under ice, and for the ability to reproduce during a brief summer. I speculate that lipid content of invertebrates from sub-alpine lakes may be unusually high than in similar organisms in warmer and more productive lakes.

Lighter δ^{13} C values in the food web and in lake trout were correlated with higher lipid proportions (but not in mountain whitefish). The lipid content of biota can influence δ^{13} C values in biota because lipid tissue has depleted δ^{13} C values compared to other tissues (DeNiro and Epstein, 1977). This has implications for organochlorine bioaccumulation in Bow Lake food web, because there was a three-way correlation between lipid content, δ^{13} C values, and organochlorine concentrations. In Bow Lake, δ^{13} C values could be causally correlated with organochlorines because increased lipid content in biota may be leading to both increased organochlorine burdens and lighter δ^{13} C values. Nevertheless, an Arctic study by Kling et al. (1992) demonstrated little difference in δ^{13} C values between heavier lipid-extracted and lighter whole samples for zooplankton (1.2‰) and salmonids (0.1-0.2‰). The difference between whole and lipid-extracted δ^{13} C values of Arctic salmonids and zooplankton is less than the standard deviation of δ^{13} C values of lake trout, mountain whitefish and zooplankton in this study. As a result the lipid content of Bow Lake biota is presumed not to have

enough effect on δ^{13} C values to cause incorrect interpretation of their dietary patterns. In Bow Lake it appears that δ^{13} C measurements are not only useful for characterizing the distribution of organochlorines, but also for characterizing lipid patterns in the food web.

In Bow Lake, Hesperodiaptomus arcticus had similar δ¹⁵N values to lake trout in 1994, but had much lower $\delta^{15}N$ values in 1995, indicating that copepod dietary patterns change over time. H. arcticus prey on other zooplankton, including rotifers and their own younger instars (Anderson, 1970; Paul and Schindler, 1994), thereby elevating their trophic level. H. arcticus also feed upon large phytoplankton (D. W. Schindler, pers. comm., University of Alberta, Edmonton, Alberta, T6G-2E9) in absence of other prey. In Bow Lake, rotifers, a common prey for H. arcticus, were rare (see Chapter I, this thesis), so Bow Lake H. arcticus are probably omnivorous. The gut contents of *H. arcticus* from Bow Lake were full of glacial silt¹⁵ (personal data), which may be ingested indiscriminately along with diatoms, algae or other zooplankton. This is borne out by high proportions of glacial silt found in the lake bottom in copepod fecal pellets (Smith and Syvitski, 1982; Smith et al., 1980), which indicates that *H. arcticus* are important in the sedimentation processes in Bow Lake. H. arcticus in Bow Lake may ingest suspended particles when rotifers or young instars are low in numbers, which would lead to widely varying $\delta^{15}N$ values. A yearround study of H. arcticus and its diet in Bow Lake would further define feeding patterns of this copepod species, and the zooplankton community ecology in Bow Lake.

¹⁵ Also called glacial flour because of its very fine texture. Glacial silt (clayey suspended sediment finer than 8ϕ) is finely ground rock and sediment from glaciers. The intense blue or green colour common in mountain lakes is due to the light-reflecting properties of suspended glacial flour. Glacial silt is continuously present year-round in Bow Lake, and is typically carried out of lakes with very little settling (Smith et al., 1980).

The ingestion of glacial silt may be an additional route of organochlorine uptake in Hesperodiaptomus arcticus¹⁶. Organochlorine compounds are hydrophobic (see Chapter I of this thesis), and sorb to suspended particles in water (Knezovich et al., 1987). Deposition of organochlorines increases with higher elevations in the mountains (Blais and Schindler, unpublished data, University of Alberta, Edmonton, Alberta, T6G-2E9), and Bow Glacier has high concentrations of many organochlorines from deposits in the recent past when organochlorine compounds were more prevalent (D. Donald, unpublished data, Environment Canada, 2365 Albert Street, Regina, SK, S4P-4K1). If glacial silt from Bow Glacier is a source of contaminants, then H. arcticus may be bioaccumulating organochlorines to higher than expected concentrations. By ingesting sediments from a possible contaminant source, along with the usual bioaccumulation routes via diet and bioconcentration routes via water, H. arcticus may be circumventing usual food web bioaccumulation patterns, and introducing high organochlorine concentrations in the Bow Lake food web. To resolve this question, further study of year-round organochlorine concentrations in zooplankton and glacial silt, and zooplankton feeding patterns is needed.

Water and sediment samples collected from across Bow Lake showed little variation in organochlorine concentrations from inflow to centre to outflow sites. This is attributable to the presence of suspended glacial silt throughout the lake¹⁷, and to the

¹⁶ A weakness of this hypothesis is that glacial silt are likely low in organic carbon, and may not be a major sink for organochlorines. Furthermore, volatilization occurring during glacial melt and river flow may reduce organochlorine inputs prior to entry into the lake (Muir, *pers. comm.*, National Water Research Institute, Environment Canada, Burlington, ON, L7R-4A6). However, this hypothesis is presented for future experimental testing.

¹⁷ As water samples were not filtered, the organochlorine values included suspended solids such as glacial silt.

well-mixed environment of Bow Lake. Toxaphene was particularly prevalent in all environmental samples (water, sediment and biota), which suggests that toxaphene is a particular concern for the Rocky Mountain aquatic environments. ΣPCB was highly concentrated in water and sediment, but was found at low concentrations in biota compared to toxaphene, SDDT and SCHL, suggesting a low bioaccumulation rate for this compound in Bow Lake. EHCH was also found at high concentrations in water but at low concentrations in Bow Lake biota and sediment, which was attributed to the low log K_{ow} values (3.2 - 3.8; Table I.1, Chapter I of this thesis). Lake trout, mountain whitefish, zooplankton and Gammarus lacustris tended to have higher concentrations of toxaphene, DDDT, DCHL and dieldrin than found in water or sediment samples, indicating that bioaccumulation of these compounds is occurring. Hesperodiaptomus arcticus had particularly high concentrations of toxaphene, SPCB and dieldrin compared with other biota. This suggests that water or suspended solids may be an important bioaccumulation route for these compounds because H. arcticus has high lipid content, high surface area-to-volume ratio and as a result, will easily bioconcentrate these organochlorines from water or bioaccumulate them from ingesting suspended particles. Furthermore, Tipulidae and Lymnaeidae, which live in benthic sediments and on rocks (personal observation), had only slightly higher concentrations of most organochlorines than found in water and sediment despite their increased exposure to sediments. These facts indicate that water or suspended solids may be more important than lake sediments as a source of organochlorines in Bow Lake biota.

CONCLUSIONS

The abbreviated food web in Bow Lake offers an opportunity to analyze organochlorine patterns in a relatively simple aquatic community. The results from this study are unusual because 1) δ^{15} N values were not correlated with

organochlorine concentrations in biota, 2) δ^{13} C values were correlated with organochlorine concentrations, and 3) some invertebrates with higher lipid content have higher organochlorine concentrations than fish (i.e. *Hesperodiaptomus arcticus* vs. lake trout and mountain whitefish, *Gammarus lacustris* vs. mountain whitefish). Organochlorine concentrations were also correlated with lipid content indicating that interactions are occurring between lipid content, δ^{13} C values, and organochlorine concentrations. Dietary patterns and lipid content are more important than trophic-related biomagnification for predicting organochlorine concentrations in lake trout in the small food web of Bow Lake.

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Table II.1. Summary data for Bow Lake biota. Average values and their standard deviations are indicated for all taxa represented by three samples or more. Taxa with two samples are indicated by ranges. Code letters are used in following tables and graphs.

Туре		n	Weight (g)	% Lipids	δ ¹³ N (‰)	δ ¹³ C (‰)	Age (уг)
Lake trout '	LT	19	1016 ± 411	6.7 ± 3.1	6.4 ± 0.5	-27.3 ± 1.8	9.6 ± 2.1
Mountain whitefish	MW	12	250 ± 238	4.3 ± 2.2	6.7 ± 0.6	-23.0 ± 1.5	7.3 ± 4.6
Mountain whitefish fry ²	MF	3	~		4.3	-22.8	<1-2
H. arcticus (* 95) ³	Н	3	~	14.9 ± 8.6	2.5 ± 0.3	-31.2 ± 4.2	~
H. arcticus ('94)	h'	ı	~	~	5.6	-31.0	~
Mixed zooplankton 4	Z	2	~	6.5 - 11.3	1.8	-31.1	~
Daphnia middendorffiana	D	l	~	~	0.68	-32.3	~
G. lacustris ('95) 5	G	2	~	5.2 - 9.9	2.7 - 2.7	-23.924.1	~
G. lacustris ('94)	g'	I	~	~	3.77	-23.9	~
Lymnaeidae	S	1	~	0.4	1.4	-25.8	~
Tipulidae	T	2	~	2.0 - 5.5	1.3 - 1.5	-24.222.8	~
Chironomidae	C	1	~	~	2.8	24.5	~
Plecoptera (1995)	St	2	~	~	3.6 - 4.2	-26.924.6	~
Plecoptera (1994)	st'	I	~	~	5.6	-23.8	~
Ephemeroptera (adults)	EA	I	~	~	4.86	-20.57	~
Ephemeroptera (nymphs)	EN	3	~	~	1.74 ± 0.3	-27.0 ± 3.0	~
Trichoptera 6	~	2	~	~	~	-26.2	~
Oligochaeta	O	1	~	~	1.23	-25.2	~
Hydracarina	~	i	~	~	~	-33.1	~
Benthic algae '94 ⁷	В1	i	~	~	-1.34	-17.98	~
Benthic algae '958	B2	I	~	~	1.27	-24.05	~

 ¹⁶ lake trout was analyzed for organochlorines.
 2 Whole mountain whitefish fry was pooled.

³ Hesperodiaptomus arcticus. 3 samples from 1995 were analyzed for organochlorines.

Only one sample of "mixed zooplankton" was analyzed for stable isotopes (two samples were analyzed for organochlorines). "Mixed zooplankton" are unsorted zooplankton samples.

⁵ Gammarus lacustris. These 1995 samples were analyzed for organochlorines.

 $^{^{6}}$ Trichoptera were removed from their cases and pooled. $\delta^{15}N$ sample was lost.

⁷ Benthic algae were collected from the west shore of Bow Lake.

Benthic algae were collected from the east shore of Bow Lake.

Table II.2. Bow Lake fish stomach contents. The number of stomachs containing each taxa found is indicated. The bracketed numbers are the relative frequency for each taxa (the percentage of total observations that a taxa was found in stomachs). The year that fish were captured, the number of stomachs inspected, and the number of empty stomachs are listed. Data for 1984, 1991 and 1992 were obtained from Environment Canada (see Methods).

Fish species	Mountain	Lake trout	Lake trout	Lake trout	Lake trout
	whitefish				
Year	1994	1994	1992	1991	1984
Total number of	12	16	23	26	30
stomachs					
Number empty	7	2	1	0	0
Dietary item:					
Chironomidae	~	7 (15.9%)	16 (23.2%)	18 (34.6%)	28 (36.4%)
Chironomidae (larvae)	~	~	15 (21.7%)	1 (1.9%)	2 (2.6%)
Gammarus lacustris	1 (7.7%)	10 (22.7%)	10 (14.5%)	13 (25.0%)	23 (29.9%)
Ephemeroptera (adults)	~	12 (27.3%)	~	~	~
Unidentified aerial insects 1	~	~	7 (11.6%)	~	~
Plecoptera	1 (7.7%)	7 (15.9%)	1 (1.5%)	2 (5.7%)	4 (5.2%)
H. arcticus ²	~	2 (4.5%)	1 (1.5%)	~	~
D. middendorffiana³	~	1 (2.3%)	4 (5.8%)	~	6 (7.8%)
Lymnaeidae	4 (30.7%)	1 (2.3%)	~	1 (3.8%)	~
Trichoptera	6 (46.2%)	~	3 (4.3%)	2 (5.8%)	11 (14.3%)
Ephemeroptera (nymphs)	1 (7.7%)	~	2 (2.9%)	~	2 (2.6%)
Mtn. Whitefish	~	4 (9.1%)	~	12 (23.2%)	~
Lake trout	~	~	2 (2.9%)	~	~
Unidentified fish	~	~	5 (7.2%)	~	~
Oligochaeta	~	~	2 (2.9%)	~	~
Araneae	~	~	~	~	1 (1.2%)
Total Observations	13 (100%)	44 (100%)	69 (100%)	52 (100%)	77 (100%)

¹ Assumed to be adult Ephemeroptera. ² Hesperodiaptomus arcticus

³ Daphnia middendorffiana

Table II.3. Summary data for organochlorines in Bow Lake biota. Average values and standard deviations are indicated for all taxa represented by three or more samples. Taxa with only two samples are indicated by ranges. Code letters are used in the following tables and graphs. Organochlorines in biota are in ng g⁻¹ wet weight.

Туре		n	Toxaphene	∑DDT	ΣPCB	∑CHL	Dieldrin	∑нсн
Lake trout	LT	16	88.2 ± 65.6	22.2 ± 19.1	7.4 ± 5.7	8.1 ± 5.8	2.3 ± 1.8	1.4 ± 0.9
Mountain whitefish	MW	12	11.3 ± 12.9	8.1 ± 6.1	2.8 ± 2.8	2.5 ± 1.8	0.7 ± 0.5	0.6 ± 0.4
H. arcticus I	Н	3	118 ± 46.6	22.5 ± 9.1	12.9 ± 13.4	12.2 ± 1.9	10.5 ± 12.8	2.0 ± 0.9
Mixed zooplankton	Z	2	37.0 - 126.5	8.1 - 20.5	1.9 - 3.7	7.3 - 15.5	4.9 - 19.2	1.9 - 4.6
G. lacustris ²	G	2	27.1 - 36.8	8.9 - 9.1	3.7 - 3.7	4.0 - 5.5	3.2 - 4.5	1.1 - 2.1
Lymnaeidae	S	I	1.7	0.6	0.5	0.6	0.0	0.03
Tipulidae	T	2	1.8 - 4.8	1.8 - 2.4	2.4 - 3.5	1.8 - 2.6	0.0 - 0.2	0.3 - 0.5

¹ Hesperodiaptomus arcticus.
² Gammarus lacustris.

<u>Table II.4</u>. Non-parametric Spearman's correlation coefficients (r_s) for physical parameters of Bow Lake fish. Asterisks indicate significance at α <0.05 (Zar, 1984). Numbers in brackets indicate number of samples.

a) Lake trout (16)

	Weight	Length	% Lipids	$\delta^{13}C$	δ15 _N
Length	0.956 *				
% Lipids	0.630 *	0.575 *			
$\delta^{13}C$	-0.366	-0.378	-0.754 *		
δ ¹⁵ N	-0.281	-0.294	-0.552 *	0.788 *	
Age	0.540 *	0.557 *	0.267	-0.032	0.201

b) Mountain whitefish (12)

	Weight	Length	% Lipids	δ ¹³ C	δ ¹⁵ N
Length	0.989 *				
% Lipids	0.405	0.397			
δ ¹³ C	0.529 *	0.527 *	0.224		
$\delta^{15}N$	0.418	0.383	0.169	0.355	
Age	0.986 *	0.975 *	0.364	0.554 *	0.415

<u>Table II.5</u>. Spearman's correlation coefficients (r_s) for stable isotope ratios, % lipids, age, and weight against organochlorines in lake trout and mountain whitefish from Bow Lake, Banff National Park. Asterisks indicate significance at α <0.05 (Zar, 1984). Numbers in brackets indicate number of samples.

a) Lake trout (16)

Organochlorines	Weight	Age	% Lipids	δ ¹⁵ N	δ ¹³ C
(ng g ⁻¹ wet weight)					
Toxaphene	0.565 *	0.534 *	0.549 *	-0.365	-0.509 *
∑DDT	0.408	0.109	0.558 *	-0.579 *	-0.544 *
∑PCB	0.656 *	0.549 *	0.651 *	-0.312	-0.532 *
ΣCHL	0.570 *	0.456 *	0.591 *	-0.332	-0.329
Dieldrin	0.316	0.367	0.549 *	-0.543 *	-0.649 *
∑нсн	0.263	0.130	0.726 *	-0.568 *	-0.726 *

b) Mountain whitefish (12)

Organochlorines	Weight	Age	% Lipids	δ15 _N	δ ¹³ C
(ng g ⁻¹ wet weight)					
Toxaphene	-0.077	-0.069	0.070	-0.405	-0.548 *
∑DDT	0.196	0.174	0.640 *	0.077	-0.333
ΣPCB	0.257	0.235	0.647 *	0.275	-0.184
∑CHL	-0.150	-0.211	0.425	0.005	-0.329
Dieldrin	0.110	0.088	0.601 *	-0.077	-0.201
∑нсн	0.418	0.439	0.749 *	0.259	-0.017

<u>Table II. 6</u>. Organochlorine concentrations in Bow Lake water and sediment. Organochlorine concentrations are in ng L⁻¹ for water and ng g⁻¹ dry weight for sediment. Toxaphene was analyzed on both the GC-ECD (CHB_{ECD}) and on the GC-MS (∑CHB_{MS}). See Methods for analysis procedures. Sampling dates and sites are indicated.

Туре	Site	Date	CHB _{ECD}	∑CHB _{MS}	∑DDT	ΣPCB	ΣCHL	Dieldrin	ΣНСН
Water	Inflow	July '95	0.31	~	0.03	0.60	0.13	0.03	0.50
	Inflow	August '95	0.14	0.16	0.05	1.90	81.0	0.04	0.48
	Centre	August '95	0.33	0.15	0.10	1.41	0.15	0.03	0.49
	Outflow	Sept. '95	0.20	0.12	0.02	1.33	0.08	0.03	0.45
Sediment	Inflow	August '95	0.23	~	0.17	1.51	0.07	< 0.01	0.03
	Centre ¹	August '95	0.22	0.16	0.33	1.74	0.06	< 0.01	0.04
	CS 2 ²	August '95	0.32	~	1.49	2.57	0.17	< 0.01	< 0.01
	CS 3 ²	August '95	0.14	0.21	0.10	2.43	0.03	< 0.01	< 0.0
	CS 4 ²	August '95	0.20	~	0.04	5.06	0.05	< 0.01	10.0 >
	CS 5 ²	August '95	0.13	~	0.07	2.84	0.03	< 0.01	< 0.01

¹ Surface sediment samples. Sediment organic carbon content is 3.1% (Donald et al., *in prep.*).
² Core slices from a core taken at the centre of the lake.

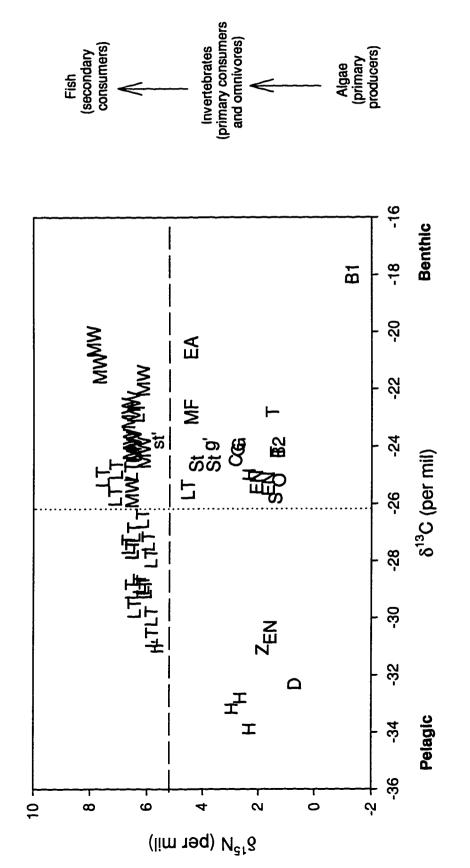


Figure II.1. Stable isotope "map" of the Bow Lake food web. δ^{13} C indicates the carbon sources of the food web, while $\delta^{15}N$ represents the trophic level. There are 3 trophic levels: fish, invertebrates, and benthic algae. Codes are from Table II.1.

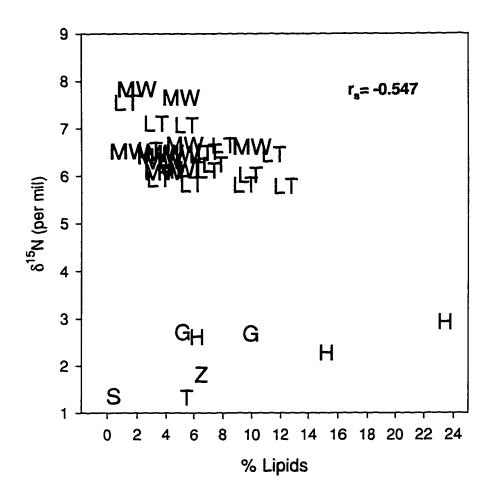


Figure II.2. Stable nitrogren isotope values vs. lipid fraction in Bow Lake biota. Codes are as given in Table II.1. The Spearman's correlation coefficient given is not significant, with significance determined at $\alpha = 0.05$ (Zar, 1984).

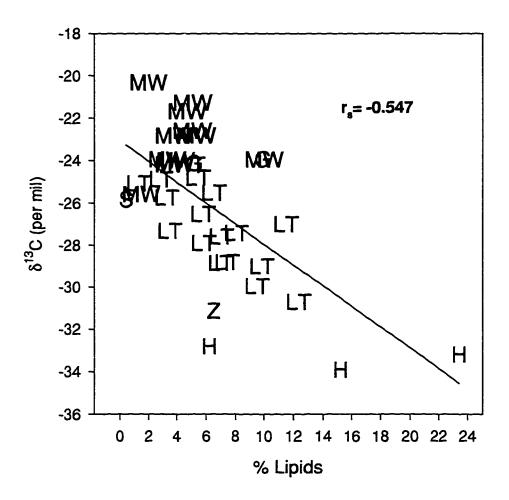


Figure II.3. Stable carbon isotope values vs. lipid fraction in Bow Lake biota. Codes are as given in Table II.1. The Spearman's correlation coefficient given is significant, with significance determined at $\alpha = 0.05$ (Zar, 1984).

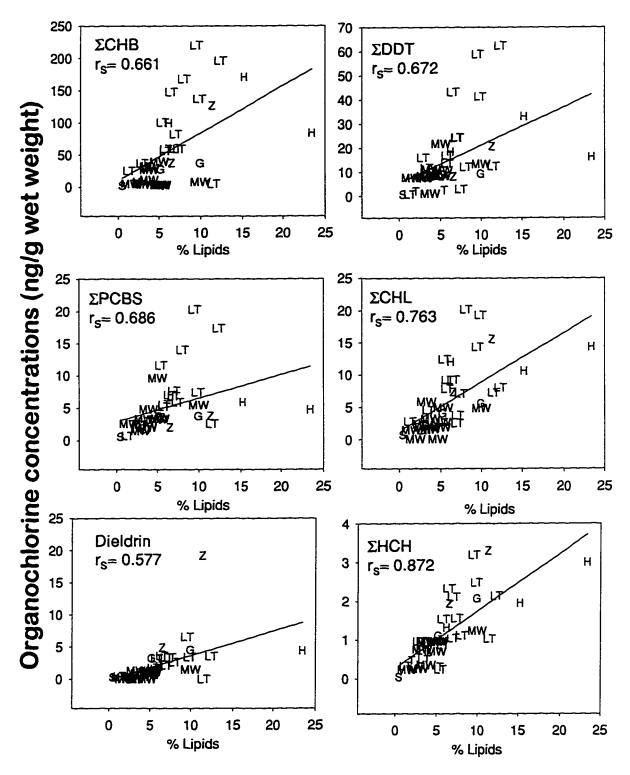


Figure II.4. Organochlorine concentrations in Bow Lake biota vs. % lipid content. Non-parametric Spearman's correlations (r_{*}) are given. Codes as given in Table II.1. Significant statistical relationships were found between organochlorine concentrations and lipid content in biota.

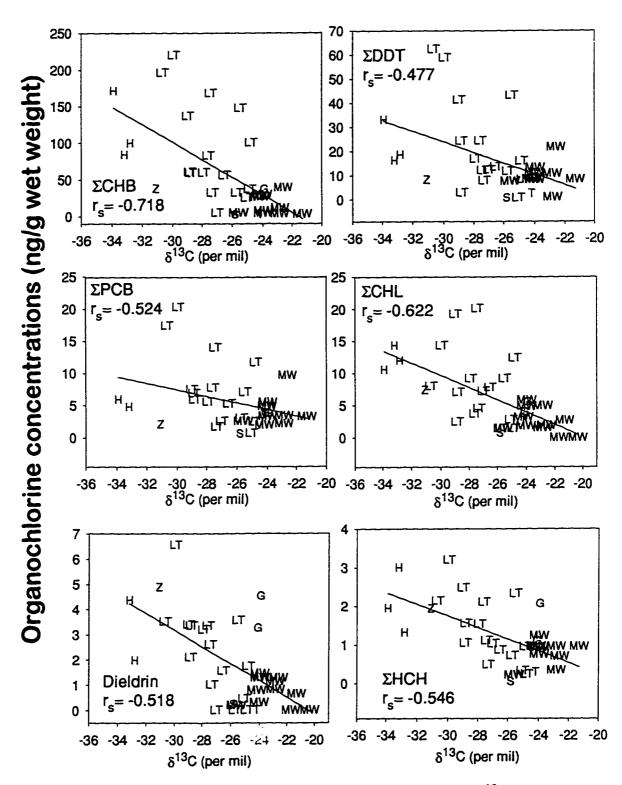


Figure II.5. Organochlorine concentrations in Bow Lake biota vs. δ^{13} C. Non-parametric Spearman's correlations (r_s) are given. Codes as given in Table II.1. Significant statistical relationships were found between δ^{13} C and organochlorine concentrations.

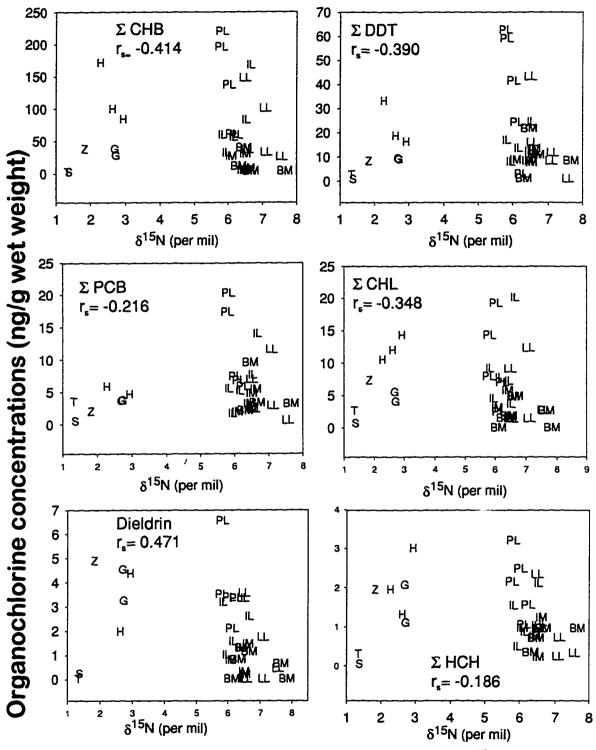


Figure II.6. Organochlorine concentrations in Bow Lake biota vs. $\delta^{15}N$. Codes as in Table II.1. Spearman's correlation coefficients are given (r_s) . No significant statistical relationships could be found between organochlorine concentrations and $\delta^{15}N$ in biota.

III. STABLE ISOTOPE RATIOS (δ^{13} C AND δ^{15} N) IN FISH MUSCLE AND FIN RAY TISSUE: POTENTIAL FOR NON-LETHAL SAMPLING OF FISH.

INTRODUCTION

Stable nitrogen and carbon isotope ratios are highly useful for characterizing trophic relationships in food webs (DeNiro and Epstein, 1980; Peterson and Fry, 1987; Fry, 1991; O'Leary et al., 1992; Chapter II, this thesis). As a result, stable nitrogen and carbon isotopes have been successfully used for studying contaminant dynamics in food webs (Cabana et al., 1994; Kidd et al., 1995a,b; Kiriluk et al., 1996; Kucklick et al., 1996, Chapter II, this thesis). Typically, measurements of stable isotope ratios of fish require muscle tissue samples that demand the sacrifice of the fish. This may be undesirable when fish populations are small, or when endangered species are studied, such as the bull trout in Alberta (Roberts, 1991). The sacrifice of fish may also present problems if repeated measurements from the same fish are required for long-term studies or if catch limits are imposed through fish management programs, common in Rocky Mountain National Parks. Sampling non-essential tissues such as fin rays or scales for isotopic analyses might permit non-destructive sampling. In this study, I examine the possibility of using fin rays for predicting stable nitrogen and carbon isotope values of fish muscle tissue.

Despite the potential usefulness of non-lethal sampling of fish for stable isotope studies, only a few published studies have examined this in freshwater fish. Estep and Vigg (1985) showed that the δ^{13} C of scales was correlated to muscle δ^{13} C in cui-ui (*Chasmistes cujus*) and there was a consistent difference of 2-3‰ between scale and muscle δ^{13} C values. Kiriluk et al. (1996) reported a similar difference in carbon signatures between scales and muscle of lake trout from Lake Ontario. Schroeder (1983a; 1983b), based on small samples (1-3 fish), noted that δ^{13} C values of fin

samples of cultured pond fish were more consistent than scales. Fin samples in Schroeder's studies were 1-2‰ heavier than muscle samples for common carp (*Cyprinus carpio*), *Tilapia aurea*, and a Tilapia hybrid (no genus given), and 0-3‰ heavier for silver carp (*Hypophthalmichthys molotrix*?). The differences in δ^{13} C values between scale and muscle tissue of those cultured pond fish varied more widely, from 1 to 5‰. Rounick and Hicks (1985) examined four New Zealand freshwater fish species, and found that anal fin δ^{13} C values varied from muscle δ^{13} C values by -0.3 to 0.3‰ and that scale δ^{13} C values differed widely by 0.4 to 1.2‰. Only Estep and Vigg (1985) compared δ^{15} N measurements and found that cui-ui scale δ^{15} N values varied widely in relation to muscle δ^{15} N values (0.2-4.0‰). However in all studies mentioned above, sample sizes were limited.

In order to use stable isotope ratios of non-essential tissue to estimate the ratios in muscle tissue of fish, the difference between isotope ratios of external tissue and fish muscle must be negligible, or constant. In this study the stable isotope nitrogen and carbon ratios of pectoral fin rays of freshwater fish were measured and compared to stable isotope values of muscle tissue. This comparison was used to determine whether stable isotope values from pectoral fin rays are significantly different from those of muscle, or whether the difference is predictable. Pectoral fin rays were used instead of scales because fin rays provide enough material for isotopic analyses, and are easy to clip from captured fish. Pectoral fin rays have been used in aging studies, so there is precedent for clipping fin rays (i.e. Barber and McFarlane, 1987). Furthermore, many fish species such as lake trout have tiny scales, making it difficult to remove a sufficient amount of scales for analyses. In this study, I compare stable isotope measurements of fin rays and muscle to determine if fin ray δ^{13} C and δ^{15} N are correlated to muscle isotope values within populations, and if so, to determine if

the relationship between fin ray and muscle stable isotopic values are consistent among populations.

METHODS:

Lake trout (*Salvelinus namaycush*) and mountain whitefish (*Prosopium williamsoni*) were collected at Bow Lake, Banff National Park, Alberta (51°45' N, 116°30'W) as a part of a study of contaminant concentrations in the food web (Chapter II, this thesis). Lake trout and lake whitefish (*Coregonus clupeaformis*) from two Yukon lakes, Fox Lake (61°14'N, 135°28'W), and Kusawa Lake (60°20'N, 136°22'W), were also sampled for these tissues. Dorsal muscle tissue (without skin) was removed, along with the first three fin rays from each fish's pectoral fin (including the tissue between the fin rays). The fin rays were cleaned with distilled water. Both muscle and fin ray samples were oven dried at 60°C for 1 week.

The dried samples were hand-ground to a fine powder, and weighed into aluminum capsules. The Bow Lake fish samples were analyzed at the Freshwater Institute, Winnipeg, and the Yukon fish samples at the Environmental Isotope Laboratory, University of Waterloo. At the Freshwater Institute, Pharmamedium, a protein nutrient standard with constant stable carbon and nitrogen isotope ratios, was used as an internal calibration standard, while at Waterloo, the in-house dogfish standard ("JKUZ Dogfish") was used. The standards were run with every 10 samples.

Dried samples were combusted in a Carlo Erba NA1500 elemental analyzer. The sample gases were then introduced into a VG Optima automated mass spectrometer with H₂ carrier gas. Water and CO₂ were removed using magnesium perchlorate and an Ascarite® column respectively, and N₂ was cryogenically trapped on a molecular sieve. The ratios of the stable isotopes were then expressed as parts per mil (parts per

thousand) difference from the standards PeeDee dolomite for δ^{13} C and air for δ^{15} N using the formulae below. The delta notation (δ) is used to indicate the range of parts per mil (%).

$$\delta^{13}$$
C or δ^{15} N = [Rsample/Rstandard - 1] x 100
where R = 13 CO₂/ 12 CO₂ for δ^{13} C
R = 15 N₂/ 14 N₂ for δ^{15} N

Statistical analyses were done using SAS software (SAS Institute Inc., Version 6.10, 1993). Data for fish from three Yukon lakes (Yukon lake trout and lake whitefish) were pooled because of the limited sample size from each lake. Linear regressions of muscle and fin ray stable isotopes were used to construct simple models for each region. The slopes and intercepts of regression models were tested for differences using small sample t-tests for parallelism and common intercepts respectively (Kleinbaum and Kupper, 1978). Fractionation differences between muscle and fin ray tissues were calculated by subtracting the two values to determine if fishes from different regions had similar magnitudes of fractionation between muscle and fin rays.

RESULTS

Fin rays were useful predictors of muscle $\delta^{13}C$. Regressions of fin ray $\delta^{13}C$ and muscle $\delta^{13}C$ for Yukon and Bow Lake fish (Figure III.1) were strongly correlated with high r^2 values (r^2 = 0.721-0.999, p= 0.002-0.014; Table III.2) and high Pearson's correlation coefficients (0.869 - 0.999). The $\delta^{13}C$ values of fin rays from all fish species were isotopically heavier than for muscle (Figure III.1). The regression slopes were not significantly different (p<0.05; Table III.2). This indicated constant and predictable changes in the $\delta^{13}C$ of fin rays and $\delta^{13}C$ of muscle. However, the intercepts of the $\delta^{13}C$ regression slopes between lake trout and mountain whitefish

from Bow Lake and between lake trout and lake whitefish from the Yukon were significantly different (p<0.05; Table III.2), leading to the conclusion that the δ^{13} C range can vary between species and between regions. However, lake trout and mountain whitefish from Bow Lake had a similar magnitude of δ^{13} C fractionation between fin rays and muscle tissue ($\approx 1.1\text{-}1.5\%$ average).

For fish from Bow Lake, the $\delta^{15}N$ of fin rays was isotopically similar to that of muscle (i.e. close to a 1:1 relationship), but in Yukon lake trout, fin rays were somewhat isotopically heavier in $\delta^{15}N$ than muscle (Figure III.2). Lake whitefish muscle samples had a very wide range (10%) of $\delta^{15}N$, but their fin rays had a narrow range of $\delta^{15}N$ (4%). Regressions of $\delta^{15}N$ in fin rays and muscle tissue (Figure III.2, Table III.3) were correlated in mountain whitefish and lake trout from Bow Lake (r^{2} = 0.643-0.997, p= 0.034-0.055) with high Pearson's correlation coefficients (0.801-0.999). Lake trout from the Yukon had a regression with low r^{2} value and was not significant (r^{2} =0.353, p=0.172), but did have somewhat high Pearson's correlation coefficient (0.717). Regression intercepts for Bow Lake mountain whitefish and lake trout were not significantly different (p> 0.05; Table III.3), leading to the conclusion that fractionation of $\delta^{15}N$ in fin rays and muscle were not significantly different in those fish species. $\delta^{15}N$ correlations between fin ray and muscle tissue do not follow a pattern similar to $\delta^{13}C$ correlations.

DISCUSSION

 $\delta^{13}C$ of fin rays and muscle tissue were well correlated, supporting the use of fin rays to predict the $\delta^{13}C$ of muscle. However, the difference between fin rays and muscle must be taken into account when using $\delta^{13}C$ of fin rays to estimate muscle tissue $\delta^{13}C$ values for a particular fish species within a particular lake. This requires subsampling in the study lake to calibrate fin ray $\delta^{13}C$ to muscle $\delta^{13}C$.

 δ^{13} C values of fin rays were consistently heavier than those of muscle tissue. This may be due to higher lipid content of muscle because lipid tissue has more depleted δ^{13} C values than other tissues. Lipid tissue has lighter δ^{13} C values because lipid synthesis in organisms leads to temperature-dependent fractionation of ¹³C/¹²C, favouring ¹³C isotopes (DeNiro and Epstein, 1977). However, in an Arctic freshwater study, lipid-extracted salmonid muscle δ¹³C values were only 0.1-0.2 ‰ heavier than whole muscle δ^{13} C values (Kling et al., 1992), which is less than the standard deviations for carbon signatures in muscle tissue in this study. Furthermore, the magnitude of δ^{13} C differences between lake trout and mountain whitefish from Bow Lake are similar, yet mountain whitefish have less lipids and weigh less than Bow Lake lake trout (Table III.1), and those two fish species have different dietary patterns (Chapter II, this thesis). The low difference between lipid-extracted and whole muscle tissues in Kling's study, and the similar magnitude of δ^{13} C differences between lake trout and mountain whitefish in Bow Lake indicate that lipid content may not be an important variable in differences in δ^{13} C values between fin ray and muscle tissue. The similarities of the δ^{13} C fractionation ranges between two fish species from the same lake, despite dietary and lipid content differences, suggests that local environmental factors may be more important than species-specific fractionation differences. Basal δ^{13} C signatures of primary producers and sediment can vary between regions (Jackson and Harkness, 1987), and may be contributing to the regional fractionation differences. Ambient temperatures can affect metabolic isotope fractionation rate in some marine fish species, and therefore, may influence the rate of ¹³C uptake in different fish tissues and the ultimate δ^{13} C values in fish (Kalish, 1991; Radtke et al., 1996). A larger study comparing δ^{13} C of fin rays and muscle tissue in a single species with a broad distribution (i.e. lake trout) may indicate whether $\delta^{13}C$ is differentially fractionated between tissues in freshwater fish due to environmental

factors such as dietary $\delta^{13}C$, stable isotope turnoer rates, or ambient temperatures, or due to physical factors within individual fish such as lipid proportion.

Use of fin rays for predicting $\delta^{15}N$ of muscle may have potential for some fish populations, but there is no overall consistency. Lake trout from Bow Lake had significant correlations of fin ray $\delta^{15}N$ with muscle $\delta^{15}N$. Only three Bow Lake mountain whitefish were analyzed, making statistical inference difficult. Lake trout from the Yukon appeared to be correlated, but there was no significant regressions. The variation in $\delta^{15}N$ in fish muscle and fin ray tissue may be due to changes in diet, which would likely affect the distribution of $\delta^{15}N$ within tissue over time, with the fin ray $\delta^{15}N$ representing older $\delta^{15}N$ values. A study of fish populations over a broad geographical range is needed to determine the extent to which $\delta^{15}N$ of fin rays can be used for predicting that of muscle.

Results from this study identify strong patterns but are not conclusive due to the small sample sizes. Fin ray and muscle tissues from a large sample of fish are needed to confirm the results from this study. Food sources of these fish should be identified, and the stable isotope values of these food items compared to both fin ray and muscle stable isotope values to measure deviations and the usefulness of using fin ray tissue to predict the fish's trophic position and dietary patterns.

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Yukon Territory and Bow Lake, Banff National Park, and their standard deviations. Average fish lipid content and weight and their Table III.1. The average δ^{13} C and δ^{15} N of fin ray and muscle tissue of lake trout, mountain whitefish and lake whitefish from the standard deviations for Bow Lake fish are included.

Lake /	.ake/ Fish							8 ¹³ Cmusche -			813Nmuscle -
Area	species	Code	=	Area species Code n %lipids W	Weight (g)	δ ¹³ C _{muscle}	8 ¹³ C _{fin ray}	8 ¹³ Cfin ray	815Nmuscle	815N _{fin ray}	815 Nin ray
Bow Lake	Lake	BLT	9	BLT 6 3.98 ± 2.23 74	748.4 ± 500	-26.50 ± 2.18	8.4 ± 500 -26.50 ± 2.18 -25.01 ± 1.67 -1.49 ± 0.81 6.81 ± 0.53 7.33 ± 0.54	-1.49 ± 0.81	6.81 ± 0.53	7.33 ± 0.54	-0.52 ± 0.24
	Mountain whitefish	BMW	3	2.88 ± 2.76	BMW 3 2.88 ± 2.76 186.8 ± 127.1 -22.30 ± 1.86 -21.13 ± 1.73 -1.16 ± 0.10 6.93 ± 0.79	-22.30 ± 1.86	-21.13 ± 1.73	-1.16 ± 0.10	6.93 ± 0.79	6.59 ± 0.94	0.34 ± 0.12
Yukon lakes ¹	Lake trout	YLT 5	S	ł	410 ± 139.9	-25.35 ± 0.80	0 ± 139.9 -25.35 ± 0.80 -24.79 ± 0.78 -0.90 ± 0.56 8.66 ± 0.41	-0.90 ± 0.56	8.66 ± 0.41	10.15 ± 0.52	-1.84 ± 0.60
	Lake whitefish	YLW 8	∞	ł	747.2 ± 207.8	-29.37 ± 1.58	747.2 ± 207.8 -29.37 ± 1.58 -25.55 ± 1.21 -3.82 ± 0.59 = 2.37	-3.82 ± 0.59	7.29 ± 2.37	8.51 ± 0.75	-1.47 ± 2.45

¹ The Yukon lakes include Kusawa Lake and Fox Lake.

<u>Table III.2</u>. Regressions and Pearson's correlation coefficients for δ^{13} C muscle and fin ray tissue. The fish codes are from Table III.1. Significant differences among regression slopes and intercepts are shown with different superscripts (p<0.05).

Fish	Intercept (β_0)	Slope (B ₁)	r ²	F- statistic	p-value	Pearson's correlation
BLT	-6.62 ^a	0.694 ^a	0.8107	17.14	0.014	0.900
BMW	-0.41 ^b	0.929 ^a	0.9995	2012.99	0.014	0.999
YLT	-0.52 ^b	0.958 ^a	0.960	72.14	0.003	0.980
YLW	-5.21 ^a	0.690 ^a	0.721	21.66	0.002	0.869

<u>Table III.3</u>. Regressions and Pearson's correlation coefficients for $\delta^{15}N$ muscle and fin ray tissue. The fish codes are from Table III.1. Significant differences among regression slopes and intercepts are shown with different superscripts (p<0.05).

Fish	Intercept (\(\beta_0\))	Slope (B ₁)	r ²	F- statistic	p-value	Pearson's correlation
BLT	1.073 ^a	0.822 a	0.643	7.21	0.055	0.801
BMW	-1.659 ^a	1.190 ^a	0.997	354.23	0.034	0.999
YLT	2.286	0.907	0.353	3.18	0.172	0.717
YLW	8.40	0.010	0.220	0.013	0.912	0.047

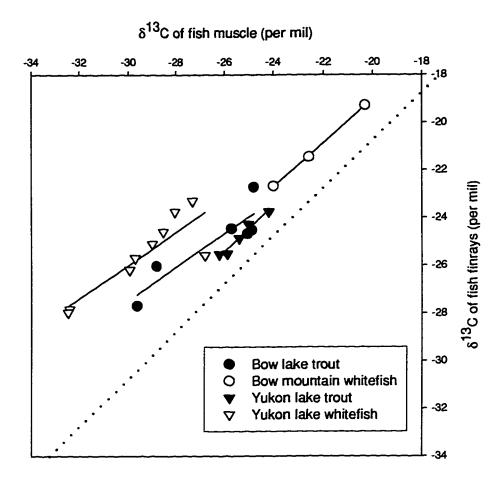
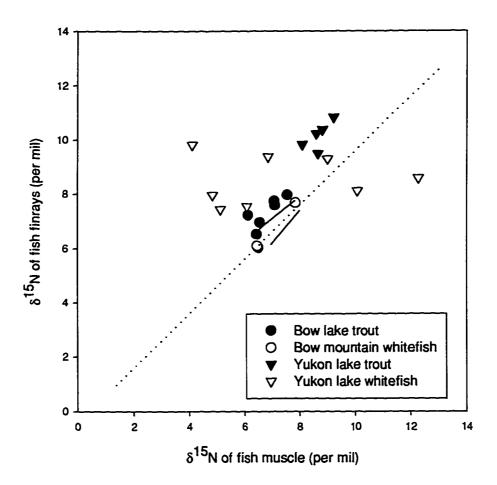


Figure III.1. $\delta^{13}C$ of fin rays vs. fish muscle tissue. Fish are from Bow Lake, Banff National Park and the Yukon Territories. The dotted line represents a 1:1correlation between fin ray and muscle $\delta^{13}C$ values. See Table III.2 for linear regression equations.



<u>Figure III.2</u>. $\delta^{15}N$ of fin rays vs. fish muscle tissue. Fish are from Bow Lake, Banff National Park and the Yukon Territories. The dotted line represents a 1:1correlation between fin ray and muscle $\delta^{15}N$ values. See Table III.3 for linear regression equations.

IV. GENERAL DISCUSSION.

The pattern of organochlorine concentrations in the food web of sub-alpine Bow Lake was significantly different from those documented in temperate and sub-arctic Yukon lakes (Cabana et al., 1994; Kidd et al., 1995; Kiriluk et al., 1996). The food web of Bow Lake was abbreviated, with only three trophic levels, as outlined by stable carbon (δ^{13} C) and nitrogen isotope (δ^{15} N) analyses (Chapter II, this thesis). This was in contrast to my original hypothesis that bioaccumulation through a long food web would lead to high concentrations of organochlorines in lake trout. In Bow Lake the dominant copepod species, Hesperodiaptomus arcticus, had unusually high concentrations of organochlorines relative to lake trout (Salvelinus namaycush) and mountain whitefish (Prosopium williamsoni). The high organochlorine concentrations in H. arcticus and some lake trout were related to lipid content, which may be high year-around in H. arcticus. Lipid content was important, as opposed to trophic position (as measured by $\delta^{15}N$), as a predictor of organochlorine concentrations in Bow Lake organisms. This was attributed to the short food chain, and the cold environments promoting increased lipid storage, therefore increasing organochlorine uptake and retention. In addition, lighter δ^{13} C values were correlated with higher organochlorine concentrations in the food web, which indicated the partitioning of organochlorine compounds in Bow Lake, with most of the bioaccumulation taking place in the pelagic portion of the Bow Lake food web (Chapter II, this thesis).

The largely benthic stomach contents of lake trout did not support the conclusion of mixed pelagic feeding patterns in lake trout (based on δ^{13} C values). This may be because lake trout were collected during the short ice-free season, leading to a limited insight into lake trout diets. However, plerocercoid larval parasite cysts on lake trout stomachs provide circumstantial evidence for inclusion of copepods in lake trout

diets, because the parasites are transmitted through feeding on copepods or other infected fish (Anthony, 1967). There are *H. arcticus* populations year-round, including under ice (D. W. Schindler, unpublished data, Department of Biological Sciences, University of Alberta, Edmonton, Alberta, T6G-2E9), so they are available as a constant source of food for lake trout especially during the winter. In brief a diet of benthic invertebrates during the summer, complemented by *H. arcticus* during the winter, may lead to high organochlorine concentrations and pelagic stable isotope values in lake trout. A year-round sampling program of invertebrates in lake trout stomachs and stable isotope analyses of organisms in Bow Lake is required to resolve the apparent inconsistency between lake trout stomach contents and their stable isotope values.

Bow Lake has high amounts of suspended glacial silt (finely ground clayey silt from the glacial inflow). In a typical glacial-fed lake, very little glacial silt is retained in the lake, but is mostly carried out of the lake with water flow (Smith et al., 1980). However, high proportions of glacial silt were found in Bow Lake sediments in the form of copepod fecal pellets (Smith and Syvitski, 1982). This was confirmed by a microscopic examination of *Hesperodiaptomus arcticus* gut contents, which were full of very fine sediment (personal data). This finding suggests that *H. arcticus* may be increasing the sedimentation of glacial silt in Bow Lake. If the glacial silt was contaminated with organochlorines, then *H. arcticus* may be bioaccumulating high concentrations of organochlorines as a result of ingesting glacial silt. Further, the presence of glacial silt throughout Bow Lake may be responsible for the constant concentrations of organochlorines observed in all sediment and water samples from the lake. Resolving this requires further study involving suspended sediment analyses (organochlorine and stable isotope analyses), and year-round collection of

zooplankton, including under ice, and glacier analyses (ice, meltwater, and glacial sediment)¹⁸.

This project was initiated because lake trout from Bow Lake had higher toxaphene concentrations than for other regional fish populations (Chapter I, this thesis), and it was hypothesized that Hesperodiaptomus arcticus in Bow Lake may be an important source of organochlorines to lake trout (Chapter II, this thesis). It would be of interest to compare Bow Lake with nearby mountain lakes to discern whether other lakes exhibit similar patterns. For example, Hector Lake lies a few kilometers south of Bow Lake in the Bow Valley, and is also fed by the Wapta Icefields in addition to water from Bow Lake via Bow River. Hector Lake is different from Bow Lake in several physical parameters, such as having a double basin, but is very similar in most respects, including chemical parameters and environmental surroundings (Smith et al., 1980; D. Donald, unpublished data, Environment Canada, 2365 Albert Street, Regina, SK, S4P-4K1; personal data). However, the organochlorine content of lake trout in Hector Lake was found to be much lower than in Bow Lake¹⁹ (Donald et al., 1993; D. Donald, unpublished data, Environment Canada). The main difference that could be discerned between the food webs of Hector and Bow Lakes was the zooplankton community composition. In Hector Lake, Hesperodiaptomus arcticus was a minor constituent (0.06 indiv. L⁻¹) of the zooplankton community, while the much smaller Diaptomus sicilis was the dominant zooplankter (1.3 indiv. L⁻¹)²⁰ (1994)

 $^{^{18}}$ Glacier and glacial meltwater analyses are currently underway with Dr. J. Blais conducting the studies (Department of Biological Sciences, University of Alberta, Edmonton, Alberta, T6G-2E9). 19 Lake trout in Hector Lake (10 fish, average weight = 979 \pm 30 g) sampled in 1993 had 8 μ g kg $^{-1}$ Σ PCB, and 14 μ g kg $^{-1}$ Σ DDT. In the same study, large lake trout from Bow Lake (10 fish, 3149 \pm 1196 g) had 43 μ g kg $^{-1}$ Σ PCB and 92 μ g kg $^{-1}$ Σ DDT, while small lake trout (10 fish, 739 \pm 337 g) had 22 μ g kg $^{-1}$ Σ PCB and 48 μ g kg $^{-1}$ Σ DDT (Donald et al., 1993). All concentrations are average wet weight values.

²⁰ In the Rocky Mountains, *Hesperodiaptomus arcticus* adult weights are approximately 64 μg dry weight (A. Hardie, *pers. comm.*, Department of Biological Sciences, University of Alberta, Edmonton,

survey, D. Donald, unpublished data, Environment Canada, 2365 Albert Street, Regina, SK, S4P-4K1). In contrast, *H. arcticus* was dominant in Bow Lake (1.7 indiv. L⁻¹; Chapter I, this thesis.) The size difference between *H. arcticus* and *D. sicilis* may be important in prey selection by lake trout, because lake trout will preferably prey on larger zooplankton (Donald et al., 1994). Alternatively, suspended glacial silt may be too large for *D. sicilis* to ingest, thereby less organochlorines may be introduced to the food web of Hector Lake. Resolving this requires further examination of the food web structure, the length of underlying food chains, and organochlorine dynamics in Hector and other neighbouring lakes.

In this study (Chapter II, this thesis), fillets with skin attached were analyzed, instead of whole fish. Lipid content of muscle is lower than for whole fish (Niimi and Oliver, 1989); therefore wet-weight values of organochlorines in fish muscle samples may be lower than in whole fish. As a result, wet-weight organochlorine content of whole fish calculated from fillet samples may be underestimated in relation to whole invertebrates analyzed for this study. Wet-weight organochlorine concentrations in muscle tissue (skin removed) of lake trout, brown trout (Salmo trutta), rainbow trout (Salmo garirdneri) and coho salmon (Oncorhynchus kisutch) collected in Lake Ontario typically were less than half of concentration values for whole fish (Niimi and Oliver, 1988). Lipid-normalization (the ratio of contaminant concentration in a sample to lipid proportion of that sample) usually removes much of the discrepancy between whole fish and fillet samples. For example, after lipid normalizing, the organochlorine values given by Niimi and Oliver (1989) of whole lake trout and muscle tissue became similar²¹. However, lipid-normalization can be problematic

AB, T6G-2E9), while *Diaptomus silicis* adult weights are $6.22 - 26.5 \mu g$ dry weight (Herzig et al., 1980).

²¹ Lake trout from Lake Ontario (n=10, lipid content =17.4 \pm 3% for whole fish, 7.3 \pm 2.2% for muscle samples) has average wet-weight values of 9966 \pm 3570 μ g kg⁻¹ Σ PCB and 160 \pm 94 μ g/kg⁻¹

because the lipid content in whole fish can vary throughout the year, but it is not known how the lipid content of muscle tissue varies in relation to whole fish (D. M. Whittle, pers. comm., Canada Centre for Inland Waters, Burlington, Ontario, L7R-4A6). For example, spawning in male lake trout causes changes in lipid reserves which influences organochlorine concentrations in whole fish, but muscle lipid content may remain unchanged. Female lake trout are more difficult to assess, because younger females exhibit less change in lipid proportions due to egg production than older females (D. M. Whittle, pers. comm.). In this study, lake trout and mountain whitefish fillets were analyzed with skin attached, including a layer of subcutaneous fat, which usually approximates whole fish values in these species (D. C. G. Muir, pers. comm., Department of Fisheries and Oceans, Freshwater Institute, 501 University Crescent, Winnipeg, MB, R3T-2N6). Nevertheless, further analyses of the Bow Lake organochlorine data set should include lipid normalized values, in addition to wet weight values, to assess the influence of lipid content on organochlorine concentrations in biota, and to remove some of the influence of lipid covariation (Hebert and Keenleyside, 1995).

Total organochlorine concentrations were used instead of individual organochlorine congeners to investigate general patterns in Bow Lake. Individual congeners within each organochlorine group (i.e. CB 153 or CB 180 instead of ∑PCB, or T2 or T12 instead of ∑CHB) have widely varying chemical properties affecting their bioaccumulation potential in biota and their distribution within the environment. An examination of all biologically important congeners and their distribution in Bow Lake using multivariate statistical analyses should lead to further insights of

DDT for whole fish, and wet-weight values of 3881 \pm 2069 μ g kg⁻¹ Σ PCB and 64 \pm 49 μ g kg⁻¹ DDT for fish muscle. When lipid normalized, whole lake trout has 57275.8 μ g kg⁻¹ Σ PCB and 919.5 μ g kg⁻¹ DDT, and muscle samples has 53164.4 μ g kg⁻¹ Σ PCB and 876.7 μ g kg⁻¹ DDT. Wet-weight data from Niimi and Oliver (1989).

organochlorine patterns in Bow Lake. Understanding the chemical attributes of each congener in relation to organochlorine patterns in biota, water and sediment will further define the organochlorine dynamics in Bow Lake.

Studies in the lakes of the Canadian Rocky Mountain Parks are facilitated by the demonstration that fin ray δ^{13} C values can be used to predict muscle stable carbon isotope ratios, because fish can be sampled without killing them. There was a significant linear correlation between the muscle and fin ray stable isotope values, with fin rays being isotopically heavier in δ^{13} C than muscle tissue. However, stable carbon isotope fractionation between muscle and fin ray tissues was significantly different for fish from different regions. For instance, Bow Lake mountain whitefish and lake trout had a difference of 1.2-1.5% between fin ray and muscle tissue, while Yukon lake whitefish and lake trout had 3.2-3.8% difference. The similarity between Bow Lake mountain whitefish and lake trout δ^{13} C fin ray - muscle differences is intriguing given that the fish species have different dietary patterns and different lipid proportions. As a result, I suggested that regional variables, such as ambient temperatures, may be important for this fractionation difference. This does not appear to be the case for δ^{13} C differences between scales and muscle tissue, because two studies, one in Nevada, USA (Estep and Vigg, 1985) and one in Ontario, Canada (Kiriluk et al. 1996) have reported very similar δ^{13} C difference (2-3%) between scales and muscle.

For fin ray and muscle $\delta^{15}N$ values, there was a broader variation than found for $\delta^{13}C$ values. However, there was indication that the $\delta^{15}N$ of fin rays can be used to predict muscle $\delta^{15}N$ values in some species such as lake trout. The variation in $\delta^{15}N$ values between fin ray and muscle tissue tended to be relatively consistent for lake trout from Bow Lake and from the Yukon, which is different than the findings of Estep and

Vigg (1985) which showed variation between $\delta^{15}N$ values of scales and muscle to be inconsistent. Before sampling, fin ray $\delta^{13}C$ and $\delta^{15}N$ values need to be "calibrated" by sub-sampling a small number of fish within each study lake to determine the local fractionation difference and correlation between muscle and fin rays. A sampling program covering a wide geographical area focusing on a few cosmopolitan fish species, such as lake trout, would contribute substantially towards the understanding of stable isotope fractionation, and environmental and physiological influences on stable isotope ratios in fish²².

²² This project is underway, with plans to obtain data from lake trout and other fish species collected by other researchers. This includes Lake Winnipeg, Manitoba, and several lakes in the Northwest Territories and in Banff National Park, Alberta.

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Appendix 1. Bow Lake data set(1994-95). Organochlorine congeners are listed (ng g-1 wet weight). All congeners are volume-corrected. F1 and F2 are % recovery of PCB 30 and OCN internal standards. Detection limits are 0.1-0.2 ng/g.

CODE	ТУРЕ	ΣСНВ	ΣDDT	o.p'-DDF	p,p'-DDE
10813	LAKE TROUT	58.98	16.72	0.26	8.10
	LAKE TROUT	219.5	58.79	0.64	22.51
10815	LAKE TROUT	59.96	24.14	0.28	12.98
10816	LAKE TROUT	82.15	24.25	0.35	9.60
10817	LAKE TROUT	147.2	43.01	0.58	16.67
10818	LAKE TROUT	195.65	62.37	0.98	28.88
10819	LAKE TROUT	55.88	13.46	0.17	< 0.01
10820	LAKE TROUT	4.53	12.29	0.03	6.62
10821	LAKE TROUT	136.2	41.18	0.70	16.73
10823	LAKE TROUT	100.1	8.18	0.33	0.00
10824	LAKE TROUT	167.6	11.95	0.36	0.68
10826	LAKE TROUT	58.70	2.76	0.19	0.19
10828	LAKE TROUT	31.85	7.74	0.20	2.76
10843	LAKE TROUT	36.59	15.76	0.16	6.43
10844	LAKE TROUT	31.92	11.59	0.18	5.37
10845	LAKE TROUT	25.07	0.68	0.06	< 0.01
10829	MOUNTAIN WHITEFISH	51.92	13.10	0.11	5.93
10830	MOUNTAIN WHITEFISH	22.81	8.31	< 0.01	3.36
10833	MOUNTAIN WHITEFISH	30.42	8.12	< 0.01	0.59
10834	MOUNTAIN WHITEFISH	24.30	10.68	0.12	5.88
10835	MOUNTAIN WHITEFISH	4.47	7.52	0.05	5.72 5.74
10836	MOUNTAIN WHITEFISH	29.52 38.99	10.99 21.57	0.07	13.99
10837 10838	MOUNTAIN WHITEFISH MOUNTAIN WHITEFISH	7.44	7.33	0.23	4.42
10836	MOUNTAIN WHITEFISH	26.15	8.47	0.12	7.32
CH1	MTN WHITEFISH (IN LAKE TROUT STOMACH)	10.94	0.92	0.07	0.00
195	HESPERODIAPTOMUS ARCTICUS	170.7	32.88	0.35	0.11
495	HESPERODIAPTOMUS ARCTICUS	83.26	16.04	0.36	4.70
595	HESPERODIAPTOMUS ARCTICUS	99.05	18.45	0.22	5.53
1395	MIXED ZOOPLANKTON	37.16	8.07	0.13	1.63
1695	MIXED ZOOPLANKTON	126.45	20.49	0.31	4.24
295	GAMMARUS LACUSTRIS	36.80	9.12	0.13	3.47
995	GAMMARUS LACUSTRIS	27.10	8.86	< 0.01	3.10
895	LYMNAEIDAE	1.73	0.55	0.03	0.17
395	TIPULIDAE	1.82	2.40	< 0.01	1.82
695	TIPULIDAE	4.48	1.84	< 0.01	1.31
S1	LAKE INFLOW SURFACE SEDIMENT	0.230	0.168	< 0.01	0.080
S3	LAKE CENTRE SURFACE SEDIMENT	0.220	0.327	< 0.01	0.089
S5	SEDIMENT CORE SLICE 2	0.320	1.492	< 0.01	0.672
S7	SEDIMENT CORE SLICE 3	0.140	0.096	< 0.01	0.007
58	SEDIMENT CORE SLICE 4	0.200	0.037	< 0.01 < 0.01	0.005
S9 W495	SEDIMENT CORE SLICE 5 JULY '95 LAKE INFLOW WATER	0.130 0.310	0.065 0.032	< 0.01	0.002 0.006
W1095	AUG 26 '95 LAKE INFLOW WATER	0.310	0.052	< 0.01	0.007
W895	AUG 26 '95 LAKE CENTRE WATER	0.330	0.034	< 0.01	0.007
W595	SEPT 14 '95 LAKE OUTFLOW WATER	0.200	0.030	< 0.01	0.002
11333	OCI 1 14 00 DAIL OUT LOVY VYATEL	0.200	0.020	- 3.01	0.002

CODE	o.p'-DDD	ם.d'-DDD	o.p'-DDT	p,p'-DDT	ΣНСН	α-HCH	в-нсн	γ-HCH	δ-НСН
10813	0.58	< 0.01	1.75	6.03	1.53	1.16	0.00	0.37	< 0.01
10814	1.35	10.09	5.91	18.29	3.19	2.11	0.00	0.81	0.28
10815	0.43	3.73	1.57	5.14	1.04	0.74	0.00	0.23	0.07
10816	0.72	4.54	2.09	6.95	2.10	1.52	0.00	0.47	0.12
10817	1.10	7.29	4.99	12.39	2.33	1.58	0.00	0.53	0.22
10818	1.40	8.42	7.56	15.13	2.14	1.43	0.00	0.41	0.30
10819	< 0.01	< 0.01	13.30	0.00	0.86	0.67	0.00	0.19	< 0.01
10820	0.42	0.00	1.27	3.94	1.02	0.79	0.00	0.23	< 0.01
10821	1.04	6.96	3.88	11.88	2.47	1.90	0.00	0.58	< 0.01
10823	0.48	< 0.01	2.26	5.11	0.23	0.00	0.00	0.23	< 0.01
10824	0.82	< 0.01	3.45	6.63	1.11	1.11	0.00	0.00	< 0.01
10826	0.55	< 0.01	1.83	0.00	1.55	1.12	0.00	0.33	0.11
10828	0.22	1.37	0.71	2.48	0.49	0.49	0.00	0.00	< 0.01
10843	0.32	3.15	0.91	4.79	0.95	0.62	0.00	0.25	0.08
10844	0.27	1.93	0.82	3.02	0.72	0.49	0.00	0.18	0.05
10845	0.15	0.00	0.48	0.00	0.32	0.26	0.00	0.06	< 0.01
10829	0.29	2.03	0.96	3.79	1.23	0.88	0.00	0.36	< 0.01
10830	0.27	1.96	0.44	2.28	0.95	0.91	0.00	0.00	0.05
10833	0.42	2.92	0.41	3.78	0.75	0.47	0.00	0.17	0.10
10834	0.34	2.15	0.41	1.78	0.95	0.66	0.00	0.22	0.06
10835	0.08	0.68	0.18	0.81	0.22	0.14	0.00	0.05	0.02
10836	0.45	1.85	0.43	2.45	0.93	0.66	0.00	0.22	0.05
10837	0.21	1.68	1.31	4.10	0.70	0.49	0.00	0.13	0.08
10838	0.26	2.23	0.34	0.00	0.25	0.00	0.00	0.16	0.08
10841	0.33	0.08	0.62	0.00	0.96	0.68	0.00	0.26	0.02
CH1	0.07	0.00	0.30 8.48	0.48	0.34 1.94	0.23 1.28	0.00	0.09 0.66	< 0.02
195 495	1.17 1.22	6.78 4.80	2.69	15.98 2.28	3.00	1.94	0.00	1.06	< 0.01
595	0.80	4.22	3.71	3.98	1.31	0.79	0.00	0.52	< 0.01
1395	1.61	2.15	1.38	1.17	1.93	0.86	0.00	0.99	0.09
1695	1.07	4.79	4.29	5.79	3.30	1.71	0.00	1.11	0.47
295	0.78	2.94	1.53	0.82	2.06	1.23	0.00	0.83	< 0.01
995	1.68	1.27	0.79	2.02	1.09	0.60	0.00	0.50	< 0.01
895	0.04	0.21	0.10	< 0.01	0.03	0.03	0.00	0.00	< 0.01
395	< 0.01	0.58	< 0.01	< 0.01	0.46	0.28	0.00	0.18	< 0.01
695	0.12	0.41	< 0.01	< 0.01	0.29	0.12	0.00	0.17	< 0.01
S1	0.006	0.082	< 0.01	< 0.01	0.058	0.033	0.000	0.015	< 0.01
S3	0.006	0.088	0.017	0.126	0.070	0.035	0.000	0.020	< 0.01
S5	0.029	0.069	0.038	0.685	0.015	0.002	0.000	0.013	0.024
S7	0.032	0.000	0.004	0.053	0.010	0.000	0.000	0.010	< 0.01
S8	0.027	0.006	< 0.01	< 0.01	0.018	0.003	0.000	0.015	< 0.01
S9	0.029	< 0.01	0.008	0.026	0.013	0.006	0.000	0.007	< 0.01
W495	< 0.01	0.001	0.016	0.009	0.501	0.366	0.000	0.135	*
W1095	0.003	0.016	0.011	0.017	0.478	0.354	0.000	0.124	*
W895	0.014	0.010	0.018	0.043	0.492	0.357	0.000	0.136	*
W595	0.003	0.008	0.004	0.006	0.445	0.317	0.004	0.125	*

CODE	ΣCHLOR	heptacir	OCSTYR	C	C1A	C1B/U6	C2/U-5	C3	C5	U3
10813	9.14	< 0.01	80.0	< 0.01	< 0.01	0.04	< 0.01	< 0.01	0.54	0.13
10814	14.28	< 0.01	< 0.01	< 0.01	< 0.01	0.27	< 0.01	< 0.01	0.23	< 0.01
10815	2.46	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10816	3.67	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10817	9.13	< 0.01	< 0.01	< 0.01	0.16	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10818	7.95	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	1.32	< 0.01
10819	7.76	< 0.01	0.06	< 0.01	< 0.01	0.04	< 0.01	< 0.01	0.45	< 0.01
10820	7.18	< 0.01	< 0.01	< 0.01	< 0.01	0.03	0.09	< 0.01	< 0.01	0.10
10821	19.23	< 0.01	0.27	< 0.01	< 0.01	< 0.01	0.23	< 0.01	1.04	< 0.01
10823	12.33	< 0.01	< 0.01	< 0.01	< 0.01	0.04	0.10	< 0.01	0.65	< 0.01
10824	20.11	< 0.01	< 0.01	< 0.01	< 0.01	0.10	< 0.01	< 0.01	1.10	0.24
10826	7.02	< 0.01	< 0.01	< 0.01	0.67	< 0.01	< 0.01	< 0.01	0.54	< 0.01
10828	4.50	< 0.01	0.06	< 0.01	0.34	< 0.01	< 0.01	< 0.01	0.01	< 0.01
10843	1.38	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10844	1.42	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10845	2.72	< 0.01	< 0.01	< 0.01	0.26	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10829	4.78	< 0.01	0.01	< 0.01	< 0.01	0.02	< 0.01	0.05	< 0.01	< 0.01
10830	2.64	< 0.01	< 0.01	< 0.01	0.12	< 0.01	< 0.01	< 0.01	0.30	< 0.01
10833	5.76	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.38	< 0.01
10834	4.88	< 0.01	0.02	< 0.01	< 0.01	0.02	< 0.01	< 0.01	0.32	< 0.01
10835	1.38	< 0.01	0.02	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.05 < 0.01	< 0.01 < 0.01
10836	1.81	0.27	< 0.01 < 0.01	< 0.01	< 0.01 < 0.01	< 0.01 < 0.01	< 0.01 < 0.01	< 0.01	< 0.01	< 0.01
10837 10838	1.84 2.25	< 0.01 < 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10836	3.12	< 0.01	0.02	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
CH1	1.50	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
195	10.50	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
495	14.28	< 0.01	1.62	< 0.01	2.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
595	11.94	< 0.01	< 0.01	< 0.01	1.36	0.37	< 0.01	< 0.01	< 0.01	< 0.01
1395	7.29	< 0.01	< 0.01	< 0.01	0.99	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
1695	15.51	< 0.01	< 0.01	< 0.01	2.47	0.82	< 0.01	< 0.01	< 0.01	< 0.01
295	5.50	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
995	3.99	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
895	0.65	< 0.01	< 0.01	< 0.01	0.07	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
395	2.57	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
695	1.80	< 0.01	< 0.01	< 0.01	0.12	0.18	< 0.01	< 0.01	< 0.01	< 0.01
S1	0.072	< 0.01	< 0.01	< 0.01	0.013	0.008	0.007	< 0.01	0.019	< 0.01
S3	0.058	< 0.01	< 0.01	< 0.01	0.008	0.003	< 0.01	< 0.01	0.013	< 0.01
S5	0.174	< 0.01	< 0.01	< 0.01	0.072	0.022	< 0.01	< 0.01	< 0.01	0.063
S7	0.027	< 0.01	< 0.01	< 0.01	0.011	< 0.01	< 0.01	< 0.01	0.006	< 0.01
S8	0.045	0.004	< 0.01	< 0.01	0.014	< 0.01	0.014	0.008	< 0.01	< 0.01
S9	0.034	< 0.01	< 0.01	< 0.01	0.009	< 0.01	0.000	< 0.01	0.022	< 0.01
W495	0.127	< 0.01	< 0.01	< 0.01	< 0.01	0.016	0.019	0.012	0.039	< 0.01
W1095	0.182	< 0.01	< 0.01	0.009	0.017	0.020	0.006	0.004	0.007	0.070
W895	0.148	0.009	< 0.01	< 0.01	0.012	0.016	0.020	0.014	< 0.01	0.029
W595	0.081	0.001	< 0.01	< 0.01	0.022	0.006	0.005	0.002	0.013	< 0.01

CODE	U1	OXYCLR	T-CHLOR	C-CHLOR	T-NONA	C-NONA	н. ЕРОХ	DIELDRIN
10813	0.03	0.90	< 0.01	< 0.01	3.61	2.61	1.20	3.18
10814	< 0.01	< 0.01	2.19	< 0.01	3.18	6.07	2.34	6.54
10815	< 0.01	< 0.01	0.49	< 0.01	1.21	< 0.01	0.76	2.08
10816	< 0.01	< 0.01	0.74	< 0.01	1.60	< 0.01	1.33	3.34
10817	< 0.01	< 0.01	1.19	< 0.01	6.42	< 0.01	1.37	3.55
10818	0.07	< 0.01	1.36	< 0.01	3.85	< 0.01	1.35	3.50
10819	0.04	0.92	< 0.01	< 0.01	3.05	2.64	0.57	1.56
10820	< 0.01	0.59	0.44	1.11	2.50	1.64	0.69	0.00
10821	0.05	1.40	1.28	3.11	6.36	4.18	1.33	3.37
10823	0.03	< 0.01	1.10	1.84	4.97	2.94	0.67	1.73
10824	0.03	1.45	1.39	3.08	7.02	4.77	0.93	2.59
10826	< 0.01	< 0.01	< 0.01	< 0.01	3.37	2.44	< 0.01	3.34
10828	< 0.01	0.36	0.56	0.64	1.30	0.85	0.39	1.00
10843	< 0.01	< 0.01	0.59	< 0.01	0.80	< 0.01	< 0.01	< 0.01
10844	< 0.01	< 0.01	0.26	< 0.01	0.67	< 0.01	0.49	< 0.01
10845	< 0.01	< 0.01	< 0.01	< 0.01	1.35	0.97	0.15	0.44
10829	< 0.01	0.26	0.23	0.55	1.58	1.40	0.47	1.39
10830	< 0.01	< 0.01	0.12	0.47	0.25	1.39	< 0.01	0.64
10833	< 0.01	0.58	0.53	1.20	1.02	1.56	0.50	1.28
10834	< 0.01	0.42	0.37	0.77	1.49	0.97	0.49	1.13
10835	< 0.01	0.12	0.08	0.17	0.43	0.42	0.09	0.19
10836	< 0.01	< 0.01	0.11	0.29	1.05	< 0.01	0.09	0.30
10837	< 0.01	< 0.01	0.26	0.50	0.53	< 0.01	0.54	1.28
10838	< 0.01	0.43	< 0.01	< 0.01	1.01	0.82	< 0.01	< 0.01 0.78
10841	< 0.01	0.42 0.21	< 0.01	< 0.01	1.39	1.28 0.26	< 0.01 < 0.01	0.78
CH1 195	< 0.01 < 0.01	1.03	0.10 1.29	0.22 2.16	0.71 1.50	2.74	1.78	2.98
495	< 0.01	1.18	1.40	3.58	1.63	< 0.01	2.87	4.35
595	0.10	0.84	0.78	1.48	3.38	2.17	1.45	1.96
1395	< 0.01	1.81	0.66	1.16	1.42	1.24	< 0.01	1.84
1695	< 0.01	1.33	1.47	2.29	3.52	3.62	< 0.01	4.77
295	< 0.01	0.75	0.55	1.39	0.88	1.22	0.70	1.46
995	< 0.01	0.33	0.35	0.79	1.17	0.81	0.54	1.63
895	< 0.01	< 0.01	< 0.01	< 0.01	0.10	< 0.01	0.48	0.09
395	< 0.01	1.54	0.19	0.19	0.42	0.22	< 0.01	0.59
695	< 0.01	0.12	0.07	0.15	0.40	0.17	0.59	0.42
S1	< 0.01	< 0.01	0.004	0.017	0.004	< 0.01	0.001	0.003
S 3	< 0.01	< 0.01	0.004	0.004	0.011	0.011	0.005	0.005
S5	< 0.01	< 0.01	0.002	0.007	0.006	< 0.01	0.003	0.005
S7	< 0.01	< 0.01	0.001	0.005	0.002	< 0.01	0.002	0.007
S8	< 0.01	< 0.01	< 0.01	< 0.01	0.001	< 0.01	0.004	0.006
S9	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.004	0.006
W495	< 0.01	0.001	0.003	0.011	0.005	0.004	0.017	0.033
W1095	< 0.01	0.003	0.005	0.012	0.003	0.004	0.022	0.037
W895	0.011	0.002	0.002	0.011	0.003	0.003	0.016	0.026
W595	< 0.01	0.001	0.003	0.007	0.002	0.002	0.017	0.025

CODE	ΣΡСΒ	7	6	8/5	19	18	17	24/27	16/32	26
10813	5.49	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.11	< 0.01	< 0.01
10814	20.25	< 0.01	< 0.01	< 0.01	0.14	< 0.01	0.12	0.18	< 0.01	< 0.01
10815	6.79	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.05	0.07	< 0.01
10816	7.64	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.06	0.07	< 0.01
10817	6.96	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10818	17.32	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.18	0.15	0.20	0.72
10819	5.17	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.07	< 0.01	0.15
10820	2.50	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.05	< 0.01	< 0.01
10821	7.39	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.32	< 0.01	< 0.01
10823	11.58	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.08	< 0.01	0.10
10824	13.08	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.17	< 0.01	< 0.01
10826	5.83	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.06	< 0.01	< 0.01
10828	1.65	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.04	< 0.01	0.12
10843	2.39	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10844	2.91	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.03	< 0.01	< 0.01
10845	0.63	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.03	< 0.01	< 0.01
10829	5.39	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.04	< 0.01	0.06
10830	3.18	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.03	< 0.01	< 0.01
10833	3.23	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10834	3.33	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.04	< 0.01	< 0.01
10835	2.53	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01 < 0.01	< 0.01	< 0.01	< 0.01
10836 10837	4.76 9.58	< 0.01 < 0.01	< 0.01 < 0.01	< 0.01 < 0.01	< 0.01	< 0.01	< 0.01	< 0.01 0.06	< 0.01 < 0.01	< 0.01
10838	1.42	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10841	1.91	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
CH1	2.10	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
195	5.43	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.09	0.21	< 0.01
495	4.68	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
595	28.35	< 0.01	0.55	< 0.01	< 0.01	< 0.01	0.36	< 0.01	0.36	0.66
1395	1.98	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
1695	3.73	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
295	3.68	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
995	3.69	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.31
895	0.49	< 0.01	< 0.01	< 0.01	< 0.01	0.03	< 0.01	< 0.01	0.03	< 0.01
395	3.49	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
695	2.42	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
S1	1.511	< 0.01	0.016	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.013	< 0.01
S3	1.737	< 0.01	0.020	< 0.01	< 0.01	0.055	< 0.01	< 0.01	0.029	< 0.01
S5	2.574	< 0.01	< 0.01	< 0.01	< 0.01	0.371	< 0.01	< 0.01	0.129	0.123
S7	2.427	0.038	0.068	< 0.01	< 0.01	0.083	< 0.01	0.555	0.028	< 0.01
S8	5.063	< 0.01	< 0.01	< 0.01	< 0.01	0.165	< 0.01	0.806	0.060	0.036
S9	2.843	< 0.01	0.152	< 0.01	0.078	0.081	< 0.01	< 0.01	0.026	< 0.01
W495	0.602	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.004	0.004
W1095	1.898	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.029	< 0.01	< 0.01
W895	1.413	< 0.01	0.030	0.009	0.008	0.034	< 0.01	0.048	0.004	0.005
W595	1.327	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.004	0.003

CODE	25	31	28	33	22	45	46	52	49	47
10813	0.00	< 0.01	0.07	< 0.01	< 0.01	< 0.01	< 0.01	0.55	< 0.01	< 0.01
10814	0.00	< 0.01	0.20	< 0.01	0.05	< 0.01	< 0.01	0.62	< 0.01	< 0.01
10815	0.00	< 0.01	0.07	< 0.01	< 0.01	< 0.01	0.20	0.17	< 0.01	< 0.01
10816	0.00	< 0.01	0.11	< 0.01	< 0.01	< 0.01	0.27	0.43	< 0.01	0.05
10817	0.00	< 0.01	0.13	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10818	0.00	< 0.01	0.39	0.21	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.31
10819	0.00	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10820	0.00	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.13	0.35	< 0.01	0.09
10821	0.07	< 0.01	0.16	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10823	< 0.01	< 0.01	0.11	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10824	< 0.01	< 0.01	0.10	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.11
10826	< 0.01	< 0.01	0.13	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.14
10828	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10843	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10844	< 0.01	< 0.01	0.04	< 0.01	< 0.01	< 0.01	< 0.01	0.09	< 0.01	< 0.01
10845	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10829	< 0.01	< 0.01	0.07	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10830	0.04	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10833	< 0.01	< 0.01	0.04	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10834	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10835	0.05	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10836	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.11	< 0.01	< 0.01	< 0.01
10837	< 0.01	< 0.01	0.04	< 0.01	< 0.01	< 0.01	0.20	0.27	< 0.01	< 0.01
10838	< 0.01	< 0.01	0.03	< 0.01	< 0.01	< 0.01	< 0.01	0.07	< 0.01	< 0.01
10841	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
CH1	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
195	< 0.01	< 0.01	< 0.01	< 0.01	0.14	< 0.01	< 0.01	< 0.01	0.16	< 0.01
495	< 0.01	< 0.01	< 0.01	< 0.01	0.17	< 0.01	< 0.01	< 0.01	0.22	< 0.01
595	0.34	0.57	0.66	0.85	0.55	< 0.01	< 0.01	0.98	0.50	0.46
1395	0.06	0.08	0.09	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.09	0.09
1695	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
295	< 0.01 < 0.01	< 0.01 < 0.01	< 0.01	< 0.01 < 0.01	< 0.01 < 0.01	< 0.01 < 0.01	< 0.01	< 0.01	< 0.01 0.38	< 0.01 < 0.01
995 895	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.02	0.02	0.02
395	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.02	< 0.02	< 0.01
695	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01		< 0.01	< 0.01	< 0.01	< 0.01
S1	< 0.01	0.142	0.255	0.037	0.023	< 0.01	< 0.01	0.035	0.020	< 0.01
S3	0.016	0.134	0.252	0.034	0.020	< 0.01	< 0.01	0.025	< 0.01	0.005
S5	< 0.01	0.273	0.894	0.089	0.056	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
S7	0.019	0.163	0.696	0.048	0.022	< 0.01	< 0.01	0.043	0.025	< 0.01
S8	0.025	0.163	0.622	0.034	0.024	0.010	< 0.01	0.098	0.119	< 0.01
S9	0.025	0.080	0.266	0.228	0.015	< 0.01	< 0.01	0.046	0.035	< 0.01
W495	< 0.01	0.028	0.086	0.011	0.011	< 0.01	< 0.01	0.018	0.007	0.009
W1095	0.003	0.015	0.106	0.013	0.011	< 0.01	< 0.01	0.059	0.009	< 0.01
W895	0.005	0.051	0.150	0.012	0.014	< 0.01	< 0.01	0.045	0.009	< 0.01
W595	< 0.01	0.035	0.136	0.012	< 0.01	< 0.01	< 0.01	0.041	0.011	0.005

CODE	48	44	42	41/71	64	40	74	70/76	66/95	56/60
10813	0.05	< 0.01	0.10	< 0.01	0.13	< 0.01	0.05	0.10	< 0.01	0.07
10814	< 0.01	0.62	0.25	< 0.01	0.40	< 0.01	0.26	0.49	< 0.01	0.23
10815	< 0.01	0.12	< 0.01	< 0.01	0.11	< 0.01	0.05	0.16	< 0.01	< 0.01
10816	< 0.01	0.26	< 0.01	< 0.01	0.16	< 0.01	0.11	0.00	< 0.01	< 0.01
10817	< 0.01	0.18	0.05	< 0.01	0.15	0.17	0.25	0.00	< 0.01	< 0.01
10818	< 0.01	< 0.01	0.04	< 0.01	0.33	< 0.01	0.38	0.39	< 0.01	0.30
10819	0.22	< 0.01	0.20	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.04
10820	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.29	< 0.01
10821	< 0.01	0.35	0.16	< 0.01	0.21	< 0.01	0.12	0.26	< 0.01	0.11
10823	< 0.01	0.33	0.11	< 0.01	0.19	< 0.01	0.16	0.25	< 0.01	0.12
10824	< 0.01	< 0.01	0.17	< 0.01	0.24	< 0.01	0.15	0.31	< 0.01	0.15
10826	< 0.01	< 0.01	0.13	< 0.01	0.18	< 0.01	0.10	< 0.01	< 0.01	0.09
10828	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10843	< 0.01	0.10	< 0.01	< 0.01	< 0.01	< 0.01	0.04	< 0.01	< 0.01	0.03
10844	< 0.01	0.09	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.08	< 0.01	0.03
10845	< 0.01	0.06	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10829	< 0.01	< 0.01	0.04	< 0.01	0.04	< 0.01	< 0.01	0.07	< 0.01	< 0.01
10830	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10833	< 0.01	0.13	0.05	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.04
10834	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.09	0.10	< 0.01	0.04
10835	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.00	< 0.01	0.05 0.11	< 0.01
10836	< 0.01	0.09	0.05	0.03	0.08	< 0.01	0.16 0.20	0.15 0.44	< 0.01	< 0.01 0.09
10837	< 0.01	< 0.01 < 0.01	< 0.01 0.04	0.05 < 0.01	0.17	< 0.01	< 0.01	< 0.01	< 0.01	0.03
10838 10841	< 0.01	< 0.01	< 0.04	< 0.01	< 0.03	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
CH1	< 0.01	< 0.01	< 0.01	< 0.01	0.09	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
195	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
495	< 0.01	0.21	< 0.01	< 0.01	0.15	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
595	< 0.01	< 0.01	0.27	< 0.01	0.30	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
1395	< 0.01	0.06	< 0.01	< 0.01	< 0.01	0.37	< 0.01	< 0.01	< 0.01	0.03
1695	< 0.01	< 0.01	0.17	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
295	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
995	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
895	< 0.01	< 0.01	0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
395	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
695	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
S1	< 0.01	0.007	< 0.01	0.005	0.005	0.009	0.233	0.029	0.007	0.012
S3	< 0.01	< 0.01	< 0.01	0.004	0.003	< 0.01	0.192	0.025	0.015	< 0.01
S5	< 0.01	0.131	< 0.01	< 0.01	< 0.01	< 0.01	0.112	0.023	< 0.01	0.005
S7	< 0.01	0.018	< 0.01	< 0.01	0.014	< 0.01	0.165	0.028	0.033	0.011
S8	1.581	0.145	< 0.01	< 0.01	0.077	0.160	0.165	0.032	0.382	0.011
S9	0.789	0.026	< 0.01	0.029	0.034	< 0.01	0.069	0.017	0.223	0.016
W495	< 0.01	0.013	< 0.01	< 0.01	0.007	< 0.01	0.087	0.011	0.148	0.002
W1095	0.061	0.013	< 0.01	< 0.01	0.004	< 0.01	0.092	0.017	0.339	< 0.01
W895	0.131	0.006	0.002	< 0.01	0.004	< 0.01	0.139	0.027	0.304	0.003
W595	0.061	0.012	< 0.01	0.004	0.004	< 0.01	0.274	0.039	0.470	0.004

CODE	91	101	99	83	97	87	85	136	110	82
10813	< 0.01	0.53	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10814	1.02	1.71	< 0.01	< 0.01	< 0.01	0.95	< 0.01	< 0.01	1.75	< 0.01
10815	0.26	0.46	0.19	< 0.01	0.25	0.24	0.80	< 0.01	0.46	< 0.01
10816	< 0.01	0.62	< 0.01	< 0.01	0.30	0.28	< 0.01	< 0.01	0.69	< 0.01
10817	< 0.01	1.07	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10818	< 0.01	1.66	< 0.01	< 0.01	0.82	0.77	0.57	< 0.01	1.51	< 0.01
10819	0.21	0.45	< 0.01	< 0.01	0.15	< 0.01	0.26	< 0.01	< 0.01	< 0.01
10820	< 0.01	< 0.01	< 0.01	< 0.01	0.14	< 0.01	< 0.01	0.10	0.37	< 0.01
10821	< 0.01	1.06	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10823	< 0.01	0.89	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.93	< 0.01
10824	< 0.01	1.12	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10826	< 0.01	0.63	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.64	< 0.01
10828	< 0.01	0.18	< 0.01	< 0.01	< 0.01	0.07	< 0.01	< 0.01	0.13	< 0.01
10843	< 0.01	0.24	< 0.01	< 0.01	0.10	0.13	0.17	0.08	< 0.01	< 0.01
10844	0.10	0.27	< 0.01	< 0.01	0.11	0.10	< 0.01	< 0.01	< 0.01	< 0.01
10845	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.07	< 0.01	< 0.01	< 0.01	< 0.01
10829	0.12	< 0.01	0.17	< 0.01	< 0.01	0.07	0.31	< 0.01	0.22	< 0.01
10830	< 0.01	< 0.01	0.04	< 0.01	0.04	0.05	< 0.01	< 0.01	< 0.01	< 0.01
10833	0.06	0.34	< 0.01	< 0.01	< 0.01	0.12	< 0.01	< 0.01	0.33	< 0.01
10834	0.09	0.26	< 0.01	< 0.01	0.11	< 0.01	< 0.01	< 0.01	0.25	< 0.01
10835	< 0.01	0.10	0.07	< 0.01	< 0.01	0.03	0.14	< 0.01	0.09	< 0.01
10836	< 0.01	< 0.01	0.24	< 0.01	0.04	< 0.01	0.19	0.07	0.21	< 0.01
10837	< 0.01	0.59	0.28	< 0.01	0.36	0.32	1.55	< 0.01	0.65	< 0.01
10838	< 0.01	0.12	< 0.01	< 0.01	0.05	0.05	< 0.01	< 0.01	< 0.01	< 0.01
10841	< 0.01	0.18	< 0.01	< 0.01	0.08	0.07	< 0.01	< 0.01	< 0.01	< 0.01
CH1	< 0.01	0.39	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
195	< 0.01	1.03	< 0.01	< 0.01	0.33	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
495	< 0.01	< 0.01	< 0.01	< 0.01	0.22	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
595	< 0.01	< 0.01	0.84	< 0.01	0.37	0.57	0.34	0.53	1.16	< 0.01
1395	< 0.01	0.22	< 0.01 < 0.01	< 0.01	< 0.01	0.08	0.07	< 0.01	< 0.01	< 0.01
1695 295	< 0.01 < 0.01	0.75 < 0.01	< 0.01	< 0.01 < 0.01	0.19 < 0.01	< 0.01	< 0.01 < 0.01	< 0.01 < 0.01	< 0.01 0.64	< 0.01
995	< 0.01	0.55	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.04	< 0.01
895	< 0.01	0.10	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.02	< 0.01
395	< 0.01	0.28	< 0.01	< 0.01	< 0.01	0.31	< 0.01	< 0.01	0.32	< 0.01
695	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.24	< 0.01
S1	< 0.01	0.027	0.017	< 0.01	< 0.01	0.012	0.019	0.009	0.246	< 0.01
S3	< 0.01	0.031	0.020	< 0.01	< 0.01	< 0.01	< 0.01	0.008	0.189	< 0.01
S5	< 0.01	0.018	0.008	< 0.01	0.003	0.008	0.015	< 0.01	0.141	< 0.01
S7	< 0.01	0.020	0.007	< 0.01	0.003	0.007	< 0.01	0.004	0.221	< 0.01
S8	< 0.01	0.023	0.006	0.008	0.004	0.010	< 0.01	0.005	0.203	< 0.01
59	< 0.01	0.019	< 0.01	< 0.01	0.010	0.013	< 0.01	0.015	0.249	< 0.01
W495	< 0.01	0.007	0.004	< 0.01	< 0.01	0.003	0.004	< 0.01	0.057	< 0.01
W1095	< 0.01	0.025	< 0.01	0.023	0.010	0.024	< 0.01	< 0.01	0.867	0.040
W895	< 0.01	0.012	0.005	< 0.01	0.004	0.006	0.009	0.003	0.136	< 0.01
W595	< 0.01	0.015	0.005	< 0.01	0.004	0.006	0.008	< 0.01	0.119	< 0.01

	454	44440	440	440	1 494	444	404	440	450	400
CODE	151	144/13	149	118	134	114	131	146	153	132
10813	0.36	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.98	< 0.01
10814	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.05	< 0.01	< 0.01	3.22	< 0.01
10815	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.24	0.92	< 0.01
10816	0.41	< 0.01	0.63	< 0.01	< 0.01	< 0.01	< 0.01	0.31	1.08	< 0.01
10817	< 0.01	< 0.01	0.98	< 0.01	< 0.01	0.13	< 0.01	< 0.01	1.57	< 0.01
10818	< 0.01	< 0.01	1.75	< 0.01	< 0.01	0.07	< 0.01	0.32	2.66	< 0.01
10819	< 0.01	< 0.01	0.44	< 0.01	< 0.01	0.55	< 0.01	0.20	0.84	< 0.01
10820	0.21	< 0.01	0.40	< 0.01	< 0.01	0.00	0.09	< 0.01	0.65	< 0.01
10821	0.44	< 0.01	< 0.01	< 0.01	< 0.01	0.08	< 0.01	0.41	< 0.01	< 0.01
10823	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.12	< 0.01	0.57	1.90	< 0.01
10824	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	1.44	< 0.01	0.64	2.34	0.96
10826	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.06	< 0.01	0.28	1.03	0.60
10828	0.04	< 0.01	0.18	< 0.01	< 0.01	0.02	< 0.01	0.06	< 0.01	0.12
10843	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.04	< 0.01	0.15	< 0.01	< 0.01
10844	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.03	< 0.01	0.12	0.51	< 0.01
10845	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.02	< 0.01	< 0.01	0.00	< 0.01
10829	0.07	< 0.01	0.35	< 0.01	< 0.01	0.01	< 0.01	0.07	0.78	0.07
10830	< 0.01	< 0.01	0.20	0.17	< 0.01	0.04	< 0.01	0.04	0.47	< 0.01
10833	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.04	< 0.01	< 0.01	0.75	< 0.01
10834	0.18	< 0.01	< 0.01	< 0.01	< 0.01	0.03	0.10	0.16	0.57	< 0.01
10835	0.05	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.12	0.42	0.06
10836	0.12	< 0.01	0.45	< 0.01	< 0.01	0.04	< 0.01	< 0.01	0.55	0.10
10837	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.39	1.11	< 0.01
10838	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.02	< 0.01	0.07	0.27	< 0.01
10841	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.49	< 0.01
CH1	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.10	0.39	< 0.01
195	< 0.01	< 0.01	0.20	< 0.01	< 0.01	0.03	< 0.01	< 0.01	1.21	< 0.01
495	< 0.01	< 0.01	< 0.01	0.32	< 0.01	< 0.01	0.83	0.12	0.70	< 0.01
595	< 0.01	< 0.01	2.62	2.08	< 0.01	0.01	< 0.01	< 0.01	2.77	< 0.01
1395	< 0.01	< 0.01	0.08	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.20	0.11
1695	< 0.01	< 0.01	0.25	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.71	< 0.01
295	< 0.01	< 0.01	0.59	0.74	< 0.01	< 0.01	< 0.01	< 0.01	0.28	< 0.01
995	< 0.01	< 0.01	0.52	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.51	< 0.01
895	< 0.01	< 0.01	0.08	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.04	< 0.01
395	< 0.01	< 0.01	0.51	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.57	< 0.01
695	< 0.01	< 0.01	0.37	0.27	< 0.01	< 0.01	< 0.01	< 0.01	0.39	< 0.01
S1	< 0.01	0.035	0.024	0.019	< 0.01	< 0.01	< 0.01	< 0.01	0.017	0.067
S3 SF	< 0.01	< 0.01	0.023	0.015	< 0.01	< 0.01	< 0.01	< 0.01	0.021	< 0.01
S5	< 0.01	< 0.01	0.007	0.006	< 0.01	< 0.01	< 0.01	< 0.01	0.007	0.066
S7	< 0.01	< 0.01	0.013	0.010	< 0.01	< 0.01	< 0.01	< 0.01	0.009	< 0.01
S8 S0	< 0.01	0.007	0.012	0.013	< 0.01	< 0.01	< 0.01	< 0.01	0.008	< 0.01 < 0.01
S9	< 0.01	< 0.01	0.036	0.033	< 0.01	< 0.01	0.009	< 0.01	0.027	
W495	< 0.01	< 0.01 < 0.01	0.005	0.010	< 0.01	< 0.01	< 0.01	< 0.01	0.004	0.015
W1095	< 0.01	0.025	0.010	0.015	0.007	< 0.01 < 0.01	< 0.01	< 0.01	0.014	0.059
W895	< 0.01		0.014	0.016	0.001		< 0.01	0.002		0.054
W595	< 0.01	< 0.01	0.008	0.008	< 0.01	< 0.01	<u> </u>	< 0.01	0.004	0.001

CODE	105	141	179	137	130/176	138	158	178/129	175	187
10813	< 0.01	< 0.01	0.07	< 0.01	< 0.01	0.61	< 0.01	< 0.01	< 0.01	0.58
10814	< 0.01	0.65	0.25	< 0.01	< 0.01	1.88	0.18	< 0.01	< 0.01	1.84
10815	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.58	< 0.01	< 0.01	< 0.01	0.50
10816	< 0.01	< 0.01	0.09	< 0.01	< 0.01	0.65	< 0.01	< 0.01	< 0.01	0.53
10817	< 0.01	0.26	0.05	< 0.01	< 0.01	0.98	0.05	< 0.01	< 0.01	< 0.01
10818	< 0.01	< 0.01	< 0.01	1.57	< 0.01	0.10	< 0.01	< 0.01	< 0.01	< 0.01
10819	< 0.01	< 0.01	0.03	< 0.01	< 0.01	0.53	0.05	< 0.01	< 0.01	< 0.01
10820	< 0.01	< 0.01	0.00	< 0.01	< 0.01	0.00	< 0.01	< 0.01	< 0.01	< 0.01
10821	< 0.01	< 0.01	0.11	< 0.01	< 0.01	1.02	0.10	< 0.01	0.14	< 0.01
10823	< 0.01	< 0.01	0.13	< 0.01	< 0.01	1.24	0.12	< 0.01	< 0.01	1.02
10824	< 0.01	< 0.01	0.17	< 0.01	< 0.01	1.35	0.15	< 0.01	< 0.01	1.37
10826	< 0.01	< 0.01	0.06	< 0.01	< 0.01	0.64	0.06	< 0.01	< 0.01	< 0.01
10828	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.20	< 0.01	< 0.01	< 0.01	0.15
10843	< 0.01	0.13	0.09	< 0.01	< 0.01	0.00	0.04	< 0.01	< 0.01	0.36
10844	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.39	0.03	< 0.01	< 0.01	< 0.01
10845	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.00	0.03	< 0.01	< 0.01	< 0.01
10829	0.08	0.05	< 0.01	< 0.01	< 0.01	0.67	0.07	< 0.01	< 0.01	0.31
10830	0.15	< 0.01	0.10	< 0.01	< 0.01	0.55	0.03	< 0.01	< 0.01	0.09
10833	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.06	0.03	< 0.01	< 0.01	< 0.01
10834	< 0.01	0.10	< 0.01	< 0.01	< 0.01	0.45	0.04	0.05	< 0.01	< 0.01
10835	< 0.01	0.08	< 0.01	< 0.01	< 0.01	0.37	< 0.01	< 0.01	< 0.01	0.19
10836	< 0.01	< 0.01	< 0.01 0.15	< 0.04	< 0.01	0.46	0.03 < 0.01	0.05 < 0.01	< 0.01	0.22 < 0.01
10837	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01 < 0.01	0.79	0.02	< 0.01	< 0.01	< 0.01
10838 10841	< 0.01 < 0.01	0.05	< 0.01	< 0.01	< 0.01	0.41	0.02	< 0.01	< 0.01	< 0.01
CH1	< 0.01	0.06	< 0.01	< 0.01	< 0.01	0.37	< 0.01	< 0.01	< 0.01	< 0.01
195	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.84	0.04	< 0.01	< 0.01	0.62
495	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.60	0.06	0.14	< 0.01	0.32
595	< 0.01	< 0.01	0.68	< 0.01	< 0.01	2.52	0.17	0.61	< 0.01	< 0.01
1395	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.16	< 0.01	< 0.01	< 0.01	< 0.01
1695	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.49	< 0.01	< 0.01	< 0.01	0.00
295	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.56	< 0.01	< 0.01	< 0.01	0.17
995	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.48	< 0.01	< 0.01	< 0.01	0.22
895	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.03	< 0.01	< 0.01	< 0.01	0.01
395	< 0.01	0.19	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.22
695	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.36	< 0.01	< 0.01	< 0.01	0.22
S1	0.007	0.007	< 0.01	< 0.01	< 0.01	0.003	0.003	0.004	< 0.01	0.080
S3	< 0.01	0.015	< 0.01	< 0.01	< 0.01	0.037	0.001	0.022	< 0.01	0.434
S5	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.006	< 0.01	< 0.01	< 0.01	0.025
S7	0.001	0.003	< 0.01	< 0.01	0.002	0.010	< 0.01	0.004	< 0.01	0.037
S8	0.004	0.003	< 0.01	< 0.01	0.005	< 0.01	< 0.01	0.003	< 0.01	< 0.01
S9	0.025	0.017	< 0.01	< 0.01	0.013	0.031	< 0.01	0.023	< 0.01	< 0.01
W495	0.001	0.001	0.018	< 0.01	0.002	< 0.01	< 0.01	0.002	< 0.01	0.018
W1095	0.002	< 0.01	< 0.01	< 0.01	< 0.01	0.017	< 0.01	0.006	< 0.01	< 0.01
W895	0.004	0.002	0.005	< 0.01	0.002	0.013	0.002	0.002	< 0.01	0.024
W595	0.001	0.002	0.005	< 0.01	0.001	0.006	0.001	0.001	< 0.01	0.015

CODE	183	128	185	174	177	171	156	201/15	172/19	180
10813	< 0.01	0.05	< 0.01	< 0.01	0.07	< 0.01	< 0.01	0.18	0.07	0.40
10814	< 0.01	0.21	< 0.01	< 0.01	0.13	< 0.01	< 0.01	0.79	0.27	1.36
10815	< 0.01	0.06	< 0.01	< 0.01	0.05	< 0.01	< 0.01	0.00	0.08	0.42
10816	< 0.01	< 0.01	< 0.01	< 0.01	0.07	< 0.01	< 0.01	0.21	< 0.01	< 0.01
10817	< 0.01	0.11	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10818	< 0.01	< 0.01	0.17	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	1.27
10819	0.17	0.06	< 0.01	< 0.01	0.04	< 0.01	< 0.01	< 0.01	0.07	< 0.01
10820	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10821	< 0.01	0.10	< 0.01	< 0.01	0.08	< 0.01	< 0.01	< 0.01	0.14	0.72
10823	0.37	0.15	< 0.01	< 0.01	0.14	0.08	< 0.01	< 0.01	0.19	0.86
10824	< 0.01	0.14	< 0.01	< 0.01	0.07	< 0.01	< 0.01	< 0.01	0.18	1.00
10826	< 0.01	0.07	< 0.01	< 0.01	0.06	< 0.01	< 0.01	< 0.01	0.08	0.40
10828	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.06	< 0.01	0.14
10843	< 0.01	0.04	< 0.01	< 0.01	0.04	< 0.01	< 0.01	< 0.01	< 0.01	0.25
10844	0.16	0.03	< 0.01	< 0.01	0.03	< 0.01	< 0.01	< 0.01	< 0.01	0.26
10845	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.20
10829	0.26	0.10	< 0.01	< 0.01	0.05	< 0.01	< 0.01	< 0.01	0.05	0.42
10830	0.08	0.09	< 0.01	0.05	0.04	< 0.01	0.05	< 0.01	0.04	0.37
10833	< 0.01	0.05	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.10	0.05	0.33
10834	< 0.01	< 0.01	< 0.01	0.06	0.03	< 0.01	< 0.01	< 0.01	0.04	0.20
10835	0.07	0.05	< 0.01	0.05	0.04	< 0.01	< 0.01	< 0.01	0.06	0.23
10836	0.14	0.06	< 0.01	0.09	0.04	< 0.01	< 0.01	0.06	< 0.01	0.27
10837	< 0.01	0.10	< 0.01	0.15	0.07	< 0.01	< 0.01	0.17	0.09	0.49
10838	< 0.01	0.03	< 0.01	0.03	0.02	< 0.01	< 0.01	< 0.01	< 0.01	0.12
10841	< 0.01	< 0.01	< 0.01	0.05	0.02	< 0.01	< 0.01	< 0.01	< 0.01	0.22
CH1	0.07	0.05	< 0.01	0.05	0.03	< 0.01	< 0.01	< 0.01	< 0.01	0.19
195	0.13	< 0.01	< 0.01	0.00	0.12	< 0.01	< 0.01	< 0.01	< 0.01	0.45
495	0.14	0.07	< 0.01	0.17	< 0.01	0.12	< 0.01	< 0.01	< 0.01	0.26
595	1.08	0.14	< 0.01	< 0.01	< 0.01	0.63	< 0.01	< 0.01	< 0.01	1.75
1395	0.05	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.06
1695	0.18	< 0.01	< 0.01	0.18	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.21
295	< 0.01	< 0.01	< 0.01	0.14	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.29
995	< 0.01	< 0.01	< 0.01	0.12	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.23
895	< 0.01	< 0.01	< 0.01	0.02	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.02
395	< 0.01	< 0.01	< 0.01	0.22	< 0.01	< 0.01	0.12	< 0.01	< 0.01	0.26
695	0.14	< 0.01		0.15	0.08	< 0.01	< 0.01	< 0.01	< 0.01	0.21
S1	0.005	0.004	< 0.01 0.017	0.011	0.022 < 0.01	< 0.01 < 0.01	< 0.01	< 0.01	< 0.01 < 0.01	0.007
S3	0.003	< 0.027		< 0.022	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.018
S5 S7	< 0.01 < 0.01	< 0.01	< 0.01	0.020	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.003
S8	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.003
S9	0.039	< 0.01	< 0.01	0.021	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.007
W495	0.001	0.001	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.002
W1095	0.007	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.005
W895	0.001	0.002	< 0.01	0.002	0.002	< 0.01	< 0.01	< 0.01	< 0.01	0.003
W595	0.001	< 0.01	< 0.01	0.001	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.002
11333	0.001	, - 5.51	<u> </u>		- 5.51	- 3.3 (<u> </u>	0.01	J. J.	U.UUZ

CODE	193	191	200	170	198	199	196/203	189	195	194
10813	< 0.01	< 0.01	0.08	0.13	< 0.01	0.07	0.07	< 0.01	< 0.01	< 0.01
10814	0.17	< 0.01	< 0.01	< 0.01	< 0.01	0.15	< 0.01	< 0.01	< 0.01	0.06
10815	< 0.01	< 0.01	0.00	< 0.01	< 0.01	0.08	0.09	< 0.01	< 0.01	0.06
10816	< 0.01	< 0.01	0.18	< 0.01	< 0.01	0.07	< 0.01	< 0.01	< 0.01	< 0.01
10817	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10818	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	0.12	< 0.01	0.05	0.04	< 0.01
10819	0.07	< 0.01	0.06	0.12	< 0.01	0.07	0.07	< 0.01	< 0.01	0.04
10820	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10821	0.10	< 0.01	0.38	0.23	< 0.01	0.11	0.10	< 0.01	< 0.01	< 0.01
10823	0.15	< 0.01	0.17	0.33	< 0.01	0.20	0.22	0.03	< 0.01	< 0.01
10824	< 0.01	< 0.01	0.17	0.27	< 0.01	0.11	0.14	< 0.01	< 0.01	0.04
10826	0.05	< 0.01	0.07	0.14	< 0.01	0.07	0.08	< 0.01	< 0.01	< 0.01
10828	< 0.01	< 0.01	0.09	0.06	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
10843	< 0.01	< 0.01	0.00	< 0.01	< 0.01	0.08	0.05	< 0.01	< 0.01	< 0.01
10844	0.05	< 0.01	0.09	< 0.01	< 0.01	0.06	0.07	< 0.01	< 0.01	< 0.01
10845	0.03	< 0.01	0.00	0.11	< 0.01	0.07	< 0.01	< 0.01	< 0.01	< 0.01
10829	0.00	< 0.01	0.10	0.23	< 0.01	0.08	0.10	< 0.01	0.05	0.09
10830	0.06	< 0.01	0.03	0.15	< 0.01	0.04	0.04	< 0.01	< 0.01	0.06
10833	0.06	< 0.01	0.00	< 0.01	< 0.01	0.00	< 0.01	< 0.01	< 0.01	0.04
10834	0.10	< 0.01	0.03	0.06	< 0.01	0.06	0.06	< 0.01	< 0.01	0.07
10835	0.05	< 0.01	0.04	0.07	< 0.01	0.08	< 0.01	< 0.01	< 0.01	< 0.01
10836	0.06	< 0.01	0.20	< 0.01	< 0.01	0.08	0.07	< 0.01	< 0.01	0.04
10837	0.10	0.03	0.31	< 0.01	< 0.01	0.12	0.12	< 0.01	< 0.01	0.08
10838	0.02	< 0.01	0.04	0.05	< 0.01	0.03	< 0.01	< 0.01	< 0.01	< 0.01
10841	0.07	< 0.01	< 0.01	0.10	< 0.01	0.06	0.05	< 0.01	< 0.01	< 0.01
CH1	0.07	< 0.01	0.04	0.09	< 0.01	0.06	0.06	< 0.01	< 0.01	< 0.01
195	0.00	< 0.01	< 0.01	< 0.01	< 0.01	0.11	0.11	< 0.01	< 0.01	< 0.01
495	0.19	< 0.01	< 0.01	0.14	< 0.01	< 0.01	0.06	< 0.01	< 0.01	0.23
595	0.62	< 0.01	0.15	0.41	< 0.01	< 0.01	< 0.01	0.08	< 0.01	0.14
1395	0.04	< 0.01	< 0.01	0.04	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
1695	0.18	< 0.01	< 0.01	0.11	< 0.01	0.10	< 0.01	< 0.01	< 0.01	< 0.01
295	0.14	< 0.01	< 0.01	0.14	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
995	0.13	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01 < 0.01	< 0.01	< 0.01	< 0.01
895	< 0.01	0.03	< 0.01	< 0.01 0.15	< 0.01 0.11	0.03 < 0.01	< 0.01	< 0.01 < 0.01	< 0.01 < 0.01	< 0.01
395		< 0.01			< 0.11					< 0.01
695	< 0.01 0.005	< 0.01	< 0.01	< 0.01 0.010	< 0.01	< 0.01 0.002	< 0.01 0.001	< 0.01	< 0.01	< 0.01
S1 S3	0.005	< 0.01	< 0.01	< 0.010	< 0.01	< 0.002	< 0.001	< 0.01	< 0.01	< 0.01
S5	0.007	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
S7	< 0.019	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
S8	< 0.01	0.003	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
S9	0.021	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
W495	0.004	< 0.01	< 0.01	0.002	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
W1095	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
W895	0.012	< 0.01	< 0.01	0.005	< 0.01	0.001	0.001	< 0.01	< 0.01	< 0.01
W595	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01

CODE	205	206	209	F1	F2	TISSUE / type	%I IPID	δ ¹³ C (per mil)
10813	< 0.01	< 0.01	< 0.01	88	62	MUSCLE	5.80	-27.90
10814	< 0.01	0.04	< 0.01	86	85	MUSCLE	9.50	-29.94
10815	< 0.01	0.47	< 0.01	55	63	MUSCLE	6.97	-28.85
10816	< 0.01	< 0.01	< 0.01	78	80	MUSCLE	7.02	-27.59
10817	< 0.01	< 0.01	< 0.01	74	79	MUSCLE	7.02	-25.51
10818	< 0.01	0.22	< 0.01	77	58	MUSCLE	12.33	-30.69
10819	< 0.01	0.02	< 0.01	87	68	MUSCLE	5.77	-26.52
10820	< 0.01	< 0.01	< 0.01	76	82	MUSCLE	11.49	-27.00
10821	< 0.01	< 0.01	< 0.01	93	76	MUSCLE	9.84	-28.99
10823	< 0.01	0.32	< 0.01	94	100	MUSCLE	5.42	-24.80
10824	< 0.01	< 0.01	< 0.01	95	84	MUSCLE	8.03	-27.42
10826	< 0.01	< 0.01	< 0.01	70	75	MUSCLE	7.42	-28.80
10828	< 0.01	< 0.01	< 0.01	78	90	MUSCLE	3.49	-27.31
10843	< 0.01	0.10	< 0.01	74	70	MUSCLE	2.86	-24.88
10844	< 0.01	< 0.01	< 0.01	74	83	MUSCLE	3.28	-25.73
10845	< 0.01	< 0.01	< 0.01	92	100	MUSCLE	1.27	-25.07
10829	< 0.01	0.07	0.04	80	75	MUSCLE	10.01	-23.95
10830	< 0.01	< 0.01	< 0.01	76	65	MUSCLE	5.06	-21.26
10833	< 0.01	< 0.01	< 0.01	77	85	MUSCLE	3.32	-23.95
10834	< 0.01	< 0.01	< 0.01	85	81	MUSCLE	5.34	-22.82
10835	< 0.01	< 0.01	< 0.01	71	72	MUSCLE	1.50	-25.57
10836	< 0.01	0.03	< 0.01	64	62	MUSCLE	3.90	-23.99
10837	< 0.01	0.05	< 0.01	76	70	MUSCLE	5.07	-22.59
10838	< 0.01	< 0.01	< 0.01	65_	81	MUSCLE	2.87	~
10841	< 0.01	< 0.01	< 0.01	65	68	MUSCLE	3.81	-24.17
CH1	< 0.01	< 0.01	< 0.01	76	80	WHOLE	3.81	-22.81
195	< 0.01	< 0.01	< 0.01	100	100	WHOLE	15.20	-33.90
495	< 0.01	< 0.01	< 0.01	100	100	WHOLE	23.40	-33.19
595	< 0.01	0.12	< 0.01	100	100	WHOLE	6.20	-32.81
1395	< 0.01	< 0.01	< 0.01	100	100	WHOLE	6.50	-31.12
1695	< 0.01	< 0.01	< 0.01	100	100	WHOLE	11.30	22.02
295	< 0.01	< 0.01	< 0.01	100	100	WHOLE	9.90	-23.93
995	< 0.01	< 0.01	< 0.01	100	100	WHOLE WHOLE	5.17	-24.12
895	< 0.01 0.22	< 0.01 < 0.01	< 0.01	100 95	100 97	WHOLE	0.40 5.46	-25.82 -24.20
395	0.22	< 0.01	< 0.01	100	100	WHOLE	2.02	-24.20
695 S1	< 0.00	< 0.01	< 0.01 < 0.01	86	100	sediment	2.02	
S3	< 0.01	< 0.01	< 0.01	86	100	sediment	~~	
S5	< 0.01	< 0.01	< 0.01	89	100	sediment		~
S7	< 0.01	< 0.01	< 0.01	89	100	sediment	~	~
S8	0.031	< 0.01	< 0.01	92	100	sediment	~	~
S9	< 0.01	< 0.01	< 0.01	85	84	sediment	~	~
W495	< 0.01	< 0.01	< 0.01	100	100	water	~~~	~
W1095	< 0.01	< 0.01	< 0.01	100	100	water	~	~
W895	< 0.01	< 0.01	< 0.01	100	100	water	~	~
W595	< 0.01	< 0.01	< 0.01	100	100	water	~	~
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CODE	δ ¹⁵ N (per mil)	SEX		LENGTH (cm	
10813	5.82	М	7	465	1265
10814	5.81	M	11	463	1241
10815	6.12	M	12	490	1303
10816	6.51	F	9	446	1033
10817	6.49	F	12	445	1061
10818	5.78	F	9	489	1396
10819	6.14	M	9	478	1241
10820	6.45	M	9	476	1287
10821	6.02	M	9	461	1218
10823	7.08	F	13	484	1288
10824	6.65	F	13	489	1494
10826	6.24	F	Œ	438	987
10828	5.93	U	7	293	291
10843	6.55	М	8	330	421
10844	7.11	М	6	319	370
10845	7.55	F	10	335	360
10829	6.61	F	16	406	827
10830	7.65	F	9	353	504
10833	6.42	F	4	195	80
10834	6.66	U	3	155	39
10835	6.52	U	2	136	24
10836	6.49	F	8	291	275
10837	6.46	M	8	260	241
10838	~	F	5	252	160
10841	6.08	U	4	195	68
CH1	6.31	~	~	~	~
195	2.28	~	~	~	~
495	2.93	~	~	~	~
595	2.62	~	~	~	~
1395	1.82		~	~	~
1695	~	~	~	~	~
295	2.69	~		~	~
995	2.72	~		~	~
895	1.36	~	~	~	~
395	1.30	~	~	~	~~
695	~	~	~_	~	~
S1	~	~	~	~	
S3	~		~	~~	
S5	~		~	~	~
S7	~		~	~~	
S8	~	~	~_	~	~
<u>S9</u>	~	~	~	~	~
W495	~	~	~_	~	
W1095	~		~	~	~
W895	~		~	~	~
W595	~	~	~	~	~