DISTRIBUTION OF PHTHALATE ESTERS IN A MARINE MAMMAL FOOD CHAIN FROM CANADA'S EASTERN ARCTIC

by

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Title of Project

Distribution of Phthalate Esters in a Marine-mammal Food Chain from Canada's Eastern Arctic

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Abstract

Phthalate Esters (PEs) are used as additives in numerous commercial products including polyvinyl chloride. Eight individual PEs (DMP, DEP, DIBP, DBP, BBP, DEHP, DnOP, and DnNP) were analyzed in adult male beluga whales (*Delphinapterus leucas),* Arctic Cod (*Boreogadus saida*), sediment, and lichen (*Cladina rangeferina*) from Eastern Hudson's Bay to gain insight into (a) the geographic range of PEs, (b) the distribution of PEs in components of a marine-mammal food-chain, and (c) the relationship between PE biomagnification and K_{OA}'s. Lichen concentrations were obtained to estimate air concentrations for assessing the beluga's exposure to PEs through breathing. This is the first comprehensive examination of PEs in arctic ecosystems and in marine mammals. The mean ΣPE concentrations (ng/g lipid eq) for beluga, cod, sediment, and lichen are 14500 (4520-46500), 18200 (9500-34800), 23900(10500-54300), 8990 (4900-16500) and 8990(4900-16500) respectively. The mean ΣPE concentrations PEs do not appear to biomagnify from cod to beluga. Trophic dilution of DEHP may be occurring as $f_{cod} > f_{beluga}$ ($p<0.05$). BMF_{LS} did not increase with K_{OA} as was hypothesized for air breathing organisms because PEs are likely being metabolized. Because of metabolism, PEs are not ideal compounds to test the hypothesis that biomagnification increases with K_{OA} in marine mammals. Mean BSAFs for belugas were greater than unity for the intermediate and higher molecular weight phthalates with the exception of DEHP. For cod, only BBP, DEHP and DnOP had mean BSAFs greater than unity. $BAAF_{LS}$ (C_{beluga}/C_{air}) were greater than unity for all phthalates except DEHP. DEHP appears to behave differently than the other PEs with comparatively high lipid equivalent concentrations in cod and air relative to other ecosystem components. One possible explanation for this distribution

pattern is the efficient metabolic elimination of DEHP by belugas, which would limit magnification from the diet and air. Unlike certain POPs, PEs did not increase with age or length in adult males. Linear regressions of concentrations versus age and length exhibited negative slopes. The levels of PEs in Hudson's Bay fish are similar to the levels in fish from an urban ecosystem. This suggests that phthalates are likely being atmospherically transported to Polar Regions.

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1.0 Introduction

The contamination of remote regions by persistent organic chemicals has been well documented. Despite the relative absence of local sources of pollutants, remote regions like the Canadian Arctic have become vulnerable to numerous, potentially toxic, chemicals that are primarily emitted in the industrial regions of mid-latitudes. Most notably, persistent organic pollutants (POPs) have been found at significant levels in the Arctic environment and have been biomagnifying in marine and terrestrial food webs (*1-5*). Although POPs can be found at high levels throughout the world, their presence in the Arctic is of particular concern for many reasons. Many features of the North's unique climate and environment (e.g. permafrost, low sedimentation rates and the lack of natural contaminant sinks) can facilitate the availability of harmful chemicals to the regions ecosystems (*6-9*). From a human health perspective, exposure to POPs tends to be amplified in the Arctic due largely in part to the high trophic levels that indigenous populations like the Inuit occupy (*10-17*). This dietary exposure to POPs has resulted in PCB blood concentrations and daily intake levels of chlordane, toxaphene, dieldrin and PCBs, that exceed Health Canada and World Health Organization guidelines respectively (*13, 14*). Compared to southern Canadians, the levels of certain POPs (DDE, PCB, mirex, HCB and chlordane) in breast milk are up to tenfold higher in Inuit from Nunavik (*15-17*). The increased vulnerability to potential toxic effects associated with POPs has engaged Inuit interest groups and circumpolar countries to become more involved in the management of harmful anthropogenic pollutants both domestically and internationally.

Due to the evidence that has been accumulated regarding the toxic, bioaccumulative and persistent behaviour of POPs since the 1960s, many industrial and

agricultural chemicals have been banned or heavily regulated. In Canada, the dirty dozen POPs (i.e. DDT, chlordane, toxaphene, mirex, aldrin, dieldrin, endrin, heptachlor, PCBs, HCB, dioxins and furans) are regulated under the Toxic Substances Management Policy (TSMP) as Track I substances. According to the Canadian Environmental Protection Act (CEPA), Track I substances are partially defined as those having bioaccumulation factors (BAFs) or bioconcentration factors (BCFs) greater than 5000. When BAFs and BCFs are not available, categorization is based on the compound's octanol water partition coefficient (K_{OW}) where chemicals with a log $K_{OW} > 5$ are considered to be bioaccumulative. Under the TSMP, Track I substances are to be 'virtually eliminated' from the environment through prohibiting any further releases and removing or managing the substance if it is already present in the environment. Similar management criteria are used by the United States and Europe and are found in the United Nations Environment Programme's (UNEP) long-range transboundary air pollution protocol (LRTAP).

These bioaccumulation criteria have recently been adopted by UNEP's 2001 Stockholm Convention on Persistent Organic Pollutants. This legally binding agreement outlines international strategies for the management of the dirty dozen POPs, but unlike the preceding policies, the Stockholm Convention is guided by the precautionary principle and allows for chemicals that threaten to exhibit "serious or irreversible damage" to be regulated even if they do not meet the criteria (*18*). Many countries have embraced the opportunity for the inclusion of chemicals into the treaty without the full scientific certainty of their bioaccumulative, toxic and persistent behaviour. Conversely, other parties like the United States have challenged this possible expansion of regulated

substances and have yet to ratify the agreement (*19*). Flexibility towards the addition of compounds to the Stockholm Convention reflects the emerging concern for the potential impacts of compounds that are now largely unregulated such as polybrominated diphenyl ethers (PBDEs), endosulphan, polychlorinated naphthalenes (PCNs) and phthalate esters, all of which are manufactured in large quantities. In adopting the precautionary principle, the signatories inherently question the current set of management criteria that is in part based on a chemical's K_{OW}.

K_{OW} is a measure of how a chemical will partition between lipid and water in an organism (i.e. its hydrophobicity) and has consistently been associated with the extent to which a chemical will bioaccumulate. This association is based predominantly on the results of studies conducted on fish that can eliminate less hydrophobic compounds to the water via their gills. Air-breathing organisms have different mechanisms of chemical uptake and elimination and thus the criteria designed to protect aquatic life may be inappropriate for the protection of terrestrial organisms and marine mammals. Both fish and mammals can be exposed to contaminants through their diet via the absorption of chemical into the gastro-intestinal tract (GIT) and common mechanisms of chemical elimination include urine excretion and fecal egestion. However, fish can also absorb and eliminate chemical through gill ventilation whereas mammals absorb and eliminate contaminants through breathing. The comparable route of uptake in mammals is via absorption into the lungs when inhaling. Chemical can be eliminated to alveolar air during exhalation (Figure 1.1). This distinction intuitively suggests that the extent to which a chemical will partition between lipid and air (which can be represented by the octanol-air partition coefficient or K_{OA}) may be a more relevant chemical property when

determining the potential for bioaccumulation in air-breathing organisms. This theory was recently supported by the findings presented in Kelly and Gobas (2001 and 2003) (*5,20*).

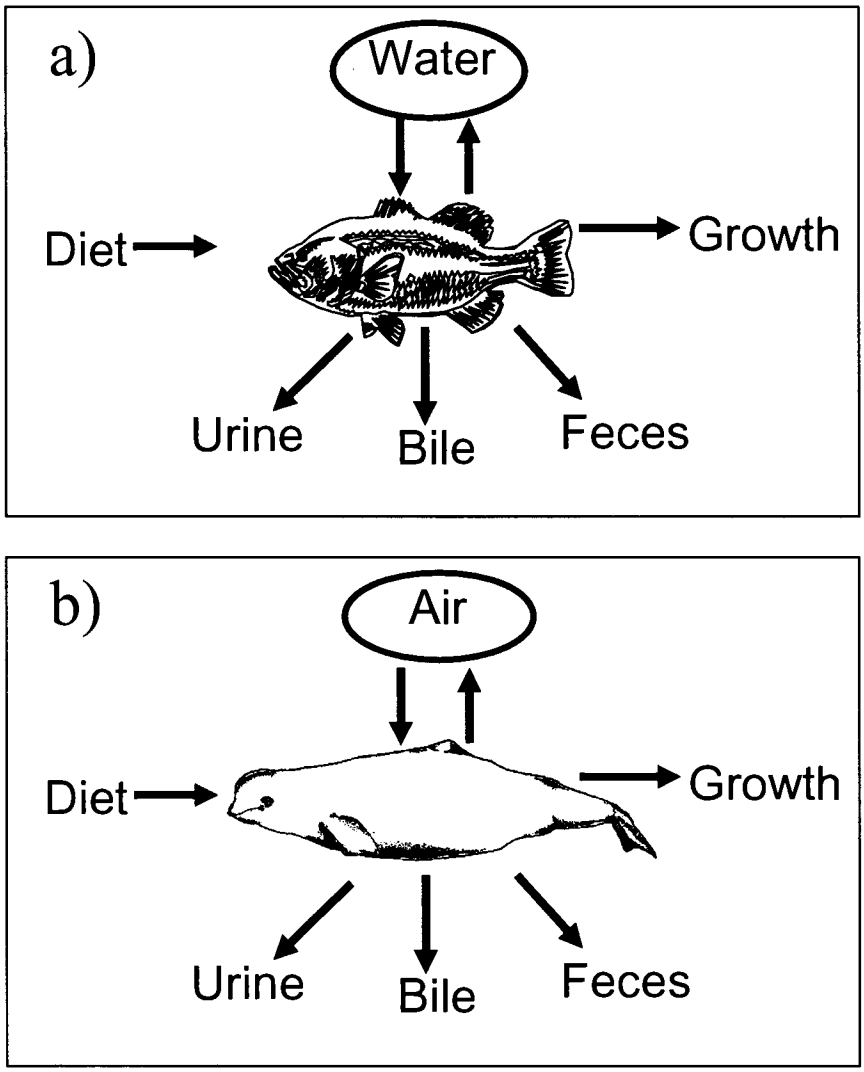


Figure 1.1 - The uptake and elimination mechanisms of chemical for a) aquatic organisms and b) air-breathing organisms

Kelly and Gobas (2001) found that chemicals with a log K_{OW}<5 were biomagnifying in the lichen-caribou-wolf food chain from the Western and Central Canadian Arctic and concluded that K_{OA} was a better indicator of biomagnification of a contaminant in terrestrial food chains, with K_{OA} being positively correlated with

biomagnification factors (BMF)(*5*). Furthermore, in Kelly and Gobas (2003), it was proposed that chemicals with a log K_{OA}>5 can biomagnify in the event of limited metabolic transformation and urine excretion (*20*).

The current study attempts to determine if a similar relationship exists for the biomagnification of contaminants in marine mammals. It is hypothesized that non-metabolized chemicals with a high K_{OA} will biomagnify in marine mammals, as it is believed that air-breathing organisms are unable to eliminate non-volatile chemicals despite a low K_{OW}, as they will partition to the animal's lipid rather than to the exhaled alveolar-air. The specific group of chemicals that will be investigated in this paper are phthalate esters.

Phthalate esters (PEs or phthalates) are a group of twenty-five compounds that are found in numerous commercial products as additives. Roughly 80% of phthalates that are manufactured are used as plasticizers in polyvinyl chloride (PVC). Phthalates act as a lubricant in PVC resulting in a more flexible product without compromising durability. Consequently phthalates are added to many vinyl products ranging from medical equipment to children's toys and construction materials. Other products in which phthalates are added include cosmetics, perfumes, adhesives and paints (*21*). It is estimated that 5.5 million tonnes of phthalates are produced globally every year largely due to the widespread use of PVC. As a result, PEs have become ubiquitous in the natural environment (*22*). One of the major concerns surrounding these chemicals is their endocrine disrupting behaviour. Some phthalates have been linked to reproductive effects and altered hormone levels (*23-27*).

Phthalate esters have a wide range of physical and chemical properties which suggest that they may not all exhibit the same fate once introduced into the environment. As a result, this study will look at eight individual phthalates that cover a spectrum of properties that could influence their bioaccumulative behaviour. The eight congeners and some of their properties are listed in Table 1.1 (*28, 29*). Their names refer to the composition of the two "R" groups that are attached to the ester groups on the cyclohexatriene ring (benzene-dicarboxylic acid) that characterizes a phthalate ester (Figure 1.2).

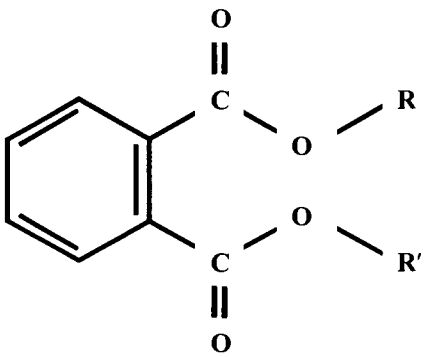


Figure 1.2 - The chemical structure of a phthalate ester

Table 1.1 - The abbreviations, molecular weight, K_{OW}, water solubility, vapour pressure and K_{OA} of the eight phthalate esters investigated in this study (Cousins and Mackay 2000, Staples et al. 1997).

Phthalate Ester	Abbr.	Molecular Weight (g/mol)	Log K_{OW}	Water Solubility (mg/L)	Vapour Pressure (Pa)	Log K_{OA}
Dimethyl	DMP	194.2	1.61	5220	0.263	7.01
Diethyl	DEP	222.2	2.54	591	6.48×10^{-2}	7.55
Diisobutyl	DIBP	278.4	4.27	9.9	4.73×10^{-3}	8.54
di-*n*-butyl	DBP	278.4	4.27	9.9	4.73×10^{-3}	8.54
Butyl benzyl	BBP	312.4	4.7	3.8	2.49×10^{-3}	8.78
Di(2-ethylhexyl)	DEHP	390.6	7.73	2.5×10^{-3}	2.52×10^{-5}	10.53
di-*n*-octyl	DnOP	390.6	7.73	2.5×10^{-3}	2.52×10^{-5}	10.53
di-*n*-nonyl	DnNP	418.6	8.6	6.0×10^{-4}	6.81×10^{-6}	11.03

The K_{OW}'s of the higher molecular weight phthalates would suggest that these chemicals are bioaccumulative in aquatic food chains due to their hydrophobicity (log K_{OW} >5). More importantly, the high K_{OA}'s indicate the potential for biomagnification in air-breathing organisms according to the above-mentioned theory, given that the K_{OA}'s for all the phthalate esters are greater than 10^5. Despite these properties, little evidence exists which suggests that phthalate esters are in fact bioaccumulative. Most phthalate esters are thought to be biotransformed before they are able to accumulate in organisms and subsequently food chains (*29*). Nevertheless, there are several gaps in PE research that have yet to be addressed specifically in regards to the geographic dispersion of the chemicals, their presence in the natural environment and the extent to which they may accumulate in high trophic level wildlife including marine mammals.

To date, there are no reported values of phthalate esters in arctic fish and wildlife and in general, there are virtually no descriptions of phthalate distribution in natural ecosystems; the majority of data that exists for phthalates originates from laboratory studies. Mackintosh et al. (2003) presented field values for a marine aquatic ecosystem on Canada's west coast and this study was the first comprehensive look at how phthalate esters are distributed in a food chain (*30*). Any additional field data are limited to isolated values of a given medium. There have also been no reported concentrations of phthalates in marine mammals.

This study will examine the distribution of eight phthalate esters in a portion of a marine ecosystem in Canada's Eastern Arctic, focusing on beluga whales (*Delphinapterus leucas)* from the Eastern Hudson's Bay population, their diet and their surrounding environment. Beluga whales are good candidates for such a study as they

are permanent residents of the North and could therefore provide us with an indication of the extent of PE distribution in a remote region.

Given the lack of field data in the Arctic, the underlying purpose of this study is to confirm whether phthalates are capable of traveling to the polar regions. More specifically, the primary objectives of this project are to (A) investigate the distribution of phthalate esters in a number of biotic and abiotic components of an Arctic ecosystem, including marine mammals, fish, sediment and air, and to (B) determine the relationship between octanol-air partioning coefficients and phthalate ester distribution in this environment. To meet the first objective, the goals are to (i) measure concentrations and assess fugacities of selected phthalate esters in *Delphinapterus leucas*, their diet as represented by Arctic cod (*Boreogadus saida)*, and the sediment with the intent of capturing information on primary routes of dietary exposure for the animals; (ii) estimate the level of selected phthalate esters in the air from the concentrations in lichen (*Cladina rangiferina)* using a vegetation-gas partitioning coefficient; and (iii) gain insight into the transfer of selected phthalate esters through the food chain by determining biomagnification factors (BMFs), biota-sediment accumulation factors (BSAFs) and biota-air accumulation factors. The second objective of the study will be met by identifying the magnitude of BMFs for a range of K_{OA}s, and to determine if a relationship between biomagnification and octanol-air partitioning exists. It is hypothesized that phthalate esters with a high K_{OA} have the potential to biomagnify in marine mammals ($C_{beluga} > C_{cod}$) if they are not metabolized. Under these conditions a positive relationship between BMFs and log K_{OA} is expected. If PEs are metabolized, this relationship will likely not hold.

A handful of studies on the diet of the beluga whale have indicated that the animals are opportunistic feeders (*31-33*). Belugas are known to consume a wide range of additional prey items ranging from fish such as capelin, halibut and herring to benthic organisms and invertebrates such as octopus, squid and shrimp although fatty acid and stable isotope analysis have suggested that Arctic Cod in particular is an important prey species (*31-33*). The trophic level of beluga is thus difficult to identify. Stable isotope and stomach content analysis of belugas from the high arctic indicate that the belugas occupy a trophic position of 3.9, whereas the trophic position of arctic cod is 2.5 or 3.6 for fish that are 14-16mm and 48-247mm respectively. Although the cod is considered a main prey species of beluga whale, lower-trophic level organisms such as those mentioned above, also represent important prey items (*32*). Analysis of the fatty acid composition of beluga blubber supports the stable-isotope evidence, indicating that cod is a significant prey species but that there are other fish and crustaceans that complete the diet (*33*). Thus despite the similarities in trophic position, arctic cod is consistently considered to be a valuable element of the beluga diet and therefore biomagnification of phthalate esters in beluga whale will be calculated using concentrations in cod.

Sediment will also be considered as a source of dietary exposure for the beluga. Beluga whales can dive to depths in excess of 400m and can dislodge benthic organisms from the bottom sediment using suction. This suction may be evidenced by the sediment and stones that are often found in the animal's stomachs. Consequently, sediment is analyzed as a potential source of contaminants in the beluga diet through either direct (intake of sediment) or indirect (intake of benthic worms that are in direct contact with the sediment) consumption.

By investigating concentrations in the diet, we are capturing information on a primary route of exposure for the marine mammals sampled: absorption of chemical via the gastro-intestinal tract (GIT). In terms of measuring additional routes of contaminant uptake, lichen samples have been analyzed for the purpose of estimating air concentrations of phthalate esters. Such measurements can provide insight into beluga whale exposure via air inhalation and provide a more complete picture of PE delivery into the Arctic environment via atmospheric transport. Concentrations for the environmental samples will be presented on a wet weight basis (ng/g wet wt) for the biota and a dry weight basis (ng/g dry wt.) for the lichen and sediment. For all matrices, a lipid equivalent concentration (ng/g lipid) and fugacity (nPa) will be presented as well.

Although this study focuses on only a small portion of an Arctic marine ecosystem, it will touch on aspects of phthalate ester research that have yet to be examined. It is hoped that the results of this study will improve our understanding of the behaviour and fate of phthalate esters in the natural environment by providing insight into the geographical distribution of these chemicals and their presence in wild, air-breathing, high trophic level organisms. Such awareness will assist in evaluating the risk associated with these widely produced chemicals, as well as other compounds, particularly in the North where an increased vulnerability to POP contaminants has been identified for subsistence-oriented Inuit peoples.

2.0 Methods

The methods used to complete this study will be presented in three separate subsections. Section 2.1 will discuss how environmental samples were collected in the field and Section 2.2 will outline the analytical methods that were used to quantify the levels of phthalate esters in the samples. Finally, Section 2.3 will summarize how the data were analyzed and normalized.

2.1 Field Sampling Methods

2.1.1 Study Area and Design

Collection of field samples occurred along the eastern coast of Hudson's Bay between the town of Umiujaq (56°05' N, 76°05' W) and the Nastapoka River estuary (56°55' N, 76°05' W) in the Northern Quebec region of Nunavik over three summers (2000-02). The samples were collected between the shore and the Nastapoka Islands, which are approximately 2km west of the coast.

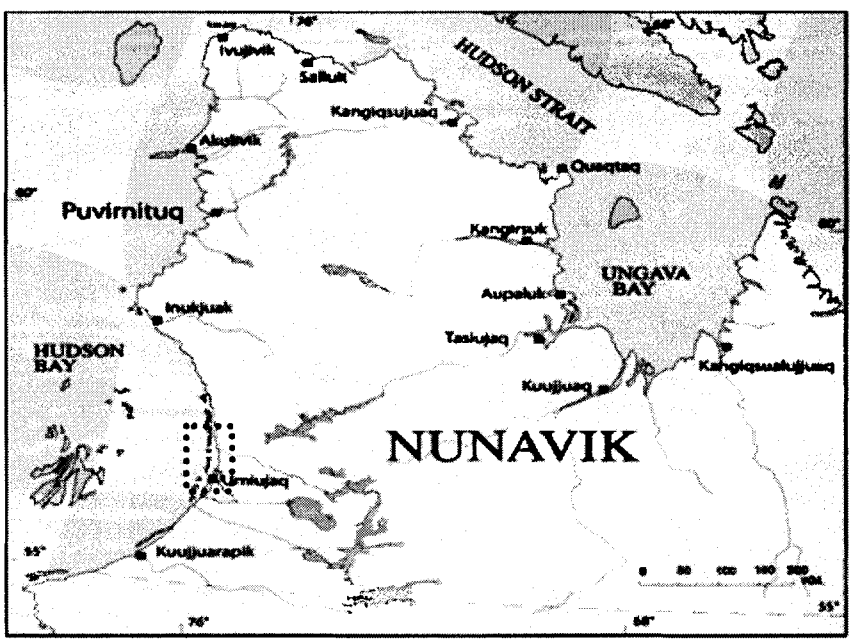


Figure 2.1 – Map of Northern Quebec (Nunavik): dashed line indicates where the study area was located. Source: Makivik Corporation.

The beluga samples were collected from whales that were harvested near the Nastapoka River estuary. The cod were caught in the Islands adjacent to Umiujaq and northwards. Sampling of the bottom sediment took place just north of Umiujaq between the Islands and the shore. Lichen was collected from un-trampled patches north of Umiujaq.

2.1.2 Preparation of Field Sampling Equipment

To minimize the possible contamination of samples during collection, sampling equipment and collection jars were baked and solvent rinsed to eliminate background levels of phthalate esters. All collection jars and relevant sampling equipment were rinsed with acetone, hexane and dichloromethane (DCM) and baked overnight (10h) at 325° C. After baking, each item was then rinsed twice with iso-octane, once with hexane and DCM, twice with methanol and then once again with DCM. Following this process the collection jars and sampling equipment were covered with aluminum foil that had been rinsed with hexane and DCM and baked at 400°C overnight.

2.1.3 Biota Sample Collection

Beluga tissue samples were obtained from animals that had been legally harvested by Inuit hunters. Consequently, a formal experimental design could not be implemented, as the sampling was limited to those whales that were hunted. Samples of blubber, liver, stomach contents (if available), lower gastro-intestinal tract and blood were obtained from each animal using clean sampling equipment with consent of the hunter, immediately after the whales were killed. In the event of a lactating female being harvested, samples of milk were also collected. The length, girth and sex of the animal were recorded for most animals during sampling in the field. The age of the animal was determined by tooth analysis at the Nunavik Research Centre in Kuujjuaq, Quebec.

Arctic cod were caught individually using a standard fishing line. Only muscle was collected for analysis. For lichen, collection jars were densely packed to maximize the weight of each sample. Table 2.1 shows the number of samples that were collected in each sampling season. All samples were collected in clean jars and kept frozen.

Table 2.1 – Number of biota samples that were collected over three summers.

Species		Year			TotalSample Size (n)	
Common Name	Latin Name	2000	2001	2002	Collected	Used in Study
Beluga	*Delphinapterus leucas*	30	14	0	44	11*
Arctic Cod	*Boreogadus saida*	0	6	6	12	12
Lichen	*Cladina rangeferina*	0	0	7	7	7

*Only samples from adult males were analyzed

2.1.4 Sediment Sample Collection

Bottom sediment from Hudson's Bay was collected using a 6" petit ponar sediment grab attached to a 200 ft. rope. The sediment was released from the ponar into a solvent-rinsed stainless steel bowl and a 75mL sample from the middle of the grab was transferred into a collection jar. The approximate location and depth of each sample was recorded and they are shown in Table 2.2.

Table 2.2 – Location and depth of sediment samples collected in August 2002 from Hudson's Bay.

Sample Name	Location	Depth (ft.)	Comments
HBS02-01	Nastapoka Islands off Umiujaq	160	Sampling location progressed northwards along the eastern side of the Nastapoka Islands from HBS02-01 to HBS02-06.
HBS02-02	Nastapoka Islands off Umiujaq	75	
HBS02-03	Nastapoka Islands N. of Umiujaq	185	
HBS02-04	Nastapoka Islands N. of Umiujaq	180	
HBS02-06	Nastapoka Islands N. of Umiujaq	99	
HBS02-07	Along shore N. of Umiujaq	65	Sampling location progressed northwards along the shore from HBS02-07 to HBS02-12.
HBS02-08	Along shore N. of Umiujaq	100	
HBS02-09	Along shore N. of Umiujaq	65	
HBS02-10	Along shore N. of Umiujaq	80	
HBS02-11	Along shore N. of Umiujaq	85	
HBS02-12	Along shore N. of Umiujaq	85	

2.2 *Analytical Methods for Determining Phthalate Ester Concentrations in Environmental Samples*

2.2.1 Preparation of Glassware and Reagents

To minimize background contamination, all laboratory glassware was cleaned and proofed before coming into contact with the samples. Dirty glassware was scrubbed with a brush under water and then dishwasher washed with distilled water. After the dishwasher, the glassware was rinsed with acetone, toluene, hexane and dichloromethane (DCM) and then baked overnight at 325-400°C. After baking, everything was rinsed with iso-octane (2X), hexane, DCM, methanol (2X) and then DCM again. At this stage, each piece of glassware in a batch was rinsed with 1:1 DCM/hexane and the rinse was collected for proofing. Once the glassware proofs were found to be free of phthalate contamination (i.e. levels were below the GC/MS detection limits) they were approved for use in sample extraction and clean-up where each piece is rinsed again with distilled hexane just prior to being used. The same protocol was followed for the cleaning of mortars, pestles and metal spatulas. The GC vials were baked overnight at 325 °C and then sonicated for 15 minutes in hexane and DCM. The nitrogen needles that were used for blowing down the samples were sonicated with toluene and DCM and the syringes that were used for spiking were rinsed numerous times with toluene between uses. Sodium sulfate and alumina were baked overnight at 325-400 °C and kept in a dessicator. As for the solvents, hexane was found to be a main source of background phthalate contamination and therefore only distilled hexane was used for sample processing.

2.2.2 Extraction and Clean-up of Biota and Sediment Samples

A summary of the clean-up and extraction process is illustrated in Figure 2.2. A 10g sub sample was used for extraction of sediment and lichen and a 5g and 0.25g sub sample

was used for fish and beluga (liver) samples respectively. The biota and sediment were homogenized in a mortar and ground up into a fine, dry powder with 15 to 20g of sodium sulfate. The powder was transferred to a round bottom flask for spiking and solvent extraction. Samples were spiked with 100ng of a phthalate ester internal standard (PE IS) which is made up of deuterated DMP, DBP and DnOP and 50ng of a PCB internal standard. The extraction, which is repeated three times, involved the addition of 20mL 1:1 DCM/hexane to the sample/sodium sulfate mixture, followed by 15 minutes of sonication in a Branson 5210 ultrasonic water bath. Following sonication, the supernatant from all three extractions were collected in a clean round bottom flask. The sample was then blown down to 2-3mL using a gentle stream of high purity nitrogen and allowed to evaporate to dryness and then reconstituted with 1 to 2mL of double-distilled hexane (dd hexane).

The concentrated samples were then quantitatively transferred onto an alumina column for chemical fractionation. The first fraction of each sample was eluted with 30mL of dd hexane and contained PCBs. This fraction was collected for analysis but the results are not presented in the current paper. The second fraction was eluted with 30mL 1:9 DCM/hexane and subsequently discarded. The third and final fraction was eluted with 30mL 1:1 DCM/dd hexane and contained the phthalate esters. This third fraction was collected in a clean jar and was blown down to 1.0mL and then quantitatively transferred to a centrifuge tube where it was concentrated even further before being transferred to a GC micro vial. In the micro vial, the sample is blown down to 0.2mL under a very gentle stream of nitrogen and then spiked with 50ng of a phthalate ester recovery standard that is made up of two deuterated phthalates (DEP-d4 and BBP-d4).

2.2.3 Preparation of alumina columns

The alumina columns were prepared using glass columns that had been cleaned and proofed. Just before packing, the columns were rinsed with distilled hexane. The deactivated alumina was then hydrated to 15% with HPLC grade water. To pack the columns, they were each filled with distilled hexane. 15g of hydrated alumina (15% H_2O w/w) was then poured into each column and allowed to settle. 1-2 cm of sodium sulfate was then added to the top of the alumina and then 10-15mL of distilled hexane was run through the column before being loaded with the sample.

2.2.4 GC/LRMS Analysis of Environmental Samples

The eight individual phthalate esters were quantified in the samples using low-resolution gas chromatography with detection by mass spectrometry at the Institute of Ocean Sciences. A Finnigan Voyager GC/MS machine was used following a program and setup described in Lin et al. (*34*).

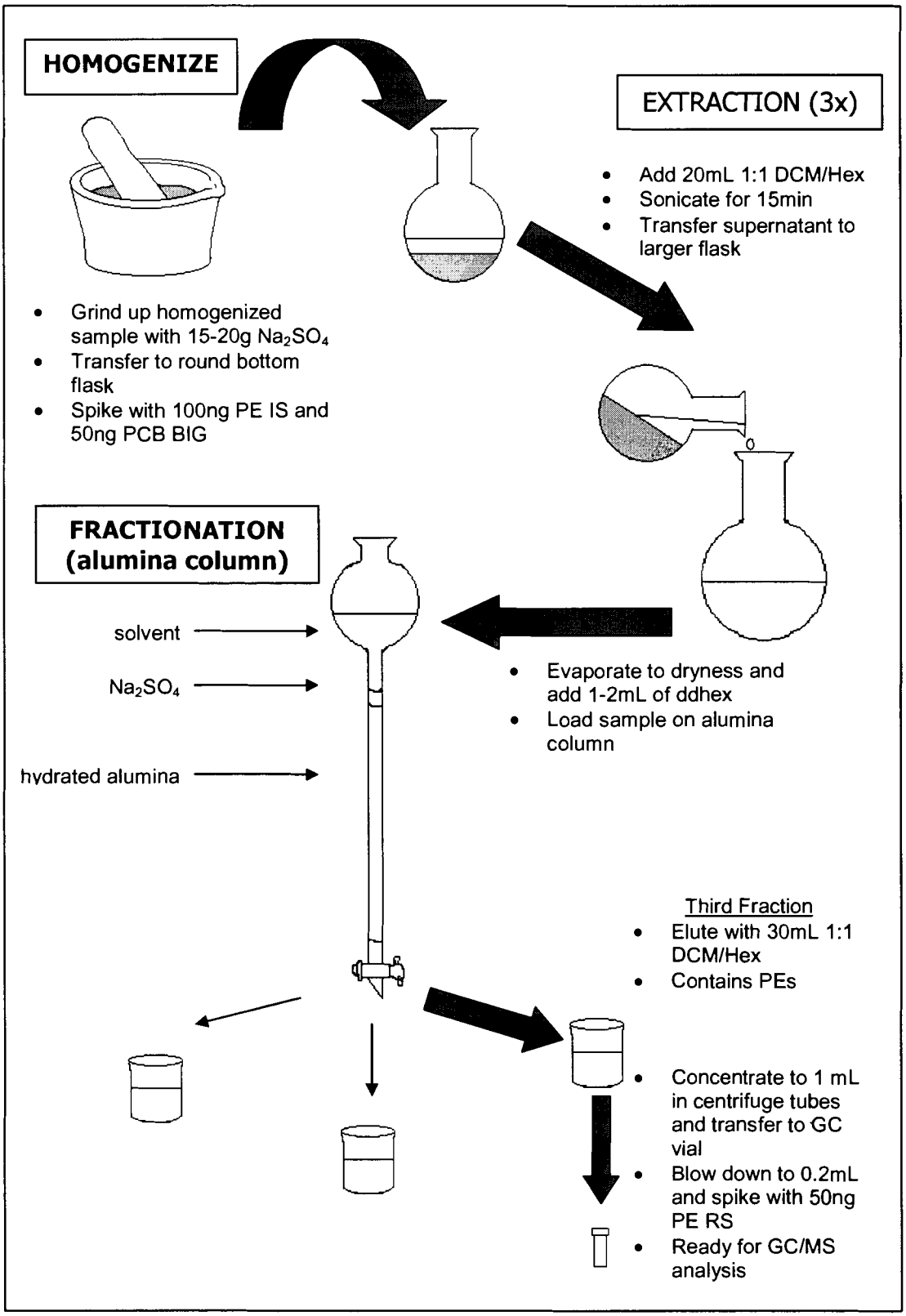


Figure 2.2– Diagram outlining the extraction and chemical fractionation process for phthalate ester analysis of environmental samples.

2.2.5 Quantification of Phthalate Esters in Environmental Samples

In calculating the concentration of phthalates esters in the biota and sediment samples, loss of chemical in extraction and clean-up, background contamination and machine variability were taken into account. Variability in the MS was accounted for by calculating relative response factors (RRFs) that are a measure of how a given chemical is detected relative to another in terms of peak area for compounds in a calibration standard with known concentrations. The first set of RRFs that were calculated related the peak area of each internal standard (IS) compound to that of the appropriate recovery standard (RS) compound (Equation 2.1). The appropriate recovery standard compound is the one with the closest molecular weight to the internal standard of interest and thus the recovery of DMP-d4 IS and DBP-d4 IS was compared to that of DEP-d4 RS, and the recovery of DnOP-d4 IS was compared to BBP-d4.

$$RRF_{IS/RS} = \left(\frac{PeakArea_{IS}}{PeakArea_{RS}}\right) \times \left(\frac{Conc_{RS}}{Conc_{IS}}\right) \qquad \text{Equation 2.1}$$

This $RRF_{IS/RS}$ was important for determining the recovery of the internal standards in the samples as it accounts for how the MS detects the internal standards (which are added to the sample prior to extraction) relative to the recovery standards (which are added prior to injection) based on the peak areas given by known concentrations. This is significant because the amount of internal standard left in the sample at the time of injection was not known whereas the amount recovery standard being injected was known (i.e. the spiking amount). Thus using the $RRF_{IS/RS}$ allowed for a more accurate quantification of the IS amounts in the environmental samples by accounting for machine

variability. The amount of IS in the samples was calculated using the following equation (Equation 2.2).

$$\text{Mass Recovered}_{IS}(ng) = \left(\frac{PeakArea_{IS}}{PeakArea_{RS}}\right) \times \left(\frac{SpikeAmount_{RS}}{avgRRF_{IS/RS}}\right) \qquad \text{Equation 2.2}$$

The percent recovery is then determined by dividing the amount detected by the amount each sample was spiked with prior to extraction (Equation 2.3).

$$\%\text{Recovery}_{IS} = \left(\frac{Mass_{IS}\ (ng)}{Spike\ Amount_{IS}}\right) \qquad \text{Equation 2.3}$$

The percent recovery of the internal standard compounds are used to evaluate the amount of test chemical that was lost during the extraction and clean-up process. The percentage of internal standard recovery can be used as an estimate for the recovery of each analyte in that it is assumed that the fraction of internal standard recovered is the same as the fraction of individual phthalates recovered. The internal standard that has the closest molecular weight to the individual phthalate of interest is used to estimate recovery. Table 2.3 shows the individual phthalates that correspond to each deuterated standard.

Table 2.3 Individual Phthalate Esters and the Internal standards used to estimate recovery. Recovery of individual PE analytes is estimated from the internal standard compound that has the most similar molecular weight.

Internal Standard	Individual Phthalate Esters
DMP-d4	DMP, DEP
DBP-d4	DIBP, DBP, BBP
DnOP-d4	DEHP, DOP, DNP

The resulting percentage is an indicator of how successful the extraction and clean-up process was. Table 2.4 and Figure 2.3 show the average percent recovery of the internal standards for the sodium sulfate blanks and the different environmental samples.

Table 2.4 Average Recoveries (%) and standard deviations of Internal Standard Compounds for Sodium Sulfate Blanks and Environmental Samples

	DMP-D4		DBP-D4		DOP-D4	
	Mean	StDev	Mean	StDev	Mean	StDev
Blanks	87.6	8.49	91.7	8.57	90.1	4.41
Beluga	74.5	3.57	99.1	9.02	79.2	7.45
Cod	75.6	7.85	89.4	7.12	70.9	6.85
Lichen	68.1	5.41	77.0	6.76	69.0	6.58
Sediment	71.6	11.1	89.8	4.03	91.5	3.76

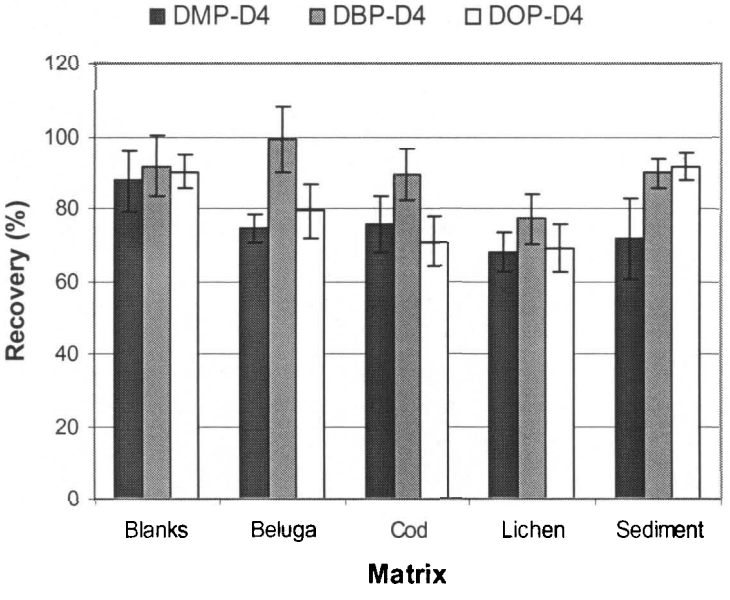


Figure 2.3 Average recovery (%) and standard deviations of internal standard compounds for sodium sulfate blanks and the environmental samples

Average recoveries ranged from 68% for DMP-d4 in lichen to 99.1% for DBP-d4 in the beluga liver samples. These recoveries indicate that the majority of chemical was recovered in the sample during extraction and fractionation.

With acceptable recoveries, the amount of each individual phthalate ester was quantified in the environmental samples. To do this, a relative response factor for each phthalate was determined relative to the appropriate internal standard compound (Equation 2.4). This factor, known as the $RRF_{PE/IS,}$ was calculated as follows:

$$RRF_{PE/IS} = \left(\frac{PeakArea_{PE}}{PeakArea_{IS}}\right) \times \left(\frac{Conc_{IS}}{Conc_{PE}}\right) \qquad \text{Equation 2.4}$$

As with the $RRF_{IS/RS}$, the $RRF_{PE/IS}$ indicates how each phthalate ester is quantified by the MS relative to the internal standard compound that has the closest molecular weight by comparing the peak areas yielded by known concentrations in the standard calibration solution. As before, this factor was intended to correct for machine variability in detecting the different compounds. With this final relative response factor, the amount of each PE in the environmental samples and the blanks were calculated using Equation 2.5.

$$PEng = \left(\frac{PeakArea_{PE}}{PeakArea_{IS}}\right) \times \left(\frac{SpikeAmount_{IS}}{avgRRF_{PE/IS}}\right) \text{ Equation 2.5}$$

The next step was to screen the concentrations against the appropriate method detection limit (MDL). MDL is the lowest signal that you can properly detect on the instrument for a given matrix. It is determined by multiplying the level of noise (measured in peak height on a chromatogram) by three, which yields the smallest peak that can be considered a signal. This signal is then converted into a concentration which is the method detection limit for the matrix. Table 2.5 shows the MDLs that were determined for the analysis of the biota and sediment samples (ng/g) on the GC/LCMS. In the event that the sample concentration was above the MDL, it was considered acceptable. Those values that were less than the MDL were not used in further analysis.

Table 2.5 Method Detection Limits (ng/g) for Liver, tissue, lichen and sediment analyzed by GC/LCMS

Phthalate Ester	Liver	Tissue	Lichen	Sediment
DMP	0.4	0.06	0.06	0.005
DEP	1.5	0.06	0.06	0.015
DIBP	0.35	0.09	0.09	0.005
DBP	1.2	1.1	1.1	0.006
BBP	1.8	0.25	0.25	0.012
DEHP	4.8	1.1	1.1	0.021
DnOP	0.6	0.08	0.08	0.024
DnNP	7.1	1.3	1.3	0.017

The sodium sulfate blanks were used to quantify background contamination. For each batch of ten samples, two blanks were processed. The average amount of each individual phthalate ester that was quantified in the two blanks was subtracted from amounts calculated in the samples from the same batch. This blank corrected mass was then used to calculate the concentration (ng/g wet weight) in the sample by dividing the mass by the wet weight of the sample. Figure 2.5 shows the average concentrations of the individual phthalate esters in the sodium sulfate blanks from the batches of all the biota and sediment samples.

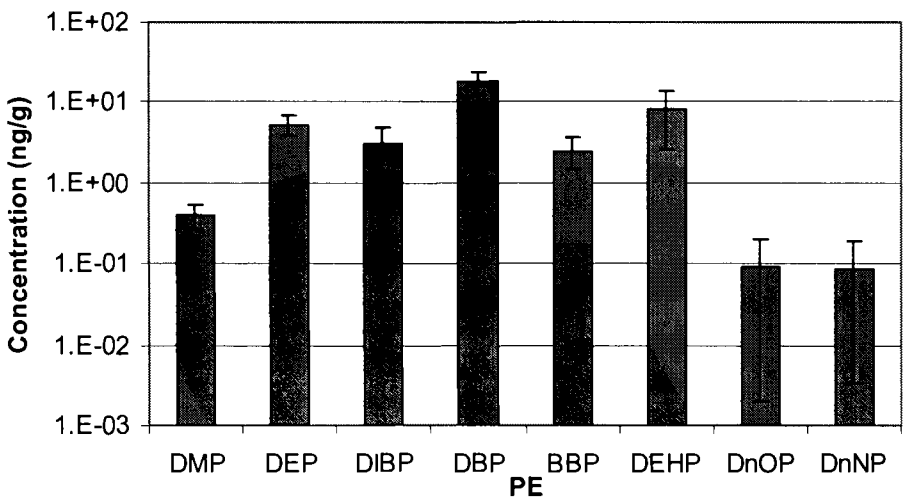


Figure 2.4 Mean Concentrations (ng/g) of Phthalate Esters in Sodium Sulfate Blanks.

By calculating recoveries of internal standards and the relative response factors, chemical loss during extraction and clean-up, and machine variability were accounted for, thus establishing quality assurance and quality control (QA/QC) of the reported data. Screening results against the method detection limit and blank correcting the data also contributed to QA/QC by accounting for the capacity of the GC/LCMS and background contamination.

All concentration results are presented on a wet weight (ng/g wet wt) or a dry weight (ng/g dry wt) basis as well as a lipid equivalent basis (ng/g lipid eq). Chemical fugacities (nPa) were also calculated.

2.2.6 Lipid Content and Organic Carbon Content Determination

The lipid content of each fish and beluga sample was determined at the Institute of Ocean Sciences (IOS) using the following method. A 5g sub-sample of tissue was ground up with 100g of baked anhydrous sodium sulfate and then transferred to a 30 x 30cm glass column. The sample was eluted with 100mL 1:1 DCM/hexane and the collected eluent was evaporated to 1mL. The sample was allowed to dry overnight in a 40°C vented oven and then weighed again after it cooled completely. From the difference between the initial and final sample weights, the percent lipid was calculated. Organic carbon content analysis in the sediment samples was conducted at IOS following the method of Van Iperen and Helder (1985) (*35*). Lipid and organic carbon content for lichen were taken from Kelly and Gobas for a different set of samples of the same species (2001) (*20*).

2.3 *Data Analysis and Normalizations*

2.3.1 Data Distribution and Statistical Analysis

In order to statistically analyze the concentration data for the environmental samples, it is necessary to determine the distribution of the data, as normality is a key assumption when performing analysis of variance (ANOVA). To test the normality of the data, the Shapiro-Wilk test was used where the null hypothesis states that the data are normally distributed. This test produces a W-statistic that ranges from 0 to 1.00, where 1.00 indicates a perfectly normal distribution. A p-value is also provided. For each individual phthalate ester and each type of media, a W-statistic was calculated for the original data and the log-transformed data using SPSS 11.5. The data set with the highest W-statistic and p-value was considered to be the most normal. For the purpose of comparison, the most commonly-found results were used. In the majority of cases, the log-transformed data had a higher W-statistic and a higher p-value (strong indication that the null hypothesis (i.e. normality) should not be rejected). Consequently, all data were assumed to be log-normally distributed and thus all concentration and fugacity values were log transformed prior to analysis and the geometric means and standard deviations of the geometric means were calculated. ANOVA was used to determine if the geometric means of the concentrations and fugacities in the different ecosystem components were statistically different by using the log of the concentrations and fugacities.

2.3.2 Lipid Equivalent Normalizations

All concentration data for the beluga whale and arctic cod samples were normalized to the lipid content. The lipid content for each beluga liver and fish muscle sample was determined and then used to calculate the amount of phthalate on a lipid equivalent basis (ng/g lipid equivalent) using the following equation.

$$C_{lipid} = \frac{C_{wet}}{L} \qquad \text{Equation 2.6}$$

Where C_{wet} is the wet weight concentration of the given phthalate (ng/g wet wt.) and L is the lipid fraction (g lipid/ g wet tissue).

The environmental concentrations in lichen were converted to dry weight concentrations prior to lipid equivalent normalizations. To calculate the concentrations on a dry weight basis, the wet weight concentration of the sample is divided by one minus the moisture content (ϕ_W). Moisture content was not determined for the specific samples used in this study. This value, as well as lipid (L) and non-lipid organic matter (NLOM) values, were taken from Kelly and Gobas (2003), for the same species of lichen – *Cladina rangiferina* (*20*).

$$C_{dry} = \frac{C_{wet}}{(1 - \%\phi_W)} \qquad \text{Equation 2.7}$$

C_{dry} (ng/g dry wt.) is then used to calculate the lipid equivalent normalized concentration (ng/g lipid equivalent).

$$C_{lipid} = \frac{C_{dry}}{(0.035NLOM + L)} \qquad \text{Equation 2.8}$$

The constant, 0.035 accounts for the different sorptive capacity of NLOM relative to that of lipid (*20*).

Phthalate ester concentrations in sediment were lipid equivalent normalized by dividing the dry weight concentration by the organic carbon content. The constant 0.35 is a proportionality constant representing the degree to which the sorptive capacity of sediment based organic carbon resembles that of lipid.

$$C_{lipid} = \frac{C_{dry}}{0.35\phi_{OC}} \qquad \text{Equation 2.9}$$

2.3.3 Estimating Air Concentrations

As mentioned previously, the reason for analyzing lichen samples was to estimate the amount of chemical in the gaseous phase. Air concentrations for the eight individual phthalates were estimated using a dimensionless vegetation-gas partition coefficient (K_{VG}), which is calculated as follows (Equation 2.10):

$$K_{VG} = (\nu_L + 0.035 \times \nu_N) K_{OA} \qquad \text{Equation 2.10}$$

Where: ν_L = lipid content on a dry weight basis
ν_N = non-lipid organic matter (NLOM) on a dry weight basis

This equation from Kelly and Gobas (2003) estimates the partitioning of chemical between the gaseous phase of air and vegetation by accounting for the sorptive capacities of the lipid and the non-lipid organic matter (NLOM) phases of the vegetation (*20*). The NLOM is multiplied by the constant 0.035 which relates the sorptive capacity of NLOM to that of lipid in the vegetation (*36*). The amount of chemical in the particulate phase of the air is not calculated, as it is assumed that the beluga whale exposure to chemical via air inhalation is only through the gaseous phase. It should still be noted however that the amount of chemical present in the gaseous and particulate phase in ambient air changes seasonally with fluctuations in temperature.

The concentration of a PE congener in air (ng/g) is equal to the concentration in lichen (ng/g dry wt) divided by K_{VG}. Air concentrations were normalized to a lipid equivalent value by multiplying the calculated concentration by the chemical's K_{OA}.

2.3.4 Fugacity Calculations

Fugacity (*f*, in Pa) was calculated for the biota, sediment and air samples. Fugacity is equal to the concentration (mol/m^3) divided by the fugacity capacity (Z, in mol/m^3 Pa) of the media, $f = C/Z$. The fugacity capacity is a measure of the ability of a given medium to "take up" the chemical of interest. The fugacity capacities of the environmental samples were calculated as:

Biota:

$$Z_{BIO} = L_{BIO} \times K_{OW} \times \rho_B \times \left(\frac{1}{H}\right) \qquad \text{Equation 2.11}$$

Sediment:

$$Z_{SED} = 0.35 \times \phi_{OC} \times K_{OW} \times \rho_{SED} \times \left(\frac{1}{H}\right) \qquad \text{Equation 2.12}$$

Lichen:

$$Z_L = K_{VG} \times Z_{AIR} \qquad \text{Equation 2.13}$$

Air:

$$Z_{AIR} = \left(\frac{1}{RT}\right) \qquad \text{Equation 2.14}$$

where: L_{BIO} = lipid fraction in tissue
K_{OW} = octanol-water partition coefficient
ρ = density (kg/L) (biota=1kg/L, sediment = 1.5kg/L, lichen = 0.64kg/L)
H = Henry's Law Constant (mol/Pa/m^3)
ϕ_{OC} = organic carbon fraction
R = universal gas constant
T = absolute temperature

2.3.5 BMF, BSAF and BAAF Calculations

Biomagnification factors (BMFs) were calculated on a wet weight and lipid equivalent basis using Equations 2.15 and 2.16 respectively.

$$BMF_W = \frac{C_{beluga}(ng/gwet\_wt.)}{C_{cod}(ng/gwet\_wt.)} \qquad \text{Equation 2.15}$$

$$BMF_L = \frac{C_{beluga}(ng/g\_lipid)}{C_{cod}(ng/g\_lipid)} \qquad \text{Equation 2.16}$$

BMF_W is the ratio between the amounts of chemical in the organism's tissue and BMF_L is the ratio of chemical in the organism's lipid. The average concentrations (χ) for each PE were used in the calculations and +/- 1 standard deviations (-1SD and +1SD) were calculated (Equation 2.17)

$$-1SD_{BMF} = \sqrt{(-1SD_{beluga}/\chi_{beluga})^2 + (-1SD_{cod}/\chi_{cod})^2} \qquad \text{Equation 2.17}$$

The same calculation is used to determine $+1SD_{BMF}$ using $+1SD_{beluga}$ and $+1SD_{cod}$.

Biota-sediment accumulation factors (BSAFs) are calculated by dividing the mean lipid normalized biota concentration by the mean organic carbon normalized sediment concentration for each PE and are presented in units of kg OC/kg lipid (Equation 2.18). Standard deviations are determined in the same manner as they were for BMFs (Equation 2.17). Due to the different sorptive capacities of lipid and organic carbon, unity (i.e. equilibrium) is represented by a value of three as the sorptive capacity of lipid is considered the same as the sorptive capacity of octanol, whereas organic carbon has a sorptive capacity that is 0.35 times that of octanol (Equation 2.19). A BSAF greater than three is thus an indication of more chemical being in the biota compared to the sediment.

$$BSAF(kgOC/kglipid) = \frac{C_{biota}(\mu g/kg\_lipid)}{C_{sed}(\mu g/kg\_OC)} \qquad \text{Equation 2.18}$$

At equilibrium:

$$fbiota = fsed$$

$$\frac{Cbiota(ng/g\_lipid)}{Csed(ng/g\_OC)} = \frac{Zlipid}{Zoc} = \frac{1.0 \times Zoct}{0.35Zoct} \cong 3 \qquad \text{Equation 2.19}$$

The distribution between air and beluga is being investigated in order to gain insight into the possible exposure to phthalates through air inhalation and the general PE partitioning throughout the system. The distribution of chemical between air and belugas is assessed by calculating a ratio between the two lipid equivalent concentrations (Equation 2.20). This ratio will be referred to as the biota-air accumulation factor (BAAF). Standard deviations are calculated as shown in Equation 2.17.

$$BAAF = \frac{C_{beluga}(\mu g/kg\_lipid\_eq.)}{C_{air}(\mu g/kg\_lipid\_eq.)} \qquad \text{Equation 2.20}$$

3.0 Results and Discussion

Overview – The results presented below indicate that Hudson's Bay is within the range of global phthalate ester dispersion. Despite being in a remote region devoid of significant local sources, PEs were detected in all components of the ecosystem that were examined: beluga, arctic cod, sediment, lichen and air. As a summary, mean ΣPEs for the different components are shown in Figure 3.1. ΣPE is equal to the sum of all phthalate esters analyzed except for DnNP which was only detected in 29 of 41 environmental samples.

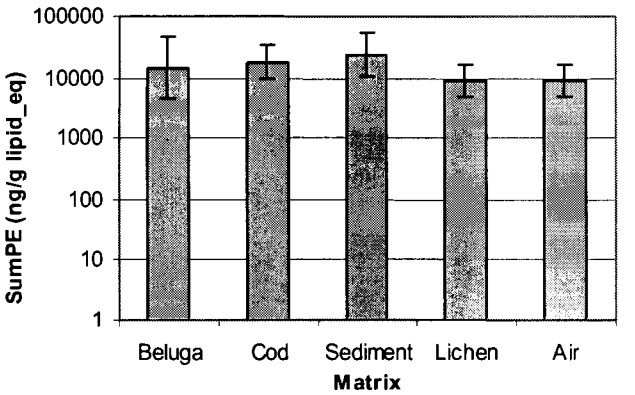


Figure 3.1 – Mean Lipid Equivalent ΣPEs (+/- 1 standard deviation) for Beluga, Arctic Cod, Sediment, Lichen and Air. ΣPE does not include DnNP.

The mean ΣPE concentrations (ng/g lipid eq) for beluga, cod, sediment, and lichen are 14500 (4520-46500), 18200 (9500-34800), 23900(10500-54300) and 8990 (4900-16500) respectively. The lipid equivalent concentration for air is the same as that for lichen. On average, the total PE concentrations are the same for all components ($p>0.05$). Congener specific results for beluga, cod, sediment, lichen and air are presented in section 3.1 followed by an analysis of the distribution of phthalate esters amongst the different matrices, as represented by biomagnification, biota-sediment distribution and biota-air distribution (sections 3.2, 3.3 and 3.4). The relationship

between biomagnification and K_{OA} is discussed in section 3.5 and section 3.6 examines the relationship between beluga contamination and the length and age of the animals. Finally, the Hudson's Bay data is compared with data from a British Columbian inlet in order to compare phthalate ester contamination between a remote and urban environment (Section 3.7).

3.1 Environmental Concentrations and Fugacities

3.1.1 Beluga

The raw wet weight concentrations in the 11 male beluga liver samples are presented in Table A.1 in Appendix A with means and standard deviations for the log transformed values shown in Figure 3.2a. The average values ranged from 2.19ng/g wet wt for DMP to 72.9 ng/g wet wt for DBP. The concentrations of DEHP were highly variable. Original DEHP concentrations ranged from 2.8 ng/g wet wt to 4150.21 ng/g wet wt. The highest value was thought to be questionable but it was confirmed by repeat analysis. Nevertheless, the value was left out when calculating the average DEHP concentration, as it was such a significant outlier. The second highest DEHP concentration was 412.45 ng/g wet wt. For DnNP, 7 out of the 11 samples analyzed yielded results that were below the method detection limit. The four remaining values ranged from 8.22 ng/g wet wt to 47.97 ng/g wet wt. Figure 3.2a, shows the mean concentrations (ng/g wet wt) for each individual phthalate ester in the beluga whales sampled.

The average lipid content for the liver samples was 1.98% +/- 0.01% (n=11). Average lipid equivalent concentrations ranged from 124ng/g lipid equivalent for DMP to 4120ng/g lipid equivalent for DBP. The average lipid equivalent concentrations for all

individual phthalates are shown in Figure 3.2b. Individual lipid equivalent concentrations for each sample are presented in Table A.2 in Appendix A.

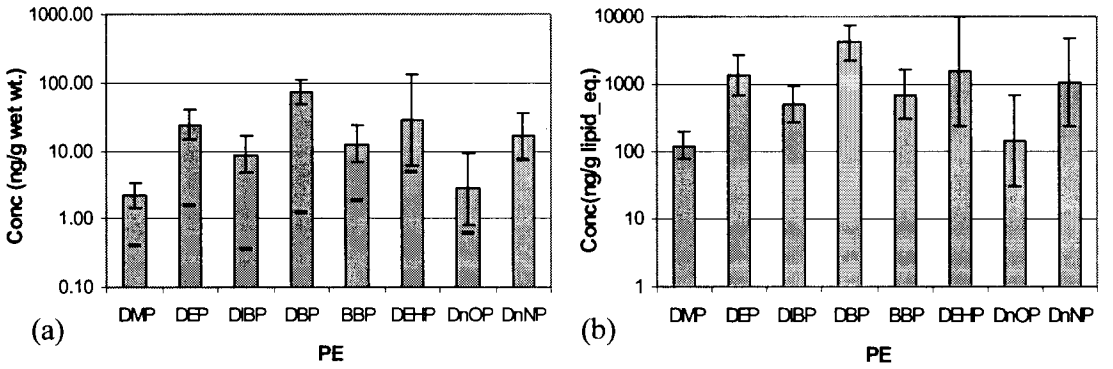


Figure 3.2 - Average Concentrations (+/- 1 standard deviation) of phthalate esters in Beluga (*Delphinapterus leucas*) expressed in (a) ng/g wet weight with MDLs (-) and (b) ng/g lipid weight

Fugacity data for beluga liver samples are shown in Table A.3. Average fugacities ranged from 0.03nPa for DnOP to 1060nPa for DBP. In general, the lower molecular weight phthalates had higher fugacities than those with molecular weights greater than 300g/mol. The mean fugacities for each individual phthalate ester in beluga are shown in Figure 3.3.

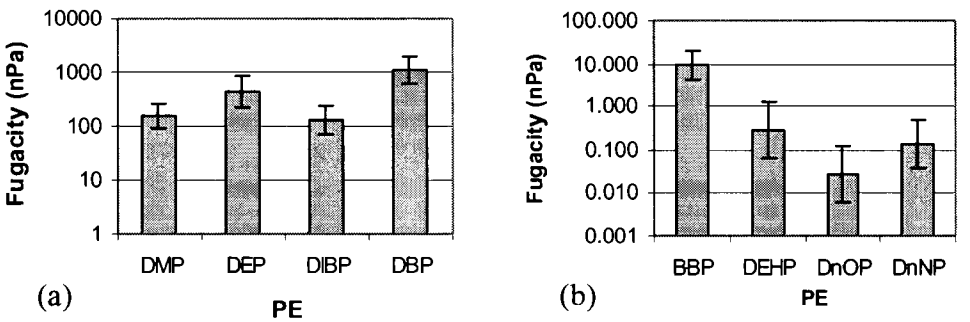


Figure 3.3 - Average Fugacities +/- 1 standard deviation (nPa) for (a) lower molecular weight phthalate esters and (b) higher molecular weight phthalate esters (>300g/mol) in Beluga (*Delphinapterus leucas*)

3.1.2 Arctic Cod

Tables A.4, A.5 and A.6 in Appendix A contain the PE concentration data for *Boreogadus saida* expressed in wet weight concentration (ng/g wet weight), lipid equivalent concentration (ng/g lipid equivalent) and fugacity (nPa) respectively.

Wet weight concentrations ranged from an average of 0.18ng/g wet wt. for DMP to 28.2ng/g wet wt. for DEHP. For DnNP, all but one of the twelve tissue samples that had been analyzed, were below the method detection limit. The average lipid content of the cod tissue was quite low at 0.24% with a standard deviation of 0.05%. For mean lipid equivalent concentrations, DMP levels were the lowest at 74.2ng/g lipid equivalent and DEHP levels were the highest at 5550ng/g lipid. The average concentrations for the individual phthalate esters for wet weight and lipid equivalent are shown in Figure 3.4 (a) and (b) respectively.

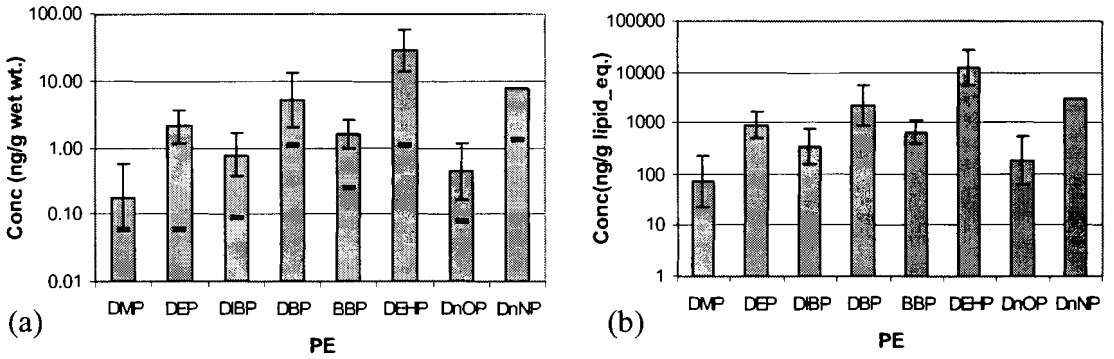


Figure 3.4 - Average Concentrations (+/- 1 standard deviation) of Phthalate Esters in Arctic Cod (*Boreogadus saida*) in (a) ng/g wet weight with MDLs (-), and (b) ng/g lipid equivalent.

Average fugacities ranged from 0.035nPa for DnOP to 566nPa for DBP. As with beluga, the lower molecular weight phthalates had higher fugacities in arctic cod compared to the phthalates with a molecular weight greater than 300g/mol. The average fugacities (+/- 1 standard deviation) for PEs in arctic cod are shown in Figure 3.5. There

are no error bars for the chemical fugacity of DnNP as there is only one data point and therefore it is uncertain if this value is representative of the population.

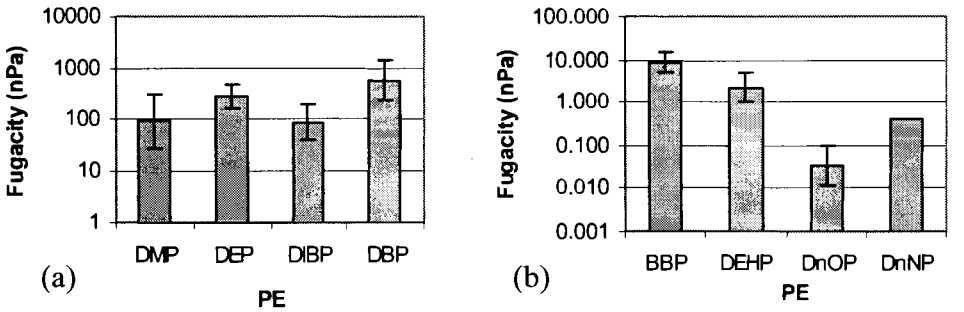


Figure 3.5 - Average fugacities +/- 1 standard deviation (nPa) for (a) lower molecular weight, and (b) higher molecular weight (>300g/mol), phthalate esters in arctic cod (*Boreogadus saida*)

3.1.3 Sediment

Concentrations of the individual phthalate esters in sediment are presented on a dry weight (ng/g dry wt) and lipid equivalent basis (ng/g lipid equivalent). Individual sediment sample data are presented in Tables A.7 and A.8 for the dry weight and lipid equivalent concentrations respectively. Original fugacity values are presented in Table A.9. Dry weight concentrations ranged from 0.019ng/g dry wt for DnNP to 6.78ng/g dry wt for DEP. Figure 3.6a, shows the mean dry weight concentrations for the eight individual phthalates.

The average organic carbon content for the Hudson's Bay sediment was 0.18% (kg OC/kg dry sediment) with a standard deviation of 0.1%. The mean lipid equivalent concentrations ranged from 32.4ng/g lipid equivalent for DnNP to 12200ng/g lipid equivalent for DEP. The mean lipid equivalent concentrations for sediment are shown in Figure 3.6b.

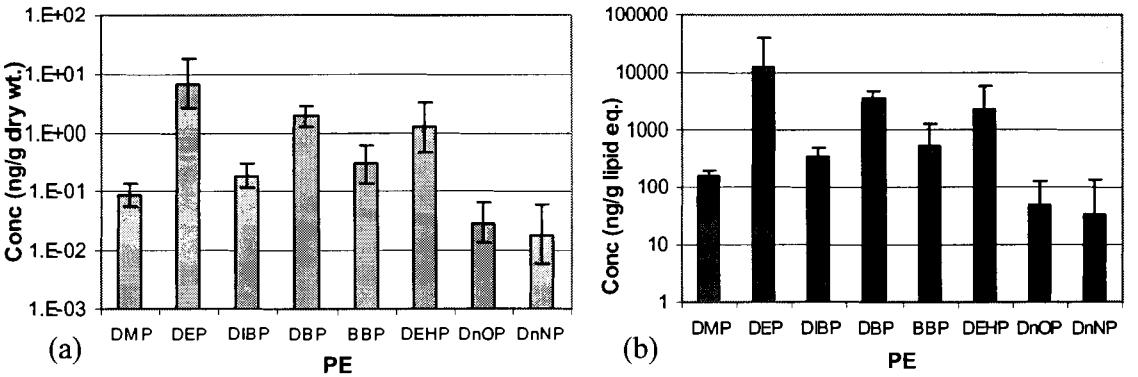


Figure 3.6 - Mean phthalate ester concentrations (+/- 1 standard deviation) in Hudson's Bay sediment in (a) ng/g dry wt, and (b) ng/g lipid equivalent

Mean chemical fugacities for phthalates in sediment exhibited the same pattern as the fugacities in the biota samples in that they were higher for the lower molecular weight phthalate esters, with DEP having the highest fugacity at 3970nPa. DnNP had the lowest fugacity of 0.004nPa. Figure 3.7 shows the mean fugacity for each individual phthalate in the sediment samples.

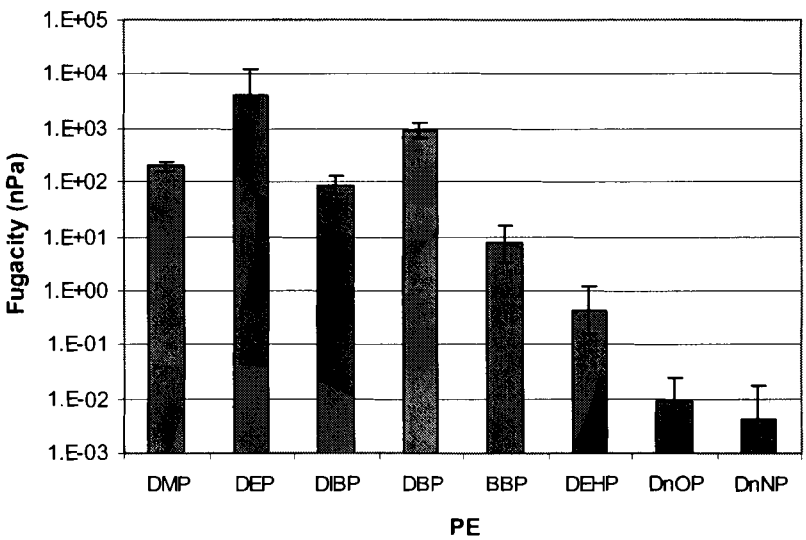


Figure 3.7 - Mean fugacities +/- 1 standard deviation (nPa) for Hudson's Bay sediment

3.1.4 Lichen

Concentration data for PEs in lichen are presented in Table A.10, A.11 and A.12 expressed in dry weight concentrations (ng/g dry wt), lipid equivalent concentrations (ng/g lipid eq) and fugacity (nPa). The mean dry weight concentrations in *Cladina rangiferina* ranged from 0.31ng/g dry wt for DMP to 187.14 ng/g dry wt for DEHP. Seven samples of lichen were analyzed, however only 4 samples had detectable levels of DnNP. The average concentrations of each phthalate ester in lichen are shown in Figure 3.8(a). Moisture content, organic carbon content and lipid content were not analyzed specifically for the samples collected in this study. The values that were used were taken from Kelly and Gobas, 2003, for the same species of lichen. The average moisture content used for *Cladina rangiferina* was 59.5% and the organic carbon and lipid content were 40.0% and 0.5% respectively on a dry weight basis (*20*).

Mean lipid equivalent concentrations ranged from 16.2ng/g lipid for DMP to 9830ng/g lipid for DEHP. Figure 3.8b shows the mean lipid equivalent values of all eight phthalate esters. Again, the mean and standard deviation for DnNP was measured from only four samples.

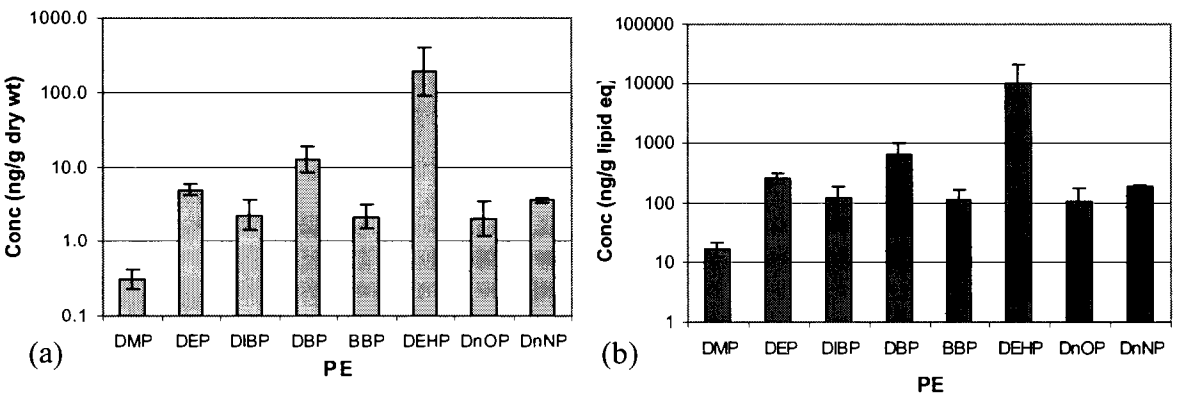


Figure 3.8 – Mean phthalate ester concentrations (+/- 1 standard deviation) in lichen (*Cladina rangiferina)* in (a) ng/g dry wt, and (b) ng/g lipid equivalent.

Mean fugacities in lichen ranged from 0.007nPa for DnNP to 55.2nPa for DEP. As in the other matrices, the fugacities of the higher molecular weight phthalate esters were smaller than the lower molecular weight phthalates. Figures 3.9(a) and (b) show the mean fugacities of each individual phthalate in lichen.

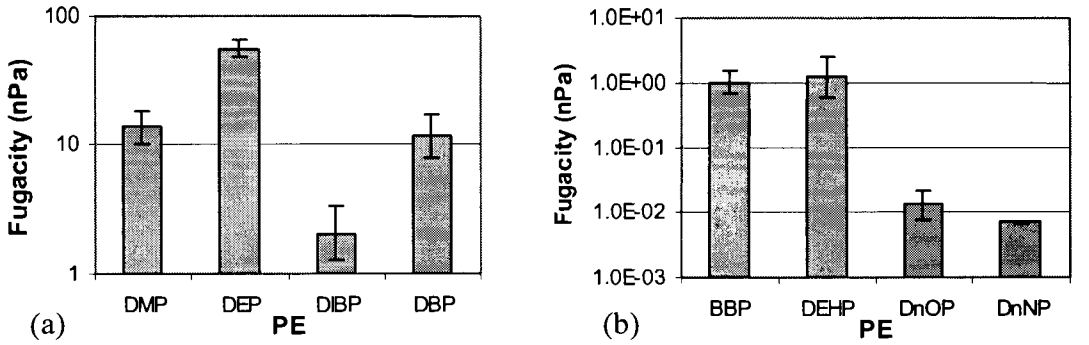


Figure 3.9 - Mean fugacities +/- 1 standard deviation (nPa) of (a) low molecular weight, and (b) high molecular weight (>300g/mol) phthalate esters in *Cladina rangeferina*.

3.1.5 Air

Concentration (ng/g), lipid equivalent concentration (ng/g lipid eq) and fugacity(nPa) data for PEs in air are presented in Table A.13, A.14 and A.15 respectively. Mean air concentrations are shown in ng/g and ng/g lipid equivalent in Figure 3.10. The sample size was 7, however for DnNP, the mean concentration was only based on 4 measurements as this phthalate was non-detectable in 3 lichen samples. Concentrations ranged from 1.76 x 10^{-9}ng/g for DnNP to 7.4 x 10^{-6}ng/g for DEP. Lipid equivalent concentrations which are equal to the regular concentration multiplied by the chemical's K_{OA} ranged from 16.2ng/g lipid for DMP to 9830ng/g lipid for DEHP.

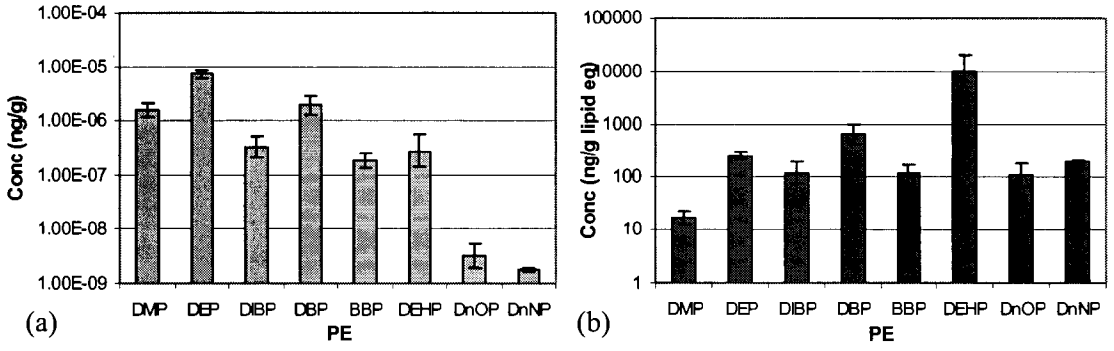


Figure 3.10 - Mean phthalate ester concentrations (+/- 1 standard deviation) in air in (a) ng/g, and (b) ng/g lipid equivalent.

The fugacities of phthalate esters in air are the same as those in lichen as equilibrium between the two matrices is assumed. Fugacity ranged from 0.007nPa for DnNP to 55.2nPa for DEP. The same pattern that was found in the other matrices was again detected in air, where the lower molecular weight chemicals had higher fugacities than the higher molecular weight chemicals. The mean fugacities for all individual phthalate esters in air are shown in Figures 3.11(a) and 3.11(b).

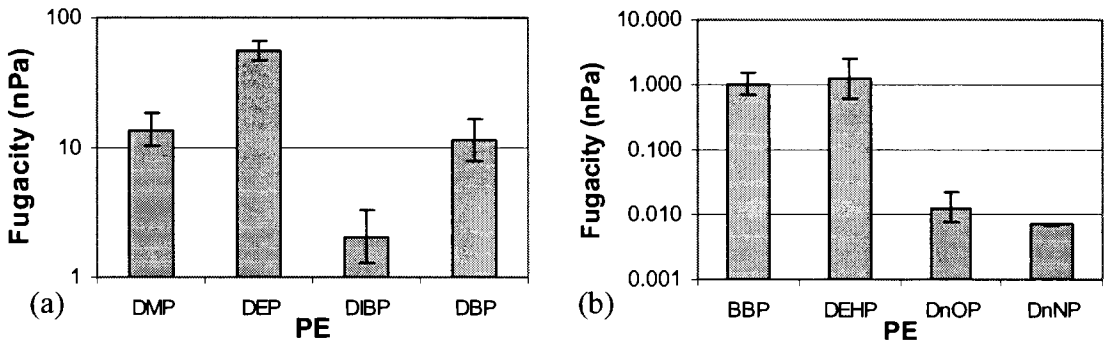


Figure 3.11 - Mean fugacity +/- 1 standard deviation (nPa) in air for (a) low molecular weight, and (b) high molecular weight (>300 g/mol) phthalate esters.

3.2 Biomagnification

The levels of phthalate esters in beluga and its diet are compared according to wet weight concentrations, lipid equivalent concentrations and fugacity using analysis of

variance (ANOVA) and biomagnification factors (BMFs). ANOVA has been used to compare the concentrations and fugacities of the beluga and fish samples. The null hypothesis states that the two means are the same. Biomagnification factors (BMFs) have been calculated for each individual phthalate ester and for all eight phthalate esters combined for the purpose of determining if the amount of chemical is increasing with trophic level. In the event that the BMF is greater than one, the chemical is magnifying up the food chain provided that the difference in the two populations is statistically significant. BMFs have been determined on both a wet weight (Equation 2.15) and lipid equivalent (Equation 2.16) basis and are presented in Table B.1 and B.2 in Appendix B. The concentrations and fugacities for beluga and cod that were presented in sections 3.1.1 and 3.1.2 are shown together in Figure 3.12.

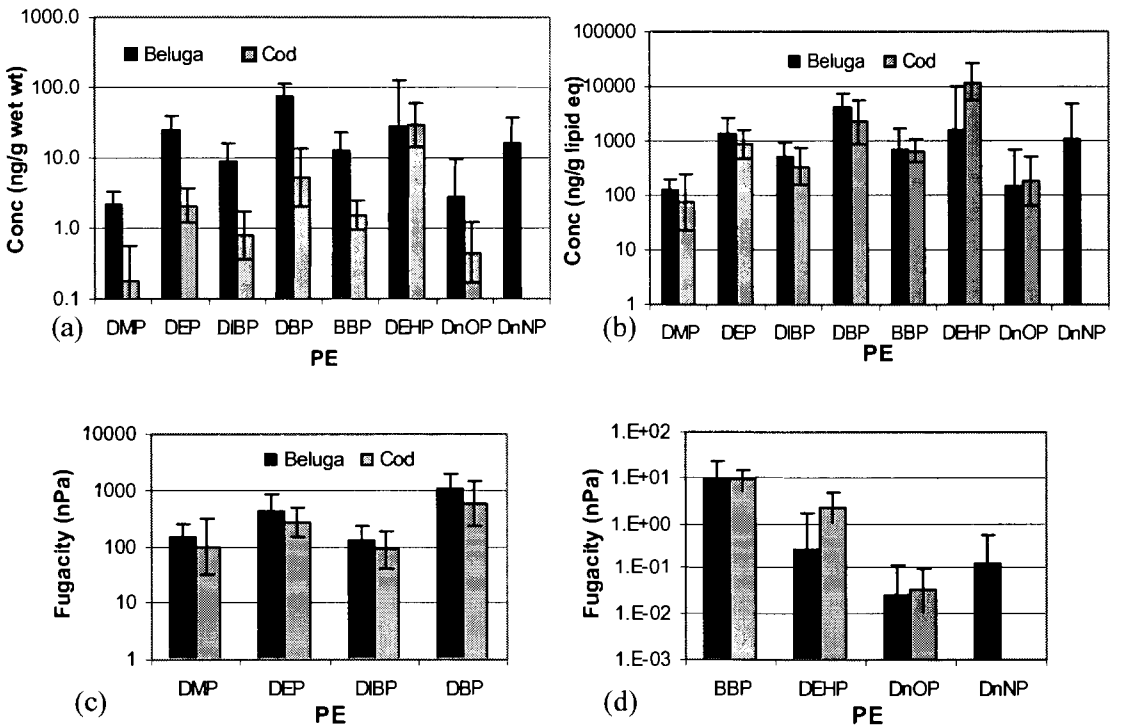


Figure 3.12 - Comparison of PE levels in *Delphinapterus leucas* (dark grey) and *Boreogadus saida* (light grey) expressed in (a) wet weight concentrations (ng/g wet wt.), (b) lipid equivalent concentrations (ng/g lipid) , and fugacity (nPa) for (c) low molecular weight, and (d) high molecular weight (>300g/mol) phthalates.

Figure 3.13a shows the BMF_ws of seven phthalate esters. The wet weight concentrations show that all PEs are biomagnifying from diet to beluga and these increases are all statistically significant except for DEHP where the null hypothesis was not rejected. BMF_ws for the wet weight concentrations ranged from 1.0 for DEHP to 14.0 for DBP. BMF was not calculated for DnNP as this compound was only detected in one cod sample. The method detection limit (MDL) for DnNP in cod tissue was 1.3 ng/g wet weight whereas the MDL was 7.1ng/g wet wt for DnNP in the beluga liver samples. Although, the exact amount cannot be quantified, the amount of DnNP in the cod samples is either equal to, or less than the MDL. This suggests that this congener is significantly lower in cod, which implies that the belugas are being exposed to DnNP from another source (e.g. air, sediment, other prey species).

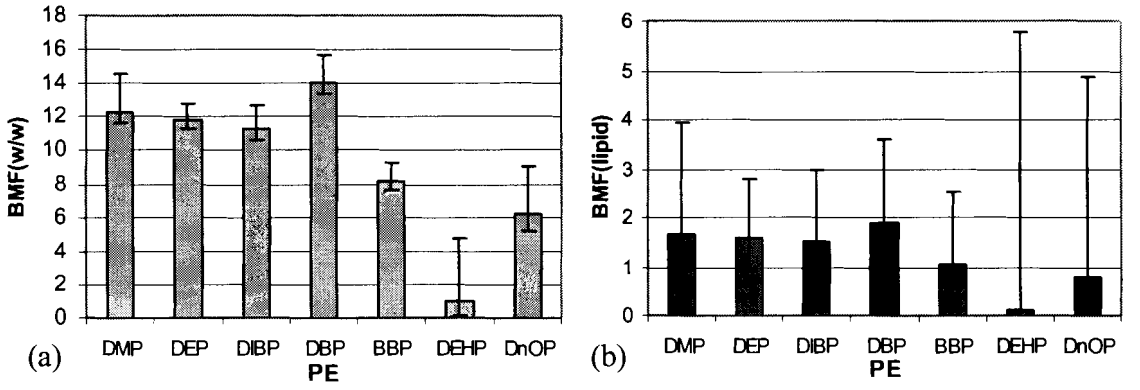


Figure 3.13 - Biomagnification factors of phthalate esters (+/- 1 standard deviation) for *Delphinapterus leucas* using (a) wet weight concentrations (ng/g wet wt), and (b) lipid equivalent concentrations (ng/g lipid eq)

The BMF_Ls that were calculated from the lipid equivalent concentrations were close to 1 (no magnification) ranging from 0.13 for DEHP to 1.87 for DBP (Figure 3.13b). The low molecular weight PEs exhibit BMF_Ls somewhat larger than 1.0. The low molecular weight phthalates are appreciably soluble in water and have low K_{OW}s which would preclude significant partitioning from the water to the cod via gill

ventilation. This could explain why the cod concentrations appear to be lower than the beluga. The higher molecular weight PEs show BMF_Ls less than 1.0. Statistically however, the BMF_Ls of all phthalates (with the exception of DEHP) were not significantly different from 1.0 ($p>0.05$). The lipid equivalent concentrations and fugacity of DEHP is statistically higher in cod than in beluga ($p<0.05$). Another exception may be DnNP, as this congener was too low to be detected in cod.

Biomagnification of total phthalate esters was determined using the mean of the sums of the concentrations (ΣPE) of seven of the eight compounds for beluga and cod. DnNP levels were not used because the majority of values were non-detectable on a wet weight basis. BMF_w for ΣPE was 6.04 (5.25 – 8.01) and the beluga concentration was significantly higher than that of cod ($p<0.05$). When lipid equivalent concentrations of ΣPE were used, the BMF_L was 0.58 (0 – 2.23) and the levels in beluga and cod are not statistically different ($p=0.65$).

From the ANOVA and biomagnification results it would appear that phthalate esters are not biomagnifying from prey (cod) to predator (beluga). The lipid equivalent concentrations in beluga and cod are approximately the same ($p > 0.05$) which would suggest that there is equilibrium partitioning of phthalates between predator and prey. The only exception is for DEHP, which is lower in beluga whales ($p<0.05$).

The same pattern emerges from the chemical fugacity data. The fugacity for DEHP is significantly higher in the prey which is an indication of trophic dilution. The unique behaviour of DEHP may be explained by the highly variable levels that were found in the beluga and cod but despite the range of data points, a significant difference between the two species was observed. DEHP is a very hydrophobic compound with a

log K_{OW} of 7.73 and it would readily be absorbed by cod. It also has a very high log K_{OA} of 10.53 and thus gill elimination and elimination to the air in fish and mammals respectively, can be expected to be low. It is likely however that the compound is being metabolically transformed at a faster rate than gill elimination or air elimination, and that the metabolic ability in mammals is greater than that in fish which could explain why the parent compound is not biomagnifying and accumulating in the beluga. Studies on rats suggested that DEHP in food is rapidly cleared from the body in some mammals and that the means of metabolic transformation of DEHP are not the same for all mammalian species(*37*). The DEHP discrepancy, could therefore be, in part, explained by congener-specific metabolic abilities. Marine mammals have illustrated congener-specific metabolic efficiencies for other compounds. The ability of cetaceans to metabolize PCBs, is dependent on the chemical structure of the congener and thus the patterns of PCBs in marine mammals typically do not reflect the patterns in their prey or the technical formulations originally released into the environment (*38*). A similar tendency could exist for phthalate esters which could clarify the dilution of DEHP with increasing trophic position.

High molecular weight phthalates (DEHP, DnOP and DnNP) exhibited trophic dilution in a marine aquatic food web along the coast of British Columbia (*39*). Although not statistically significant, DnOP concentrations and fugacities in the Arctic ecosystem appeared to be slightly higher in the beluga's diet. The notable exception is DnNP. DnNP concentrations are lower than the MDL for cod (1.3ng/g wet wt $\approx$ 545ng/g lipid eq) which may be indicative of trophic dilution within the aquatic food web, exclusive of mammals. The mean DnNP concentration in belugas is 1050ng/g lipid eq. and thus

trophic dilution of DnNP from cod to beluga is not likely to be possible on average. Whereas, the capacity to biotransform DEHP in marine mammals is probably high, the opposite might be true for DnNP resulting in the potential biomagnification of this compound from prey to mammalian predator.

The similarity in trophic positions of cod (3.6) and beluga (3.9) may be another reason for the lack of a difference between the predator and the prey as this difference in trophic position may not be significant enough to detect biomagnification. A review of cod-beluga BMFs of certain POPs from other study areas does show that biomagnification between these two species is possible. Muir et al. (1992) reported BMF_Ls for toxaphene, Σchlordane, ΣDDT, PCBs and HCB that were 4.6, 9.4, 9.8, 8.0, and 3.0 respectively for Arctic cod and beluga (*2*). Consequently, it is more likely that other processes (i.e. metabolic transformation) prevent PEs from biomagnifying. An additional explanation could be the dietary make-up of the belugas sampled. As previously mentioned, belugas are opportunistic feeders and there is no way of knowing the exact make-up of their individual diets. It is possible that lower trophic species were being consumed more regularly in this particular sub-sample of the population, in which case, the beluga trophic level would be similar or even lower than that of cod.

3.3 Biota-sediment distribution

Figure 3.14 compares the concentrations (ng/g lipid eq) and fugacities (nPa) of PEs in beluga, cod and sediment (sections 3.1.1, 3.1.2, and 3.1.3). Figure 3.14 suggests that lipid equivalent concentrations and fugacities for the low and intermediate molecular weight phthalates are similar for sediment, beluga and cod except for DEP which appears to have higher levels in the sediment when compared with the biota. DEHP seems to

have comparable levels in both beluga and sediment whereas the average DEHP concentration and fugacity in cod appears to be higher than that in sediment. For DnOP and DnNP, concentrations and fugacities appear to be lower in the sediment compared to both the beluga and cod levels.

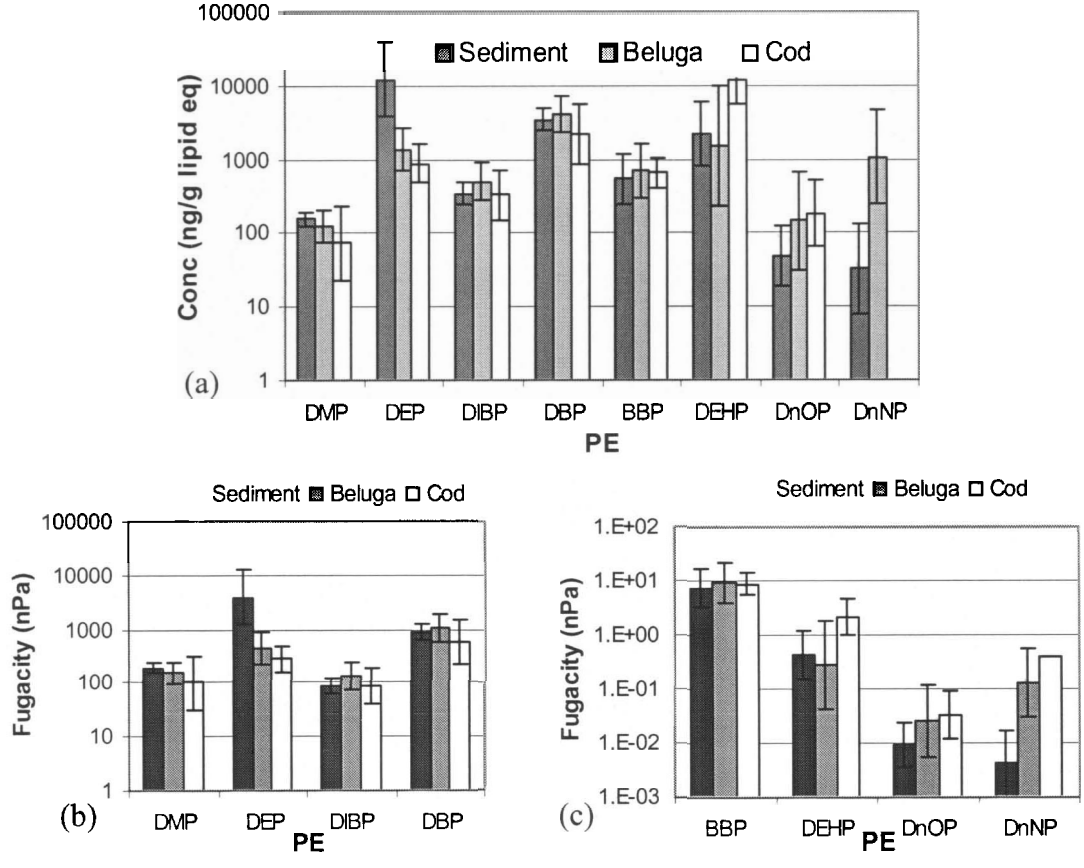


Figure 3.14 - Levels of PEs in *Delphinapterus leucas*, *Boreogadus saida* and Hudson's Bay sediment expressed in (a) lipid equivalent concentrations, and fugacity for (b) lower molecular weight, and (c) higher molecular weight (>300g/mol) phthalates.

ANOVA results that compare the biota and sediment concentrations (ng/g lipid equivalent) agree with many of the observations from the data. For the low and intermediate phthalates, with the exception of DEP, the lipid equivalent concentrations in sediment and biota are not significantly different ($p>0.05$) ($C_{sed} \cong C_{beluga} \cong C_{cod}$). DEP lipid equivalent concentrations are significantly greater ($p<0.05$) in sediment compared to

both cod and beluga ($C_{sed} > C_{beluga} \cong C_{cod}$). DEHP and DnOP concentrations (ng/g lipid eq) were significantly higher in cod ($p<0.05$) than in sediment but there was no significant difference between the beluga and sediment concentrations ($C_{cod} > C_{sed} \cong C_{beluga}$). Finally, DnNP concentrations (ng/g lipid) were significantly higher in beluga compared to sediment ($p<0.05$); this congener was not compared for cod and sediment due to a high number of non-detectable values in the analysis of the cod samples. ANOVA of fugacities yielded the same results.

BSAFs for beluga and cod are presented in Table B.3 and B.4 in Appendix B and are graphed together in Figure 3.15. Beluga BSAFs ranged from 0.32 kg OC/kg lipid (0 – 2.66) for DEP to 111 kg OC/kg lipid (110 – 115) for DnNP. The low molecular weight phthalates (DMP and DEP) had BSAFs less than three. The intermediate and high molecular weight phthalates all had BSAFs greater than three, except for DEHP, suggesting that these congeners are higher in beluga. This does not entirely agree with the ANOVA results from the lipid equivalent concentrations (ng/g lipid eq) because the BSAFs are not calculated using the lipid equivalent concentration for sediment. Rather the organic carbon normalized concentrations (ng/gOC) are used to determine the ratio between biota and sediment.

For cod, BSAFs ranged from 0.2 kg OC/kg lipid (0-2.49) for DEP to 15.5 kg OC/kg lipid (14.6-18.3) for DEHP (for all values see Table B.4). Unlike with beluga, the BSAF for DEHP in cod showed little variability and is convincingly greater than unity. For the remaining phthalates, the BSAFs exhibit a range that is similar to the beluga results but are lower than unity for DIBP and DBP.

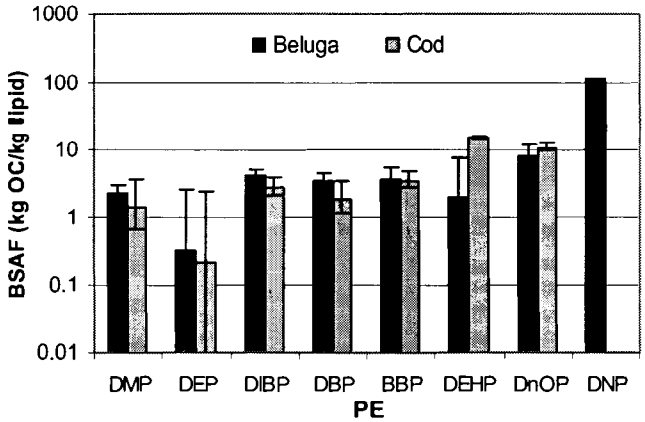


Figure 3.15 - BSAFs (kgOC/kglipid) of phthalate esters for *Delphinapterus leucas* and *Boreogadus saida*.

The analysis of the levels of PEs in beluga and sediment, and the BSAFs suggest that in general, the PE concentrations in beluga are similar to those in sediment with the exception of DEP and DnNP. DEP was significantly higher in the sediment and DnNP was significantly lower in the sediment. The pattern of BSAFs for beluga-sediment distribution in Figure 3.15, somewhat agrees with the observations from Mackintosh (*39*) where BSAFs between the sediment and the biota in an urban aquatic food-web exhibited a parabolic relationship with molecular weight and K_{OW} with the lowest BSAFs in DMP, the highest for mid-molecular weight phthalates and then a drop off for higher molecular weight PEs (See Figure B.1 in Appendix B). A parabolic relationship is observed from DMP to DEHP, in that only the mid-molecular weight compounds have BSAFs greater than unity, but the highest molecular weight compounds (DnOP and DnNP) do not follow for the Hudson's Bay samples. Another main difference between the Arctic food-web and the urban food web is that BSAFs for the mid and high molecular weight phthalates (except for DEHP) in the Hudson's Bay samples, were significantly greater than unity.

In the Mackintosh study, BSAFs were consistently less than 3 ranging from 0.0008kg OC/ kg lipid to just over 1kg OC/kg lipid, which is likely due to a sediment-water disequilibrium (*39*). In natural ecosystems, equilibrium between water and bottom sediment is in some cases not established for hydrophobic organic chemicals because of organic carbon decomposition (*40*). Rather, there tends to be a higher chemical fugacity in the sediment with a more pronounced disequilibrium for lower K_{OW} compounds. The disequilibrium is also related to the organic carbon content of the sediments. In Hudson's Bay, the organic carbon content is very low (0.18% +/- 0.1%) and thus it is difficult for a significant disequilibrium to exist. This could explain why the BSAFs are higher for several phthalate esters. To gain a better understanding of the sediment-water distribution in this particular ecosystem, water concentrations need to be determined.

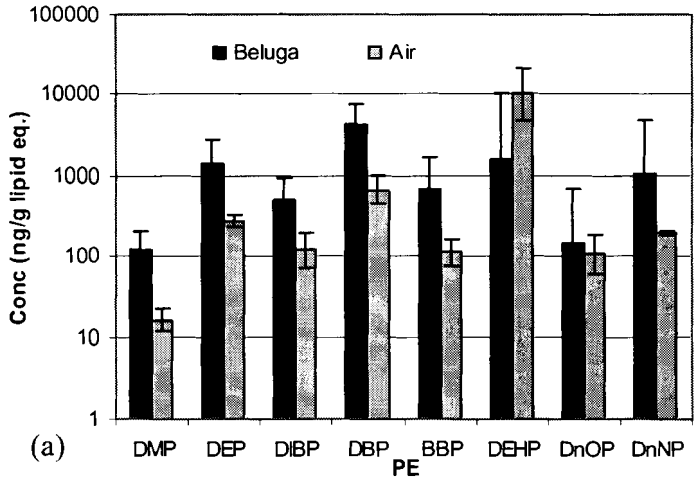
Belugas ingest sediment using suction while scavenging for benthic organisms. Consequently, the sediment can be considered a component of the beluga diet in terms of the chemical in the sediment being available for absorption in the gastro-intestinal tract. From this perspective it would appear as though the sediment could be a source of phthalate ester contamination for the animals, in addition to the cod, especially for DEP which has a higher fugacity and will thus be subject to a stronger fugacity gradient in the gut.

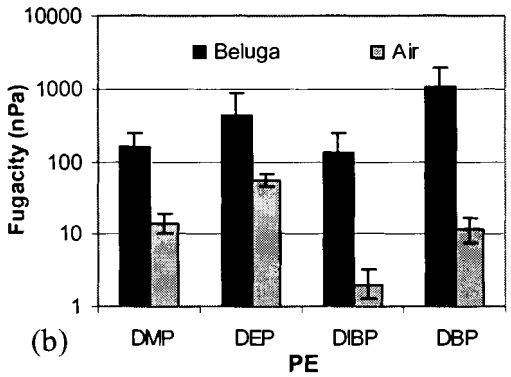
The BSAFs for cod are not greater than unity for DiBP and DBP but are for DEHP, DnOP and DnNP. Unlike in beluga, DEHP is significantly higher in cod with a BSAF of 15.5 kg OC/kg lipid (14.6 – 18.3).

3.4 Biota-air distribution

Figure 3.16 presents the beluga and air concentrations (ng/g lipid eq) and fugacities (nPa), which are compared using ANOVA, and a ratio of beluga and air concentrations (BAAFs). BAAFs are presented in Table B.5 in Appendix A. In Figure 3.16 the lipid equivalent concentrations and fugacities of phthalates in beluga appear to be higher than in air with the exception of DEHP. DEHP seems to have the opposite relationship due to the comparatively high lipid equivalent concentration in air. ANOVA tests of the lipid equivalent concentrations, confirm that all phthalates are significantly higher in beluga ($p<0.05$) than in air except for DEHP, in which case the null hypothesis could not be rejected. When statistically comparing fugacities in the two matrices, f_{beluga} was significantly higher than f_{air} for all phthalates except DEHP and DnOP. DEHP is on average higher in air than beluga but the difference was not significant ($p=0.1$).

The ratio between beluga and air concentrations (BAAFs) are shown in Figure 3.17; they ranged from 0.15 (0-5.8) for DEHP to 7.66 (7.19 to 8.37) for DMP. The range of BAAFs (+/- 1 standard deviation) is greater than unity for all phthalates except DEHP and DnOP. The large error bars for DEHP and DnOP are due to the highly variable beluga and air concentration data.





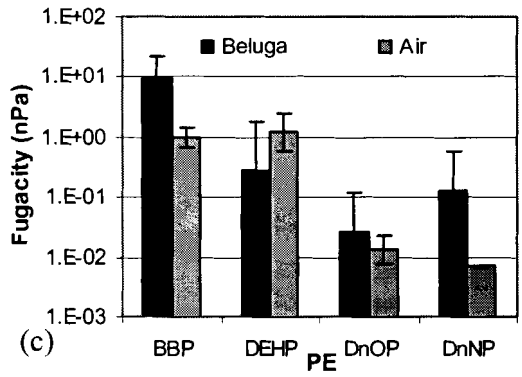


Figure 3.16 - Levels of Phthalate Esters (+/- 1 standard deviation) in *Delphinapterus leucas* and air expressed in terms of (a) lipid equivalent concentrations (ng/g lipid_eq.), and fugacity (nPa) for (b) lower molecular weight, and (c) higher molecular weight (>300g/mol) phthalates.

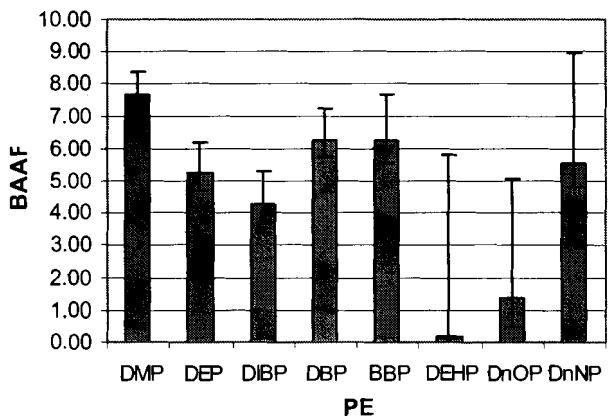


Figure 3.17 - Biota-air accumulation factors (+/- 1 standard deviation) for *Delphinapterus leucas* and air.

There does not appear to be a pattern in the BAAFs although it may be important to note that DEHP levels are comparatively high in air, as they were in cod. DEHP is the most heavily produced phthalate (*41*) and thus it would not be surprising for this compound to be found at relatively higher concentrations in the environment but on average f_{air} and f_{cod} were both higher than f_{beluga} which was not the case for the remaining phthalates. As mentioned in the discussion on biomagnification, it may be that belugas are efficient at metabolizing this congener once it is exposed to the chemical through the diet and air-inhalation. Efficient DEHP elimination was observed for other mammals (*38*).

Nevertheless, the fugacity of phthalate esters in air is noteworthy particularly for the higher molecular weight compounds and it is likely that air inhalation may be responsible for some PE contamination in belugas especially given that the K_{OA}s for these compounds are so high.

3.5 Relationship between K_{OA} and PE accumulation in Beluga

One of the hypotheses of this study was that phthalate ester biomagnification would increase with higher K_{OA}'s, provided that metabolic transformation is not occurring. Phthalate esters have very high octanol-air partitioning coefficients ranging from 10^7 to 10^{11} and thus it is hypothesized that once accumulated by the animals, the chemical will not be readily eliminated to the air through exhalation. This can result in biomagnification in beluga that is positively correlated with K_{OA}. A plot of BMF_Ls (C_{beluga}/C_{cod}) against log K_{OA} is shown in Figure 3.18.

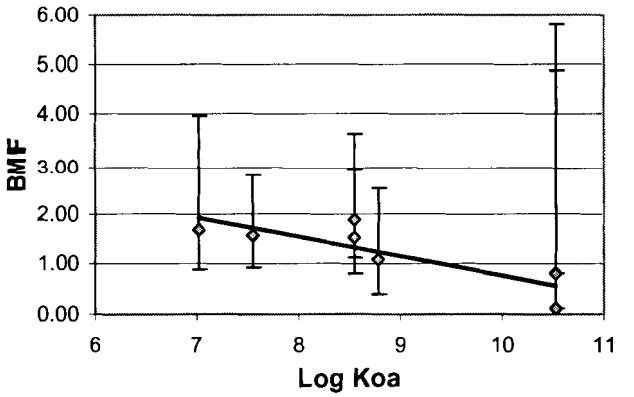


Figure 3.18 – Phthalate Ester Lipid Equivalent Biomagnification factors (+/- 1 standard deviation) for *Delphinapterus leucas* against log K_{OA} with a linear trend line (y =- 0.378x + 4.552).

The resulting relationship is the opposite of what was hypothesized. The slope of the regression line is negative (- 0.378) suggesting that biomagnification of PEs decreases with increasing K_{OA}. The R^2 for this relationship is 0.70, which convincingly indicates that these two parameters are not positively correlated. In not being positively correlated with K_{OA}, BMF is implicitly not positively related to K_{OW} and molecular weight as these properties are all positively correlated.

It is believed that the results are not in agreement with what was theoretically expected due to the metabolic transformation of phthalate esters which counteracts the magnification effect in the gut. The theory for the accumulation of chemicals in air-breathing organisms was conditional on the absence of significant rates of metabolism. Due to the apparent loss of chemical through metabolism, phthalate esters do not seem to be good candidates for testing this hypothesis.

Nevertheless, the relationship in Figure 3.18 agrees with the results presented in section 3.2 where the concentrations in the prey species (cod), relative to that of the predator (beluga), increased with higher molecular weights. Molecular weight and K_{OA} are positively correlated and thus the negative correlation between biomagnification and

octanol-air partitioning would be expected for this group of phthalate esters. This relationship may be due to the different rates of metabolic transfer relative to other routes of elimination. The lower molecular weight PEs do not appear to be very quickly metabolized with regard to their elimination rates whereas, high molecular weight PEs are broken down at significant rates compared to their elimination rates.

3.6 PE Concentrations versus Beluga Age and Length

When beluga samples were collected, the length of most animals was documented and the age was later determined from tooth analysis. For the 11 samples that were analyzed in this study, the length of 6 animals and the age of 6 animals were obtained. Both variables (age and length) were obtained for 5 animals (Table 3.1). The relationship between length and age is defined by the Gompertz equation *(42)*.

$$Length(cm) = A\exp(-B\exp(-kt))$$

where: A = asymptotic length(cm)
B = rate constant
k = rate constant
t = time (yrs)

Constants A, B and k have been defined for the Eastern Hudson's Bay beluga population by Doidge (1990) as 349, 0.28 and 0.71 respectively (*42*). The equation and the given constants, would suggest that the belugas being analyzed would not grow beyond 349cm, but as can be seen in Table 3.1, 5 of the 6 animals measured exceeded this length by up to 51cm. Consequently, this equation will not be used to determine missing age and length information.

Table 3.1 – Age (yrs) and Length (cm) of Male Beluga Whales

Beluga Sample ID	Age (years)	Length (cm)
PE394	12	350
PE395	38	390
PE396	34	400
PE397	13	370
PE398	n/a	383
PE402	21	n/a
PE404	19	330

For persistent organic pollutants, it is common for tissue concentrations to increase with age in marine mammals that have relatively long life-spans. This is especially true for males as they are not able to depurate themselves periodically through lactation. Consequently, the highest levels of POPs are typically found in older males (*31,43*). This study looked specifically at older males under the assumption that this group of the population would be most susceptible to accumulating high concentrations of phthalate esters. The length of beluga whales is believed to increase rapidly when the animal is approaching sexual maturity (approximately 8 years for males) and then growth begins to taper off asymptotically. During the rapid growth phase, growth dilution can be a significant means of 'chemical loss' after weaning (at approximately 1.5 years) as the amount of chemical in the animal gets redistributed over a larger body mass. As growth slows down however, POPs concentrations will increase with age (*44*).

These trends however are for POPs which are not readily metabolized in cetaceans. In the event of metabolic transformation, this relationship will not necessarily hold. Figure 3.19 shows ΣPEs (ng/g lipid) against age and length and Figures 3.20 and 3.21 show concentrations of individual PEs on a lipid weight basis as a function of the age and length respectively of seven sampled belugas. P-values for the regressions in

Figures 3.19, 3.20 and 3.21 are shown in Table 3.2. DnNP is not plotted as there were limited detectable concentrations for the animals that had been aged and measured.

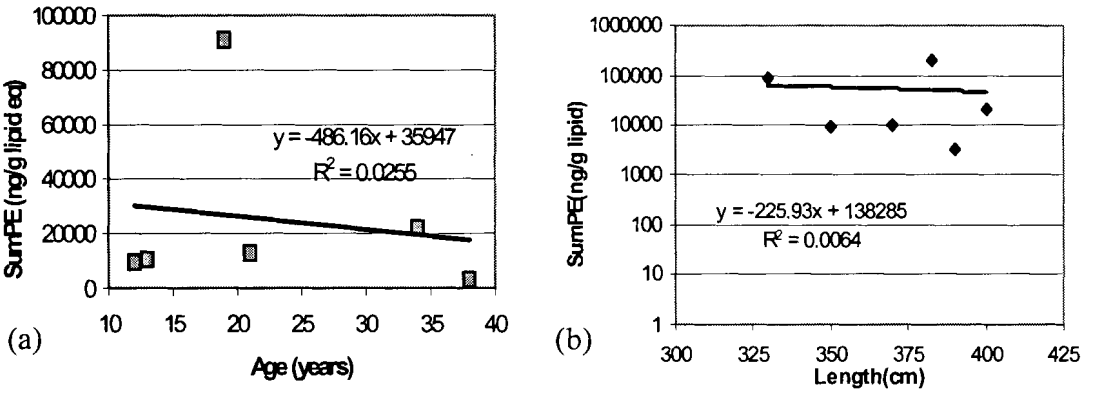


Figure 3.19 – ΣPE in Beluga Whales (*Delphinapterus leucas)* as a function of (a) age (years) and, (b) length (cm).

Table 3.2 – P-values for the Regressions of Phthalate Ester Concentrations (ng/g lipid eq) against Age (yrs) and Length (cm) for ΣPE and Individual Phthalate Esters

	p-value	
	Age	**Length**
ΣPE	0.76	0.88
DMP	0.55	0.08
DEP	0.46	0.05
DIBP	0.24	0.08
DBP	0.51	0.08
BBP	0.43	0.05
DEHP	0.88	0.26
DnOP	0.87	0.15

ΣPE shows no relationship with age or length as indicated by the low R^2 values and high p-values, although there appears to be an outlier in each regression. In Figure 3.19a, a 19 year old beluga has very high levels of PE contaminants although the relationship does not dramatically improve if this data point is left out of the regression ($R^2 = 0.03$). In Figure 3.19b, the highest concentration comes from the animal with a length of 383cm. This animal had a very high concentration of DEHP that was left out of

mean concentration and mean fugacity calculations, as well as ANOVA analysis as it was believed to be an outlier. This high DEHP level significantly increases the ΣPE concentration and thus it is considered an outlier in this case as well.

The slopes and r^2 values for the linear trend lines of each regression are shown on the graphs in Figures 3.20 and 3.21. Generally, the relationships of these regressions are negative for all PEs where a slope of zero indicates no change with age and length. A decrease in PE concentrations with length might indicate a loss of chemical through growth dilution although this phenomenon is more common before the age of sexual maturity (8 years for males) when growth is rapid (*44*). Nevertheless, growth dilution could be observed in the event of limited PE accumulation. Decreasing concentrations with age could be indicative of increased metabolic ability or changes in the diet as the animals age. If older animals switch to higher trophic level prey items than a decrease in concentrations with age could be observed.

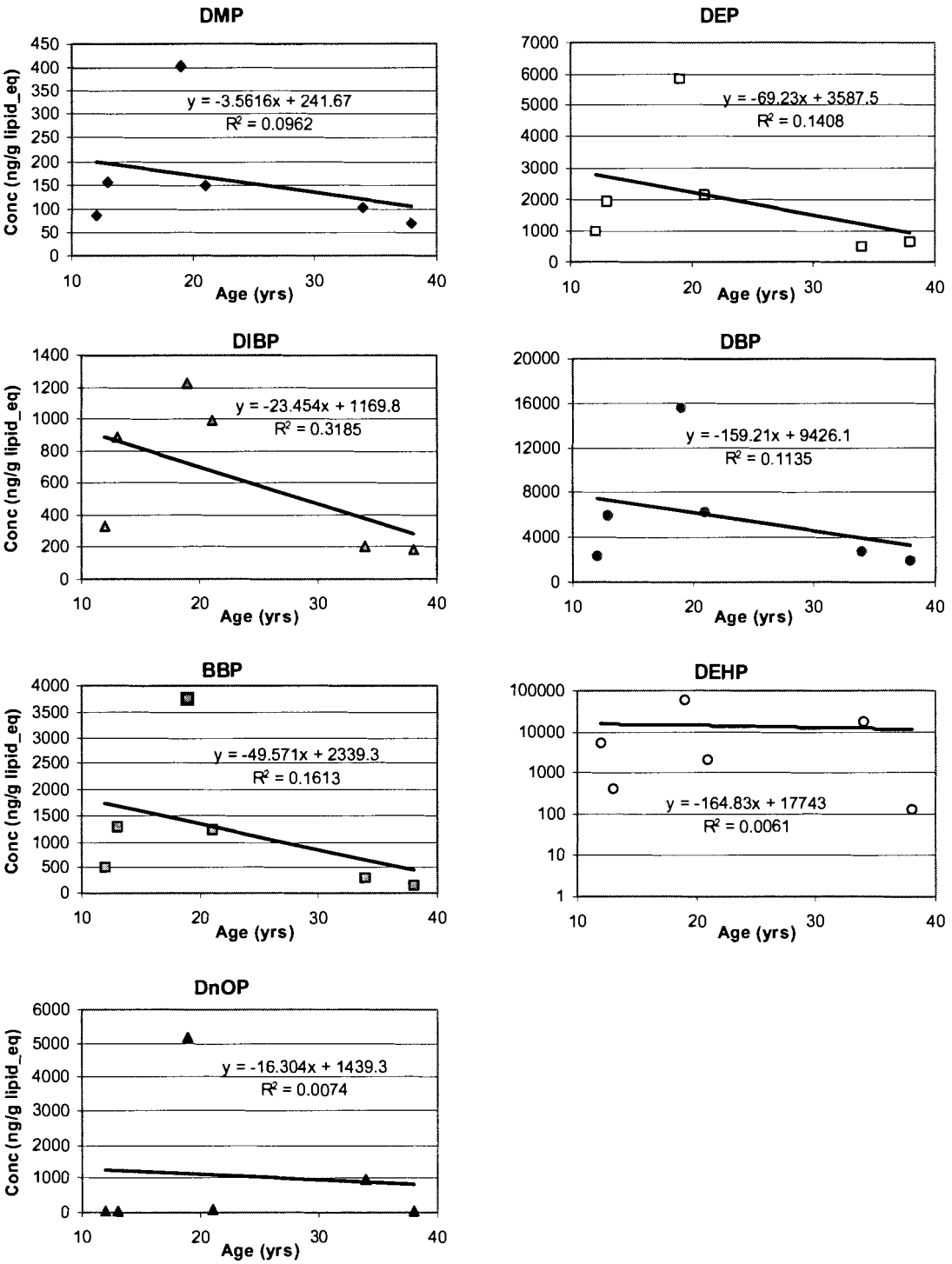


Figure 3.20 – PE concentrations (ng/g lipid) of *Delphinapterus leucas* as a function of age (years)

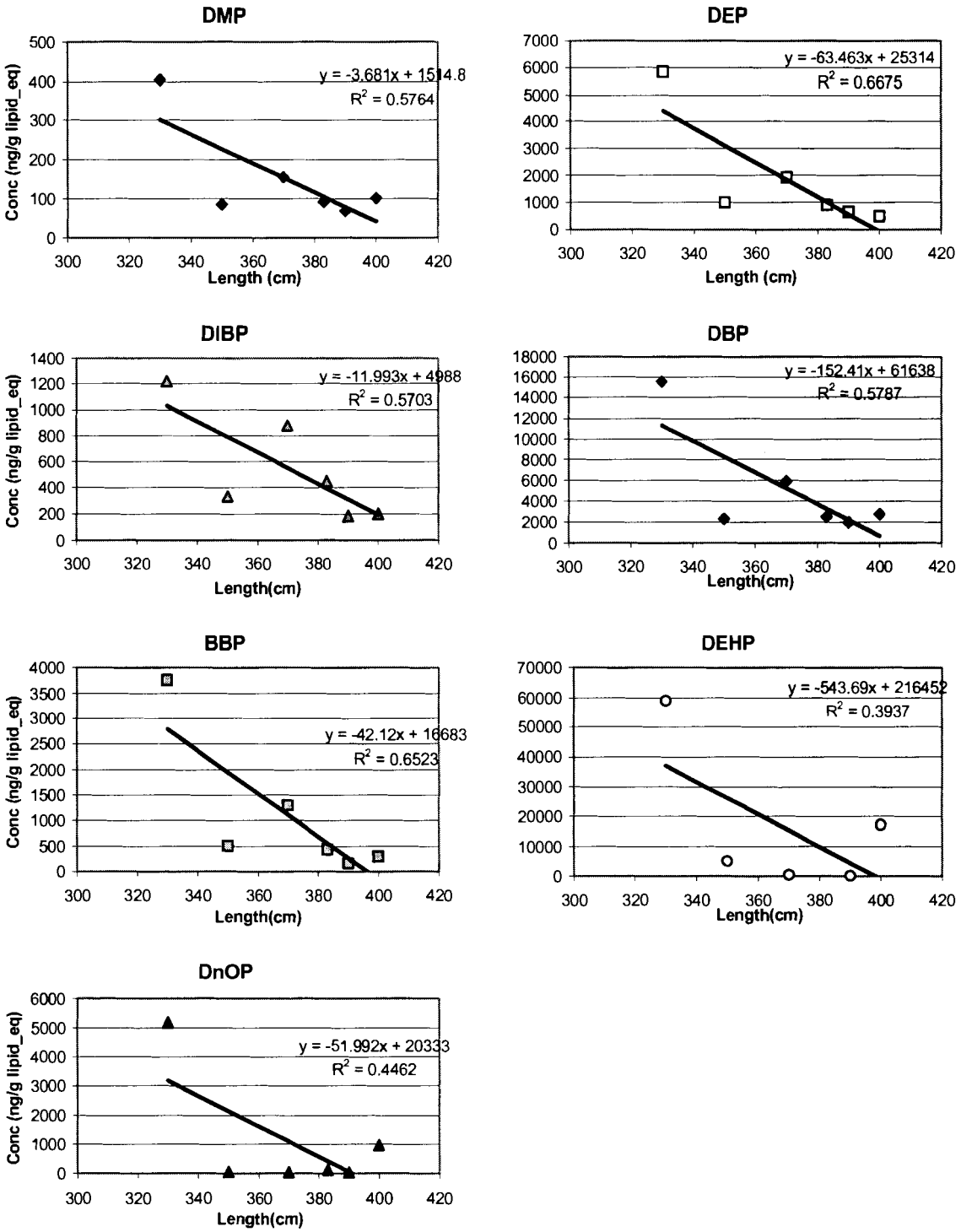


Figure 3.21 – PE concentrations (ng/g lipid) of *Delphinapterus leucas* as a function of length (cm)

Overall, the relationship between concentration and length is stronger than the relationship with age. The average R^2 value for the regressions in Figure 3.20 is 0.12 versus an average of 0.55 for the regressions in Figure 3.21. Also, the p-values for the concentration-length regression are considerably lower than the p-values for the concentration-age regressions. The low R^2 value in the concentrations-age regressions could be explained by the comparatively high PE concentrations in whale PE404. This animal has the highest concentrations of the six whales but is one of the youngest (19 years). The high levels in this animal could be a symptom of poor physical condition resulting from starvation. The lipid content in the liver of this animal is 0.26% compared to an average lipid content of 2.15% for the other whales. This is likely an indication of a malnourished animal with depleted lipid reserves which could explain its high levels of PEs. When lipid content is low, the amount of chemical becomes more concentrated. If the concentrations are plotted as a function of age without this animal, the average R^2 value goes up to 0.3. When this sample is omitted from the concentration-length regressions, the average R^2 value goes down to 0.19 and the relationship between the two parameters (i.e. slope) becomes positive for DnOP and DEHP.

To better characterize the changes in concentration with age and length, a greater sample size would need to be used. From the data that was examined, it can likely be concluded that PEs do not magnify with time in the same way that POPs do, otherwise we would see an indication of chemical increases with age and length whereas in the majority of cases, large negative slopes were found.

3.7 Arctic versus Urban Contamination of Phthalate Esters

In this section, concentrations and fugacities of phthalate esters in Hudson's Bay are compared to those in False Creek to assess the difference between a remote Arctic ecosystem and an urban ecosystem that has a history of industrial activity and subject to continued inputs from urban sources. It is often assumed that remote regions are less contaminated by anthropogenic pollutants due to the absence of any prevalent local sources. This is not always the case as many contaminants that are emitted at mid-latitudes, are introduced into the Polar Regions by way of atmospheric, fluvial and oceanic transport pathways *(13)*.

To compare the urban and arctic environments, levels of PEs in fish and sediment are compared using False Creek data from Mackintosh (2002) and the data presented in this study *(38)*. Lipid equivalent concentrations and fugacities of PEs in Pacific Staghorn Sculpin *(Leptocottus armatus)* are contrasted with the Arctic Cod data from section 3.1.2. Sculpin is being used for comparison as this species holds a trophic level in its food-web (i.e. 3.51) that is similar to that of Arctic Cod (i.e. 3.6 for adult fish). Figure 3.22 illustrates the average concentrations and fugacities for these two fish species.

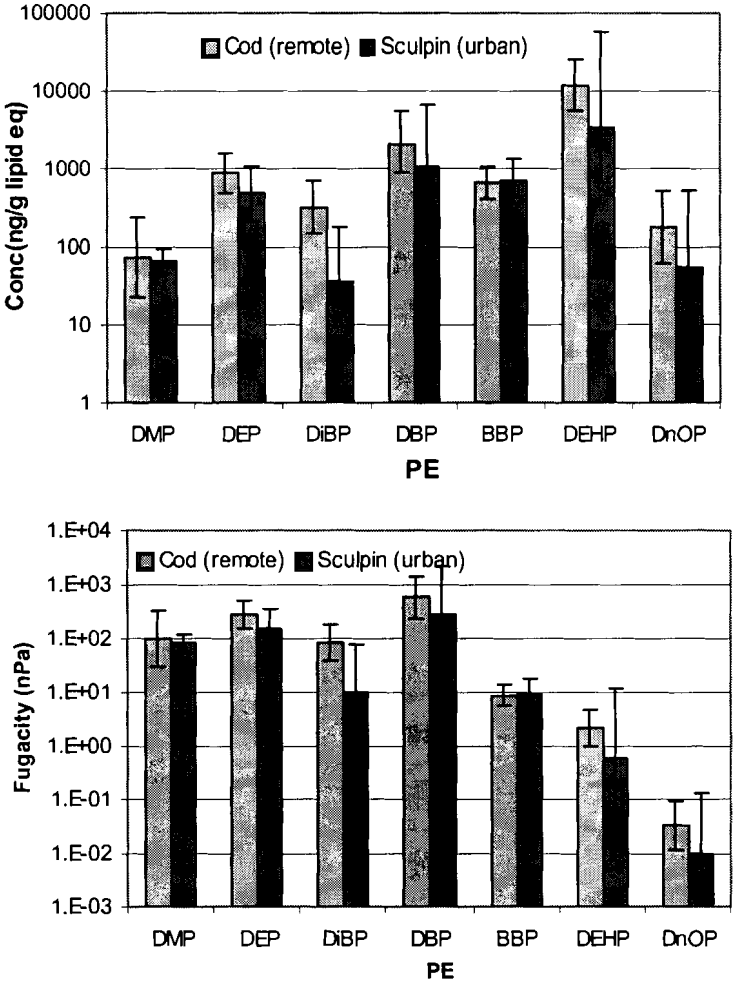


Figure 3.22 – Mean PE levels (+/- 1 standard deviation) for Pacific Staghorn Sculpin (*Leptocottus armatus*) from False Creek (data from Mackintosh, 2002) and Arctic Cod (*Boreogadus saida*) from Eastern Hudson's Bay, expressed in (a) lipid equivalent concentration (ng/g lipid), and (b) fugacity (nPa).

From this figure it appears as though the PE levels in Arctic Cod are slightly higher than in Sculpin for most compounds. ANOVA indicated that the DIBP concentration (ng/g lipid) and fugacity is significantly higher ($p<0.05$) in Arctic Cod when compared to that in sculpin. There was no significant difference between the two fish species for the other phthalate esters ($p>0.05$).

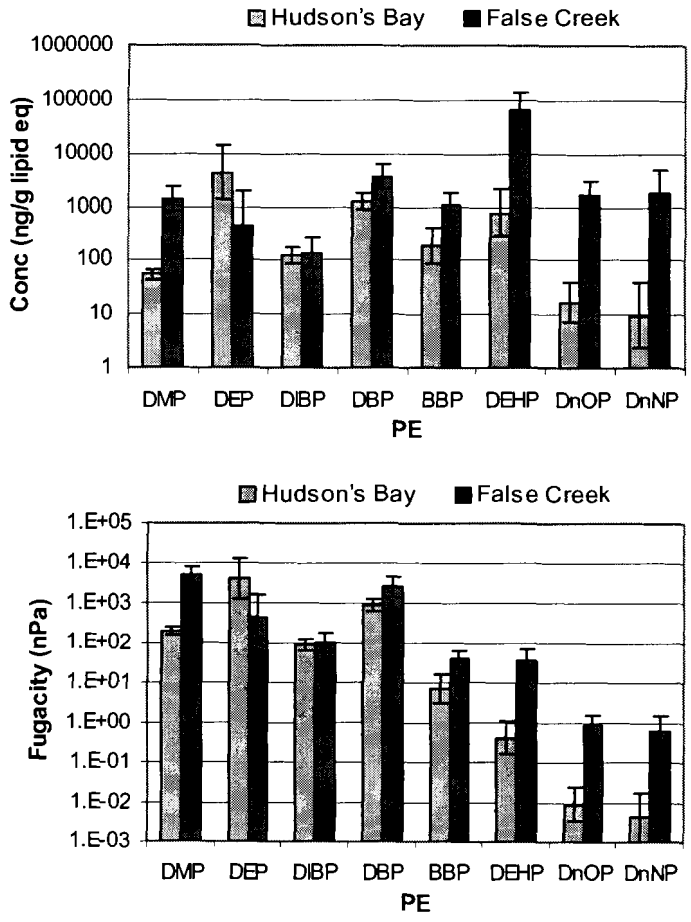


Figure 3.23 - Mean PE levels (+/- 1 standard deviation) for bottom sediment from False Creek (data from Mackintosh, 2002) and Eastern Hudson's Bay, expressed in (a) organic carbon normalized concentration (ng/g OC), and (b) fugacity (nPa).

A comparison of bottom sediment from the two study areas is shown in Figure 3.23. DEP sediment concentrations (ng/g OC) and fugacities (nPa) are significantly higher in Hudson's Bay ($p<0.05$). There was no significant difference in DIBP concentrations (ng/gOC) and fugacities (nPa) between the two sites ($p>0.05$) and for DMP, DBP, BBP, DEHP, DnOP and DnNP concentrations (ng/gOC) and fugacities (nPa) were significantly higher in False Creek sediment ($p<0.05$).

These two comparisons illustrate that the levels of PEs in the remote aquatic food-chain appear to be approximately the same. In some cases concentrations of some PEs in

the Arctic are higher than that in the urban inlet based on an examination of a third trophic level fish species. The relationship between the sediment levels in Hudson's Bay and False Creek appear to be dependant on the congener. Concentrations are higher in Hudson's Bay for DEP and similar for intermediate PEs (DIBP). Conversely, DMP and the higher molecular weight compounds are not as pronounced in the north when compared to False Creek.

Although, this is a limited comparison, this information strongly suggests that phthalate esters are being transported northwards from mid-latitudes because the concentrations in these two, very different ecosystems are not drastically different, which would be expected if estimates were based on the proximity of industrial activities. We cannot be certain what proportion of PE contamination in the Arctic is from foreign regions but considering that there are only a handful of potential local sources along Hudson's Bay (e.g. small landfills), it would appear as though phthalate esters are behaving in a similar way as certain POPs, in that these chemicals are making their way into the Arctic and sub-arctic environments.

4.0 Conclusions and Possible Research Extensions

This study was the first comprehensive look at the distribution of phthalate esters in the Arctic and in marine mammals. From the comparison with PE concentrations in an urban food web, it can be initially concluded that phthalate esters are in fact contaminating sub-arctic environments and that the geographic distribution of these chemicals appears to be ubiquitous, having similar levels of PEs in the North. In terms of distribution through this small section of the ecosystem, it does not appear as though phthalate esters are biomagnifying in beluga whales as the concentrations were, for the most part, similar to those in cod and sediment. Regressions of the concentration and age data, indicate that the body burden of the animals seem to decrease with time, at least within the age group that was analyzed. This is likely due the metabolic transformation of the chemicals. From a human health perspective, this lack of biomagnification would suggest that the Inuit population may not be more vulnerable than Southern Canadians to PE exposure through the diet because of the higher trophic level they occupy.

DEHP had the most unique pattern of distribution with higher fugacities in cod and air compared to beluga. This distribution was unique because lipid equivalent concentrations and fugacities in beluga were otherwise higher than air and the same as cod. This could be the result of efficient DEHP metabolism in the beluga. Loss through metabolism generally precludes an increase in biomagnification with K_{OA} and K_{OW}. Consequently, phthalate esters proved not to be ideal candidates for testing the hypothesis regarding bioaccumulation in air-breathing organisms.

This study represents a preliminary look at the presence of phthalate esters in an arctic marine ecosystem. Further research may clarify some of the findings presented

here. First, it would be interesting to compare the PE data with levels of a compound that is known to biomagnify (i.e. PCBs) in the same group of samples. This information would confirm whether or not the comparable levels in beluga and cod are due to similar trophic levels or metabolic transformation that counteracts magnification in the gut.

The apparent metabolic transformation of phthalate esters in beluga whales should not necessarily be interpreted as a positive outcome. Often times the metabolites of a parent compound can be equally, or more, detrimental in terms of toxicity and persistence. As an example, PCB and DDT metabolites are emerging as an important class of environmental contaminants having been linked to toxic and estrogenic effects *(45)*. Therefore, another important addition to this study would be to examine the levels of monoesters, which are the metabolites of phthalate esters, in the beluga and cod samples. Perhaps these by-products, which are believed to be more toxic than the parent compounds, are magnifying in the food chain.

Appendix A
Raw Concentration and Fugacity Data

Table A.1 Original Wet Weight Concentrations (ng/g wet wt) of PEs in Beluga Whale (Delphinapterus leucas) samples (n=11).

Sample ID	DMP	DEP	DIBP	DBP	BBP	DEHP	DnOP	DnNP
PE394	1.79	20.8	6.92	48.4	10.6	108	1.15	ND
PE395	1.52	14.3	4.00	43.5	3.59	2.80	0.99	ND
PE396	2.43	11.8	4.82	66.2	7.15	412	23.30	48.0
PE397	3.35	41.5	19.2	128	28.1	9.36	0.86	ND
PE398	2.02	20.0	9.99	56.2	9.73	4150*	2.72	ND
PE399	4.50	50.6	19.8	140	24.5	29.2	2.51	9.92
PE400	3.30	44.3	12.7	101	15.6	6.04	20.1	8.22
PE401	2.14	32.6	12.8	87.5	18.3	49.1	2.70	ND
PE402	2.05	29.7	13.7	86.7	16.9	27.8	1.47	ND
PE403	2.13	21.9	9.97	79.2	19.7	26.4	1.31	ND
PE404	1.05	15.1	3.15	40.1	9.72	152	13.4	19.4

Table A.2 Original Lipid Equivalent Weight Concentrations (ng/g lipid) of PEs in Beluga Whale (*Delphinapterus leucas*) samples (n=11).

Sample ID	DMP	DEP	DIBP	DBP	BBP	DEHP	DnOP	DnNP
PE394	85.7	998	332	2320	510	5160	55.1	ND
PE395	69.1	653	182	1980	164	127	45.2	ND
PE396	101	488	200	2750	297	17100	968	1990
PE397	154	1910	883	5910	1290	431	39.6	ND
PE398	90.9	900	449	2530	438	187000*	122	ND
PE399	179	2011	786	5560	972	1160	99.8	394
PE400	130	1740	501	3980	613	237	789	323
PE401	91.4	1390	544	3730	782	2100	115	ND
PE402	147	2140	988	6250	1220	2000	106	ND
PE403	127	1310	596	4740	1180	1570	78.3	ND
PE404	405	5850	1220	15500	3760	59000	5190	7520

**significant outlier and left out of further analysis*

Table A.3 Fugacity (nPa) of PEs in Beluga Whale (*Delphinapterus leucas*) Samples (n=11)

Sample ID	DMP	DEP	DIBP	DBP	BBP	DEHP	DnOP	DnNP
PE394	106	316	85.2	596	6.68	0.972	0.0104	ND
PE395	85.5	207	46.8	509	2.15	0.024	0.0085	ND
PE396	125	155	51.4	706	3.89	3.23	0.182	0.255
PE397	191	606	226	1520	16.9	0.081	0.0075	ND
PE398	112	285	115	648	5.73	n/a	0.0230	ND
PE399	221	637	202	1430	12.7	0.219	0.0188	0.0504
PE400	160	552	128	1020	8.02	0.045	0.149	0.0413
PE401	113	441	140	957	10.2	0.395	0.0217	ND
PE402	182	676	253	1600	16.0	0.377	0.0199	ND
PE403	158	415	153	1220	15.5	0.297	0.0147	ND
PE404	500	1850	313	3990	49.3	11.1	0.977	0.961

Table A.4 Original Wet Weight Concentrations (ng/g wet wt) of PEs in Arctic Cod (*Boreogadus saida*) samples (n=12).

Sample ID	DMP	DEP	DIBP	DBP	BBP	DEHP	DnOP	DnNP
PE378	0.0676	1.67	0.211	2.34	1.21	32.3	5.08	7.65
PE379	0.0564	1.54	0.491	2.61	0.657	36.0	0.426	ND
PE380	0.0848	1.34	0.355	3.47	1.64	12.2	0.281	ND
PE381	0.0469	0.957	0.312	1.20	2.71	26.1	0.432	ND
PE382	0.0863	1.12	0.857	3.98	1.09	128	0.813	ND
PE383	0.0843	2.16	0.706	2.22	0.750	10.5	0.172	ND
PE384	0.423	3.12	1.42	15.2	2.17	39.2	0.143	ND
PE385	1.03	2.63	1.69	25.2	1.58	47.6	0.542	ND
PE386	0.727	7.58	1.53	6.68	2.12	34.9	0.359	ND
PE387	0.754	3.14	1.36	8.36	1.58	37.6	0.977	ND
PE388	0.419	2.45	1.78	15.0	2.86	26.2	0.277	ND
PE389	0.367	1.91	1.41	9.34	2.16	10.5	ND	ND

Table A.5 Original Lipid Equivalent Concentrations (ng/g lipid) for Arctic Cod (*Boreogadus saida*) samples (n=12).

Sample ID	DMP	DEP	DIBP	DBP	BBP	DEHP	DnOP	DnNP
PE378	26.7	659	83.3	924	477	12800	2000	3020
PE379	31.7	863	276	1470	369	20200	239	ND
PE380	35.3	555	148	1440	683	5060	117	ND
PE381	19.1	389	127	486	1100	10600	176	ND
PE382	42.3	548	420	1950	535	63000	399	ND
PE383	29.5	756	247	777	263	3690	60.1	ND
PE384	121	892	406	4350	621	11200	40.8	ND
PE385	367	943	606	9030	566	17000	194	ND
PE386	364	3790	763	3340	1060	17500	180	ND
PE387	377	1570	679	4180	791	18800	489	ND
PE388	204	1190	865	7300	1390	12700	134	ND
PE389	167	873	643	4260	988	4780	ND	ND

Table A.6 Fugacity (nPa) of PEs in Arctic Cod (Boreogadus saida) samples (n=12).

Sample ID	DMP	DEP	DIBP	DBP	BBP	DEHP	DnOP	DnNP
PE378	33.0	209	21.4	237	6.24	2.40	0.3771	0.386
PE379	39.2	273	70.8	376	4.83	3.81	0.0451	ND
PE380	43.6	176	37.9	370	8.94	0.952	0.0220	ND
PE381	23.6	123	32.5	125	14.4	1.99	0.0331	ND
PE382	52.3	174	108	500	7.00	11.9	0.0751	ND
PE383	36.5	240	63.5	199	3.44	0.694	0.0113	ND
PE384	149	282	104	1120	8.13	2.11	0.00769	ND
PE385	454	299	156	2320	7.40	3.21	0.0365	ND
PE386	450	1200	196	856	13.9	3.29	0.0338	ND
PE387	466	496	174	1070	10.4	3.54	0.0920	ND
PE388	252	376	222	1870	18.2	2.40	0.0253	ND
PE389	207	277	165	1090	12.9	0.899	ND	ND

Table A.7 Original Dry Weight Concentrations (ng/g dry wt) of PEs in Hudson's Bay sediment samples (n=11).

Sample ID	DMP	DEP	DIBP	DBP	BBP	DEHP	DnOP	DnNP
PE361	0.21	8.83	0.311	3.23	0.659	1.32	0.0425	0.0748
PE362	0.05	12.3	0.103	1.19	0.303	0.587	ND	0.0467
PE363	0.07	15.0	0.107	1.30	0.242	1.39	ND	0.0370
PE364	0.04	36.8	0.075	1.12	0.445	0.990	0.141	0.231
PE365	0.07	12.7	0.205	1.91	0.242	0.573	0.0103	0.0118
PE366	0.10	3.00	0.280	2.35	0.175	16.7	0.0334	0.0272
PE367	0.08	1.66	0.262	2.17	0.340	0.986	0.0172	0.0069
PE368	0.15	16.8	0.317	2.67	0.569	1.97	0.0595	0.0243
PE369	0.09	4.41	0.216	1.84	0.190	0.594	0.0183	0.0038
PE370	0.10	2.48	0.307	3.14	1.24	0.705	0.0308	0.0083
PE371	0.07	3.41	0.157	1.42	0.070	1.97	0.0255	0.0177

Table A.8 Original Lipid Equivalent Concentrations (ng/g OC) of PEs in Hudson's Bay sediment samples (n=11).

Sample ID	DMP	DEP	DIBP	DBP	BBP	DEHP	DnOP	DnNP
PE361	184	7640	269	2800	571	1150	36.8	64.8
PE362	172	40700	343	3940	1000	1940	ND	154
PE363	163	32600	233	2840	527	3020	ND	80.6
PE364	155	138000	281	4180	1670	3710	527	865
PE365	145	25500	411	3820	484	1140	20.6	23.7
PE366	163	4900	458	3850	287	27300	54.6	44.5
PE367	199	4130	653	5420	848	2460	43.0	17.3
PE368	110	12500	237	1990	425	1470	44.4	18.1
PE369	97.3	4760	233	1990	205	640	19.7	4.07
PE370	169	4300	532	5440	2160	1220	53.6	14.4
PE371	194	9270	426	3870	189	5340	69.3	48.1

Table A.9 Fucagity (nPa) of PEs in Hudson's Bay sediment samples (n=11).

Sample ID	DMP	DEP	DIBP	DBP	BBP	DEHP	DnOP	DnNP
PE361	228	2420	69.1	717	7.47	0.216	0.007	0.008
PE362	212	12900	87.9	1010	13.1	0.366	N/D	0.0197
PE363	201	10300	59.7	729	6.90	0.569	N/D	0.0103
PE364	192	43600	72.2	1073	21.82	0.698	0.0992	0.111
PE365	180	8060	105	981	6.34	0.216	0.0039	0.0030
PE366	201	1553	118	989	3.76	5.134	0.0103	0.0057
PE367	246	1310	167.5	1390	11.10	0.463	0.0081	0.0022
PE368	136	3960	60.7	511	5.56	0.276	0.0084	0.0023
PE369	120	1510	59.7	510	2.69	0.121	0.0037	0.0005
PE370	208	1360	137	1397	28.29	0.231	0.0101	0.0018
PE371	240	2940	109	994	2.48	1.01	0.0131	0.0061

Table A.10 Original Dry Weight Concentrations (ng/g dry wt) of PEs in Lichen (*Cladina rangeferina)* samples (n=7).

Sample ID	DMP	DEP	DIBP	DBP	BBP	DEHP	DnOP	DnNP
PE405	0.230	4.88	1.58	10.4	1.69	262	2.76	ND
PE406	0.218	4.00	1.81	8.93	1.60	534	0.772	ND
PE407	0.402	6.39	4.25	25.6	4.25	109	2.98	3.38
PE408	0.239	5.72	4.37	16.6	1.66	177	1.31	ND
PE409	0.363	5.32	1.28	8.21	1.70	401	2.43	3.46
PE410	0.450	4.21	2.26	12.8	2.31	102	2.73	3.59
PE411	0.371	4.85	2.13	12.4	2.99	84.6	3.03	3.91

Table A.11 Original Lipid Equivalent Concentrations (ng/g lipid equivalent) of PEs in Lichen (*Cladina rangeferina*) samples (n=7).

Sample ID	DMP	DEP	DIBP	DBP	BBP	DEHP	DnOP	DnNP
PE405	12.1	257	83.3	545	89.0	13800	145	ND
PE406	11.5	210	95.2	470	84.2	28100	40.6	ND
PE407	21.1	336	223	1350	224	5750	157	178
PE408	12.6	301	230	875	87.5	9320	69.1	ND
PE409	19.1	280	67.2	432	89.5	21100	128	182
PE410	23.7	221	119	675	122	5370	143	189
PE411	19.5	255	112	651	158	4450	160	206

Table A.12 Fugacity (nPa) of PEs in Lichen (*Cladina rangeferina*) samples (n=7).

Sample ID	DMP	DEP	DIBP	DBP	BBP	DEHP	DnOP	DnNP
PE405	10.1	53.9	1.43	9.35	0.78	1.73	0.02	N/D
PE406	9.6	44.2	1.63	8.07	0.74	3.52	0.01	N/D
PE407	17.6	70.7	3.84	23.14	1.97	0.72	0.02	0.01
PE408	10.5	63.3	3.95	15.02	0.77	1.17	0.01	N/D
PE409	15.9	58.8	1.15	7.42	0.79	2.64	0.02	0.01
PE410	19.7	46.5	2.04	11.59	1.07	0.67	0.02	0.01
PE411	16.3	53.6	1.93	11.17	1.39	0.56	0.02	0.01

Table A.13 Original Estimated Concentrations (ng/g) for Air (n=7).

Sample ID**	DMP	DEP	DIBP	DBP	BBP	DEHP	DnOP	DnNP
PE405	1.18E-06	7.24E-06	2.40E-07	1.57E-06	1.48E-07	4.07E-07	4.29E-09	ND
PE406	1.12E-06	5.93E-06	2.75E-07	1.36E-06	1.40E-07	8.30E-07	1.20E-09	ND
PE407	2.07E-06	9.48E-06	6.45E-07	3.89E-06	3.71E-07	1.70E-07	4.63E-09	1.66E-09
PE408	1.23E-06	8.49E-06	6.63E-07	2.52E-06	1.45E-07	2.75E-07	2.04E-09	ND
PE409	1.87E-06	7.89E-06	1.94E-07	1.25E-06	1.49E-07	6.23E-07	3.78E-09	1.70E-09
PE410	2.31E-06	6.24E-06	3.42E-07	1.95E-06	2.02E-07	1.59E-07	4.23E-09	1.76E-09
PE411	1.91E-06	7.19E-06	3.24E-07	1.88E-06	2.61E-07	1.31E-07	4.71E-09	1.92E-09

Table A.14Original Estimated Lipid Equivalent Concentrations (ng/g lipid equivalent) for Air (n=7).

Sample ID**	DMP	DEP	DIBP	DBP	BBP	DEHP	DnOP	DnNP
PE405	12.1	257	83.3	545	89.0	13800	145	ND
PE406	11.5	210	95.2	470	84.2	28100	40.6	ND
PE407	21.1	336	223	1350	224	5750	157	178
PE408	12.6	301	230	875	87.5	9320	69.1	ND
PE409	19.1	280	67.2	432	89.5	21100	128	182
PE410	23.7	221	119	675	122	5370	143	189
PE411	19.5	255	112	651	158	4450	160	206

Table A.15Fugacity (nPa) of PEs in Air (n=7).

Sample ID**	DMP	DEP	DIBP	DBP	BBP	DEHP	DnOP	DnNP
PE405	10.1	53.9	1.43	9.35	0.78	1.73	0.02	N/D
PE406	9.55	44.2	1.63	8.07	0.74	3.52	0.01	N/D
PE407	17.6	70.7	3.84	23.1	1.97	0.72	0.02	0.01
PE408	10.5	63.3	3.95	15.0	0.77	1.17	0.01	N/D
PE409	15.9	58.8	1.15	7.42	0.79	2.64	0.02	0.01
PE410	19.7	46.5	2.04	11.6	1.07	0.67	0.02	0.01
PE411	16.3	53.6	1.93	11.2	1.39	0.56	0.02	0.01

***Sample IDs are from lichen samples that were used to estimate air concentrations and fugacities*

Appendix B
Biomagnification Factors

Biota Sediment Accumulation Factors

Biota Air Accumulation Factors

Table B.1 PE Wet Weight Biomagnification Factors (BMF_Ws) with negative (SD-) and positive (+SD) standard deviations

Phthalate Ester	BMF_W	SD-	SD+
DMP	12.3	0.760	2.23
DEP	11.8	0.577	0.980
DIBP	11.3	0.700	1.40
DBP	14.0	0.698	1.63
BBP	8.2	0.588	1.02
DEHP	1.0	0.933	3.74
DnOP	6.2	0.943	2.92
DnNP	-	-	-

Table B.2 PE Lipid Equivalent Biomagnification Factors (BMFLs) with negative (SD-) and positive (+SD) standard deviations

Phthalate Ester	BMF_L	SD-	SD+
DMP	1.67	0.785	2.27
DEP	1.57	0.660	1.24
DIBP	1.52	0.710	1.45
DBP	1.87	0.748	1.71
BBP	1.07	0.686	1.46
DEHP	0.13	1.00	5.66
DnOP	0.79	1.02	4.09
DnNP	-	-	-

Table B.3 PE Biota Sediment Accumulation Factors (kgOC/kg Lipid) for Beluga Whale (*Delphinapterus leucas*) with negative (SD-) and positive (+SD) standard deviations

Phthalate Ester	BSAF	SD-	SD+
DMP	2.29	0.432	0.668
DEP	0.324	0.320	2.34
DIBP	4.19	0.552	0.962
DBP	3.42	0.531	0.899
BBP	3.71	0.788	1.78
DEHP	1.97	1.06	5.82
DnOP	8.92	0.999	3.99
DnNP	110.8	1.08	4.60

Table B.4 PE Biota Sediment Accumulation Factors (kgOC/kg Lipid) for Arctic Cod (*Boreogadus saida*) with negative (SD-) and positive (+SD) standard deviations

Phthalate Ester	BSAF	SD-	SD+
DMP	1.37	0.715	2.20
DEP	0.207	0.200	2.28
DIBP	2.75	0.619	1.25
DBP	1.83	0.669	1.57
BBP	3.45	0.668	1.36
DEHP	15.6	0.834	2.11
DnOP	11.2	0.898	2.47

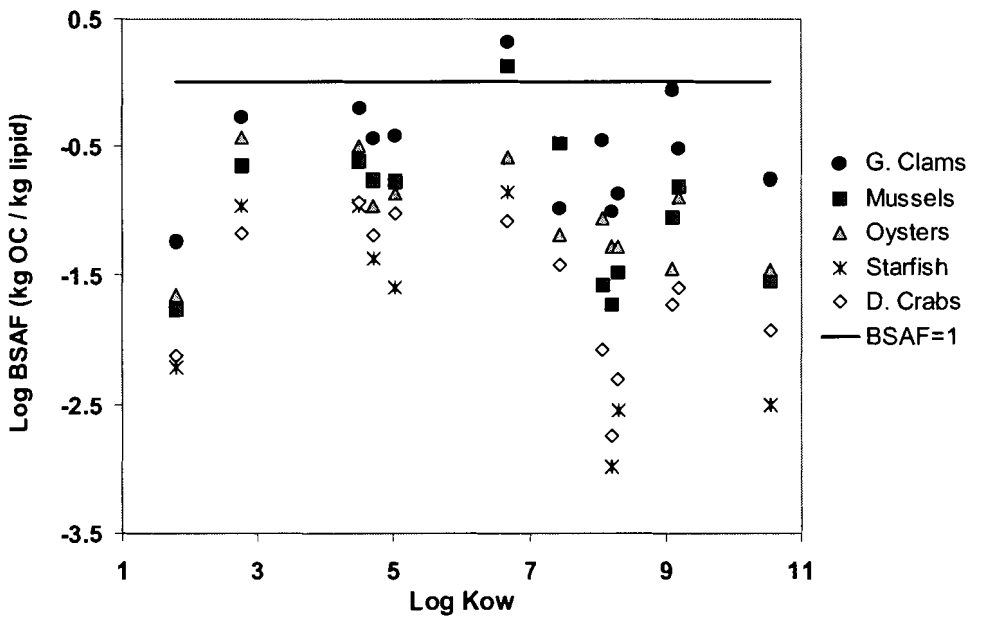


Figure B.1 Biota - Sediment Accumulation Factors (kg OC / kg lipid) on a Logarithmic Scale versus Log Octanol - Seawater Partition Coefficients for Phthalate Esters in Benthic Marine Biota from False Creek Harbour (Taken from Mackintosh, 2002).

Table B.5 PE Lipid Equivalent Biota Air Accumulation Factors (BAAFs) for Beluga Whale (*Delphinapterus leucas*) with negative (SD-) and positive (+SD) standard deviations

Phthalate Ester	BAAF	SD-	SD+
DMP	7.66	0.462	0.712
DEP	5.23	0.512	0.973
DIBP	4.28	0.596	1.05
DBP	6.26	0.550	0.932
BBP	6.23	0.649	1.39
DEHP	0.154	0.990	5.64
DnOP	1.37	0.884	3.71
DnNP	5.58	0.775	3.40

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