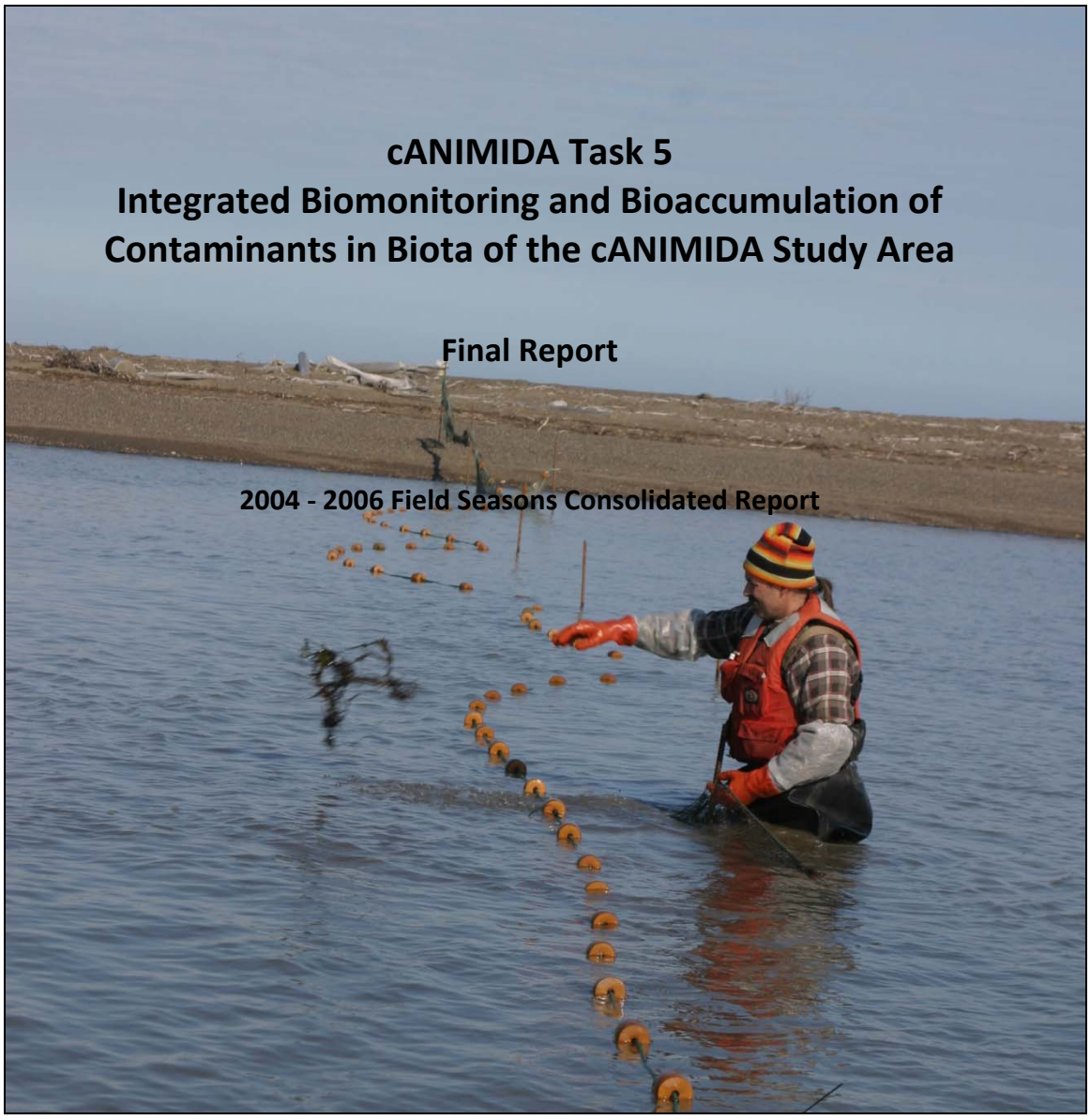
OCS Study MMS 2009-037

cANIMIDA Task 5
Integrated Biomonitoring and Bioaccumulation of Contaminants in Biota of the cANIMIDA Study Area

Final Report

2004 - 2006 Field Seasons Consolidated Report



Submitted to:
Minerals Management Service
Alaska OCS Office
Anchorage, Alaska

Submitted by:
Neff & Associates LLC
20 Templewood Drive
Duxbury, Massachusetts 02332

U.S. Department of the Interior
Minerals Management Service
Alaska Outer Continental Shelf Region

Contract No.
M04PC00020

This page intentionally left blank

Task 5
Integrated Biomonitoring and Bioaccumulation of Contaminants in Biota of the cANIMIDA Study Area

Final Report

2004-2006 Field Seasons Consolidated Report

October 2009

Prepared for:
Minerals Management Service
Alaska OCS Office
Anchorage, Alaska

Prepared by:
Jerry M. Neff, Principal Investigator
Neff & Associates LLC
20 Templewood Drive
Duxbury, Massachusetts 02332

John H. Trefry
Dept. of Marine and Environmental Systems
Florida Institute of Technology
150 West University Blvd.
Melbourne, FL 32901

Gregory Durell
Battelle
397 Washington Street
Duxbury, Massachusetts 02332

(e-mail of corresponding author: neffjm@comcast.net)

This study was funded by the United States Department of the Interior, Minerals Management Service, Alaska Outer Continental Shelf Region, Anchorage, Alaska, under Contract No. M04PC00020 as part of the MMS Alaska Environmental Studies Program.

This page intentionally left blank

Table of Contents

LIST OF TABLES

LIST OF FIGURES

APPENDICES ON ATTACHED CD

A. Summary of Biological Samples Collected in 2004, 2005, and 2006 for Analysis of Hydrocarbons, Metals, and Biomarkers
B. Summary of Analytical Methods
C. Summary of Quality Control (QC) Results
D. Summary of Chemistry Data
    1. Organic Chemicals: Polycyclic Aromatic Hydrocarbons, Saturated Hydrocarbons, Petroleum Biomarkers (Steranes/Triterpanes) in Tissues
    2. Metals: Ag, Al, As, Ba, Be, Cd, Co, Cr, Cu, Fe, Hg, Ni, Pb, Sb, Se, Tl, V, Zn
    3. Biological Biomarkers: Fluorescing Aromatic Compounds in Fish Bile; Cytochrome P450A1 Staining in Fish Tissues.
E. Field Survey Reports for 2004, 2005, and 2006
F. cANIMIDA Indicator Matrix for Decision Making

This page intentionally left blank

EXECUTIVE SUMMARY

The continuation of the Arctic Nearshore Impact Monitoring in the Development Area (cANIMIDA) Program was designed as an extension of the ANIMIDA Program. It was designed to monitor changes in the distribution and concentrations of selected chemicals in the nearshore Alaskan Beaufort Sea environment and determine if any changes observed were related to offshore oil and gas development activities. Task 5 of cANIMIDA focuses on an assessment of concentrations of petroleum hydrocarbons, including polycyclic aromatic hydrocarbons (PAH), saturated hydrocarbons (SHC), and sterane/triterpane (StTr) petroleum biomarkers, and 13 to 19 metals in soft tissues of representative species of marine bivalve mollusks, crustaceans, and fish from the offshore areas in the Alaskan Beaufort Sea near ongoing or planned oil and gas development activities. This report describes the results of chemical and biochemical monitoring of marine animals collected from the study area during the summers of 2004, 2005, and 2006.

There were five objectives for Task 5 of the cANIMIDA Program:

Objective 1 – Improve and validate the Contractor's proposed conceptual model of bioaccumulation and trophic interaction in cANIMIDA biota

Objective 2 – Measure bioaccumulation in selected species by co-collection and analysis of indigenous bivalves, benthic amphipods, and fish and deployment, retrieval, and analysis of caged bivalves and SPMDs

Objective 3 - Compare bioaccumulation data to published results for the same or similar species outside the cANIMIDA area

Objective 4 - Develop a strategy for longer-term upper trophic contaminant monitoring

Objective 5 - Develop a strategy and rationale for future Boulder Patch contaminant monitoring in conjunction with Task Order 006

Biota samples (amphipods, isopods, clams, and fish) were collected and mussels and semipermeable membrane devices (SPMDs) were deployed and then recovered at several sampling sites near the Northstar production facility, the Liberty prospect, and at reference locations, mostly east (up-current) of development activities. The samples were analyzed for a complete suite of PAH (naphthalene through benzo(ghi)perylene and selected alkyl homologues) and 19 metals (silver, aluminum, arsenic, barium, beryllium, cadmium, cobalt, chromium, copper, iron, mercury, manganese, nickel, lead, antimony, selenium, thallium, vanadium, and zinc); fish samples were not analyzed for aluminum, beryllium, cobalt, antimony, and thallium. Clam, amphipod, and isopod tissues also were analyzed for SHC and StTr crude oil biomarkers. Fish tissues were evaluated histochemically for cytochrome P450 mixed function oxygenase (CYP1A) activity and fish bile was analyzed for fluorescent aromatic compounds (FAC) as evidence of PAH exposure.

Concentrations of hydrocarbons (PAH, SHC, StTr) and all metals analyzed were relatively low in all three sampling years in tissues of several species of fish, amphipods, isopods, and clams collected in the Northstar and Liberty prospect areas and in reference areas, and in mussels deployed in cages at several Beaufort Sea locations (Tables ES-1 through ES-4). PAH concentrations in all biota were higher in 2004 and 2006 than in 2005 (Tables ES-1 and ES-2). Northstar is the only area in Federal waters of the Alaska outer continental shelf with oil production. PAH concentrations usually were similar in resident marine animals and deployed mussels and SPMDs near Northstar and at other stations in all three years.

Table ES–1. Mean Concentrations of total polycyclic aromatic hydrocarbons (TPAH), total saturated hydrocarbons (TSHC), pristane, and steranes/triterpanes (StTr) in tissues of indigenous fish, clams, amphipods, isopods, and mysids collected at several locations in the Alaskan Beaufort Sea in 2004 through 2006. BSMP is the Beaufort Sea Monitoring Program area where monitoring was performed in the ANIMIDA Program.

Taxon	**Analytes**	**Location**	**Mean (SD) TPAH Concentration (ng/g)**		
			2004	**2005**	**2006**
Fish (8 species)	TPAH	Northstar	38.8 ± 20.7	9.44 ± 6.85	**52.4 ± 12.8**
		Liberty	47.8 ± 25.3	10.1 ± 6.45	**24.8 ± 6.4**
		Tigvariak Island	30.4 ± 7.7	No data	No data
Amphipods	TPAH	Northstar	83.3 ± 52.7	13.8 ± 8.96	41.3 ± 27.4
		Liberty	73.6 ± 10.3	39.5 ± 10.1	81.5 ± 23.9
		BSMP	49.5 ± 16.7	23.6 ± 9.24	60.9 ± 14.4
	TSHC	Northstar	31,458 ± 2784	18,003 ± 1746	26,681 ± 17,987
		Liberty	26,203 ± 2010	44,625 ± 14,523	85,152 ± 41,029
		BSMP	28,704 ± 3948	26,914 ± 9980	43,679 ± 31,008
	Pristane	Northstar	26,968 ± 3237	16,598 ± 8736	24,749 ± 18,785
		Liberty	24,634 ± 2488	42,127 ± 17,377	81,071 ± 47,234
		BSMP	27,254 ± 5160	24,644 ± 14,816	39,986 ± 3397
	StTr	Northstar	12.7 ± 4.37	0.57 ± 0.25	1.09 ± 2.40
		Liberty	8.10 ± 0.40	4.88 ± 2.27	8.38 ± 6.53
		BSMP	3.70 ± 1.93	0.52 ± 0.32	13.4 ± 17.8
Isopods	TPAH	Northstar	No data	No data	67.6 ± 12.8
		Liberty	No data	67.0 ± 9.83	88.5
		BSMP	No data	73.37 ± 4.36	114
Mysids	TPAH	Northstar	No data	No data	89.3 ± 24.4
Clams	TPAH	Liberty	91.85	No data	141 ± 57.8
		BSMP	97.1 ± 52.5	38.4 ± 12.3	No data
	TSHC	Liberty	1644	No data	5276 ± 666
		BSMP	1922 ± 621	1510 ± 634	No Data
	Pristane	Liberty	80.2	No data	96.6 ± 36,0
		BSMP	152 ± 40.6	434 ± 629	No data
	StTr	Liberty	8.99	No data	0
		BSMP	8.40 ± 6.20	1.72 ± 2.98	No data

Table ES-2. Mean Concentrations of total polycyclic aromatic hydrocarbons (TPAH), total saturated hydrocarbons (TSHC), pristane, and steranes/triterpanes (StTr) in tissues of reference and deployed mussels and of TPAH in blank and deployed SPMDs used to monitor hydrocarbon concentrations in the water column of the Alaskan Beaufort Sea in 2004 through 2006.

Matrix	Analytes	Location	Mean (SD) TPAH Concentration (ng/g dry wt or ng/SPMD)		
			2004	2005	2006
Deployed Mussels	TPAH	Reference	227 ± 34.9	32.8 ± 27.0	164 ± 36.2
		Northstar	148 ± 45.8	13.0	91.6 ± 5.18
		Liberty	92.8	24.8 ± 14.7	134 ± 6.74
		BSMP	157 ± 46.8	31.5 ± 1.46	52.7
	TSHC	Reference	6051 ± 522	3632 ± 151	23,159 ± 2294
		Northstar	7624 ± 2494	3381	21,024 ± 3599
		Liberty	5725	2689 ± 384	16,040 ± 3145
		BSMP	6246 ± 123	3137 ± 380	16,033
	Pristane	Reference	637 ± 63.4	270 ± 10.9	627 ± 86.0
		Northstar	365 ± 79.5	273	671 ± 180
		Liberty	337	146 ± 29.1	1153 ± 214
		BSMP	413 ± 162	290 ± 106	850
	StTr	Reference	12.2 ± 3.79	5.87 ± 5.87	ND
		Northstar	27.2 ± 16.2	6.9	ND
		Liberty	13.5	ND	ND
		BSMP	14.1 ± 6.43	ND	ND
SPMDs	TPAH	Blank	699 ± 55.0	No data	No data
		Northstar	750 ± 182	No data	No data
		Liberty	945	No data	No data
		BSMP	606 ± 155	No data	No data

ND, not detected.

There were no consistent patterns of concentrations of any metals in tissues of indigenous animals from the different sampling locations. Metals concentrations in soft tissues of fish, amphipods, and clams from all Beaufort Sea sampling locations combined (Table ES-3) and in deployed mussels from Beaufort Sea and reference stations (Table ES-4) were similar in 2004, 2005, and 2006. Concentrations of most metals were lower in fish than in amphipods or clams. Aluminum, iron, and manganese concentrations tended to be higher in clams than in crustaceans, possibly indicating that the clams are retaining sediment particles in the gut and gills. Copper concentrations were higher in amphipods than in clams and fish, probably reflecting the presence of copper-containing respiratory pigments in many crustaceans. Metals concentrations were similar in undeployed reference mussels and mussels that had been deployed in the Beaufort Sea, indicating little exchange of tissue metals during deployment. Concentrations of most metals were similar in indigenous clams and deployed mussels.

Table ES–3. Range of concentrations of metals in soft tissues of combined fish species (up to eight species), amphipods, and clams from all sampling locations in the western Beaufort Sea in 2004, 2005, and 2006.

Metal	Taxon	Concentration Range, All Stations (µg/g dry wt)		
		2004	2005	2006
Silver (Ag)	Fish	0.01 – 0.35	0.01 – 0.35	<0.01 – 0.11
	Amphipods	1.85 – 5.96	1.61 – 4.04	0.95 – 3.01
	Clams	0.09 – 0.13	0.05 – 0.14	0.06 – 0.07
Aluminum (Al)	Amphipods	164 – 740	95.8 – 379	124 – 894
	Clams	721 – 2150	98.5 – 1320	973 – 1010
Arsenic (As)	Fish	1.12 – 16.2	0.81 – 7.35	1.54 – 6.27
	Amphipods	6.39 – 15.9	5.01 – 17.4	4.97 – 16.9
	Clams	8.22 – 15.2	11.3 – 17.4	10.7 – 11.6
Barium (Ba)	Fish	0.30 – 14.2	0.50 – 46.6	1.09 – 9.54
	Amphipods	10.7 – 50.4	11.6 – 40.7	12.0 – 59.0
	Clams	10.8 – 22.5	7.90 – 39.5	12.4 – 13.7
Berylium (Be)	Amphipods	0.01 – 0.02	0.01 – 0.03	0.01
	Clams	0.04 – 0.05	0.02 – 0.08	0.04 – 0.07
Cadmium (Cd)	Fish	0.02 – 0.27	0.01 – 0.37	0.02 – 0.27
	Amphipods	0.43 – 2.05	0.62 – 2.37	0.39 – 1.74
	Clams	0.53 – 5.85	1.13 – 9.55	6.86 – 8.69
Chromium (Cr)	Fish	0.05 – 1.14	0.03 – 3.81	0.08 – 0.58
	Amphipods	0.35 – 0.96	0.41 – 0.73	0.18 – 1.86
	Clams	2.28 -4.41	0.91 – 5.15	2.49 – 2.82
Cobalt (Co)	Amphipods	0.95 – 3.48	1.38 – 2.88	1.29 – 1.81
	Clams	1.11 – 3.92	0.75 – 1.68	1.03 - 1.22
Copper (Cu)	Fish	1.50 – 18.2	1.10 – 21.2	1.80 – 7.80
	Amphipods	108 – 333	100 – 200	114 – 203
	Clams	11.2 – 18.5	11.1 – 17.1	10.2
Iron (Fe)	Fish	37.2 – 424	19.4 – 1250	38.7 – 249
	Amphipods	103 – 439	168 – 280	137 – 477
	Clams	1040 – 3640	771 – 2110	1550 – 1630
Mercury (Hg)	Fish	0.02 – 0.45	0.03 – 0.27	0.05 – 0.20
	Amphipods	0.04 – 0.19	<0.01 – 0.12	0.04 – 0.16
	Clams	0.06 – 0.08	0.03 – 0.08	0.07
Manganese (Mn)	Amphipods	10.3 – 99.4	0.84 – 5.2	21.6 – 71.1
	Clams	68.3 – 637	65.5 – 205	47.1 – 73.3
Nickel (Ni)	Fish	0.05 – 0.92	0.03 – 1.82	0.19 – 0.76
	Amphipods	1.05 – 5.56	0.84 – 5.20	2.31 – 3.92
	Clams	1.92 – 5.28	2.04 – 5.34	3.66 – 4.02
Lead (Pb)	Fish	0.02 – 0.57	0.03 – 0.79	0.01 – 0.13
	Amphipods	0.10 – 4.39	0.05 – 0.31	0.07 – 0.25
	Clams	0.61 – 1.16	0.18 – 1.24	0.20 – 0.73
Antimony (Sb)	Amphipods	0.01 – 0.03	0.02 – 0.04	0.01 – 0.04
	Clams	0.02 – 0.03	0.03 – 0.06	0.01 – 0.05
Selenium (Se)	Fish	1.57 – 5.66	0.94 – 3.64	2.13 – 5.24

Table ES–3. Range of concentrations of metals in soft tissues of combined fish species (up to eight species), amphipods, and clams from all sampling locations in the western Beaufort Sea in 2004, 2005, and 2006, continued.

Metal	Taxon	Concentration Range, All Stations (µg/g dry wt)		
		2004	2005	2006
Thallium (Tl)	Amphipods	0.01 – 0.03	0.01 – 0.03	0.01 – 0.02
	Clams	0.02 – 0.03	0.02 – 0.03	0.01 – 0.02
Vanadium (V)	Fish	0.05 – 3.38	0.08 – 3.92	0.25 – 5.02
	Amphipods	1.16 – 3.48	0.52 – 1.74	0.86 – 1.99
	Clams	3.34 – 6.91	0.97 – 4.74	4.23 – 4.74
Zinc (Zn)	Fish	36.0 – 109	4.09 – 119	47.2 – 105
	Amphipods	94.8 – 214	79.4 – 156	74.4 – 145
	Clams	68.7 – 88.5	57.8 – 79.3	76.3 – 81.4

There were no consistent trends in concentrations of hydrocarbons or metals in fish tissues. Demersal species, such as four horn sculpin tended to contain higher concentrations of hydrocarbons and metals than the more pelagic species. The anadromous species, arctic char, tended to contain lower concentrations of metals and hydrocarbons than the other species did, perhaps because they spend more time offshore.

Table ES–4. Range of concentrations of metals in reference mussels and mussels deployed at several stations in the Beaufort Sea in 2004, 2005, and 2006.

Metal	Concentration Range, All Stations (µg/g dry wt)		
	2004	2005	2006
Silver (Ag)	0.08 – 0.13	0.07 – 0.11	0.05 – 0.10
Aluminum (Al)	130 – 2030	306 – 1010	275 – 1910
Arsenic (As)	6.91 – 11.5	10.2 – 13.1	8.63 – 11.7
Barium (Ba)	3.10 – 20.0	5.40 – 10.8	2.92 – 17.9
Berylium (Be)	0.01 – 0.04	0.01 – 0.04	0.01 – 0.04
Cadmium (Cd)	1.51 – 2.92	3.61 – 5.43	2.88 – 5.27
Cobalt (Co)	0.59 – 1.04	0.46 – 0.85	0.51 – 1.56
Chromium (Cr)	0.99 – 3.72	1.46 – 2.35	1.11 – 7.32
Copper (Cu)	5.50 – 9.40	6.50 – 8.10	5.40 – 8.20
Iron (Fe)	198 – 1230	265 – 847	366 – 2010
Mercury (Hg)	0.07 – 0.12	0.05 – 0.45	0.06 – 0.12
Manganese (Mn)	5.8 – 25.6	8.5 – 12.8	11.0 – 61.5
Nickel (Ni)	1.07 – 2.39	1.27 – 2.86	1.57 – 4.94
Lead (Pb)	0.50 – 1.10	0.52 – 0.90	0.51 – 0.93
Antimony (Sb)	0.01 – 0.02	0.01 – 0.02	0.01
Thallium (Tl)	0.01	0.02 – 0.04	0.01 – 0.02
Vanadium (V)	0.98 – 4.65	0.91 – 2.15	1.71 – 5.46
Zinc (Zn)	46.8 – 131	76.0 – 113	85.9 – 111

The concentrations of metals and TPAH in the tissues of most of the bivalve mollusks, crustaceans, and fish sampled in this program were in the range expected for the same or similar species in relatively unpolluted marine environments throughout the world (Table ES-5). Concentrations of different metals vary widely in tissues of different taxa of marine animals. Zinc is particularly abundant in mollusks and copper often is abundant in crustaceans. Mercury tends to be more abundant in fish tissues, particularly muscle, than in tissues of marine invertebrates. PAH usually are more abundant in mollusks than in crustaceans and fish. Fish and crustaceans have a well-developed, inducible cytochrome P450 mixed-function oxygenase system that rapidly metabolizes bioaccumulated PAH, facilitating their excretion. This pattern of distribution of metals and TPAH was evident in bivalve mollusks, crustaceans, and fish in the Alaskan Beaufort Sea.

Table ES–5. Concentrations of several metals and total polycyclic aromatic hydrocarbons (TPAH) in whole or muscle tissues of marine bivalve mollusks, crustaceans, and fish from unpolluted marine environments throughout the world. Concentrations are μg/g dry wt. From Neff (2002a).

Chemical	Bivalve Mollusks		Crustaceans		Fish	
	Geomean	Range	Geomean	Range	Geomean	Range
Arsenic	11	0.13 – 214	15	<0.1 – 270	6.1	0.05 – 450
Barium	4.4	0.09 – 179	3.4	0.02 – 202	0.13	0.007 – 49
Cadmium	1.18	0.05 – 26.1	1.85	0.14 – 117	0.10	0.001 – 5.8
Mercury	0.17	0.004 – 11.7	0.45	0.02 – 6.2	0.77	0.01 – 115
Chromium	3.5[a]	0.1 – 10.0	2.6[a]	0.12 – 10.1	2.1[a]	0.03 – 5.8
Copper	51.8[a]	6.4 – 150	75.4[a]	8.8 – 241	3.9[a]	0.6 – 26
Lead	4.5[a]	<0.1 – 21.4	4.5[a]	0.03 – 17.5	7.09[a]	0.02 – 55.9
Zinc	290[a]	40 – 1315	68.6[a]	23.9 – 96.5	28.8[a]	4.1 – 58.8
TPAH	0.65	0.003 – 1729	0.17	0.004 – 13.4	0.19	0.002 – 23.4

[a] arithmetic mean.

An unexpected observation was the extremely high concentrations of pristane in tissues of indigenous amphipods. Pristane is an abundant aliphatic hydrocarbon in petroleum, peat, and in some species of marine plants and animals. Copepods, particularly some species in the genera *Calanus* and *Neocalanus,* contain percent levels of pristane in their lipids. Calanoid copepods bioaccumulate phytol, a monounsaturated diterpenyl alcohol that is esterified with chlorophyll, in their phytoplankton food and convert it to pristane; the pristane accumulates to high concentrations in oil droplets in the copepods (Avigan and Blumer, 1968). The benthic amphipods may be bioaccumulating pristane from ingestion of detritus and copepod feces. Indigenous clams and deployed mussels contained much lower concentrations of pristane than amphipods did, indicating a different diet.

CYP1A staining and bile FAC analysis were evaluated as biomarkers of petroleum exposure in several species of fish collected in the Beaufort Sea in 2004 and 2005. CYP1A staining was very light in several tissues of five species of fish collected at sampling sites in 2004 and 2005. Usually, CYP1A activity was lower in fish from Northstar than in those collected near Liberty or at other sampling sites. Bile FAC concentrations were similar in fish from Northstar and Liberty in both 2004 and 2005. These results are consistent with the tissue

residue data indicating very low-level exposure to PAH, possibly of petroleum, at all stations.

Some of the Beaufort Sea fish, crustaceans, and clams contained concentrations of one or more metals higher than the median value for mussels and oysters collected in the National Status and Trends Mussel Watch Program (Table ES-6). These elevated concentrations probably are natural and attributable to species differences and to inputs of metal-laden sediments to the Beaufort Sea from river runoff, particularly during the spring breakup (Rember and Trefry, 2004), or upwelling of metal-rich deep water from the Beaufort Sea continental slope.

Table ES–6. Concentration ranges of several metals in fish, mussels, amphipods, and clams collected in the Beaufort Sea between 2000 and 2006 as part of the ANIMIDA and cANIMIDA Programs, compared to the National Status and Trends median concentration ranges for mussels or oysters collected in US coastal waters between 1986 and 2003 (From O'Connor and Lauenstein, 2006). Concentrations are μg/g dry wt (ppm).

Metal	Fish	Mussel	Amphipod	Clam	NS&T Medians
Silver (Ag)	0.01 – 0.35	0.05 – 2.5	0.8 – 4.0	0.04 – 0.13	---
Aluminum (Al)	---	131 – 2000	96 - 1200	98 - 2200	---
Arsenic (As)	0.55 – 16	6.2 – 13	4.0 – 17	8 - 16	8.1 – 9.6
Barium (Ba)	0.30 – 47	2.9 – 20	7.4 - 59	7 - 40	---
Berylium (Be)	---	0.01 – 0.06	0.01 – 0.03	0.03 – 0.08	---
Cadmium (Cd)	0.01 – 0.37	0.29 – 5.4	0.3 – 2.4	0.53 – 13	2.1 – 2.9
Cobalt (Co)	---	0.46 – 2.6	0.6 – 2.9	0.8 – 4.0	---
Chromium (Cr)	0.04 – 3.8	0.65 – 7.3	---	---	---
Copper (Cu)	1.1 – 21	5.4– 9.40	41 - 210	7.0 - 24	(8.0 – 10)[a] (91 – 140)[b]
Iron (Fe)	19 - 1200	200 – 2000	100 - 950	770 - 3600	---
Mercury (Hg)	0.02 – 0.5	0.01 – 0.45	0.02 – 0.19	0.0 – 0.13	0.09 – 0.11
Manganese (Mn)	---	5.8 – 260	10 - 71	47 - 640	---
Nickel (Ni)	0.03 – 4.4	1.1 – 4.9	0.8 – 6.7	1.92 – 5.34	1.6 – 2.2
Lead (Pb)	0.01 – 2.6	0.22 – 1.1	0.05 – 0.7	0.18 – 1.9	0.63 – 0.98
Antimony (Sb)	---	0.01 – 0.03	0.01 – 0.04	0.01 – 0.06	---
Selenium (Se)	0.94 – 5.7	---	---	---	2.3 – 3.0
Tellurium (Tl)	---	0.01 – 0.04	0.01 – 0.03	0.01 – 0.03	---
Vanadium (V)	0.05 – 5.1	0.91 – 6.7	0.5 – 3.4	1.3 – 6.9	---
Zinc (Zn)	36.0 – 120	47 – 130	54 - 170	62 - 130	(110 – 140)[a] (1600 – 2400)[b]

[a] Mussels; [b] Oysters

The coastal marine environment of the Alaskan Beaufort Sea is nutrient-poor with relatively short near-shore and offshore food webs (Figure ES-1). Crustaceans, including the species monitored in this program, are the most important primary consumers. Crusteacean zooplanton, especially calanoid copepods and euphausiids, are particularly important to top consumers in the local food web. Calanoid copepods and euphausiids are consumed by bowhead whales during their spring and fall migrations through the Beaufort Sea

development area. These zooplankton also are consumed by arctic cod, which in turn are a major part of the diet of beluga whales, ringed seals, and several marine bird species. Because zooplankton, particularly large copepods and euphausiids, are so important in the Beaufort Sea food web, they should be collected and analyzed for contaminants in any future monitoring program. Any contaminants released in large amounts from offshore oil and gas activities are likely to bioaccumulate in the local food web and spread through the ecosystem. None of the chemicals monitored in this investigation (petroleum hydrocarbons and several metals), except possibly mercury as methylmercury, biomagnify through marine food webs (Neff, 2002a,b); therefore, concentrations of metals and hydrocarbons are not likely to be high in tissues of top consumers, such as whales, seals, and polar bears.

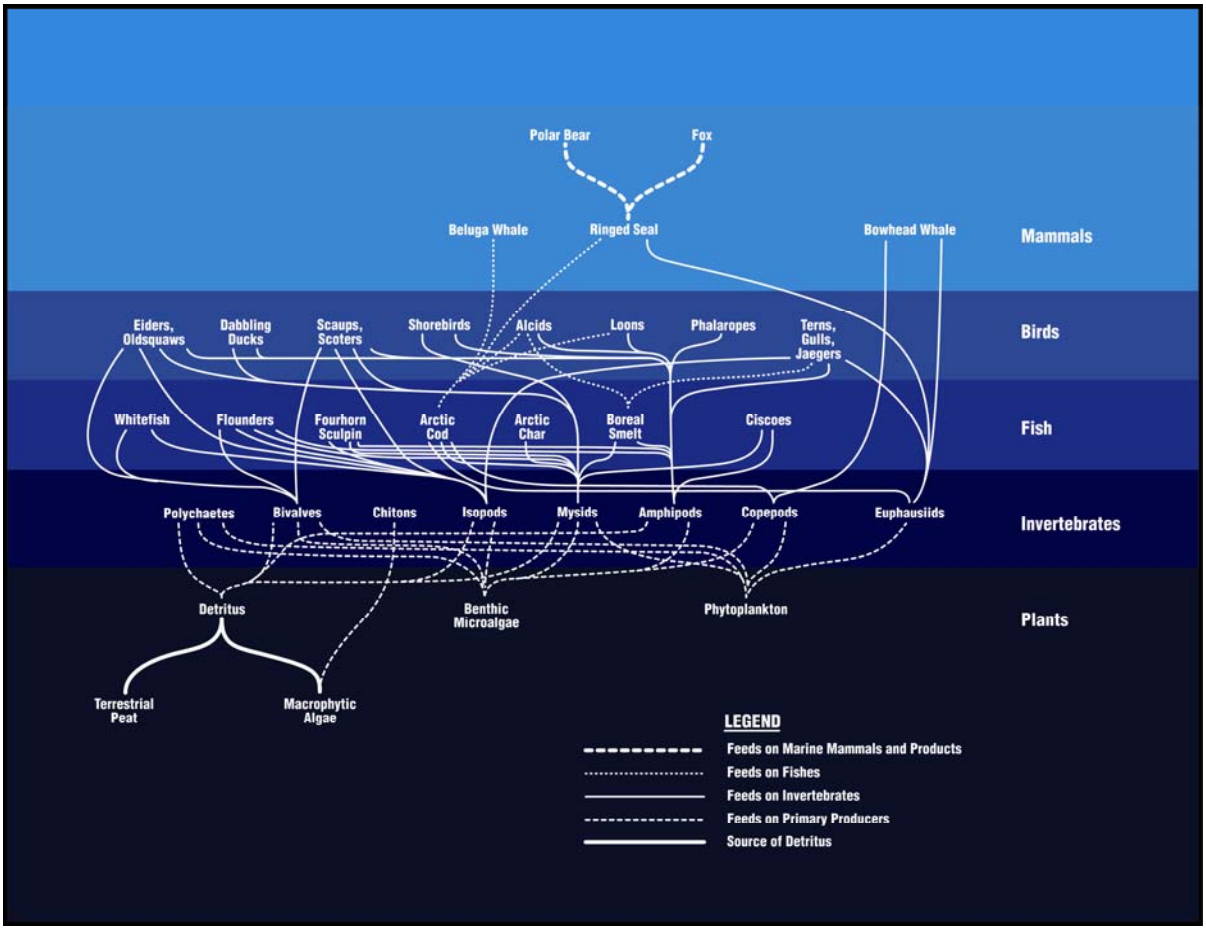


Figure ES–1. A diagram of the Beaufort/Chukchi Sea food web model, showing carbon pathways from terrestrial peat and primary production to top keystone species of marine birds and mammals. Modified from MMS (1990).

This study has shown that oil development activities in the Beaufort Sea are not contributing ecologically significant amounts of petroleum hydrocarbons and metals to the near-shore marine food web of the Alaskan Beaufort Sea. Mean TPAH concentrations in fish, deployed mussels, amphipods, isopods, mysids, and clams collected from all stations in 2004, 2005, and 2006, as well as fish collected in 2001 and amphipods, clams, and deployed mussels

collected in 2000 and 2002 in the ANIMIDA Program, ranged from 9.44 ng/g dry wt to 157 ng/g (parts per billion) (Tables ES-1 and ES-2). By comparison, the median concentration of TPAH in tissues of mussels and oysters collected in coastal waters of the United States in 2002/2003 and analyzed in the National Status and Trends Mussel Watch Program was 220 ng/g dry wt (O'Connor and Lauenstein, 2006). The lowest concentration of TPAH measured in mussels and oysters in the Mussel Watch Program was 7.3 ng/g. Thus, invertebrates and fish from the Beaufort Sea contain low concentrations of TPAH, compared to mussels and oysters (the greatest bioaccumulators) from the other US states.

The PAH profiles in tissues of fish and invertebrates from the Beaufort Sea is consistent with a mixed petrogenic/pyrogenic source; petrogenic PAH probably are derived from aerial deposition, oil and gas operations in the vicinity, and runoff from land (Steinhauer and Boehm, 1992). TPAH concentrations were not higher in tissues of marine animals from the vicinity of the Northstar Development, the only facility that produced crude oil from Federal waters of the Beaufort Sea, than in the same species from other locations. The concentrations of individual and total PAH in tissues of Beaufort Sea marine animals are well below concentrations that would represent a health risk to marine animals and the animals, including man, that might consume them (Neff, 2002a).

Concentrations of 18 metals in tissues of several species of fish, amphipods, isopods, clams, and deployed mussels collected in the Northstar development and the Liberty prospect areas and in other reference areas were similar in 2004, 2005, and 2006 (Tables ES-3 and ES-4). A few metals concentrations were slightly higher in marine animals from Northstar than in the same species from the other sampling sites; however, there were no consistent metal, year, or species patterns. Metals concentrations were in the range reported for the same or similar species from other locations throughout the world (Table ES-5), and are below concentrations that would pose a health risk to the marine animals or their consumers, including man (Neff, 2002a). There is no evidence that metals from the development and production activities at Northstar are entering the Beaufort Sea food web.

This page intentionally left blank

1.0 INTRODUCTION

1.1 Project Background

There is concern that offshore oil and gas development and production activities planned or currently under way on the continental shelf of the Alaskan Beaufort Sea may cause harm to the local marine environment and, in particular, the biological resources that depend on it. Coastal indigenous people are concerned about long-term effects of these offshore development activities on the subsistence biological resources, particularly migrating bowhead whales and near-shore fish populations.

The complex environment in this region has been studied for more than three decades by the scientific community, mostly under contract to the U.S. Dept of the Interior, Minerals Management Service (MMS), and the oil industry; however, it is essential to continue environmental monitoring to gain a better understanding of the environmental impacts of on-going production activities at the Northstar site and planned development of the Liberty prospect.

Baseline conditions in the area were characterized in the 1980s in the MMS Beaufort Sea Monitoring Program (Boehm et al., 1990). In 1999, MMS initiated Phase I of the Arctic Nearshore Impact Monitoring in the Development Area (ANIMIDA) Program to begin the process of monitoring the environmental impacts of development of the Northstar and Liberty prospects. In 2000, Phase II of the ANIMIDA program continued the activities of Phase I for an additional four years. The last field sampling for ANIMIDA was performed in 2003. The results of biological monitoring in ANIMIDA were summarized in two reports: Task 2 (Brown et al., 2005); and Task 8 (Spies et al., 2003).

A new monitoring program, called Continuation of Arctic Nearshore Impact Monitoring in Development Area (cANIMIDA), was initiated in 2004 and fieldwork began during the summer of 2004. Task 5, "Integrated Biomonitoring and Bioaccumulation of Contaminants in Biota of the cANIMIDA Study Area", is a component of cANIMIDA and was designed based on the results of Phase I and II of ANIMIDA. This report summarizes and interprets data collected during cANIMIDA (2004-2006) on metals, hydrocarbons, and biological exposure biomarkers in marine animals collected in the vicinity of the Northstar production island and the Liberty prospect.

1.2 Objectives

Task 5 contains five distinct, yet interrelated objectives (Appendix F). The fieldwork and analysis for 2004 through 2006 focused on Objective 2, but all project objectives are addressed in this report.

Objective 1: Improve and validate the Contractor's proposed conceptual model of bioaccumulation and trophic interaction in cANIMIDA biota.

A conceptual food web model is a useful tool for designing scientifically defensible studies of the sources and distribution of chemical contaminants in a marine ecosystem such as the

near-shore Alaskan Beaufort Sea. It provides the basis for the development of testable null hypotheses about relationships between physical and chemical disturbances to the marine environment by offshore oil development and production operations and changes to marine populations and ecosystems. A conceptual food web model is particularly important for identifying possible interactions between development activities and species of marine mammals and birds that cannot be sampled directly because of regulatory or cultural constraints. The food web model also helps identify the marine plants and animals that are at risk and that should be sampled if evidence is found of bioaccumulation of contaminants at lower trophic levels in the local food web.

The preliminary conceptual model focuses on food chain interaction of polycyclic aromatic hydrocarbons (PAH) and several metals, which occur naturally in soils, sediments, and biota in and around the Alaskan Beaufort Sea area and also may be introduced into the local marine environment from offshore oil and gas activities. Significant amounts of PAH and metals are introduced, associated with suspended sediment, to the Beaufort Sea annually via river runoff, and possibly from oil and gas operations onshore and offshore (Naidu et al., 2001; Steinhauer and Boehm, 1992; Rember and Trefry, 2004); smaller amounts of PAH and metals also are introduced to coastal soils and surface waters of the North American arctic in wet and dry deposition from the atmosphere (Heidam, 1984; Halsall et al., 1997; Bard, 1999; MacDonald et al., 2000). Most of the PAH and metals in the arctic aerosol over the Beaufort Sea are from remote combustion sources, particularly from southeast Asia.

The major sources of PAH and metals inputs to the continental shelf from oil and gas operations usually are permitted discharges of drilling mud/cuttings and produced water (Neff, 2002a, 2005). However, produced water from production operations in US waters of the Beaufort Sea is reinjected and recent drilling mud and cuttings discharges have been limited to winter discharges onto the ice in deep water, minimizing inputs from these sources.

PAH and metals also are introduced to the atmosphere in diesel exhaust and fugitive emissions from petroleum production, waste gas flaring, treatment, storage, and transportation facilities. Much of these atmospheric emissions are deposited on the sea surface within a few kilometers of the emission source. Accidental releases of wastewater, drilling mud/cuttings, and petroleum products from exploration, development, and production operations may introduce small amounts of PAH and metals to the Beaufort Sea. The primary sources of metals to the Beaufort Sea sediments associated with oil development activities are derived from the construction of causeways and drilling islands, emplacement of pipelines, and drilling mud/cuttings discharges. (Northern Technical Services, 1982; Naidu et al., 2001; Neff, 2002b).

Marine invertebrates and fish living in the Beaufort Sea are able to bioaccumulate metals and PAH from ambient seawater, sediments, and from their food (Neff, 2002a). The concentrations of metals and PAH in tissues of marine organisms are assumed to be at equilibrium with concentrations of bioavailable forms of the chemicals in the ambient water, sediments, and foods. Benthic and demersal invertebrates and fish absorb pollutants primarily from sediment pore water, overlying bottom water, and the ingestion of sediment particles and food. Benthic and demersal carnivores often bioaccumulate contaminants

primarily from their food. Marine birds and mammals also bioaccumulate contaminants primarily from their food. Thus, a local increase in metals and PAH in water or sediments due to discharges from oil and gas operations may be reflected by an increase in the concentration of the contaminants in tissues of marine animals from the local area. Because dissolved metals and PAH are the most bioavailable forms to marine organisms, concentrations of total metals and PAH in sediments often do not reflect concentrations in the food web. PAH and a fraction of the metals in soft tissues of marine organisms also are more biavailable to consumers than are these contaminants in sediments. Several metals (e.g., inorganic mercury in fish and mammal liver, and cadmium in kidney of several taxa of marine animals) are sequestered at high concentrations in certain tissues of marine animals; these sequestered metals are not bioavailable or toxic (Neff, 2002a).

None of the target contaminants are known to biomagnify in marine food webs, with the possible exception of mercury as methylmercury and possibly arsenic as arsenobetaine (Neff, 1997, 2002a,b). However, all the target contaminants are transferred efficiently through marine food webs (concentrations are the same or lower in the predator tissues than in the prey tissues). Arctic marine animals are about as sensitive as temperate and tropical species to petroleum and metals toxicity (Neff, 2002a; Perkins et al., 2003). Thus, bioaccumulation of these contaminants may cause adverse effects in the Beaufort Sea food web, if environmental concentrations of bioavailable forms are high enough.

This conceptual model of bioaccumulation, trophic transfer, and effects was used as a basis for developing the objectives and study design for this task. A significant component of Task 5 is to expand the information required to protect indigenous populations and the subsistence resources upon which they rely from damage resulting from chemical contamination associated with offshore oil and gas operations. The conceptual food web model will be used in the integrated final report for the cANIMIDA Program to evaluate the bioaccumulation and food web transfer of metals and PAH derived from offshore oil and gas development activities in the Beaufort Sea. Sediments (representing the primary contaminant reservoir), benthic invertebrates (a lower trophic level), and fish (usually a higher trophic level) were analyzed for several metals and PAH that are associated with oil development activities. An attempt was made to identify any contribution from oil development activities to contaminant levels in sediments and tissues of marine animals. Contaminant concentrations in organisms at lower trophic levels will be coupled with data obtained in Tasks 2, 3, and 4 in order to examine the uptake of dissolved and sediment-associated contaminants into the food chain and to predict potential levels of contamination of foods of higher trophic level animals, such as marine birds and mammals.

Objective 2: Measure bioaccumulation in selected species, including indigenous bivalves, benthic amphipods, and fish, and in caged bivalves and SPMDs.

Battelle collected and analyzed marine organisms from sampling sites in the vicinity of the Northstar development, along the coast southeast of Northstar, including the Boulder Patch, and in the vicinity and up current (southeast) of the Liberty prospect (the latter considered reference sites). Indigenous species of fish (8 species), benthic bivalve mollusks (*Astarte montagui* and *Cyrtodaria kurriana*), and demersal/benthic amphipods (*Anonyx* sp.), the same

species as collected during the ANIMIDA program, were collected during cANIMIDA. Caged mussels (*Mytlius trossulus*) and semipermeable membrane devices (SPMDs) were deployed at several sites between Northstar and Liberty during the 2004 summer survey. Only caged mussels were deployed during the 2005 and 2006 surveys.

Benthic invertebrates, particularly bivalve mollusks, and fish are unevenly distributed in coastal and offshore waters of the Beaufort Sea. Previous experience in ANIMIDA was used to select locations most likely to yield sufficient numbers of target species for a statistically rigorous analytical program. Every reasonable effort was made to collect all target species from several nearfield (near Northstar and Liberty) and farfield (reference) locations to aid in identifying tissue residues of contaminants derived from oil development operations. Marine animal samples were analyzed for petroleum hydrocarbons (polycyclic aromatic hydrocarbons, saturated hydrocarbons, and sterane/triterpane petroleum biomarkers) and 18 metals to meat Objective 2.

Objective 3: Compare bioaccumulation data from the Beaufort Sea to published data for same or similar species outside the cANIMIDA area.

There is a large and rapidly growing body of scientific literature focusing on the bioaccumulation and concentrations of metals and PAH in marine organisms from clean and contaminated estuarine and marine habitats. Much of this literature was assembled and interpreted recently, with a focus on potential effects of offshore oil and gas development and production discharges (Neff, 2002a). The US National Status and Trends Mussel Watch Program and similar programs in Europe have compiled large databases of contaminant concentrations in mussels, oysters, and sediments from coastal waters of the United States and Europe (Daskalakis and O'Connor, 1995; Beliaeff et al., 1998; O'Connor, 2002; O'Connor and Lauenstein, 2006). Concentrations of metals and PAH determined as part of the ANIMIDA and cANIMIDA programs in sediments and biota of the Alaskan Beaufort Sea will be compared to historic data for the same and similar species and sediment types from elsewhere in the world. Historic Beaufort Sea Monitoring Program data from ANIMIDA also will be included in this analysis in an effort to determine if there are any observable long-term trends of change in concentrations of metals and PAH in marine tissues and sediments from the Alaskan Beaufort Sea.

Objective 4: Develop a strategy for longer-term upper trophic level contaminant monitoring.

Several species of invertebrates and fish proposed for bioaccumulation monitoring in Task 5 are foods or surrogates for preferred foods of several upper trophic level marine birds and mammals in the coastal and offshore food web of the Beaufort Sea. For instance, bowhead whales (*Balaena mysticetus*) feed on pelagic euphausids, mysids, copepods, and amphipods (Lowry and Frost, 1984; Richardson et al., 1995; Hoekstra et al., 2002). The amphipods monitored in this study are suitable surrogates for bowhead whale foods. Gray whales (*Eschrichtius robustus*), which feed in small numbers in the Beaufort Sea during the summer, do consume large amounts of benthic amphipods, their preferred prey (Oliver and Slattery, 1985). Some ice-associated seals, such as the bearded seal (*Erignathus barbatus*) and ribbon

seal (*Phoca faciata*), which feed in coastal waters of the Beaufort Sea in some seasons, feed heavily on benthic invertebrates and demersal fish, such as shrimp, crabs, arctic cod, and sculpins (Wynne, 1997). Ringed seals (*Phoca hispida*), the favorite food of polar bears, feed in different seasons on isopods, amphipods, euphausiids, mysids, and arctic cod. Beluga whales (*Delphinapterus leucas*) migrate annually through the Alaskan Beaufort Sea, usually further offshore than bowhead whales, between summer feeding areas in the Canadian Beaufort Sea and wintering areas in the Bering Sea and feed heavily on pelagic and demersal fish, particularly arctic cod (Treacy, 2002). The protected spectacled eider (*Somateria fischeri*) which breeds along the arctic coastal plain from Barrow to the Canadian border, feeds on benthic mollusks and crustaceans that they gather in shallow coastal waters (<30 m) (Dau and Kistchinski, 1977). Several species of marine birds in the Alaskan Beaufort Sea feed heavily on arctic cod, which, in turn, feed mainly on mysids and amphipods (Frost and Lowry, 1984).

Thus, the results of the tissue monitoring in Task 5 can be used to conceptually model contaminant concentrations in the food web leading to top trophic level Beaufort Sea birds and mammals. Predicted bioaccumulation of contaminants from food can be compared to historic data on contaminant levels in upper trophic level birds and mammals in the region to determine if foods are an important source of contaminant residues in their tissues. There is some information available about contaminant concentrations, particularly metals, in tissues of Beaufort Sea bowhead whales. However, there are few data for other species of marine mammals and birds, or for PAH in upper trophic level species in general (Neff, 2002a).

Objective 5: Develop a strategy and rationale for future Boulder Patch contaminant monitoring in conjunction with Task 6.

The Boulder Patch is a unique marine ecosystem offshore from the Endicott Development and northwest of the Liberty prospect in Stefansson Sound (Dunton et al., 2003a,b, 2004). The boulder patch community is dominated by arctic kelp (*Laminaria solidungula*) that may be sensitive to increased suspended sediment loads and chemical contaminants from offshore development activities, particularly at Liberty. Considerable information has been acquired on the ecology of the macroalgal community in the Boulder Patch during ANIMIDA (Dunton et al., 2003a,b, 2004), with an emphasis on effects of suspended sediments on primary production. The work is continuing in cANIMIDA as Task 6, "Monitoring the Boulder Patch as Part of the cANIMIDA Program." However, little is known about the bioaccumulation of and sensitivity of the macroalgal community and associated marine fauna to metal and PAH contaminants that may come from nearby oil development activities. Attempts were made during the summer 2005 and 2006 field surveys to collect samples of Boulder Patch biota for chemical analysis. Amphipods were captured in the Boulder Patch in 2005 and mussels were deployed in this area during the 2006 field season.

Task 5 Monitoring Hypotheses

The cANIMIDA program was designed to address a series of scientific questions concerning the potential impacts of the Northstar and Liberty developments. Each testable hypothesis is intended to guide the design of the sampling and analysis plan. The Task 5 biomonitoring and bioaccumulation study is based on four hypotheses:

H1: Baseline concentrations of PAHs, metals, and exposure/response biomarkers in biota from the Northstar and Liberty areas of the Beaufort Sea are not a result of oil and gas industry activities.

H2: Oil and gas industry activities in the Northstar and Liberty prospects will not result in an increase in tissue concentrations of PAHs, metals and exposure/response biomarkers in biota from the Northstar and Liberty areas.

H3: Concentrations of metals and PAH in tissues of indigenous benthic invertebrates and demersal fish from the Northstar and Liberty areas are not different from the regional background, which reflects concentrations of bioavailable contaminants from natural and anthropogenic sources.

H4: Concentrations of metals and PAH in caged mussels and of PAH in SPMDs following a minimum 21-day deployment near Northstar oil and gas activities will reflect concentrations of regional contaminants in the water column of the Beaufort Sea.

1.3 Study Area

The cANIMIDA study area will be described briefly here because the physical characteristics of the Beaufort Sea have been described in detail elsewhere, including the Environmental Impact Statements for the Oil and Gas Lease Sales for Beaufort Sea Planning areas 87, 97, 124, 144, and 202 (MMS 1984, 1987, 1990, 1995, and 2006, respectively).

The Beaufort Sea is the southern part of the Arctic Ocean off the coasts of Alaska and the Yukon and Northwest Territories of Canada. The Alaskan Beaufort Sea extends from the Barrow Sea Valley at Point Barrow eastward approximately 600 km to the Alaska-Canadian border and northward for a distance of 322 km (200 miles) (Figure 1-1). The continental shelf off Alaska, measured from the shoreline to the 200-m isobath, is about 80 km wide, among the narrowest in the circumpolar Arctic (National Research Council, 2003), with an average depth of 37 m. The continental slope in the northern Beaufort Sea extends to a depth of more than 1000 m. The major bathymetric features on the continental shelf are barrier islands and shoals, the boundaries of which migrate from year to year, due mainly to spring freshets from rivers and winter ice scour (Weingartner et al., 2005).

There are two distinct circulation patterns in the Alaskan Beaufort Sea. Water currents on the inner shelf in water depths of less than 40 meters are strongly wind driven and have been shown to undergo dramatic seasonal changes due mainly to buildup of land-fast ice. Because the principal wind direction during the summer ice-free season is from the east, nearshore flow is generally from east to west (National Research Council, 2003).

Residual water currents on the outer continental shelf (water depths greater than 40 meters) are dominated by the Beaufort Gyre, flowing toward the west, and the Beaufort Undercurrent, which transports Pacific/Bering water eastward to the Canadian Beaufort Sea (MMS 1995; National Research Council, 2003; Weingartner et al., 2005).

Sea ice can cover the Beaufort Sea shelf year-round; however, the inner shelf (and the entire shelf in recent years) is ice-free during summers from late June to early October (Weingartner, 2006). Land-fast ice begins to form in October and extends 20 to 40 km offshore until mid-June. Ice ridges form as the ice extends offshore. Ice keels often form beneath the ridges and can gouge the seafloor if the ice moves inshore during spring breakup.

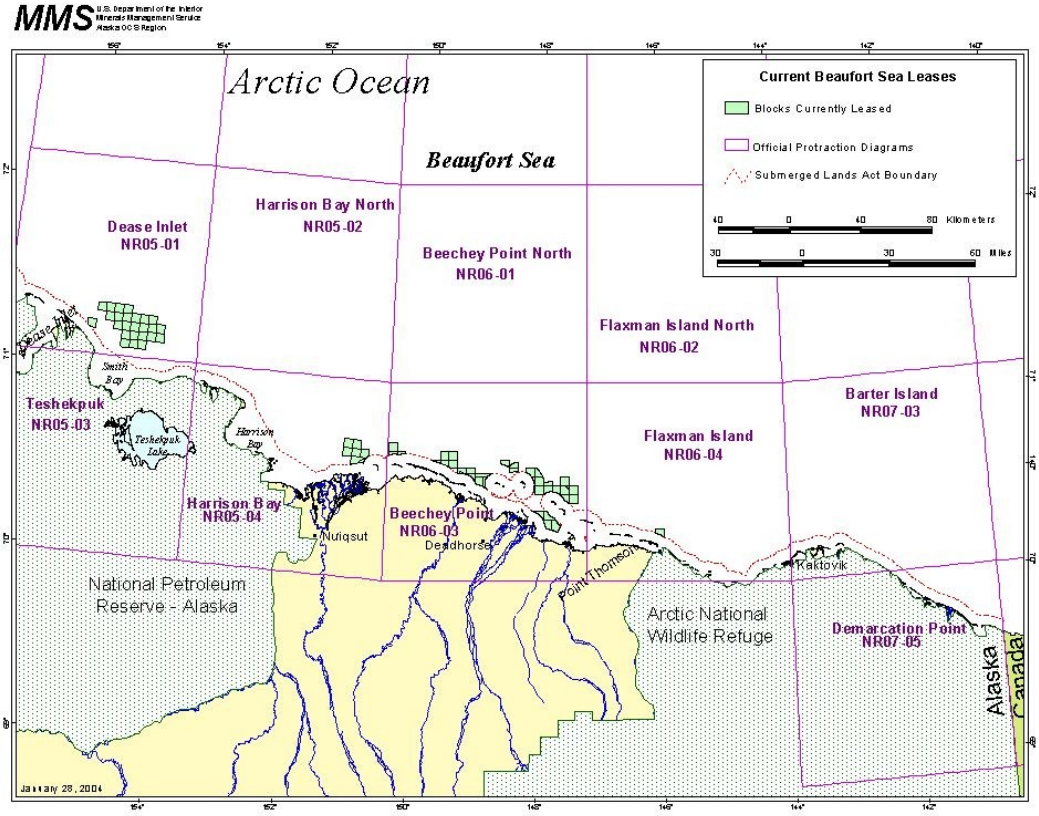


Figure 1-1. Map of the Alaskan Beaufort Sea showing offshore lease areas.

The Northstar Production facility and the Liberty prospect are located in the shallow, coastal waters of the Alaskan Beaufort Sea, and are being operated (Northstar) and developed (Liberty) by BP Exploration Alaska, Inc (BPXA). The Northstar development is located seaward of the barrier islands about 10 km northwest of Prudhoe Bay in a water depth of about 12 m. It is on an artificial island constructed partly on the remains of the old Seal Island development built and evaluated by Shell Oil Company in 1984-5 (Figure 1-2).

Although the Northstar Island is in Alaska State waters, several slant wells were drilled into Federal waters. State and Federal leases were allocated 82.16 percent and 17.84 percent, respectively, of total unitized production. The first production began from Northstar in 2001. Total oil production rose from 1,265,883 barrels (201,275 $m^{3;}$ bbl: 1 bbl = 42 gallons, 159 liters, or 0.159 m^3) in 2001 to 25,079,017 bbl (3,987,564 m^3) in 2004 and then declined to 10,606,113 bbl (1,686,372 m^3) in 2008. Average total unitized daily crude oil production has declined by about 54 percent from a high of 68,522 bbl/day (10,895 m^3/day) in 2004 to 31,660 bbl/day (5,034 m^3/day) in 2008 (Figure 1-3).

Figure 1-2. The Northstar Development on an artificial island in Alaska State waters.

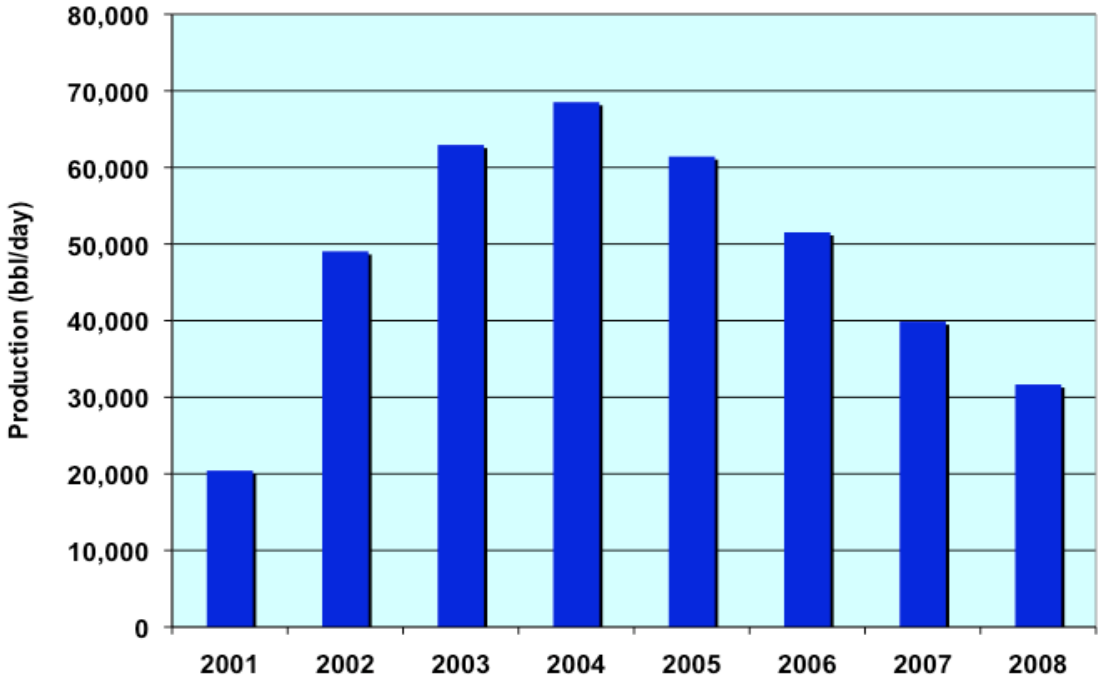


Figure 1-3. Mean total unitized (combined State and Federal) production rate per day of crude oil from the Northstar production facility from inception of production in 2001 through 2008. From MMS Alaska Office.

The Liberty Prospect is inside the barrier islands about 10 km offshore in Foggy Island Bay, at a water depth of about 7 m. It is approximately 50 km southeast of the Northstar development and about 10 km from the Endicott Causeway, near Tern Island, where Shell Oil Co. drilled in 1982 (Figure 1-4). BP Exploration (Alaska), Inc. (BPXA) drilled the discovery well in the Liberty Prospect in 1997. The original development plan for Liberty called for development drilling from an artificial island, similar to that at Northstar. In August 2005, BPXA announced that it will develop the Liberty Prospect by ultra-extended-reach drilling (uERD) from an expansion of the existing Endicott satellite drilling island (SDI), mitigating potential offshore environmental impacts to the Boulder Patch, marine mammals, and concerns of the North Slope Inupiat communities related to the bowhead whale and subsistence whaling. The Final Development and Production Plan for Liberty was submitted to MMS in April 2007 (BP Exploration (Alaska), Inc., 2007).

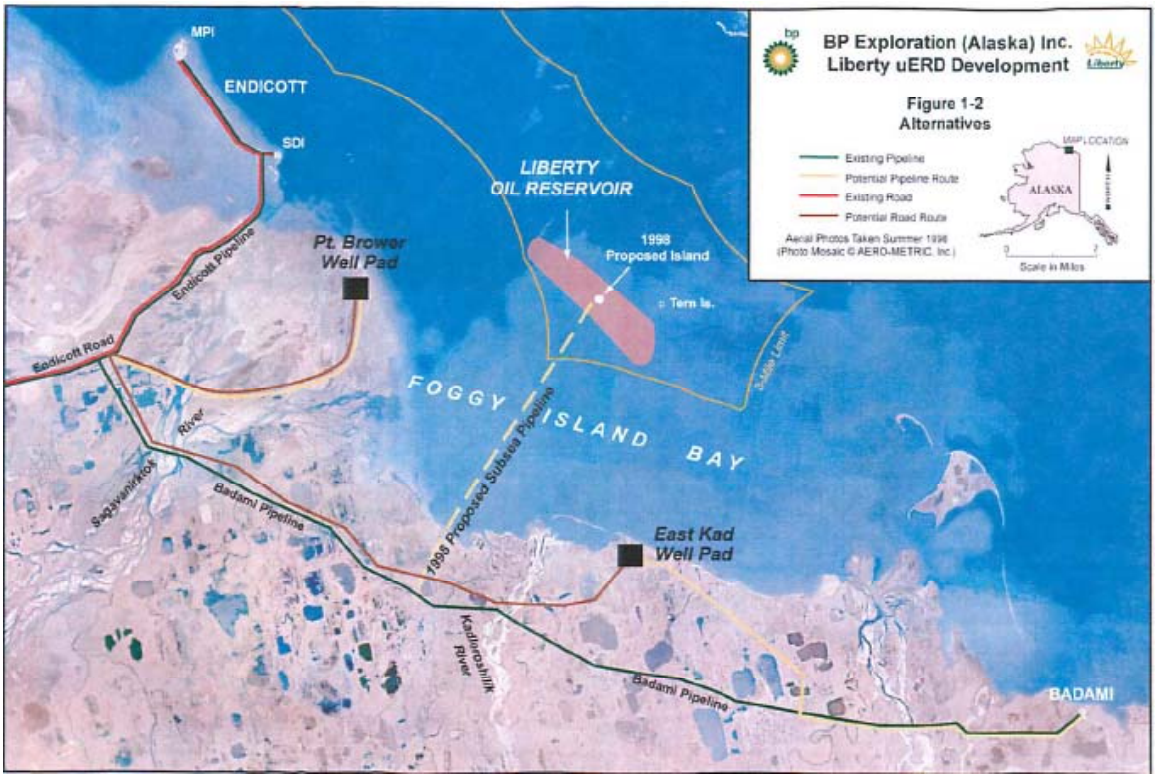


Figure 1-4. The Liberty prospect area of the Beaufort Sea, showing the alternative development strategies considered by BPXA. The Liberty reservoir will be developed from the Endicott SDI by ultra-extended-reach drilling (uERD). From MMS (2007).

Results of ANIMIDA (1999-2002)

In Task 2 of ANIMIDA, metals and hydrocarbons were measured in indigenous bivalves, amphipods, deployed caged mussels, and semi-permeable membrane devices (SPMDs) at several sites between the Northstar development and the Liberty prospect area. Task 2 was performed during the summers of 1999 (Phase 1), 2000, 2001, and 2002; results of this

component of ANIMIDA are summarized in a Final Report, Brown et al. (2005): "Hydrocarbon and Metal Characterization of Sediments, Bivalves, and Amphipods in the ANIMIDA Study Area".

In Task 8 of ANIMIDA, five species of indigenous fish were analyzed for metals and hydrocarbons, the PAH exposure biomarkers, cytochrome P4501A (CYP1A) and bile fluorescent aromatic compounds (FACs), as well as well as polychlorinated biphenyls (PCBs) and several chlorinated hydrocarbon pesticides. Sampling for Task 8 was performed at several sites in and east (upcurrent) of the development area in 2001. The results of Task 8 are summarized in a Final Report: Spies et al. (2003), "Baseline Characterization of Anthropogenic Contaminants in Biota Associated with the Alaska OCS Liberty and Northstar Oil and Gas Production Unit in the Nearshore Beaufort Sea".

Indigenous clams (*Astarte montagui*) and amphipods (*Anonyx nugax*) collected during the ANIMIDA program, contained low concentrations of saturated and polycyclic aromatic hydrocarbons (PAH) in their soft tissues, indicating that the low concentrations of hydrocarbons in local sediments and water had a limited bioavailability for these taxa. Metal concentrations in indigenous clams and amphipods were in the range of normal (background) values with no obvious indications of higher concentrations near the Northstar development site. The mean concentrations of Ba, Cu, Pb, V, and Zn in clams were quite uniform, with no detectable shifts between 1986 and 2000. However, the standard deviation for a given metal within an individual year could be quite large, severely limiting the statistical discrimination of year-to-year differences in tissue metal concentrations. There were no significant changes in concentrations of Ba, Cd, Cu, Pb, V, and Zn in amphipods collected in 1999, 2000, and 2002, indicating a lack of inputs of bioavailable metals to the Beaufort Sea from the Northstar development. The metals data for amphipods provide a good baseline for monitoring changes in inputs of metals and hydrocarbons to the nearshore Beaufort Sea from oil and gas development activities (Brown et al., 2005).

PAH concentrations also were low and variable in fish tissue, with total PAH concentrations ranging from 0 to 55.3 ug/kg (wet weight) and there were no evident relationships between PAH concentration and fish weight or sampling site (Spies et al., 2003). Concentrations of higher molecular weight PAH and PAH exposure biomarkers (CYP1A and bile FAC) were higher in some fish collected at Stump Island (southeast of Northstar) and Point Brower (near the Endicott causeway) than in the same or other species collected elsewhere, indicating possible exposure to low levels of petroleum or pyrogenic PAH, or other inducers, such as PCBs or chlorinated pesticides; however, these results require verification because increases in biomarker scores can be triggered by chemicals other than PAH (Kopponen et al., 1993; Stegeman et al., 2001). Additionally, the samples analyzed may not be representative of the entire study area.

2.0 METHODS

2.1 Field Collection

The sampling procedures used during the cANIMIDA surveys (2004 through 2006) were consistent with those used during the ANIMIDA sampling surveys. The cANIMIDA Field Survey Reports are provided in Appendix E and contain a more detailed description of the day-to-day survey activities. Individual sample collection logs from the cANIMIDA surveys are included in the Field Survey Reports (Appendix E).

The Summer 2004 field sampling was performed from July 28 to August 17, 2004; 3.5 days were lost to adverse weather conditions. The Summer 2005 survey was conducted from July 26, 2005 to August 14, 2005, with only 1 day lost to weather. The Summer 2006 survey took place from July 24 to August 12, 2006; only 1 travel day was lost at the end of the survey due to fog.

Five different types of biological samples were collected during cANIMIDA; semi-permeable membrane devices (SPMDs) were utilized only in 2004. Indigenous clams (*Astarte montagui* and *Cyrtodaria kurriana*) (Bernard, 1979), amphipods (usually *Anonyx nugax*) (Fisk et al., 2001), and fish were collected locally. Caged blue mussels (*Mytilus trossulus*) were collected in-state from Port Chatham. During all three years, isopods (*Saduria sabini*) (Percy, 1983) were collected whenever available; mysids (*Mysis* sp) were collected opportunistically in 2006. Station locations where each species was collected in 2004, 2005, and 2006 are summarized in Appendix A, Tables A-1, A-2, and A-3. The storage and preservation requirements for these samples are listed in Appendix A Table A-4. The number of each taxon collected in each year is summarized in Table 2-1.

Table 2-1. Summary of number of stations sampled for each type of biological sample in 2004, 2005, and 2006. More than 1 sample of a particular taxon was collected from some stations in 1 or more years. Fish samples contained 5 to 31 individuals of up to 8 species.

Taxon	Number of Stations where Samples were Collected		
	2004	2005	2006
Amphipod	11	10	18
Clam	5	6	2
Mussel/SPMD[a]	7[b]	8[c]	9[c]
Isopod	2	6	6
Mysid	0	0	3
Fish (up to 8 spp.)	5	2	2

[a] SPMDs deployed only in 2004;
[b] Two SPMDs and 1 container of mussels were deployed at 6 locations, and 3 reference samples of mussels were collected from Port Chatham;
[c] Two containers of mussels were deployed at each station, and 2 reference samples of mussels were collected from Port Chatham.

Biological samples were collected in 7 areas, each containing 1 to 11 sampling stations, in 2 or 3 years of the cANIMIDA Program (Table 2-2). Each biological sample, except for some large fish, was a composite of several individuals of the same species. Samples were grouped

by taxon and year as shown in Table 2-2 for statistidal analysis. Thus, the number of area replicates was highly variable for different areas, species, and years. Only, amphipods, bivalves, and fish were collected at 1 or more stations in all 3 years (Table 2-3). This limits our ability to perform statistical comparisons of chemical concentrations in individual species among the 3 years of the program.

Table 2-2. Numbers of invertebrate and fish samples analyzed in each year of the program. All samples, except for larger fish, were composites several individuals sufficient to make 10-20 g of homogenate. Fish (8 species) were composited by species. Fish sampled in 2004 and 2005 were subsampled for CYP1A and bile metabolite analysis. SPMDs were deployed with mussels in 2004. Analytical data for each taxon were grouped by collection area for statistical comparisons.

Area	Year	Amphipod	Clam	Mussel	Fish	Isopod
BSMP	2004	4	3	4	0	1
	2005	6	5	4	0	4
	2006	7	0	2	0	1
Liberty	2004	2	2	2	3[a]	0
	2005	3	1	6	18	3
	2006	4	2	8	20	1
Northstar	2004	6	0	6	34[b]	1
	2005	3	0	2	17	0
	2006	10	0	4	19	6[c]
Pt. Chatham (Ref)	2004	3	0	0	0	0
	2005	2	0	0	0	0
	2006	0	0	3	0	0
Tigvariak Island	2004	0	0	0	30[b]	0
Prudhoe	2005	0	0	2	0	0
West Dock	2006	2	0	2	0	1

[a] Five samples were collected from one station; three were used for PAH/metals analysis and two were used for CYP1A analysis.
[b] Organ tissues were collected for CYP1A and bile analysis from some fish samples.
[c] Includes three each of isopods and mysids.

Table 2-3. Stations where biological samples were collected in 2 or 3 years of the cANIMIDA Program. Species collected each year are listed as: A – amphipod; B – bivalve; F – fish; I – isopod; M – mussel; NS – no biological sample collected; and S - SPMD.

Station ID	**Station Type**	**2004**	**2005**	**2006**
3A	BSMP	B, M, S	B	NS
4A	BSMP	A	A	A
L08	Liberty	B	B, I, M	A, B, I, M
N03	Northstar	A, I	A, M	A, M
N06	Northstar	M, S	NS	A
N11	Northstar	A	A	A, M
N18	Northstar	A	A	NS
SIS	Northstar	F	F	F

2.2 Fish Collection, Processing, and Sample Storage

2.2.1 Summer 2004

Fish samples were collected at five locations during the 2004 Survey: Point Brower (Liberty Station PBS), Stump Island (Northstar Station SIS), Tigvariak Island (Station TGV), near the Liberty development area (Station L14), and near Northstar (Station N18) (Appendix A Table A-1, Figures 2-1 through 2-3). Fyke nets were used for sample collection at Point Brower, Stump Island and Tigvariak Island. Nets were deployed for 2 days at both Point Brower and Stump Island, and captured fish were collected and processed daily. A fyke net was deployed at Tigvariak Island for 1 night and 1 day; fish were collected once in the morning and again that afternoon. Fish were collected by trawling at both Liberty and Northstar.

2.2.2 Summer 2005

During the 2005 sampling survey, fish were collected only from Point Brower (Station PB) and Stump Island (Station SIS) with fyke nets. The 2005 biota sampling locations are shown in Figure 2-4; the total numbers sampled at each location are summarized in Appendix A Table A-2. Fish caught near Point Brower are considered to be from the Liberty Area (Figure 2-5) and those from near Stump Island are considered to be Northstar samples (Figure 2-6) for the purpose of data comparison.

2.2.3 Summer 2006

Fish were collected in the summer of 2006 at the same locations and using the same methods as in 2005. All 2006 biota sampling locations are illustrated in Figures 2-7 through 2-9.

2.2.4 Fish Processing

Fish samples were processed at the British Petroleum (BP) Seawater Treatment Plant (STP) Conex building the same day collected by methods defined in the Field Sampling and Logistics Plans (Appendix E). Samples were processed for whole body tissue chemistry (hydrocarbon and metal concentrations). Subsamples of gill, gut, kidney and liver were removed and preserved in 10 percent formalin for immunohistochemical staining for cytochrome P4501A (CYP1A) activity; bile was removed and frozen for analysis of fluorescent aromatic compounds (FACs) (PAH metabolites).

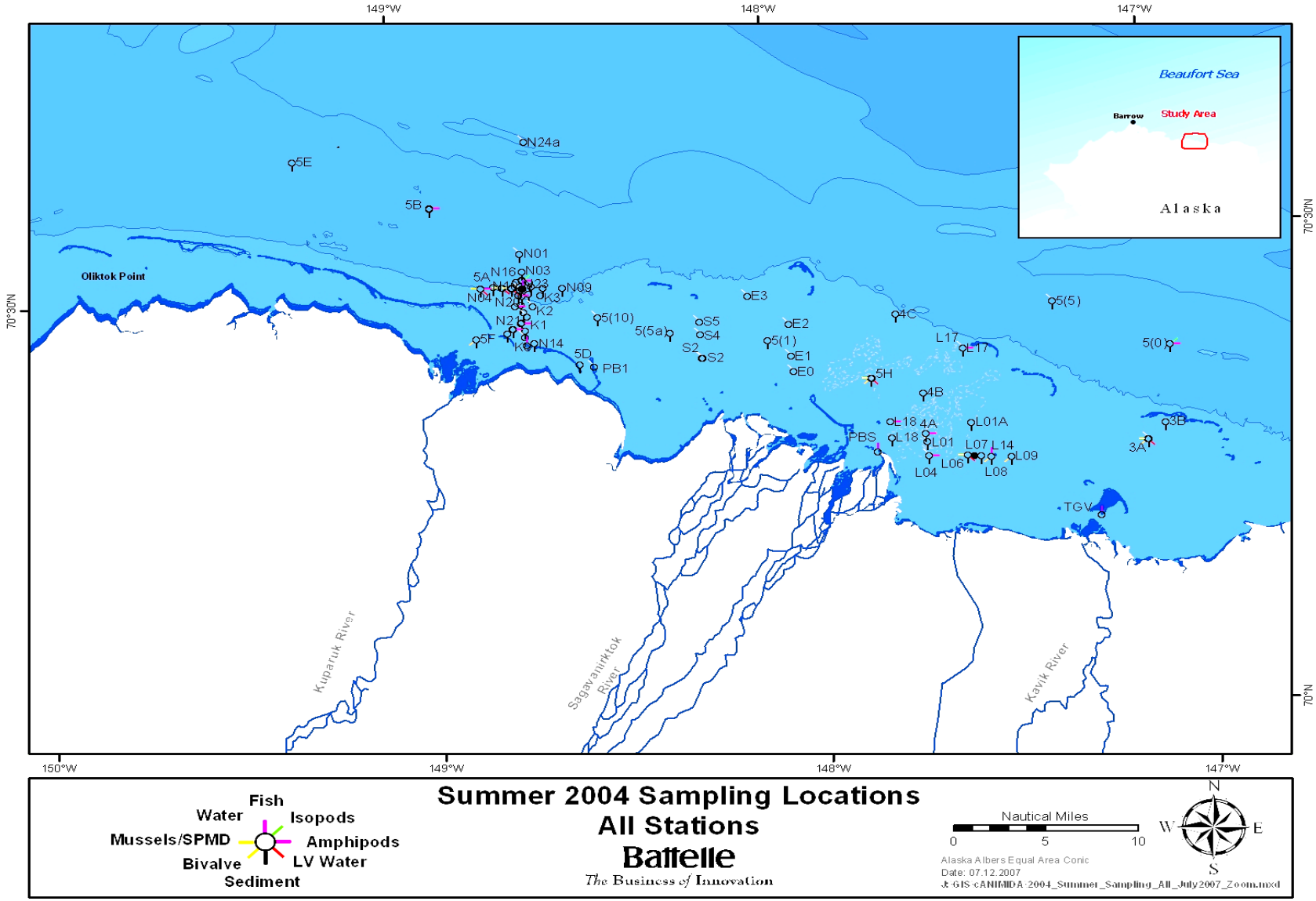


Figure 2-1. Sampling stations for biological samples and mussel/SPMD deployments in 2004

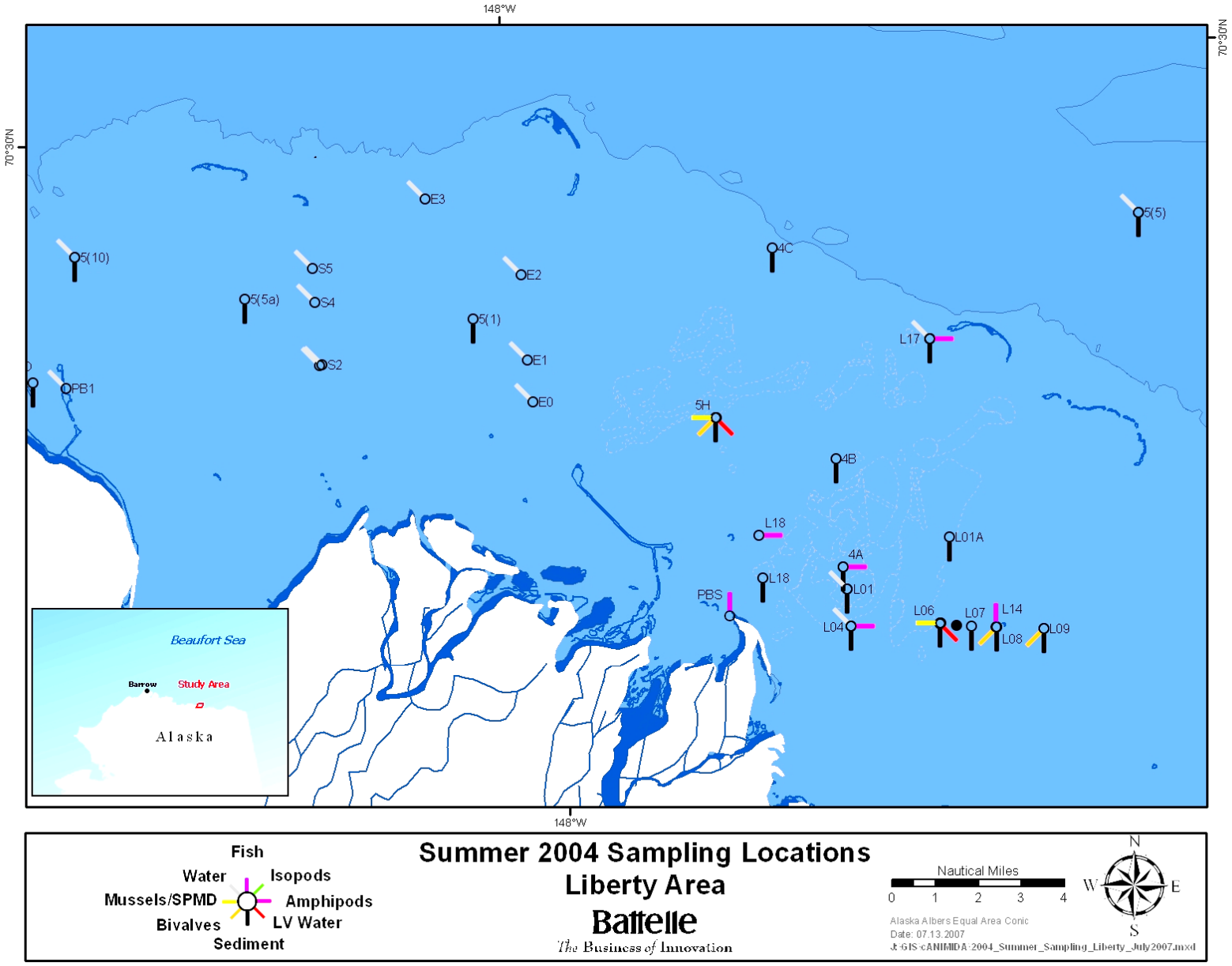


Figure 2-2. Sampling stations for biological samples and mussel/SPMD deployments near the Liberty Prospect for 2004.

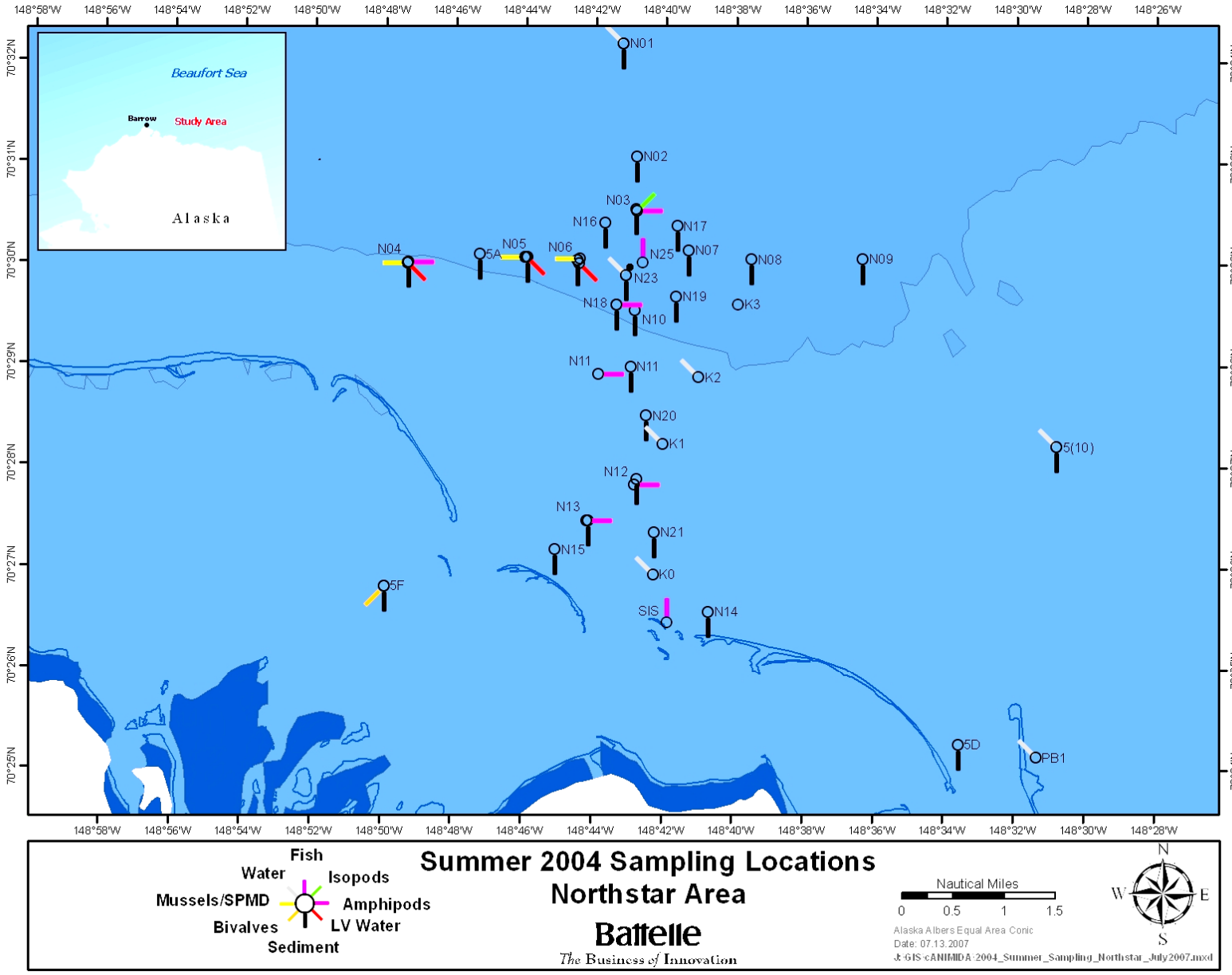


Figure 2-3. Sampling stations for biological samples and mussel/SPMD deployments near Northstar for 2004.

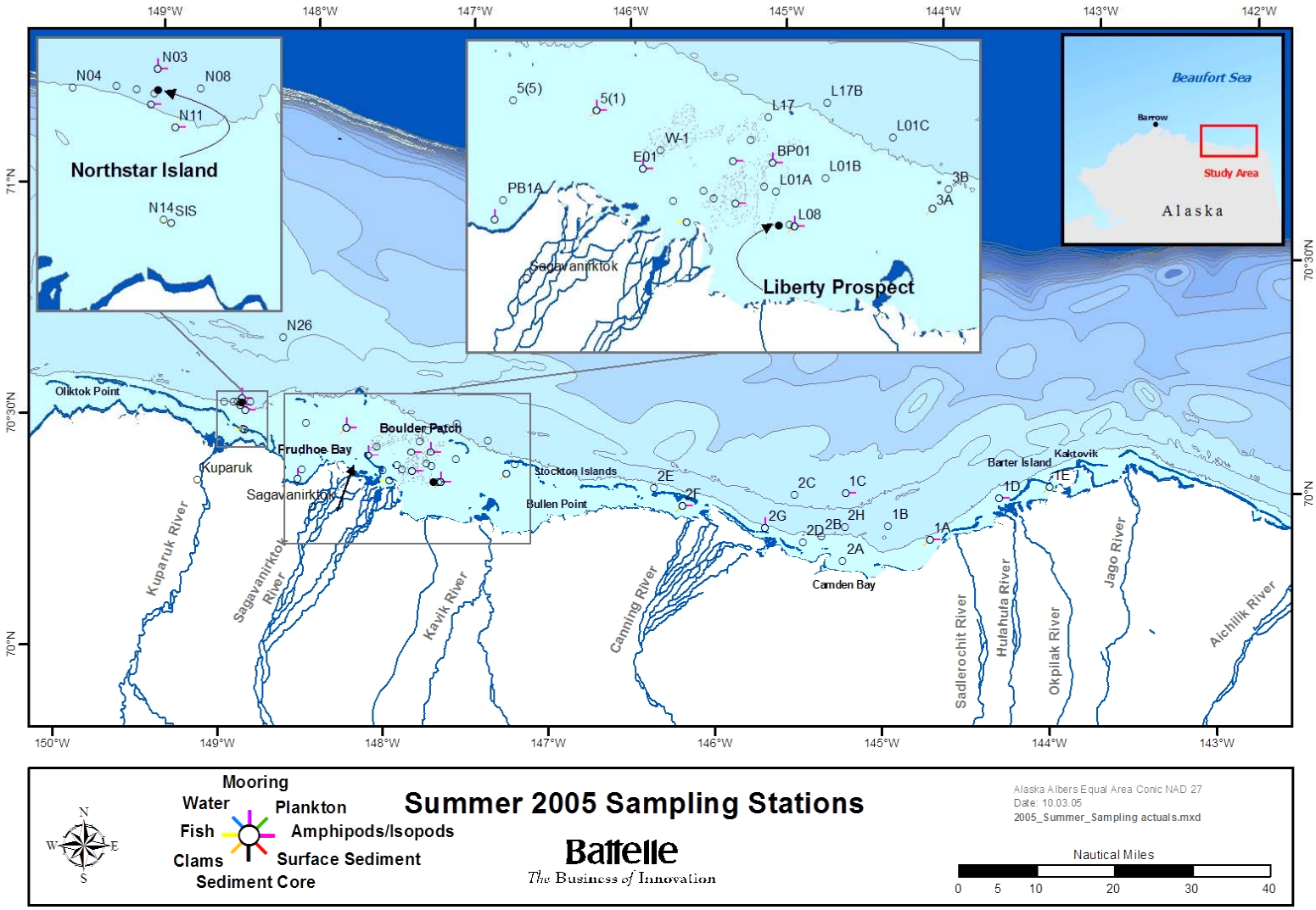


Figure 2-4. Sampling stations for biological samples for 2005.

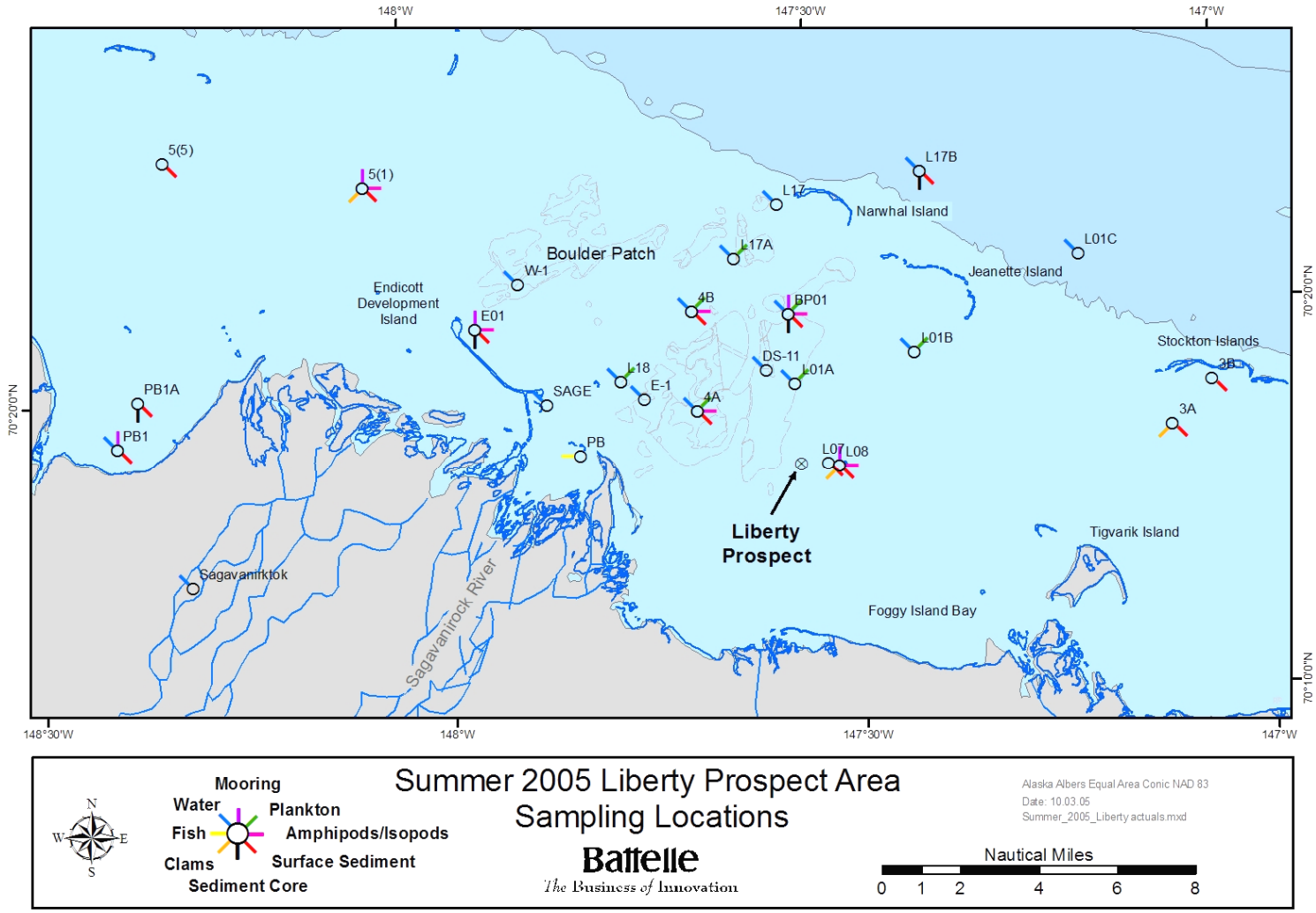


Figure 2-5. Sampling stations for biological samples near the Liberty Prospect for 2005.

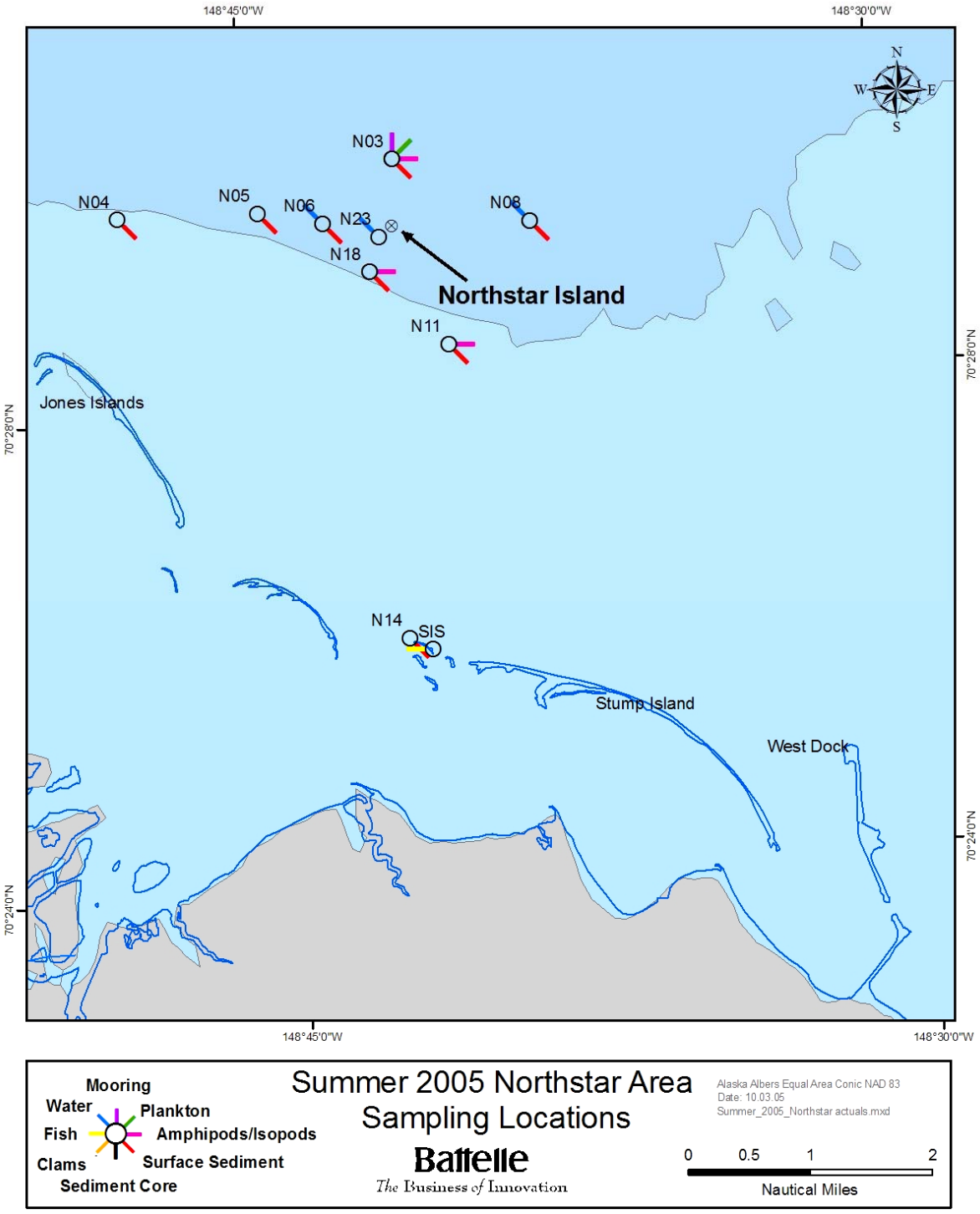


Figure 2-6. Sampling stations for biological samples near the Northstar Development for 2005.

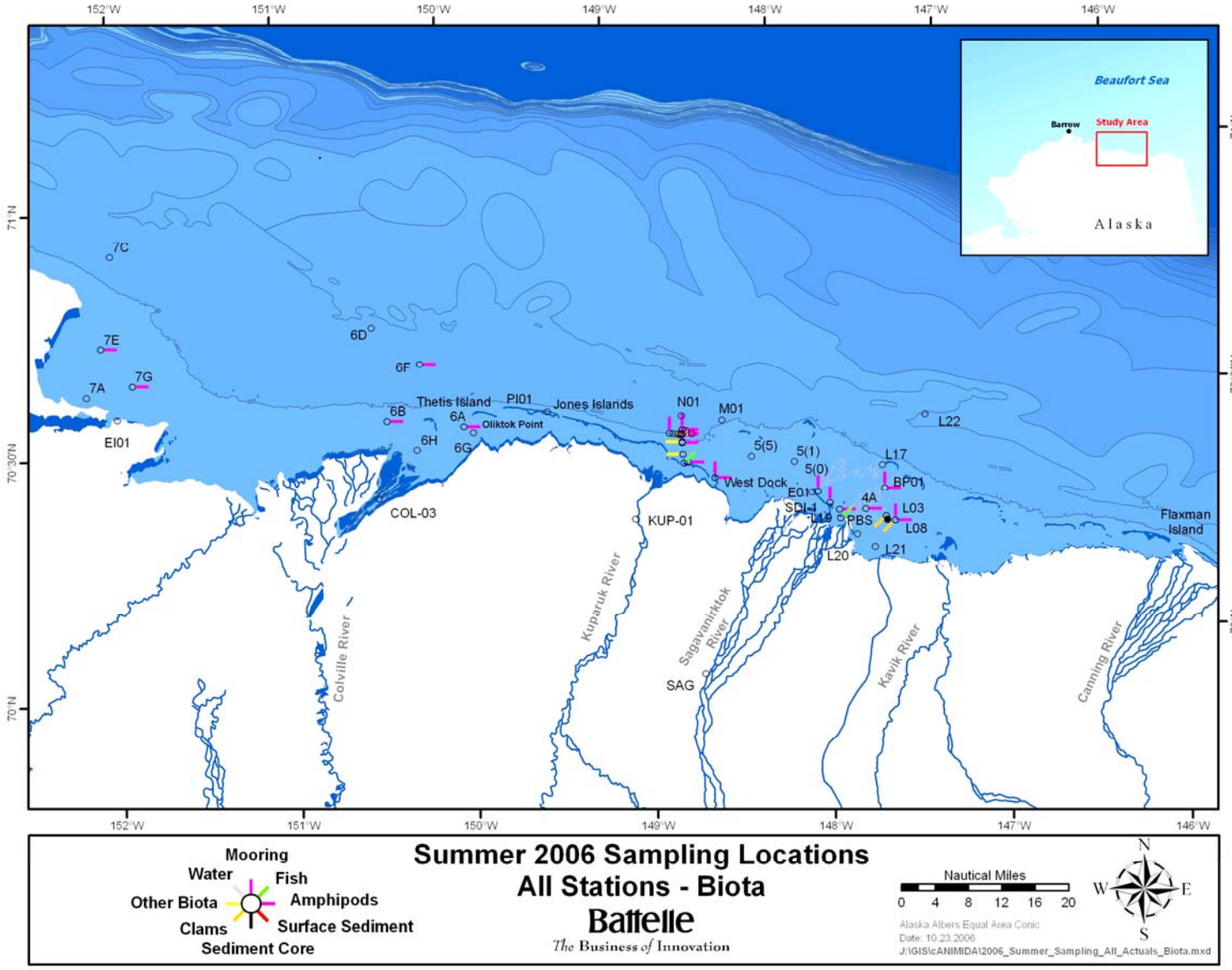


Figure 2-7. Sampling stations for biological samples for 2006.

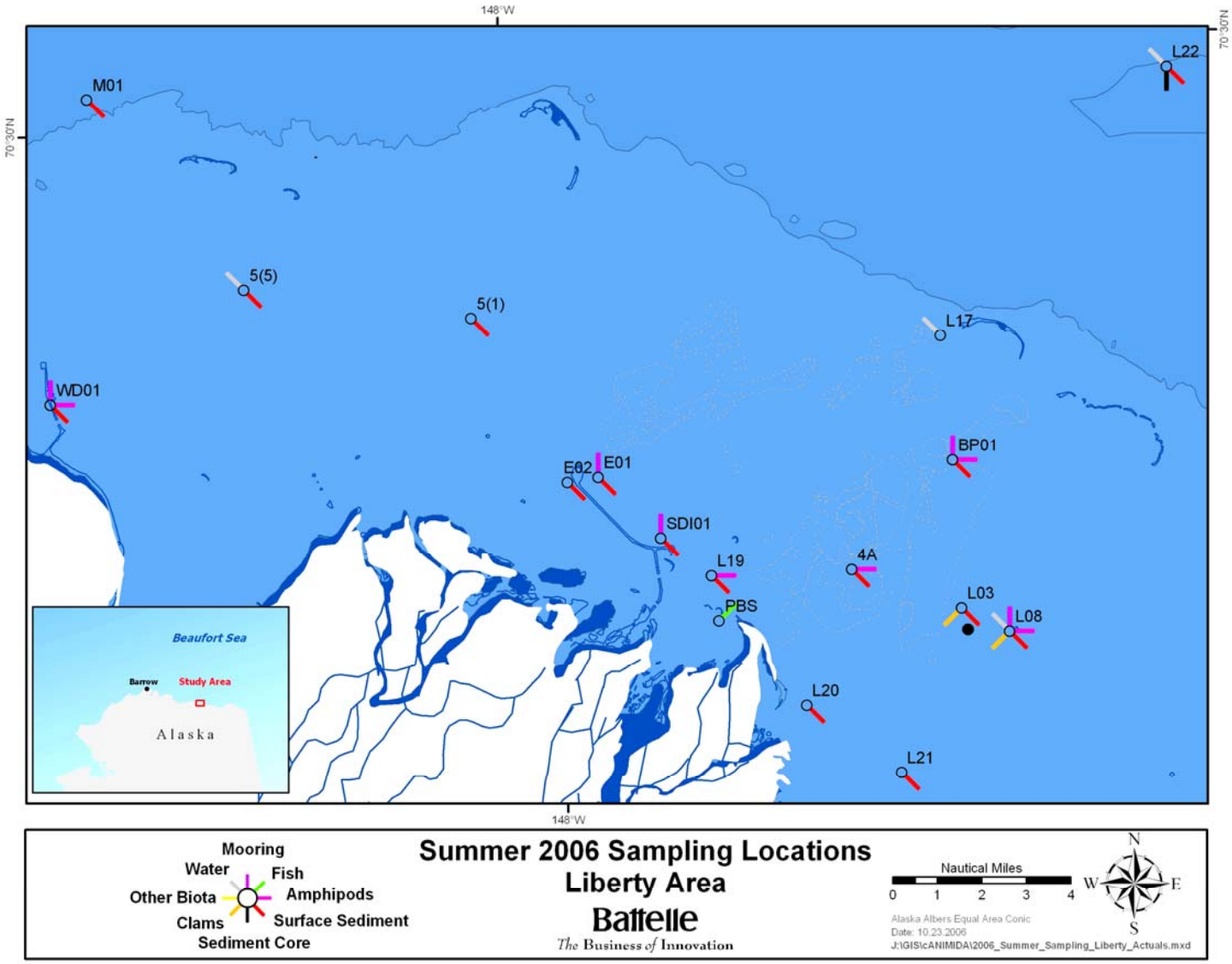


Figure 2-8. Sampling stations for biological samples near the Liberty Prospect for 2006.

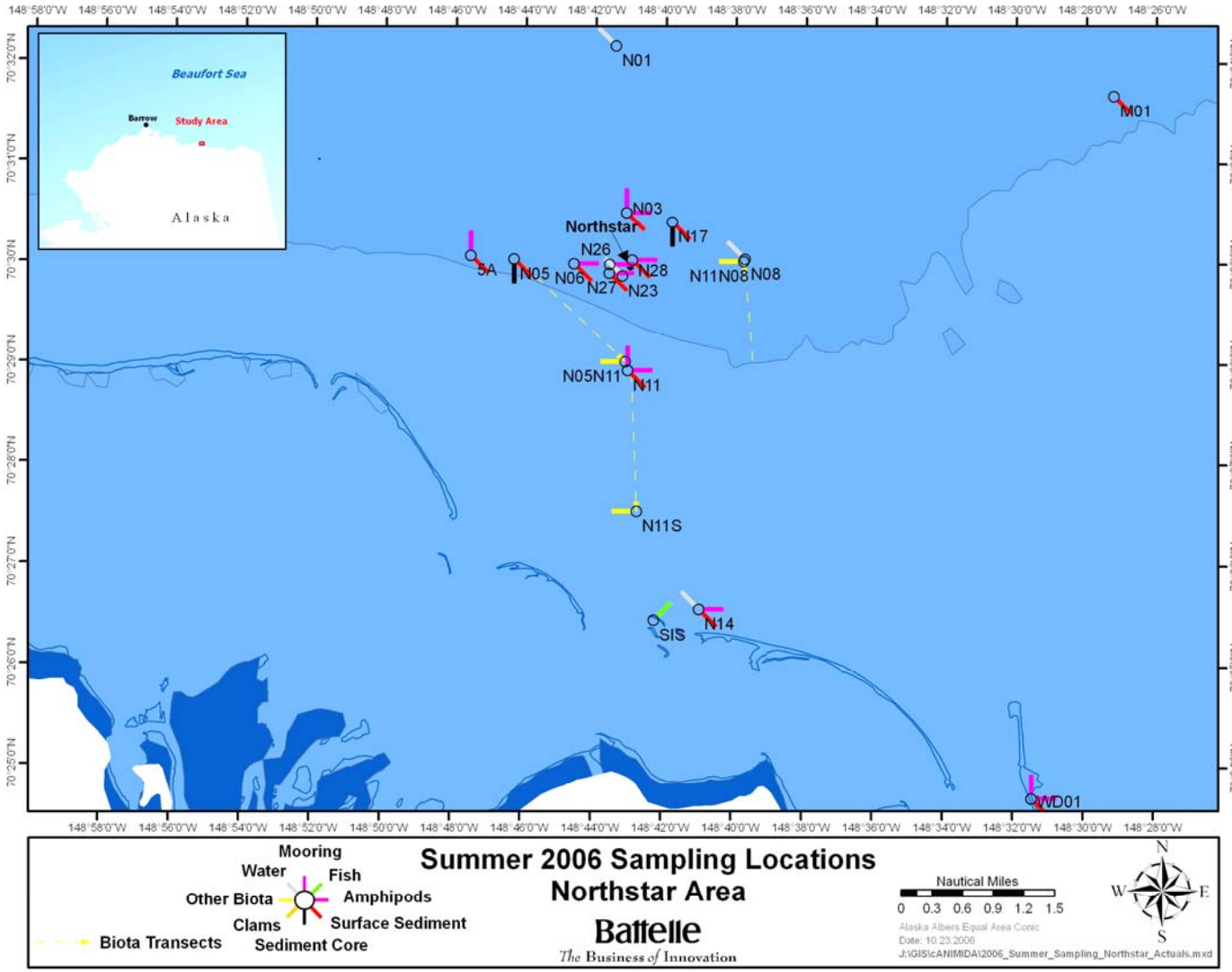


Figure 2-9. Sampling stations for biological samples near the Northstar Development for 2005.

2.3 Mussel and SPMD (2004 Only) Deployment, Retrieval, Processing, and Storage

2.3.1 Summer 2004

A total of six subsurface moorings were deployed in the study area in 2004 (Appendix A Table A-1); three locations were within the Northstar area, one was near Liberty, and two were at historical Beaufort Sea Monitoring Program (BSMP) stations (Figures 2-1 through 2-3). Each mooring consisted of one mussel cage containing approximately 40 mussels in a Nytex tube, two SPMD cages (each cage contained two SPMDs), and an acoustic pinger (Figure 2-10). All the mussels in each cage were composited to make a single homogeneous sample. The SPMDs from each site also were composited. The mussels *(Mytilus trossulus)* were obtained from Port Chatham, AK, on July 27 by Mark Savoi of KLI, Anchorage. The mussels were transported to Prudhoe Bay wrapped in wet paper towels, and stored on ice. The mussels were then acclimated to local conditions by placing them in a Rubbermaid tub filled with Beaufort Sea water and aerated with a battery driven air pump. Surface floats were not used in order to minimize ice entanglement; each mooring had a secondary anchor and a ~100 meter drag line to aid in retrieval.

Three samples of 'time zero' mussels were collected and used as trip blanks (MZ-1, 2, and 3) and reference samples. These mussels were transferred to glass jars and frozen three days after collection in Port Chatham. Moorings were deployed for 14 days at the Liberty station (L06) and one of the BSMP stations (3A), for 15 days at a second BSMP station (5H), and for 17 days at the three Northstar stations (N04, N05, and N25). The field sampling plan calls for the SPMDs and mussels to be deployed a minimum of 21 days; however, due to logistical considerations and time constraints, the moorings were retrieved after a shorter time interval. In subsequent years, the deployment time for the mussels was 13 to 14 days each year.

Upon retrieval, the mussels were counted to ascertain if any had been lost or died and then removed to a pre-cleaned glass jar. Mussel samples were stored frozen until shipment to the Battelle analytical laboratory in Duxbury, Massachusetts. The SPMDs were inspected visually for tears and bio-fouling before being carefully removed and transferred to a pre-cleaned glass jar. The SPMDs were stored and shipped frozen.

Several problems were encountered in using deployed mussels and SPMDs as passive samplers of hydrocarbons in the Beaufort Sea water column. Cold ambient temperatures, short deployment times, and elevated zero-time concentrations of target analytes in both SMPDs and mussels, rendered results difficult to interpret. This problems are discussed in detail in Section 4.

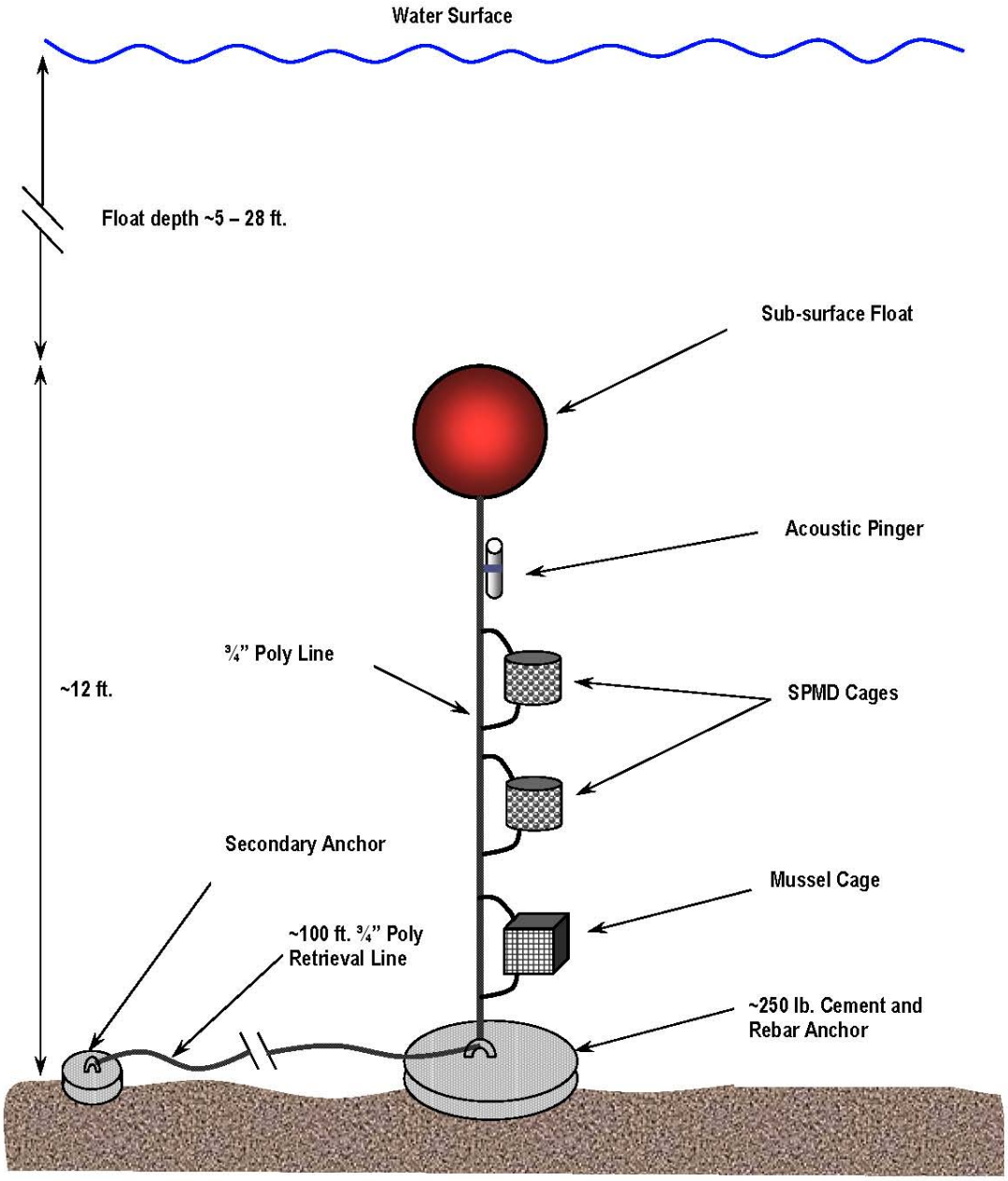


Figure 2-10. Mussel and SPMD mooring diagram.

2.3.2 Summer 2005

Because of technical problems with the SPMDs (high blank concentrations of PAH), they did not provide much useful data in addition to that provided by the deployed mussels. Therefore, they were not deployed in 2005 and 2006.

Seven subsurface moorings containing caged mussels were deployed and recovered in the study area during the 2005 survey (Appendix A Table A-2). Moorings were located at 1 station near the Northstar production facility (Station N03), in Prudhoe Bay (Station PB1), and in the Boulder Patch (Station BP01), and at two stations were located near the Liberty Prospect (Stations E01 and L08), and in the historic Beaufort Sea Monitoring Program (BSMP) area (Stations 2G and 5[1]). Three moorings were deployed in the (BSMP) area, but only 2 were recovered. The mussels *(Mytilus trossulus)* were obtained from Port Chatham, AK, on July 26 by Mark Savoi of KLI, Anchorage.

Two samples of 'time zero' mussels were collected and used as trip blanks and reference samples. One blank was collected by immediately freezing a sub-set of mussels upon collection in Port Chatham. The second blank consisted of mussels that had been acclimated and transported to the Beaufort Sea; the mussels in this blank were sacrificed when mussels were deployed at stations 2G and 2E.

Moorings were deployed for 13 days at station E01 and in Prudhoe Bay. All other moorings were collected after a 14-day deployment.

2.3.3 Summer 2006

During the 2006 survey, 8 subsurface moorings containing caged mussels were deployed in the study area (Appendix A Table A-3). Moorings were located in the following areas: the Liberty Prospect, including the Boulder Patch (Stations PB01, E01, and L08), near West Dock (Station WD01), near SDI (Station SDI-1), and at a historic BSMP station (Station 5A); two moorings were deployed near Northstar (Stations N03 and N11). The mussels were obtained from Port Chatham, Alaska, on July 24, 2006.

Three samples of 'time zero' (reference) mussels were collected and used as trip blanks. One blank was obtained by immediately freezing a sub-set of mussels upon collection in Port Chatham. The mussels in the remaining two blanks were sacrificed when mussels were deployed at the Boulder Patch and at West Dock.

Moorings were deployed for 13 days at stations 5A, N03, N11, and West Dock. All other moorings were collected after a 14-day deployment.

2.4 Clam, Amphipod, Isopod, and Mysid Collection and Storage

2.4.1 Summer 2004

Nineteen samples of indigenous marine invertebrates, consisting of clams (*Cyrtodaria kurriana* and *Astarte montagui*), amphipods (*Anonyx nugax*), or isopods (*Saduria sabini*), were collected from 16 stations during the Summer 2004 field survey (Table 2-1; Appendix A Table A-1; Figures 2-1 through 2-3). Isopod samples were collected from the Northstar development area

and from a BSMP station. Amphipod samples were collected from the Northstar, Liberty, and BSMP areas; clams were collected from the Liberty and BSMP areas. Baited minnow and amphipod traps were used to capture amphipods and isopods. Upon retrieval of the traps, the volume of organisms collected was estimated and tissues were transferred to pre-cleaned glass containers and stored frozen.

2.4.2 Summer 2005

The same methods were employed in 2005 to capture indigenous marine invertebrates as had been used previously. A total of 24 indigenous biota samples were collected from the study area (Appendix A Table A-2; Figure 2-4, 2-5, and 2-6). Isopods and bivalves were collected from Liberty and BSMP areas. Amphipods were collected near Liberty, Northstar, and from several BSMP locations. Clams were collected from Station L08 at Liberty.

2.4.3 Summer 2006

Thirty-four indigenous biota samples were collected from the cANIMIDA study area during the 2006 field sampling survey (Appendix A Table A-3). Amphipod collections were quite successful and organisms were acquired from 17 stations throughout the study area (Figures 2-7 through 2-9). Bivalve samples were obtained from two stations in the Liberty area and mysids (*Mysis* sp) were collected from three stations in the vicinity of the Northstar development. Isopods were collected near Northstar, Liberty, West Dock, and from a historical BSMP station.

2.5 Source Materials Collection

Approximately one liter of Northstar crude oil was received for chemical analysis during the Summer 2004 Survey. Source oil was not collected during the 2005 or 2006 surveys; however, peat samples were collected during the 2006 survey from the Colville, Kuparuk, and Sagavanirktok Rivers, as well as from Pingok Island.

2.6 Sample Shipment

Following completion of the surveys, samples were inventoried, kept frozen, and packed in coolers for overnight shipment from the Prudhoe Bay Airport (Deadhorse) using Federal Express or another air freight courier. Inventory included counting all the samples to ensure that all samples were collected and safely returned to the custody area, documenting all samples, and preparing chain-of-custody (COC) forms for all samples. Samples were shipped to the Battelle Duxbury facility for chemical analysis. At all times after collection, sample integrity and custody was maintained (Battelle Standard Operating Procedures (SOP) 6-040 and 5-210). Custody seals were used on all shipping containers (i.e., coolers) to maintain custodial security while the samples were in the possession of a third party (e.g., air freight courier). Samples were shipped to the analytical laboratory packed with cold blue ice. Upon receipt, samples were logged into the Laboratory Information Management System (LIMS) and assigned unique Battelle Duxbury identification numbers.

2.7 Field Quality Control

2.7.1 Equipment Decontamination

Field equipment was thoroughly decontaminated between sampling stations by scrubbing with soap and water, rinsing with seawater, rinsing with de-ionized water, and a doing a final solvent rinse with methanol.

2.7.2 Blanks

Equipment blanks were collected during sediment, biota, and water collection in order to characterize any potential contamination that may have been introduced during sampling. A deck blank was collected during the surveys for metals and organics analysis. This blank was collected at Station L01A (2004) and at Station 2B (2005) by placing two clean, pre-labeled, open sampling jars on the deck during the collection of one sample. The jar was subsequently closed and handled in the same manner as the remaining samples. These deck blanks were not thought to be particularly useful after having been stored in a freezer for several weeks and were not analyzed.

Aqueous equipment blanks were collected at two locations, L01A and N21, during the Summer 2004 sampling and at 2B during the Summer 2005 sampling, and at SDI, West Dock, and N26 during the summer of 2006. At each location, a blank was collected for metals and organics analysis by rinsing equipment with high-purity deionized or MilliQ water and collecting the rinsate into two pre-cleaned sample jars. A clean stainless steel or Teflon® funnel was used to facilitate water collection. Prior to the blank collection, all equipment was decontaminated and the equipment blanks were then stored and handled in the same manner as the samples.

Two additional 'equipment wipe' samples were collected during the 2004 and 2005 surveys. One sample represented the diesel fuel used on the *1273* and a second represented hydraulic oil used on the vessel. The purpose of these samples was to, if required, distinguish any sample contamination that may have been derived from the ship's fuel or hydraulic systems.

2.7.3 Field Replicates and Trip Blanks

The only tissue sample replicates collected during the cANIMIDA surveys were the time-zero mussels. The mussel blanks from 2004 were collected and analyzed in triplicate and those in 2005 and 2006 were collected an analyzed in duplicate when adequate volume was available. These samples also served as trip blanks since they were collected after the transit from Port Chatham to Deadhorse. Two SPMD blanks were collected and analyzed for PAH (2004 only). One was a lab blank, which was shipped directly from the supplier to Battelle Duxbury Operations and was not removed from the sealed shipping container (paint can) until immediately prior to extraction. The second SPMD blank was a trip blank that was removed from its shipping container and transferred to a sample collection jar on July 30th, 2004, at the time of SPMD and mussel deployment.

2.7.4 Documentation

Field scientists maintained field notes and station logs in a field logbook. Sampling information, including species collected, numbers of organisms, and sampling location were recorded. Any

exceptions to procedures and technical difficulties also were noted. All the sampling information, including copies of station and sampling logs, are included in the Survey Field Reports (Appendix A).

2.8 Analytical Methods

The following section contains a general description of the analytical methods employed for tissue samples during the cANIMIDA program. Analysis of hydrocarbons in tissue samples was performed at Battelle Duxbury Operations under the supervision of Gregory Durell. A metals analysis in tissues was performed in the laboratory of Dr. John Trefry at Florida Institute of Oceanography. Additional information, such as specific data quality objectives, quality control sample definitions, and detailed analytical methodology is included in Appendix B: Summary of Analytical Methods.

2.8.1 Hydrocarbon and Metal Analysis

A detailed description of the methods for preparation of tissue samples for analysis and the analytical methods for metals and hydrocarbons in tissues are included in Appendix B. A summary of analytical methods is presented here.

2.8.1.1 Tissue Sample Selection for Chemical Analysis

Summer 2004
A total of 28 fish samples (representing 7 species and 41 individual fish), 9 deployed mussel, 14 SPMDs, and 4 clam samples, and 8 amphipod samples from the 2004 field survey were selected for analysis (Appendix A Tables A-5 through A-7).

Summer 2005
A total of 21 fish samples (representing 5 species), 9 deployed mussel samples, 8 clam samples, 11 amphipod samples, and 7 isopod samples from the 2005 field survey were selected for analysis (Appendix A Tables A-8 through A-10).

Summer 2006
A total of 18 fish samples (representing 5 species), 11 deployed mussel samples, 2 clam samples, 25 amphipod samples, 6 isopod samples, and 3 mysid samples from the 2006 field survey were selected for analysis (Appendix A Tables A-11 through A-13).

2.8.2 Tissue Sample Preparation and Analysis

2.8.2.1 Tissue Preparation

All tissues were stored frozen until just prior to preparation. Whole mussels and clams were rinsed with deionized or MilliQ-treated water to remove any sediment from the outside of the shell and were shucked with a titanium or ceramic knife. Crustaceans and fish were partially thawed before homogenization. Whole fish were homogenized; if fish were too large to fit into the sample jars, they were cut into smaller pieces with a solvent- and acid-rinsed ceramic or titanium knife prior to homogenization. Most samples, except some large fish, were composited by species collected at each station and year. Fish also were composited by species. The number of replicate samples from each sampling area and year is summarized in Table 2-2. All tissue

samples were homogenized in a Tissuemizer® with a titanium probe. If a sufficient mass of homogenized tissue was available in a sample, 15 to 20 grams of homogenized tissue was removed to a pre-cleaned glass jar, frozen, and shipped to Florida Institute of Technology (FIT) for metals analysis.

2.8.2.2 Analysis of Organic Compounds

A detailed description of methods used for extraction, cleanup, and analysis of target organic compounds in tissue samples is in Section 2.0 of Appendix B. The homogenized tissue aliquot retained for organic analysis was spiked with a surrogate internal standard solution (SIS), combined with a drying agent (typically desiccated sodium sulfate) and methylene chloride and serially extracted. The extracts were collected, concentrated, and then purified using a combination of alumina, silica gel, and liquid chromatography techniques. The purified extracts were then spiked with a recovery internal standard (RIS) prior to analysis for PAH and biomarkers (S/T) by gas-chromatography/mass-spectrometry (GC/MS) or SHC analysis by GC/flame ionization detection (FID).

Three classes of organic analytes were quantified in tissue samples: polycyclic aromatic hydrocarbons (PAH), saturated hydrocarbons (SHC), and sterane/triterpane petroleum biomarkers (StTr). PAH were measured in all tissue samples and in SPMDs; however, there was insufficient tissue mass in 3 clam samples collected in 2005 and they were analyzed for just metals. Mussel, clam, and amphipod samples also were analyzed for SHC and StTr. Target organic analytes for PAH, SHC, and StTr are summarized in Tables 2-4, 2-5, and 2-6.

Table 2-4. Target polycyclic aromatic hydrocarbons (PAH) analyzed in tissue samples.

Compound	Internal Standard and Surrogate Reference	Compound	Internal Standard and Surrogate Reference
Naphthalene	A/1	Benzo[a]anthracene	B/3
C1-Naphthalenes	A/2	Chrysene	B/3
C2-Naphthalenes	A/2	C1-Chrysenes	B/3
C3-Naphthalenes	A/2	C2-Chrysenes	B/3
C4-Naphthalenes	A/2	C3-Chrysenes	B/3
Acenaphthylene	A/2	C4-Chrysenes	B/3
Acenaphthene	A/2	Benzo[b]fluoranthene	B/4
Biphenyl	A/2	Benzo[k]fluoranthene	B/4
Fluorene	A/2	Benzo[e]pyrene	B/4
C1-Fluorenes	A/2	Benzo[a]pyrene	B/4
C2-Fluorenes	A/2	Perylene	B/4
C3-Fluorenes	A/2	Indeno[1,2,3-c,d]pyrene	B/4
Anthracene	A/3	Dibenzo[a,h]anthracene	B/4
Phenanthrene	A/3	Benzo[g,h,i]perylene	B/4
C1-Phenanthrenes/Anthracenes	A/3		
C2-Phenanthrenes/Anthracenes	A/3		

Table 2–4. Target polycyclic aromatic hydrocarbons (PAH) analyzed in tissue samples, continued

Compound	Internal Standard and Surrogate Reference	Compound	Internal Standard and Surrogate Reference
C3-Phenanthrenes/Anthracenes	A/3		
C4-Phenanthrenes/Anthracenes	A/3	Surrogate Compounds	
Dibenzothiophene	A/3	Naphthalene-d8	A/1
C1-Dibenzothiophenes	A/3	Acenaphthene-d10	A/2
C2-Dibenzothiophenes	A/3	Phenanthrene-d10	A/3
C3-Dibenzothiophenes	A/3	Benzo(a)pyrene-d12	B/4
Fluoranthene	A/3		
Pyrene	A/3	Internal Standard	
C1-Fluoranthenes/Pyrenes	A/3	Fluorene-d10	A
C2-Fluoranthenes/Pyrenes	A/3	Chrysene-d12	B
C3-Fluoranthenes/Pyrenes	A/3		

Table 2-5. Target saturated hydrocarbons (SHC), including the n-C9 through n-C40 normal alkanes, and the isoprenoids pristane, phytane, and 1470, analyzed in mussel, clam, and amphipod samples.

Target Saturated Hydrocarbon (SHC)			
nC_8 (optional)	1650	nC_{23}	nC_{33}
nC_9	nC_{16}	nC_{24}	nC_{34}
nC_{10}	nC_{17}	nC_{25}	nC_{35}
nC_{11}	Pristane	nC_{26}	nC_{36}
nC_{12}	nC_{18}	nC_{27}	nC_{37}
nC_{13}	Phytane	nC_{28}	nC_{38}
1380	nC_{19}	nC_{29}	nC_{39}
nC_{14}	nC_{20}	nC_{30}	nC_{40}
1470	nC_{21}	nC_{31}	Total SHC
nC_{15}	nC_{22}	nC_{32}	

Table 2-6. Target steranes and triterpanes (StTr) analyzed in mussel, clam, and amphipod samples.

Steranes/Triterpanes		Reporting Code
Analyte Name	Common Name[b]	
13β,17α-diacholestane-20S	Diacholestane	S4
13β,17α-diacholestane-20R	Diacholestane	S5
5α,14α,17α,24-methylcholestane-20R	Methylcholestane	S24
5α,14α,17α,24-ethylcholestane-20S	Ethylcholestane	S25
5α,14α,17α,24-ethylcholestane-20R	Ethylcholestane	S28
C_{23} diterpane	Diterpane	T4

Table 2–6. Target steranes and triterpanes (StTr) analyzed in mussel, clam, and amphipod samples continued.

Steranes/Triterpanes		Reporting Code
Analyte Name	**Common Name[b]**	
C_{29} tricyclictriterpane	Tricyclictriterpane	T9
C_{29} tricyclictriterpane	Tricyclictriterpane	T10
18α(H)-22,29,30-trisnorhopane-TS	Trisnorhopane (TS)	T11
17α(H)-22,29,30-trisnorhopane-TM	Trisnorhopane (TM)	T12
17α(H),21β(H)-30-norhopane	Norhopane	T15
18α(H)-oleanane	Oleanane	T18
17α(H),21β(H)-hopane	Hopane	T19
22S-17α(H),21β(H)-30-homohopane	Homohopane	T21
22R-17α(H),21β(H)-30-homohopane	Homohopane	T22
*5a, 14a, 17a-Cholestane	5a, 14a, 17a-Cholestane	S17
*17b(H), 21b(H)-Hopane	17b(H), 21b(H)-Hopane	T23

**Compound used in calibration, but not reported*

2.8.2.3 Analysis of Metals in Tissue Samples

Upon arrival at FIT, the tissue sub-samples submitted for metals analysis were thawed and thoroughly mixed with a Teflon® spatula. Samples were split into two aliquots, one to be digested wet for Hg determination and the other to be freeze dried and digested for the analysis of the remaining metal analytes. The freeze-dried aliquots were re-weighed to determine the percent moisture and the desiccated tissue was then digested by a sequential addition of concentrated, high-purity nitric acid (HNO_3), hydrogen peroxide (H_2O_2), and hydrochloric acid (HCl). The acid extract was then diluted with de-ionized, distilled water (DDW) and stored in polyethylene vials until analysis.

Tissue subsamples designated for mercury analysis were digested with concentrated, high-purity nitric acid (HNO_3) and sulfuric acid (H_2SO_4) and refluxed in the sealed extraction tubes. Upon completion of digestion, the acid solutions were diluted with DDW and stored in polyethylene screw-cap vials until analysis.

Metal concentrations in the digested tissue samples and associated quality control samples were determined by FAAS, GFAAS (Zeeman or Continuum background correction), CVAAS, or ICP-MS (Appendix B Table B-6). The analytical methods are all based on USEPA methods described for Series 7000 (FAAS and GFAAS), Series 7470 (CVAAS), and Series 6010A (ICP/MS) (USEPA, 1991). The metals analyzed in tissue samples and their method detection limits (MDL) are summarized in Table 2-7.

Table 2-7. Metals analyzed in tissue samples and their method detection limits (MDL). Concentrations are µg/g dry wt (parts per million). Se was not analyzed in invertebrate tissue samples; Al, Be, Co, Mn, and Tl were not analyzed in fish tissues.

Metal	MDL	Metal	MDL
Ag – Silver	0.004	Hg – Mercury	0.001
Al – Aluminum	2.3	Mn – Manganese	1.1
As – Arsenic	0.012	Ni – Nickel	0.004
Ba – Barium	0.01	Pb – Lead	0.001
Be – Beryllium	0.001	Sb – Antimony	0.001
Cd – Cadmium	0.001	Se – Selenium	0.03
Co – Cobalt	0.001	Tl – Thallium	0.001
Cr – Chromium	0.01	V – Vanadium	0.002
Cu – Copper	0.7	Zn – Zinc	0.4
Fe – Iron	2.5		

2.9 Data Reduction and Statistical Analysis of Metals and PAH Concentrations in Tissues

2.9.1 Data Processing

Data processing and analysis tasks were performed using the SAS® Software System (version 9.1.3) in batch programming mode. SAS® is a data management, statistical, and graphical system that is used widely as the recognized standard by academic, government and medical/health industries worldwide. Data were translated from Microsoft® Excel files to SAS® data sets, and all analyses were performed within the SAS® system.

Statistical analyses were performed on subsets of the original data archived in a project-specific dataset. The subset was selected for key contaminants of concern. For example, total PAH (TPAH) was used in computations and test statistics in place of individual PAH to reduce the size of the original dataset without loss of important information. TPAH concentrations were estimated as summations of individual analytes (e.g., 40+ individual PAH or congener groups) and, where non-detected analytes were encountered, zeros were used to represent the individual analyte concentration. PAH analyte concentrations were retained for potential diagnostic analyses to aid in identification of the type and potential sources of PAH found in the tissues. The following sections provide brief descriptions of the statistical analyses performed.

2.9.2 Data Transformations

Data were transformed and/or normalized prior to analysis to meet certain statistical test assumptions. These data modifications are performed routinely in environmental investigations of chemical contamination and are used to increase accuracy and aid in statistical interpretation of data. Prior to inference testing, chemical data distributions were examined for normality using a Shapiro-Wilk test (Shapiro and Wilk 1965). Data that were not normally distributed were log-normalized and the distributions were retested for normality. Log-normalized data that failed normality retesting were rank transformed prior to hypothesis testing.

2.9.3 Statistical Analysis Design

TPAH and metals concentrations were analyzed in tissues of several species of fish, indigenous marine invertebrates (clams, amphipods, isopods), and deployed mussels each summer for three years (2004, 2005, 2006). Fish were captured at Northstar and Liberty during all three years, and at Tigvariak Island only in 2004; Tigvariak Island was used in 2004 as a reference site. Table 2-8 lists the frequency of sampling and locations where fish, transplanted mussels, and indigenous invertebrate samples were collected and analyzed for metals and PAH.

As a consequence of capture success, sampling frequency, and location, the statistical design for fish supports a variety of null hypotheses that can be addressed as:

- There is no significant difference in analyte concentration in fish tissues (all species combined) between the three sampling locations in 2004;
- There is no significant difference in analyte concentration in fish tissues (all species combined) between Northstar and Liberty in 2004, 2005, and 2006;

Table 2-8. Frequency and location of fish and invertebrate samples, analyzed for metals and PAH, and subjected to hypothesis testing.

Organism	Analyte Group	Liberty	Northstar	Tigvariak Island	BSMP	Prudhoe Bay	SDI	TimeZero Reference	West Dock
Fish	Metals	2004 2005 2006	2004 2005 2006	2004					
	PAH	2004 2005 2006	2004 2005 2006	2004					
Mussels	Metals	2004 2005 2006	2004 2005 2006		2004 2005 2006	2005	2006	2004 2005 2006	2006
	PAH	2004 2005 2006	2004 2005 2006		2004 2005 2006	2005	2006	2004 2005 2006	2006
Amphipods	Metals	2004 2005	2004 2005 2006		2004 2005 2006				2006
	PAH	2004 2005 2006	2004 2005 2006		2004 2005 2006				2006
Clams	Metals	2004 2005 2006			2004 2005				
	PAH	2004 2006			2004 2005				
Isopods	Metals	2005			2005				
	PAH	2005 2006	2006		2005 2006				2006

- There is no significant difference in analyte concentration in different species of fish among the three sampling locations in 2004;
- There is no significant difference in analyte concentration in different species of fish from Northstar and Liberty in 2004, 2005, and 2006.

Similar null hypotheses can be proposed for the deployed mussels and indigenous marine invertebrates and statistical comparisons follow these examples.

Dependent variables examined are dry weight tissue concentrations of Total PAH (TPAH) and 13 metals. Metal concentrations reported as less than the method detection limit (MDL) are assigned a value of ½ the MDL for subsequent statistical evaluations. All target PAH analytes are summed to create a TPAH. Non-detected analytes used in the generation of TPAH are given a value of zero and, when all analytes in a given group are less than the MDL, ½ of the largest MDL for the group is used to represent the TPAH. It is useful to note that the total number of fish captured varied greatly among locations and sampling events. Additionally, there is substantial variability in the fish species composition over time and space, limiting the number fish species that can be comfortably used in statistical comparisons. Isopods and mysids were collected at only a few sampling locations, limiting ability to perform statistical comparisons.

2.9.4 Inference Testing

The experimental design is not completely balanced and thus, straightforward hypothesis testing using an Analysis of Variance (ANOVA) is not appropriate. The General Linear Model (GLM), an application of the analysis of variance (ANOVA) statistic, was used to elucidate significant differences in chemical concentrations while addressing the unbalanced nature of the data. In the GLM, a continuous response, or dependent variable (e.g., TPAH) is measured under experimental conditions identified by classification, or independent variables (e.g., location and time). The variation in the response is explained as being due to effects in the classification, with random error accounting for the remaining variation (Searle 1971). The experimental design permitted the implementation of a one-way GLM testing location effects in 2004, and a two-way GLM testing location and temporal effects for several locations and sampling periods. Prior to GLM processing, data were examined for normality using a Shaprio-Wilk test (Shapiro and Wilk, 1965; Snedecor and Cochran 1980). Data that were non-normally distributed were subjected to standard transformation/standardization procedures (e.g., log of analyte tissue concentration). Contaminant data that could not be transformed into a normal distribution were examined using a non-parametric GLM technique (Kruskal-Wallis). Non-normal data were assigned rankings, based on concentration of contaminant and the ranks were processed using the GLM procedure. Each contaminant was examined for significant differences using the following logic. Normally distributed data and non-normally distributed data successfully normalized were processed using the GLM. Data that could not be normalized were examined as ranked data under the same procedure.

Tissue concentrations found to be significantly different at the $p <= 0.05$ level were further examined with a Duncan's multiple range test (Duncan, 1975; Miller, 1981). The Duncan's test is a result-guided test that compares the treatment (effects) means, while controlling the comparison-wise error rate, and was used to differentiate significant differences ($\alpha = 0.05$) between treatment groups (e.g., location, or year).

2.10 Bile Fluorescing Aromatic Compounds (FACs)

Bile samples were collected from several species of fish collected in 2004 and 2005, but not 2006. The Geochemical and Environmental Research Group (GERG) at Texas A & M University performed the analysis of bile fluorescing aromatic compounds (FACs) in fish. The analyses were performed under the direction of research associate Stephen Sweet.

2.10.1 Fish Bile Samples

Fifty-six (56) fish bile samples were sent to GERG for analysis in the fall of 2004 (Appendix A Table A-14). The samples included 22 bile samples from mixed species of fish collected at Northstar, 19 bile samples from mixed species of fish from Liberty, and 15 bile samples from mixed species of fish from Tigvariak Island. All the samples were received frozen and intact. A majority of the samples (37 of 56) contained a total bile volume of at least 50 µl. Nineteen (19) samples had a total volume less that 50 µl, requiring dilution with a 0.85% sodium chloride (NaCl) solution to achieve a final volume of 50 µl. Appendix A Table A-14 summarizes the sample inventory, volume of each sample, and dilution factor for each sample. Six samples contained less than 10 µl of total sample volume and were analyzed in addition to the 50 requested samples for PAH metabolites and total protein.

Thirty (30) samples were sent to GERG in the fall of 2005 for analysis (Appendix A Table A15). The samples included 15 samples each from Northstar and Liberty of bile from mixed species of fish. The majority of the samples in the sample delivery group (27 of 30) contained a total bile volume of at least 50 µl. Three (3) samples had a total volume less that 50 µl, which required the dilution (using 0.85% NaCl solution) of the samples to achieve a final volume of approximately 60 µl. Appendix A Table A-15 summarizes the sample inventory, volume for each sample, and dilution factor for each of the samples.

2.10.2 Sample Analysis

The analysis of the fish bile samples for PAH metabolites was conducted in accordance with GERG SOP 0302, Revision 1 and all data were reviewed for quality. FAC concentrations were quantified by HPLC- fluorescence spectroscopy, with the detector excitation and emission wavelengths optimized for separate analysis of naphthalene-equivalent, phenanthrene-equivalent, and benzo(a)pyrene-equivalent metabolites. Total protein concentration was determined by the modified Lowry method (colorimetric assay) using a spectrophotometer.

The PAH-equivalent metabolite concentrations were reported in µg/ml or µg/g, assuming the density of the bile is 1 g/ml. Method detection limits (MDL) were approximately 0.6, 0.1, and 0.05 µg/ml (or µg/g) for naphthalene, phenanthrene, and benzo(a)pyrene metabolites, respectively. PAH metabolite concentrations also were normalized to protein concentration and reported as ng metabolites/µg bile protein. Additional details relevant to the analytical methods are presented in Appendix B.

2.11 Cytochrome P4501A (CYP1A) Analysis

2.11.1 Sample Preparation and Analysis

Immunohistochemical staining of fish tissues for cytochrome P450 1A (CYP1A) activity was performed at Woods Hole Oceanographic Institution under the direction of Dr. John Stegeman. Fish samples collected in 2004 and 2005 for CYP1A analysis by immunohistochemistry are summarized in Appendix A Tables A-16 and A-17. CYP1A analysis was not performed on fish samples collected in 2006. Fish tissues or whole fish (small specimens) were fixed in formalin in the field and were sent to Woods Hole Oceanographic Institution (WHOI) where they were sub-sectioned into histology cassettes for embedding. In some cases, whole fish taken in the field were further dissected and recognizable tissues were removed and sectioned into cassettes. With some small animals, the entire peritoneal cavity and head were split along the midline axis and arranged so that both split faces would be at the cut surface of the paraffin block. Not every tissue was present in every sample. Sections of tissues in cassettes were maintained in 10% neutral buffered formalin, embedded in paraffin, sectioned and analyzed immunohistochemically for the presence of CYP1A.

Embedded tissues were analyzed immunohistochemically for the presence of CYP1A by the same methods used during ANIMIDA (Smolowitz et al., 1991). Briefly, serial sections of embedded tissues were incubated with monoclonal antibody MAb 1-12-3 or with nonspecific purified mouse myeloma protein (MOPC31, IgG1, Sigma). Color development was achieved using 2% 3-amino-9-ethylcarbazole and 1% hydrogen peroxide. Sections were then counterstained with Mayer's hematoxylin. Several analytical reagents were components of the Signet™ (Medford, MA) murine immunoperoxidase kit.

Slides were examined with a Zeiss Axioskop™ microscope and relative staining intensities were determined subjectively by comparing the staining of samples to that of control and highly induced scup and winter flounder liver sections included in each run. Nonspecific staining, if present, was determined by comparison with MOPC31 stained sections. Staining occurrence was scored as:

- 0-no staining (or equal to MOPC31 staining)
- 1-rare- few cells staining
- 2-many cells staining
- 3-multifocal and diffuse (all cells staining)

The intensity of staining was scored as:

- 0-none (or equal to MOPC31 staining)
- 1-mild
- 2-moderate
- 3- medium
- 4-strong
- 5-very strong

Intermediate scores also were assigned as appropriate, resulting in a more continuous response curve (e.g., 1.5 for cell occurrence greater than a few but less than many). A scaled product of staining occurrence times the staining intensity (O x I) was determined for each cell type.

Other observations were made, in addition to the CYP1A scoring, including sex (if gonad present), degree of vacuolation in liver (1 low to 5 high), presence of abnormalities in tissues scored, presence of parasites (primarily *Trichodina*) and other signs of infection. These observations are included in the raw data scoring sheets, but were not carried through data reduction or otherwise analyzed (See Appendix D3: Biological Biomarkers: Fluorescing Aromatic Compounds in Fish Bile; Cytochrome P450A1 Staining in Fish Tissues.) Pathologies noted were not confirmed by a qualified histopathologist. *Trichodina* assessment is easy and certain to identify, but the assignment of the cyst/nodular appearance of some gills to myxosporidial infection is tentative and outside the scope of this analysis. All observations of CYP1A staining and other conditions were done in a blind study without knowledge of sample identity.

2.11.2 Data Reduction and Analysis

After microscopic scoring was completed, animal staining scores were assigned using the Battelle ID to field sample/station/species ID. Species-wise one way ANOVA was performed for all species with an $n \geq 3$ occurring at 2 or more stations, and for which CYP1A staining was detectable in the analyzed tissue/cell type. Mean, standard deviations, and standard errors of the mean were determined for scaled scores for each cell type within the four species meeting the requirement for ANOVA analysis. Means were compared ad hoc by ANOVA, using the Tukey's Honestly Significant Difference (Tukey-Kramer) test statistic for unequal sample sizes. The data sorted by species was transferred and analyzed using SuperAnova™ statistical program.

2.12 Laboratory Quality Control

2.12.1 Hydrocarbon Analysis

The data quality objectives (DQOs) used for ANIMIDA Task 2 and Task 8 also were used for Task 5 of cANIMIDA. These DQOs were included in the laboratory QAPPs for the cANIMIDA program and they ensure that the data are of the quality necessary to attain the project goals. The DQOs for the PAH, S/T, and SHC analyses are summarized in Table 2-9. The types of quality control samples utilized during cANIMIDA include:

- Procedural Blanks (PB)
- Laboratory Control Spikes (LCS)
- Replicates (DUP or TRP)
- Standard Reference Material (SRM)
- North Slope Crude (NSC) control oil
- Northstar control oil (CO)

Table 2-9. Data Quality Objectives (DQOs) and criteria for organic analysis QC samples.

Sample Type	Minimum Frequency	Acceptance Criteria/Corrective Action
Initial Calibration	Prior to every instrument sequence for GC/MS analysis and as needed for GC/FID analysis	PAH and SHC: 5 point curve, %RSD < 35% for all target compounds, 90% must be < 25%. S/T: 4 point curve, %RSD < 25% for all target compounds. Instrument maintenance and recalibration.
Continuing Calibration	After every 12 samples or 16 hours, whichever is more frequent, and at end of instrument sequence	PAH and SHC: %D < 35% for all CCC target compounds; 90% must be < 25%. S/T: %D < 25% for all compounds. Instrument maintenance and recalibration.
Oil Reference Standard	With each instrument sequence (One North Slope Crude)	North Slope Crude < 35% D from laboratory mean for target compounds (use surrogate corrected values) except for compounds below the reporting limit.
Procedural Blank	One per batch	No compound to exceed 5 times the MDL unless sample amount is > 10X blank amount. Re-extract, re-analysis, and/or qualify data with a "B" if value is <5x blank concentration.[1]
Laboratory Control Sample (LCS)	One per batch	PAH and SHC: Recovery between 35 and 125% for PAH, and 45 to 125% for SHC S/T: Not applicable. Re-analysis.[1]
Instrument SRM (1491)	One per instrument sequence (PAH only)	Values must be <15% difference of true value for all certified compounds
Tissue SRM	One per batch as appropriate (PAH only)	Values must be within 30% of the true value on average for all compounds, not to exceed 35% of true value for more than 30% of the compounds
Duplicate Analysis	One per batch	RPD < 30% for all compounds >10 times the MDL; Mean RPD<30% Qualify data.[1]
Surrogate Standards	Every sample	Recovery between 45 and 125% - (35% for d_8-naphthalene). Re-extract, re-analysis, and/or qualify data.[1]

[1] Project Manager will determine if re-analyses are necessary.

These types of samples are defined, as are the DQOs, in Appendix B of this report. In addition, surrogate internal stanrard (SIS) compounds were added to each sample and the recoveries monitored to assess performance. Other quality control measures included solvent and standard checks, initial calibration checks and continuing calibration verification checks. Selected samples, both sediment and tissue, from the ANIMIDA program were analyzed during the cANIMIDA program as part of a laboratory comparison study. These additional measures are further explained in Appendix B.

2.12.2 Metals Analysis

QC measures associated with metals analysis in tissue sample include balance and instrument calibration, as well as analysis of matrix spikes, analytical duplicates, standard reference materials (SRM), and procedural blanks. Sample preparation batches contained no more than 20 samples; a procedural blank, SRM, duplicate, and matrix-spike were each performed at a frequency of at least 1 per 20 field samples. DQOs for these QC measurements are provided in Table 2-10. The QC measures are further defined in Appendix B.

Table 2-10. Data Quality Objectives (DQO) and Criteria for Metals in Tissues.

Sample Type	Minimum Frequency	DQO/Acceptance Criteria
Initial Calibration	Prior to every batch of samples	3- to 5-point curve depending on the element and a blank. Standard Curve correlation coefficient r $\geq$0.999 for all analytes
Continuing Calibration	Must end every analytical sequence; for flame, repeat all standards every 5 samples; for graphite furnace and ICP/MS recheck standard after every 8 to 10 samples	%RSD <15% for all analytes
Standard Reference Materials	One per batch of 20 samples	Values must be within 20% of accepted values for >85% of the certified analytes and within 25% for Hg.
Method Blank	One per batch of 20 samples	No more than 2 analytes to exceed 5 times MDL unless analyte not detected in associated samples
Matrix Spike and Spike Method Blank	One per batch of 20 samples	%RSD 80 to 120%
Laboratory Duplicate	One per batch of 20 samples	RPD <25% for 65% of the analytes

2.12.3 Bile Fluorescing Aromatic Compounds (FACs) Analysis

Method detection limits (MDL) were approximately 0.6, 0.1, and 0.05 μg/ml (or μg/g) for naphthalene, phenanthrene, and benzo(a)pyrene metabolites, respectively. Criteria for specific QC samples (i.e. SRM, duplicate) are discussed below. The criterion for the relative percent difference (RPD) between duplicate samples are less than 25% for analytes that are above 3 times the method detection limits (MDL).

The GERG Standard Bile Reference Material (Bile Reference Standard II) was run in duplicate prior to each analytical batch. The Bile Reference Standard II is a fish bile composite with PAH metabolite concentrations of 380, 110, and 1.5 μg/ml for naphthalene-, phenanthrene-, and benzo(a)pyrene (BaP)-equivalent metabolites, respectively. The Bile Reference Standards were

run in duplicate for protein determination and reported separately for QA purposes. The QA criteria for the RPD between duplicate samples is less than 25% for the duplicate analyses and the protein content of the Bile Reference Material II must be within 25% of the average value of the reference material.

2.12.4 Cytochrome P4501A (CYP1A) Analysis

Several measures were taken to ensure the data obtained from the CYP1A analyses were valid. Internal standards were included in each staining to assure consistency and quality of a run, and to determine maximum scaled staining score (occurrence 3 X intensity 5=15) and minimum (0) staining. Duplicate slides for all samples were stained with MOPC31 to determine if nonspecific staining was present. As part of the standard Signet protocol, slides were presoaked in 3% H_2O_2 to eliminate endogenous peroxidase activity. Scoring of samples was performed blind. Only the Battelle ID number was known during scoring. Species and station IDs were determined post scoring. The correlation of subjectively determined CYP1A immunohistochemical staining scores with the independent and nonsubjective protein immunoblotting densities of hepatic microsomes from the same livers has been demonstrated by WHOI (Woodin et al, 1997).

2.13 Data Reporting

The data collected during cANIMIDA have been compiled into the ANIMIDA database, which was delivered to Battelle by Harvard Design and Mapping (HDM) at the inception of cANIMIDA. HDM developed the database under the ANIMIDA contract and was responsible for loading all the ANIMIDA data. Battelle refined the structure of the database, where possible and was responsible for loading all the cANIMIDA data, and have delivered the refined and combined ANIMIDA/cANIMIDA database to MMS.

Upon the completion of chemical analyses of fish and invertebrate tissues, the data were summarized and statistically analyzed. The data from the 2004 summer field season were discussed at the Information Transfer Meeting (ITM) in Anchorage, Alaska, in February 2005; an annual report containing only the 2004 data was not submitted to MMS. A preliminary, interim draft report for Task 5, summarizing the 2004 and 2005 analytical results and initial interpretations, was submitted to MMS in February 2006. Some of the results of Task 5 were presented at American Society of Limnology and Oceanography Marine Sciences Meeting and the 11th ITM meeting in 2008. The information and interpretations presented in this Final Report supersede those previously submitted. Interpretations discussed in this report also will be incorporated in the cANIMIDA Task 1 Synthesis Report.

3.0 RESULTS

3.1 Hydrocarbons in Tissues of Marine Animals

3.1.1 Fish

3.1.1.1 Fish Species Sampled

Eight species of fish were collected one or more times in 2004, 2005, and 2006 at different locations in the study area in the Beaufort Sea for analysis of hydrocarbons and metals in whole tissues and measurement of the biomarkers of PAH exposure, cytochrome P450A mixed function oxygenase (CYP1A) activity in selected tissues and bile fluorescent aromatic compounds (FACs). CYP1A and bile FAC analyses were preformed on fish tissue samples collected in 2004 and 2005, but not 2006. The following species were analyzed for PAH, 13 metals, and PAH exposure biomarkers:

- Arctic char (*Salvelinus alpinus*)
- Arctic cisco (*Coregonus autumnalis*)
- Least cisco (*C. sardinella*)
- Broad whitefish (*C. nasus*)
- Humpback broad whitefish (*C. pidschian*)
- Arctic cod (*Boreogadus saida*)
- Arctic flounder (*Liopsetta glacialis*)
- Four horn sculpin (*Myoxocephalus quadricornis*)

Species identifications are from Thorsteinson and Wilson (2006). Fish identified as arctic char (*S. alpinus*) may be a northern form of dolly varden (*S. malma*). Arctic cisco, four horn sculpin, and least cisco were collected in most locations and years. The other five species were collected less frequently.

A total of 28 fish of 7 species were collected and selected for PAH and metals analysis in 2004 at Northstar, Liberty, and Tigvariak Island (considered a reference site) (Appendix A Table A-7). In 2005, a total of 21 fish of 5 species from 2 stations (Stump Island near Northstar and Point Brower near Liberty) were selected for PAH and metals analysis (Appendix A Table A-10). In 2006, 20 fish of 5 species from the same two stations as in 2005 were selected for analysis (Appendix A Table A-13).

3.1.1.2 PAH in Fish Tissues

Summer 2004

A total of 97 fish were collected in 2004 (Appendix A Table A-1) and 28 samples 7 species of fish were selected for PAH analysis (Table 3-1). Concentrations of TPAH varied widely in whole fish collected from the vicinity of Liberty, Northstar, and the reference area at Tigvariak Island (Table 3-1, Figure 3-1). One arctic cod sample from Northstar contained 154.6 ng/g dry wt (parts per billion) phenanthrene and one arctic cod sample from Liberty contained 64.3 ng/g C_1-fluorenes. These values were considered anomalously high, compared to concentrations of

other PAH in the same samples and were not included in the overall averages. The overall mean TPAH concentration in all fish samples from the three sites was 39.9 ± 20.75 ng/g dry wt.

Table 3-1. Summary of total PAH concentrations in fish collected in 2004. Concentrations are ng/g dry wt (parts per billion).

Location	Species	Number of Samples	Minimum	Maximum	Mean	Standard Deviation
Liberty	Arctic Char	1	75.44	75.44	75.44	
	Arctic Cisco	2	15.16	40.39	27.78	17.84
	Arctic Cod	2 (7 fish)	83.55	92.05	87.80	6.01
	Broad Whitefish	2	43.83	46.2	45.02	1.68
	Least Cisco	2	19.39	42.52	30.96	16.36
	Four Horn Sculpin	2	32.29	35.04	33.67	1.94
Northstar	Arctic Cisco	3	24.51	36.13	30.87	5.89
	Arctic Cod	2 (6 fish)	33.8	70.12	51.96	25.68
	Least Cisco	2	20.95	25.94	23.45	3.53
	Four Horn Sculpin	2	21.64	41.48	31.56	14.03
	Arctic cod, Sculpin	1 (4 fish)	81.19	81.19	81.19	
Tigvariak Island	Arctic Char	2	20.5	31.59	26.05	7.84
	Arctic Flounder	2	29.65	34.81	32.23	3.65
	Least Cisco	2	17.28	35.45	26.37	12.85
	Four Horn Sculpin	2	35.49	38.77	37.13	2.32
All Samples		28	15.16	92.05	39.90	20.75

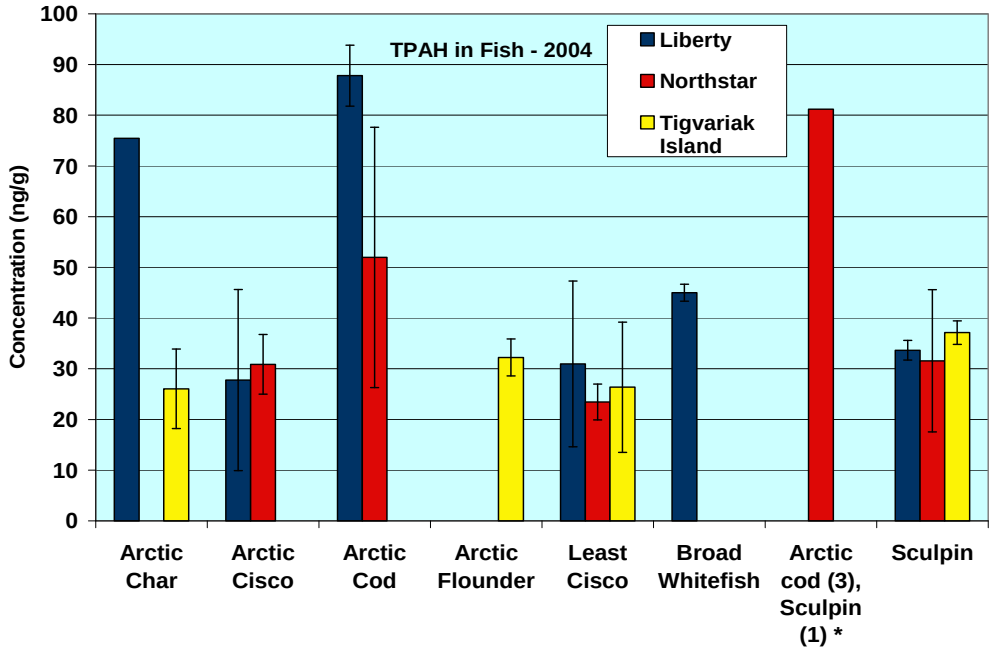


Figure 3-1. Mean TPAH concentrations in fish collected in 2004. Samples containing anomalous concentrations of phenanthrene and C1-fluorene were not included.

Highest mean TPAH concentrations were in tissues of arctic cod from Liberty and Northstar. No arctic cod were collected at Tigvariak Island. Mean tissue TPAH concentrations in fish from Tigvariak Island (considered a reference site) (36.08 ± 7.7 ng/g) were lower than mean TPAH concentrations in fish from Liberty and Northstar (47.8 ± 25.3 and 38.8 ± 20.7 ng/g, respectively), but the differences were not statistically significant. There also was not a significant difference in TPAH concentration in any of the fish species analyzed among the three sampling locations.

The PAH composition and relative concentrations of individual PAH in fish tissues varied among fish species and sampling sites. The most abundant PAH group in all fish from all locations was the naphthalenes (Figure 3-2). C_2- or C_3-naphthalenes were most abundant in most fish tissue samples, reflecting the greater bioaccumulation and slower release of alkylnaphthalenes than naphthalene. Phenanthrene and alkylphenanthrenes often were the second most abundant PAH group in fish tissues; however, some fish contained little or no phenanthrene. Fluorenes also were abundant in some fish. The abundance of low molecular weight, two- and three-ring PAH, including alkyl homologues, is consistent with a petroleum source for the PAH in fish tissues. However, some fish also contained traces of high molecular weight, predominantly pyrogenic PAH.

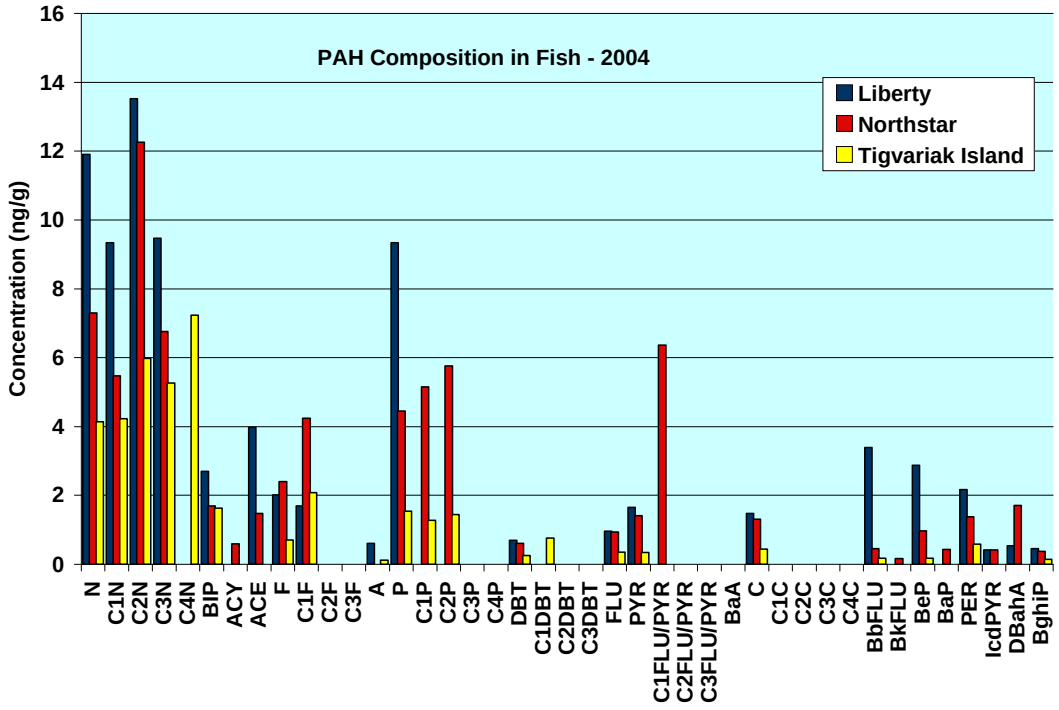


Figure 3-2. Profiles of mean concentrations of individual PAH in tissues of fish from Tigvariak (reference), Northstar, and Liberty collected in 2004. Samples containing anomalous concentrations of phenanthrene or C1-fluorene were not included.

The mean concentration for each PAH in all fish from the three regions, Tigvariak, Northstar, and Liberty, was plotted to aid in evaluating possible sources of PAH in the fish tissues (Figure 3-2). Mean PAH profiles were roughly similar in fish from the three locations, suggesting that sources of the PAH in the fish tissues were similar. Low molecular weight alkyl naphthalenes and fluorenes were the most abundant PAH in most fish tissue samples. There were low (usually less than 5 ng/g) concentrations of higher molecular weight PAH in fish from all locations. These results suggest that the PAH in the tissues of fish from all three locations were primarily from petrogenic sources, with a minor addition of pyrogenic (combustion sourced) PAH.

Summer 2005

Only 35 fish were collected in 2005, 17 from Northstar and 18 from Liberty (Appendix A Table A-2). A total of 21 fish tissue samples were analyzed for PAH (Table 3-2). Three species were collected and analyzed from each site: arctic cisco, arctic flounder, and humpback broad whitefish at Liberty and arctic char, four horn sculpin, and humpback broad whitefish at Northstar.

Table 3-2. Summary of total PAH concentrations in fish collected in 2005. Concentrations are ng/g dry wt (parts per billion).

Location	Species	Number of samples	Minimum	Maximum	Mean	Standard Deviation
Point Brower (Liberty)	Arctic Cisco	3	4.45	21.71	13.35	8.64
	Arctic Flounder	2	8.41	17.13	12.77	6.17
	Humpback Broad Whitefish	5	2.92	14.64	7.13	4.92
Northstar	Arctic Char	3	1.5	4.87	2.85	1.78
	Four Horn Sculpin	2	6.5	12.92	9.71	4.54
	Humpback Broad Whitefish	5	5.24	23.62	13.28	6.93
All Samples		20	1.5	23.62	5.16	6.49

Concentrations of TPAH were quite low in all fish, ranging from 1.5 ng/g dry wt TPAH in an arctic char to 23.6 ng/g in a humpback broad whitefish, both from Northstar. Mean TPAH concentrations ranged from 2.85 ng/g in Northstar arctic char to 13.35 ng/g in Liberty arctic cisco (Table 3-2, Figure 3-3). Mean concentrations of TPAH in humpback broad whitefish, the only species common to both collection sites, were 7.13 ± 4.92 ng/g at Liberty and 13.28 ± 6.93 ng/g at Northstar, not significantly different because of the large standard deviations. The mean concentration of TPAH in all fish collected at both sites in 2005 was 5.16 ± 6.49 ng/g.

Naphthalenes were the most abundant PAH in most fish tissue samples, and many samples also contained lesser amounts of phenanthrene, benzo(e)pyrene, and perylene (Figure 3-4). Fish captured from the Northstar area also contained detectable levels of fluorene, fluoranthene, pyrene, chrysene, benzo(a)pyrene, and benzo(g,h,i)perylene. These results indicate that the PAH in fish tissue from both areas were primarily from petrogenic sources, with a minor addition of pyrogenic and biogenic (i.e., perylene) PAH.

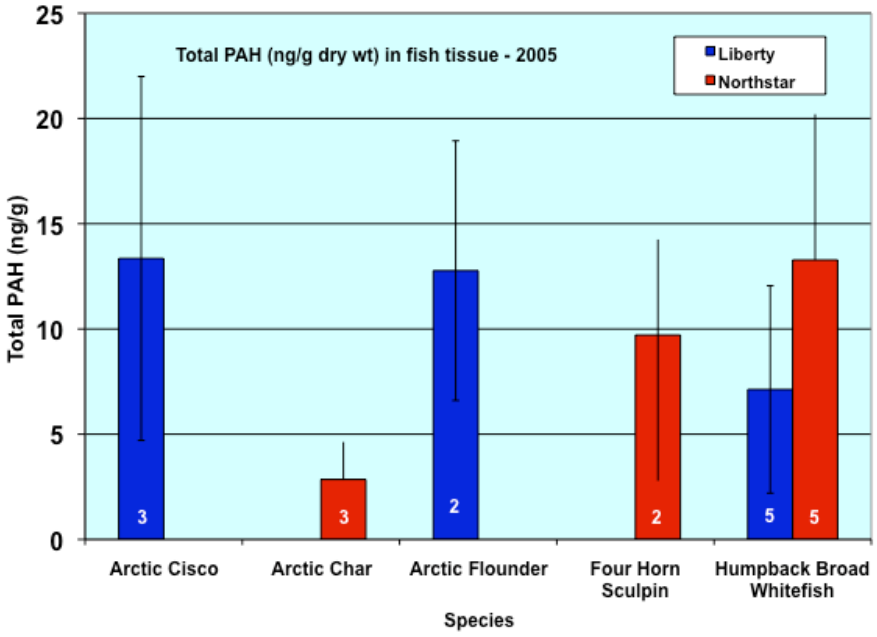


Figure 3-3. Mean concentrations of total PAH (TPAH) in fish collected from Northstar and Liberty in 2005. Vertical bars are ± standard deviation. Number of replicates is given in bars.

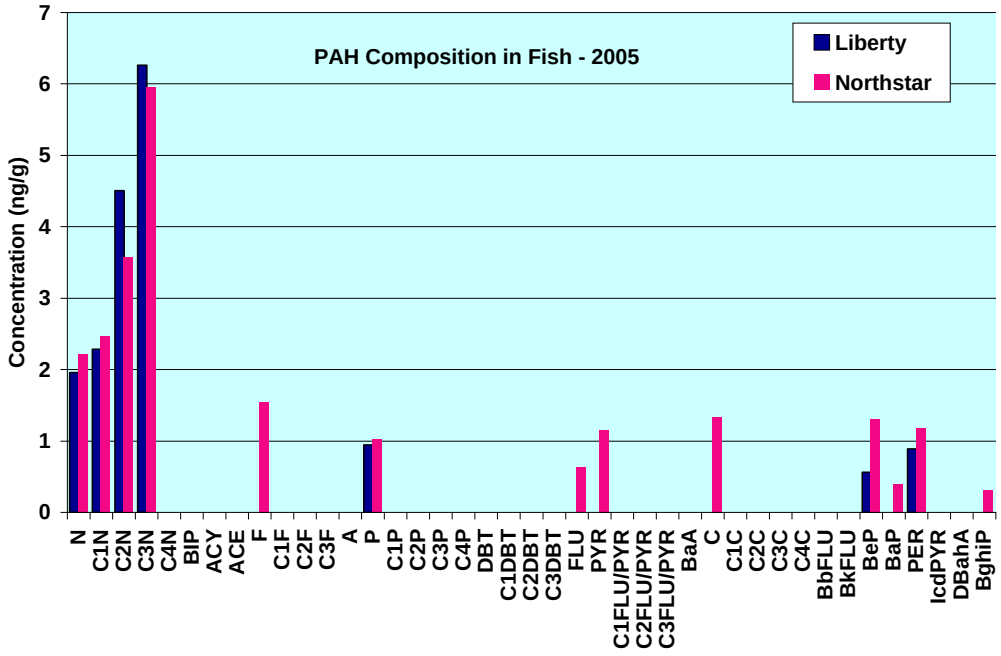


Figure 3-4. Profiles of mean concentrations of individual PAH in tissues of fish from Northstar and Liberty collected in 2005.

Summer 2006

Thirty-nine (39) fish were collected in 2006, 19 from Northstar and 20 from Liberty (Appendix A Table A-3). A total of 20 tissue samples from 5 species collected in 2006 were analyzed (Table 3-3). Samples of arctic flounder, four horn sculpin, and least cisco from each site were analyzed. Humpback broad whitefish and broad whitefish from the Liberty area also were analyzed.

Table 3-3. Summary of total PAH concentrations in fish collected in 2006. Concentrations are ng/g dry wt (parts per billion).

Location	Species	Number of Samples	Minimum	Maximum	Mean	Standard Deviation
Liberty	Arctic Flounder	1	15.73	15.73	NA	NA
	Four Horn Sculpin	3	18.92	36.49	26.86	8.9
	Broad Whitefish	1	21.86	21.86	NA	NA
	Humpback Broad Whitefish	3	15.99	30.71	25.31	8.11
	Least Cisco	3	24.01	28.86	26.38	2.43
Northstar	Arctic Flounder	1	53.15	53.15	NA	NA
	Four Horn Sculpin	2	44.63	63.13	56.93	10.66
	Least Cisco	6	27.22	59.02	47.57	17.67
All Samples		20	15.73	63.13	35.55	16.52

The mean TPAH concentration in all fish collected from Northstar and Liberty in 2006 was 35.55 ± 16.52 ng/g dry wt. (Table 3-3). TPAH concentrations in fish tissue ranged from 15.7 ng/g in an arctic flounder collected near Liberty to 63 ng/g in a four horn sculpin captured near Northstar. The highest mean concentration of TPAH was 56.9 ng/g in four horn sculpin collected from the Northstar area (Table 3-3, Figure 3-5). The TPAH concentration (56.9 ± 10.7 ng/g) in four horn sculpin collected at Northstar in 2006 was significantly higher than the TPAH concentration (26.8 ± 8.9 ng/g) in the same species from Liberty ($p = 0.02$). There were no other significant differences in TPAH concentration in the other species of fish or for all species combined collected at Northstar and Liberty in 2005 and 2006.

Naphthalenes and phenanthrenes were the most abundant PAH in most fish tissue samples; many samples also contained lesser amounts of biphenyl, acenaphthene, fluorene and C1-fluorene, dibenzothiophene and C1-dibenzothiophene, fluoranthene, pyrene, chrysene, benzo(e)pyrene, indeno(c,d)pyrene, and benzo(g,h,i)perylene (Figure 3-6). Fish captured from the Northstar area also contained detectable levels of C3-fluorenes, C2-dibenzothiophenes, C1-fluoranthenes/ pyrenes, benzo(b)fluoranthene, benzo(a)pyrene, and perylene. As in the previous two years, the PAH distribution in fish tissue is indicative of a petrogenic source with minor contributions of pyrogenic and biogenic PAH.

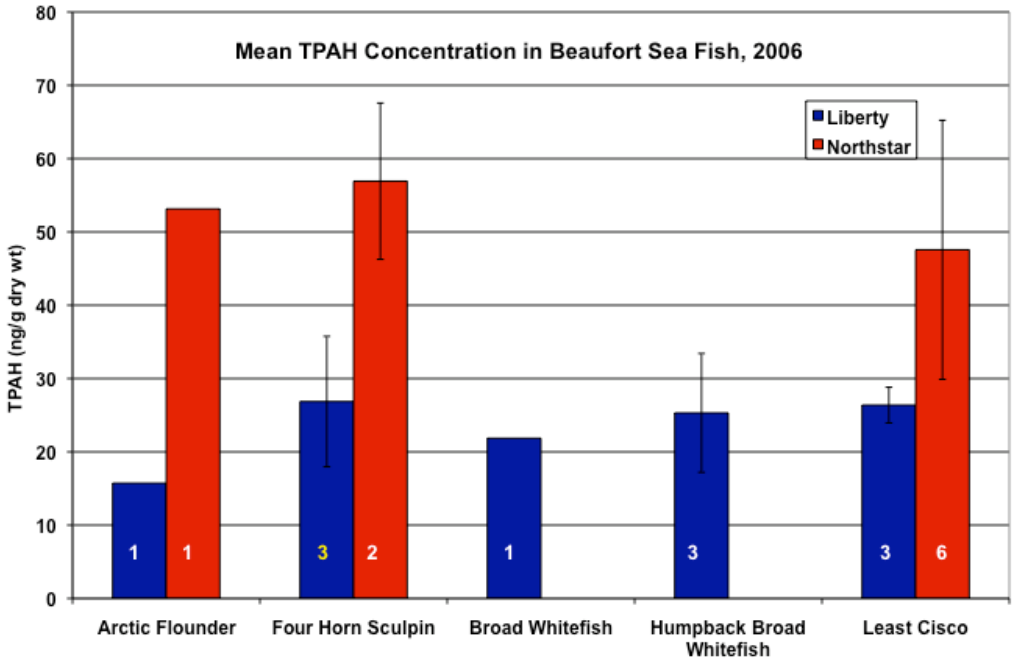


Figure 3-5. Mean concentrations of TPAH in fish collected at Northstar and Liberty in 2006. Vertical bars are ± standard deviation. Number of replicates is given in bars.

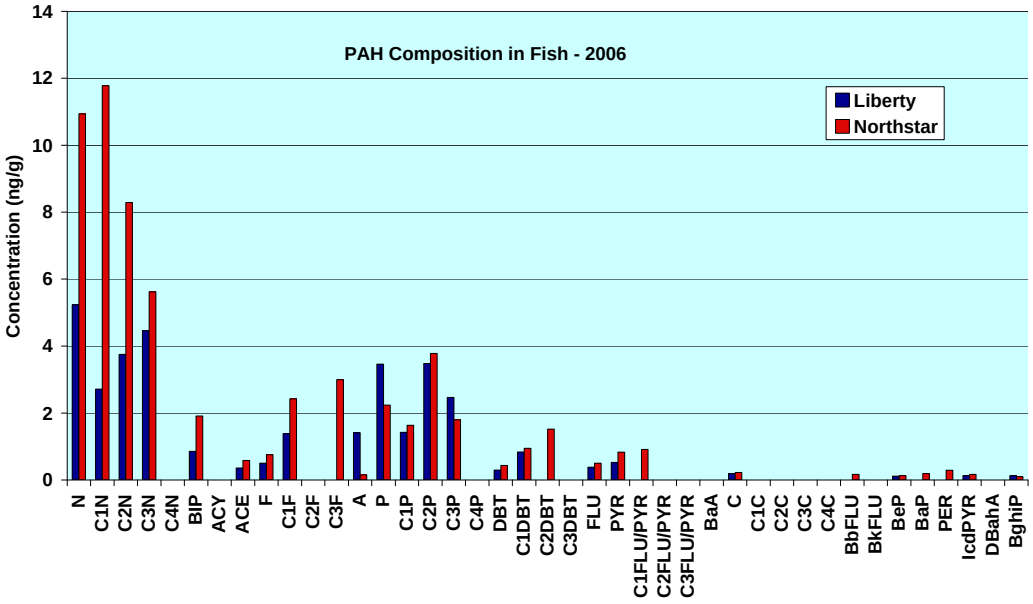


Figure 3-6. Profiles of mean concentrations of individual PAH in tissues of fish from Northstar and Liberty collected in 2006.

The relative abundance of naphthalene and alkylnaphthalenes in the 2004 and 2006 samples suggests either a light to medium refined product as the source of fish tissue PAH, or that the fish are able to metabolize and excrete higher molecular weight PAH more rapidly than low molecular weight PAH. Northstar crude oil is a light crude oil (ºAPI = 40.8 @ 15ºC); the most abundant PAH are alkylnaphthalenes, the concentrations of which are nearly five times higher than those of the alkylphenanthrenes. Thus, some of the PAH in the fish tissues could be from Northstar crude oil. However, more likely sources include refined products (e.g., light and middle distillate fuels) and coastal peat (Steinhauer and Boehm, 1992).

The mean TPAH concentrations in fish tissue in the cANIMIDA area for all three years are presented in Figure 3-7. The overall mean concentration of TPAH in all fish tissue samples collected and analyzed in 2005 was significantly lower ($p < 0.0001$) than the overall mean for all fish collected and analyzed in 2004; the mean fish tissue TPAH concentration also was significantly lower ($p<0.0001$) in all fish combined in 2005 than in 2006 (Figure 3-7). There were no significant differences in fish tissue TPAH concentrations between Northstar and Liberty in 2004 and 2005; however, fish collected at Northstar in 2006 contained a significantly higher mean TPAH concentration than fish collected at Liberty in the same year ($p<0.0001$).

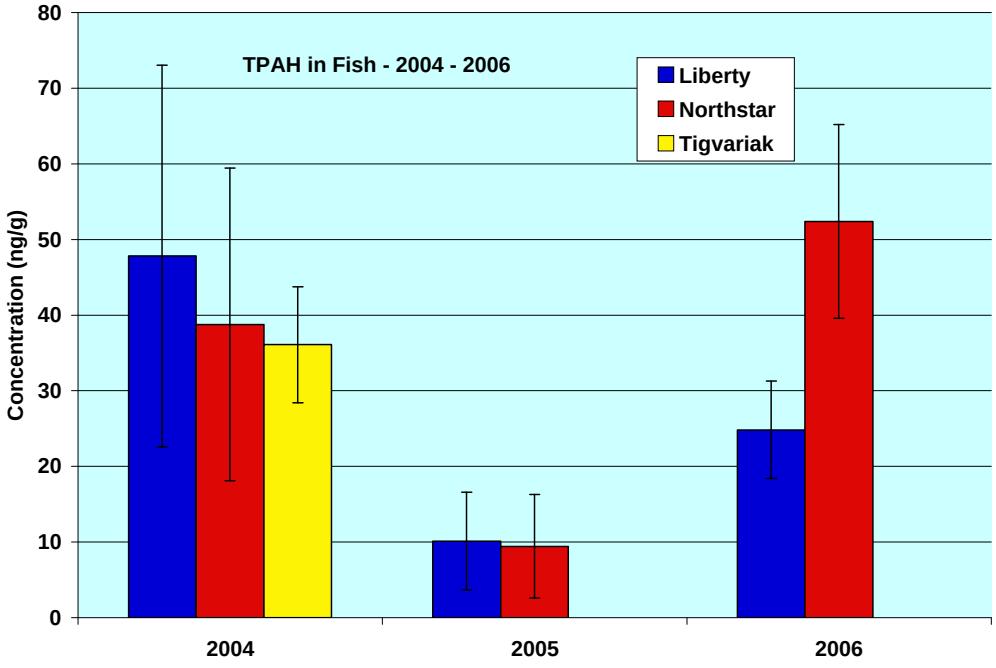


Figure 3-7. TPAH concentrations in fish from Liberty, Northstar, and Tigvariak in 2004, 2005, and 2006.

The drop in TPAH concentrations between 2004 and 2005 was investigated further during the analysis of the 2006 samples to determine whether this drop was due to an actual decrease in TPAH in the fish tissues or was related to sample processing and analysis. Two samples that had been analyzed previously, one from 2004 and one from 2005, were selected to undergo re-extraction and re-analysis. These results were then compared to the original analyses and it was determined that the PAH data for fish tissue from 2005 may have a slight low bias; some some PAH that were not detected during the initial analysis of 2005 samples that were detected near the method detection limits during the subsequent analysis. However, the reanalysis generated similar TPAH concentrations as the original analysis, indicating that for the most part the measured concentrations accurately reflect the field sample concentrations. This comparison is discussed in greater detail in Appendix C.

3.1.1.3 Comparison to PAH in Fish from ANIMIDA (2001)

Five species of fish were collected and analyzed for PAH in 2001 from 5 locations in the Beaufort Sea as part of Task 8 of the ANIMIDA program (Spies et al., 2003). The 5 species included arctic cod, arctic cisco humback broad whitefish, broad whitefish, and four horn sculpin, all collected and analyzed at least once in Task 5 of the cANIMIDA program. The sampling sites included a site near Northstar, Stump Island (SIS) west of West Dock, Point Brower (PB) east of the Endicott Causeway, a site in the Liberty prospect, and Bullen Point west of Tigvariak Island. Bullen Point was considered a reference site. Stump Island was grouped with Northstar stations and Point Brower was grouped with Liberty stations in the analysis of the cANIMIDA Task 5 data; the same grouping will be done in the summary of the ANIMIDA Task 8 data.

Spies et al. (2003) reported tissue PAH residue data in wet weight and presented it as lipid-normalized wet weight data. The average % water in fish tissues was about 75%, so the wet-weight data were converted to dry weight by multiplying by 4 so the 2001 data could be compared to the fish residue data from 2004, 2005, and 2006. Mean TPAH concentrations in fish collected at Northstar and Liberty in 2001 are 50.0 ± 57.0 and 64.1 ± 65.9 ng/g dry wt, respectively (Table 3-4), similar to the mean TPAH concentration in fish collected in 2001 from the reference site at Bullen Point: 59.5 ± 49.7 ng/g (n = 19). Mean TPAH concentrations in fish collected in 2001 were higher than mean TPAH concentrations in fish collected at Northstar and Liberty in 2004, 2005, and 2006 (Table 3-4). The high standard deviations in TPAH concentrations in fish collected in 2001 may be related to the large numbers of fish samples analyzed in 2001, compared to 2004 through 2006.

Table 3-4. Comparison of mean (plus standard deviation) TPAH concentrations in tissues of fish collected at Northstar and Liberty in 2001 (ANIMIDA) and 2004, 2005, and 2006 (cANIMIDA). n = number of samples analyzed. Concentrations are ng/g dry wt.

Year	Northstar		Liberty	
	n	Mean (SD)	n	Mean (SD)
2001	43	50.0 (± 57.0)	37	64.1 (± 65.9)
2004	10	38.8 (± 20.7)	11	47.8 (± 25.3)
2005	10	9.44 (± 6.85)	10	10.1 (± 6.45)
2006	7	52.4 (± 12.8)	11	24.8 (± 6.40)

There are no consistent temporal patterns in TPAH concentrations in fish from either Northstar or Liberty. The mean TPAH concentrations in fish from Northstar are essentially the same in 2001 and 2006, with lower mean concentrations in 2004 and 2005. The mean TPAH concentration in fish from Liberty is highest in 2006 and declines to a low in 2005.

There were no statistically significant differences in mean TPAH or low molecular weight PAH concentrations in fish collected in 2001 from the 5 sampling sites (Spies et al., 2003). High molecular weight PAH concentrations were higher in fish, particularly four horn sculpin, from Stump Island (grouped with Northstar in this analysis) near West Dock. PAH concentrations were not higher in tissues of four horn sculpin than in tissues of other fish species collected at Northstar and Liberty in 2004, 2005, and 2006 (Tables 3-1, 3-2, and 3-3). The PAH assemblage in all fish samples from all 5 years was consistent with a primarily petrogenic source with a small contribution of pyrogenic high molecular weight PAH. The fish tissue PAH residue data for 2001 through 2006 also are consistent with chronic, low-level exposure to PAH in water, sediments, and food throughout the study area, with no clear point source of bioavailable PAH in the coastal environment.

3.1.1.4 Cytochrome P4501A (CYP1A) in Fish Tissues

Summer 2004

Only 4 species occurred with an abundance of three or more at more than one site, allowing results of CPY1A assays to be compared by ANOVA; arctic cisco, least cisco, arctic cod and four horn sculpin. Only arctic cisco and least cisco exhibited significant differences in mean CYP1A staining among the 3 sites, and only for hepatocytes. For both species, CYP1A staining was observed in fish from Liberty (Point Brower) but not in fish of the same species from Northstar or Tigvariak (for least cisco) (Table 3-5). All other cell types in the arctic cisco and all the cell types in all other species exhibited no staining or light staining and no significant site related differences among staining scores. The highest mean scaled score for CYP1A staining of 2.0 ± 0.9 was in hepatocytes of Arctic Cisco from Liberty; this is a very low relative score, as compared to levels seen in the highly induced positive control fish livers (scaled score =15). CYP1A staining, when present, was observed primarily in hepatocytes, gill pillar cells, kidney tubules and gut epithelium. No significant station related differences were seen for mean CYP1A staining in gill pillar cells, kidney tubules, or gut epithelium. The highest individual staining (scaled score = 4.5) was seen in hepatocytes of Four Horn Sculpin from Tigvariak Island, although the mean hepatocyte staining for that station (1.5 ± 1.87) was not significantly different from the staining seen at the Liberty site (0.5 ± 1.2).

Table 3-5. Immunohistochemical staining for CYP1A in tissues of fish collected in 2004 at three Beaufort Sea locations. Scaled score range is 0 – 15. n = number of samples.

Fish Species	Station	Tissue	CYP1A Staining		Sig. Diff.
			n	Mean Scaled Score ± SD	
Arctic cisco	Liberty	Hepatocyte	3	2.00 ± 0.87	Liberty > Northstar
	Northstar	Hepatocyte	8	0.0	
Least cisco	Liberty	Hepatocyte	5	0.60 ± 0.82	Liberty > Tigvariak, Northstar
	Northstar	Hepatocyte	8	0.0	
	Tigvariak	Hepatocyte	7	0.0	
Least cisco	Liberty	Kidney tubule	5	0.20 ± 0.45	NS
	Northstar	Kidney tubule	8	0.0	
	Tigvariak	Kidney tubule	7	0.21 ± 0.39	
Arctic cod	Northstar	Hepatocyte	4	0.0	NS
	Liberty	Hepatcoyte	3	0.0	
	Tigvariak	Hepatocyte	6	0.17 ± 0.41	
Arctic cod	Northstar	Gill pillar cells	4	0.0	NS
	Liberty	Gill pillar cells	2	0.50 ± 0.71	
	Tigvariak	Gill pillar cells	6	0.25 ± 0.42	
Arctic cod	Northstar	Gut epithelium	4	0.0	NS
	Liberty	Gut epithelium	3	1.50 ± 2.60	
	Tigvariak	Gut epithelium	6	1.75 ± 1.54	
Four Horn Sculpin	Liberty	Hepatocyte	6	0.50 ± 1.22	NS
	Tigvariak	Hepatocyte	10	1.50 ± 1.87	
Four Horn Sculpin	Liberty	Gill pillar cells	6	0.0	NS
	Tigvariak	Gill pillar cells	10	0.70 ± 1.16	
Four Horn Sculpin	Liberty	Kidney tubule	5	0.0	NS
	Tigvariak	Kidney tubule	7	0.21 ± 0.57	

NS, no significant difference.

Gill pillar cells of a single arctic char collected from Tigvariak Island (not included in the comparisons in Table 3-5) had a scaled score of 6.0. The only other tissue in that fish with CYP1A staining was gut vascular endothelium with a scaled score of 2.0. The 2 other arctic char collected in 2004, 1 each from Tigvariak Island and Point Brower, had light staining (1.0 – 1.5) in a few tissues. There was no staining of gut vascular epithelium, gonad vascular endothelium, gill vascular endothelium, gill epithelium, and bile duct in any specimens of the 4 species of fish from the 3 sites sampled in 2004.

Summer 2005

A total of 32 fish samples collected in 2005 were stained for CYP1A activity. All samples were from Liberty and Northstar and included arctic char, arctic cisco, arctic flounder, four horn sculpin, and humpback broad whitefish. Only humpback broad whitefish were present at abundance equal to or greater than 3 in samples from both field stations. Statistical comparisons were made between the 2 sites for mean scaled CYP1A staining in hepatocytes and kidney tubules of this species. These were the only cell types in this species that exhibited staining

sufficient for statistical comparison. CYP1A staining in hepatocytes of humpback broad whitefish was significantly higher ($p \leq 0.05$) in fish from Northstar than from Point Brower (Table 3-6). Kidney tubule staining was slightly, but not significantly, higher in fish from Northstar than from Point Brower.

Hepatocyte, liver endothelium, kidney endothelium, and kidney tubule staining also was observed in arctic char, arctic cisco, and four horn sculpin. Mean scaled scores for hepatocytes, liver endothelium, and kidney tubules were higher in these 3 species than in humpback broad whitefish (Table 3-6). No staining was seen in gut epithelium or bile duct from any species at either site, and none in gill tissues, except for mild gill pillar cell staining in one arctic char from Northstar. The highest staining observed, in arctic char and arctic cisco hepatocytes (occurrence x intensity = 4.5), was moderate compared to the staining of the highly induced scup and winter flounder standards (occurrence x intensity =15). These results suggest that these 4 species of fish were bioaccumulating small amounts of CYP1A-inducing chemicals, possibly PAH, at both Liberty and Northstar. The lack of staining of gill tissues indicates that most bioaccumulation probably was from food. There was no staining in any arctic flounder tissues (2 fish samples); these fish apparently were exposed to lower concentrations of inducers than required to induce CYP1A activity.

Table 3-6. Immunohistochemical staining for CYP1A in tissues of fish collected in 2005 for the cANIMIDA Program. Scaled score range is 0 – 15. n = number of samples.

Fish Species	Station	Tissue	CYP1A Staining		Sig. Diff.
			n	Mean Scaled Score ± SD	
Humpback broad whitefish	Northstar	Hepatocyte	9	1.5 ± 1.3	Northstar > Liberty
	Liberty	Hepatocyte	7	0.21 ± 0.57	
Humpback broad whitefish	Northstar	Liver endothelium	9	0.11 ± 0.33	NS
	Liberty	Liver endothelium	7	0	
Humpback broad whitefish	Nortstar	Kidney tubules	9	1.6 ± 0.96	NS
	Liberty	Kidney tubules	7	1.2 ± 0.94	
Arctic char	Northstar	Hepatocyte	4	1.13 ± 2.25	---
	Northstar	Liver endothelium	4	1.88 ± 1.93	---
	Northstar	Kidney endothelium	4	0.5 ± 1.0	---
Arctic cisco	Liberty	Hepatocyte	7	1.29 ± 1.82	---
	Liberty	Kidney tubule	7	2.46 ± 1.48	---
Four horn sculpin	Northstar	Hepatocyte	2	1.5 ± 2.1	---
	Liberty	Hepatocyte	1	3.0	---

NS, no significant difference.

3.1.1.5 Comparison to CYP1A Staining in Fish from ANIMIDA (2001)

In 2001, fish tissues from 5 sites (2 sites near Northstar, 2 near Liberty, and 1 reference site east of Liberty) were stained for CYP1A activity (Spies et al., 2003). There was low to moderate staining of hepatocytes and gut mucus epithelium, but not other tissues, in most fish from all 5 sites. Hepatocyte staining scores were highest in four horn sculpin from Point Brower (near

Liberty) and Stump Island (near Northstar). Staining scores in gut mucous epithelium were highest in four horn sculpin from Point Brower and Bullen Point (reference site). Highest mean staining score was in gut mucous epithelium of four horn sculpin from Point Brower (mean score ~ 6.0). CYP1A staining intensity in arctic cod hepatocytes was correlated to concentrations of PCBs in whole fish tissues. CYP1A staining intensity in four horn sculpin gut mucus epithelium was correlated to concentrations of total low molecular weight PAH in the whole fish tissues. There were no other statistically significant correlations.

The CYP1A staining data for 2001, 2004, and 2005 are consistent in showing low-level staining in several fish tissues, with no significant trends among fish species or sampling locations. Fish throughout the ANIMIDA study area apparently are being exposed to very low levels of CYP1A-inducing chemicals, possibly of several types and sources, including petroleum PAH.

3.1.1.6 Fluorescing Aromatic Compounds (FACs) in Fish Bile

Summer 2004

Concentrations of FACs were low in most of the 56 fish bile samples analyzed in 2004 (Table 3-7; Figure 3-8). The protein-normalized metabolite concentrations have a much smaller range of variation, particularly for samples that had relatively higher naphthalene-equivalent metabolite concentrations. The protein-normalized naphthalene-equivalent concentrations ranged from 1.9 to 57 μg/mg protein. Normalized values for phenanthrene and BaP equivalents ranged from 0.14 to 77 μg/mg protein and 0.01 to 0.23 μg/mg protein, respectively (Table 3-7).

Summer 2005

Concentrations of naphthalene-equivalent and phenanthrene-equivalent bile FACs were lower in most of the 30 fish bile samples analyzed in 2005 than in those analyzed in 2004 (Table 3-8 ; Figure 3-9). Benzo-a-pyrene-equivalent concentrations in bile of 2005 were slightly higher than in bile of 2004 fish. Protein-normalized naphthalene-equivalent bile FACs ranged from 0.8 to 36.2 μg/g protein, normalized phenanthrene equivalents ranged from 0.1 to 5.2 ng/μg protein, and BaP equivalents ranged from 0.03 to 0.38 μg/g protein (Table 3-8). There was not a significant difference in concentrations of the three bile FACs concentrations in fish from Liberty and Northstar. As with the 2004 FAC data, the protein-normalized bile FAC concentrations in fish collected near Northstar and Liberty in 2005 were similar to bile FAC concentrations in fish from contaminated and clean marine areas of the Pacific Northwest (Meador et al., 2008; Myers et al., 2008).

Table 3-7. Bile Metabolite Concentrations (FACs) for fish collected during the 2004 cANIMIDA field survey.

	Parameter	Bile Metabolites (µg/g or µg/mL)			Protein Normalized Metabolite Concentration (µg/mg protein)		
		Naphthalene	Phenanthrene	Benzo(a)pyrene	Naphthalene	Phenanthrene	Benzo(a)pyrene
Liberty	Mean ± SD	53 ± 26	20 ± 46	0.19 ± 0.09	29 ± 15	9 ± 17	0.11 ± 0.05
	Range	6 - 120	0.86 - 210	0.03 - 0.35	3.0 - 57	0.5 - 77	0.01 - 0.23
Northstar	Mean ± SD	35 ± 37	5 ± 4	0.11 ± 0.11	22 ± 11	3 ± 2	0.08 ± 0.04
	Range	1 - 150	0 - 20	0.04 - 0.52	1.9 - 56	0.1 - 9	0.01 - 0.22
Tigvariak	Mean ± SD	44 ± 59	6 ± 8.6	0.13 ± 0.19	17 ± 11	2 ± 1	0.06 ± 0.05
	Range	4 - 190	1 - 31	0.03 - 0.77	3 - 44	0.7 - 5	0.01 - 0.19
All fish	Mean ± SD	43 ± 41	10 ± 28	0.15 ± 0.13	23 ± 13	5 ± 10	0.09 ± 0.05
	Range	1 - 190	0 - 210	0.03 - 0.77	2 - 57	0.1 - 77	0.01 - 0.23

Table 3-8. Bile Metabolite Concentrations (FACs) for fish collected during the 2005 cANIMIDA field survey.

Location	Parameter	Bile Metabolites (µg/g or µg/mL)			Protein Normalized Metabolite Concentration (µg/mg protein)		
		Naphthalene	Phenanthrene	Benzo(a)pyrene	Naphthalene	Phenanthrene	Benzo(a)pyrene
Liberty	Mean ± SD	36 ±24	6 ± 4	0.3 ± 0.1	15 ± 9	2.4 ± 1.5	0.16 ± 0.08
	Range	1 - 95	0.2 - 13	0.2 - 0.8	0.8 - 35.5	0.1 - 4.7	0.05 - 0.37
Northstar	Mean ± SD	46 ± 37	7 ± 5	0.2 ± 0.1	22.0 ± 7.9	3.6 ± 1.1	0.12 ± 0.08
	Range	5 - 150	1 - 23	0 - 0.4	9.4 - 36.2	2.1 - 5.2	0.03 - 0.38
All fish	Mean ± SD	41 ± 31	6 ± 5	0.3 ± 0.1	18.5 ± 8.9	3.0 ± 1.4	0.14 ± 0.08
	Range	1 - 150	0.2 - 23	0 - 0.8	0.8 - 36.2	0.1 - 5.2	0.03 - 0.38

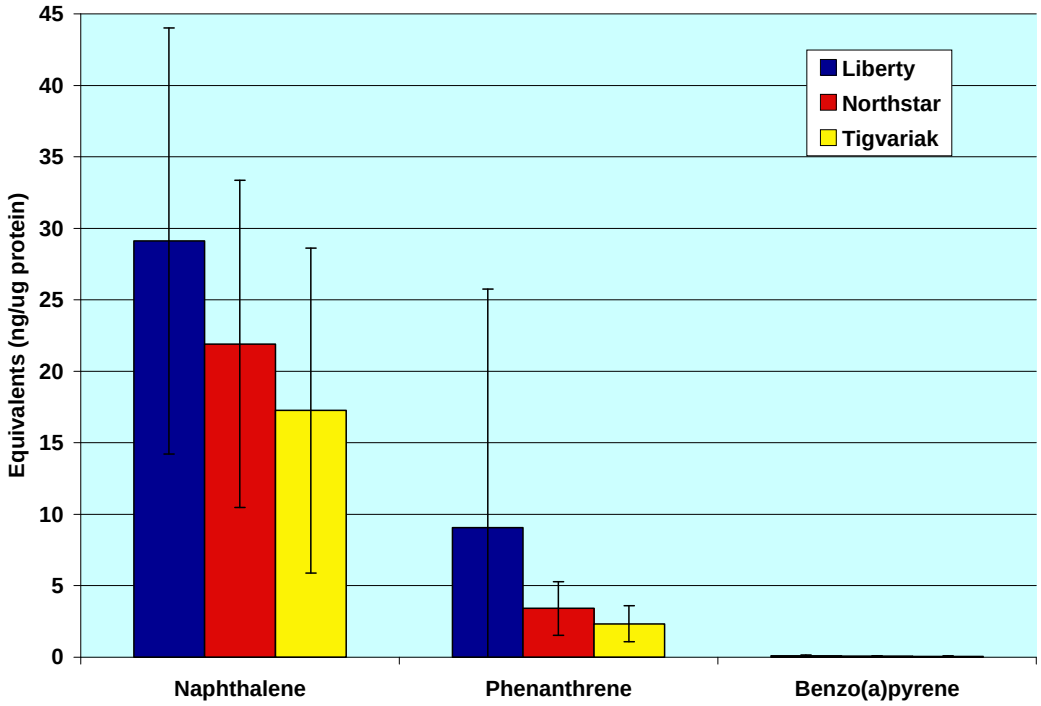


Figure 3-8. Mean and SD concentrations of fluorescent aromatic compounds (FACs) in bile of fish collected from three locations in the Beaufort Sea in 2004. Concentrations of naphthalene-, phenanthrene-, and benzo(a)pyrene-equivalent FACs are ng/µg bile protein (= µg/mg protein).

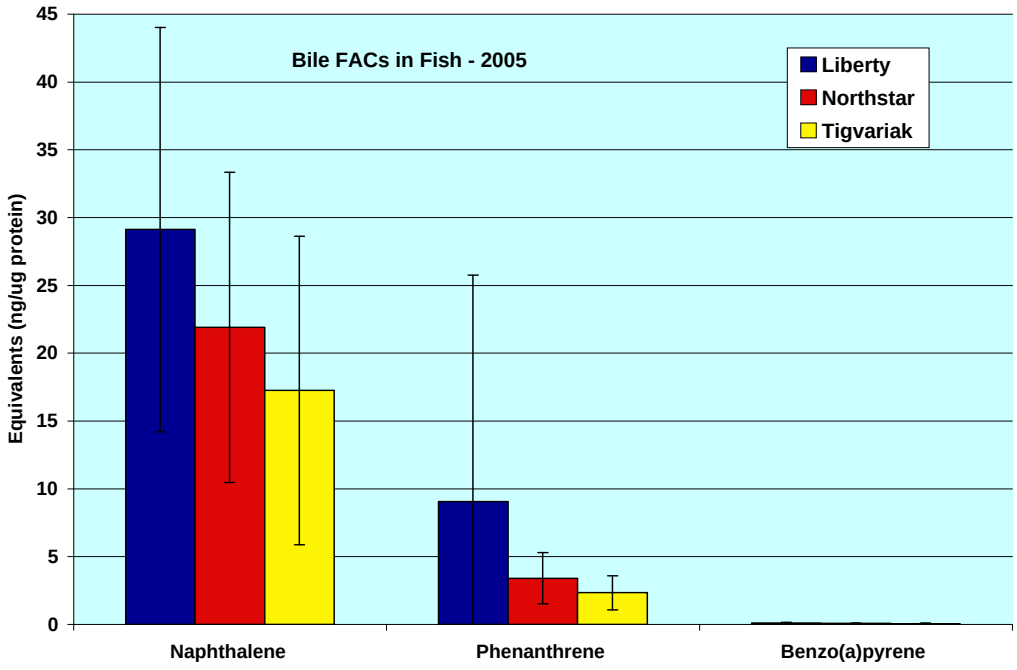


Figure 3-9. Mean and SD concentrations of fluorescent aromatic compounds (FACs) in bile of fish collected from three locations in the Beaufort Sea in 2005. Concentrations of naphthalene, phenanthrene, and benzo(a)pyrene equivalents are ng/µg bile protein (= µg/mg\ protein).

Phenanthrene-equivalent FACs are the most useful indicators of exposure to petrogenic and pyrogenic PAH (Meador et al., 2008). Phenanthrene-equivalent FAC concentrations measured in bile of Beaufort Sea fish collected in 2004 and 2005 were similar to concentrations reported by Meador et al. (2008) in 167 bile samples collected from 167 juvenile chinook salmon collected over several years from 42 locations in the Pacific Northwest (mean ± sd, 5.0 ± 6.6 µg/mg protein), a concentration that Meador et al. (2008) showed was associated with exposure to moderate concentrations of low molecular weight PAH in the water or food. Concentrations of phenanthrene-equivalent FACs in several species of fish from Prince William Sound, site of the 1989 *Exxon Valdez* oil spill, decreases from 1 to 12 µg/mg in spill path areas in 1989 to 1 to 5.5 µg/mg in spill-path and non spill-path areas of the sound in 1990 through 2001 (Hom et al., 1996; Jewett et al., 2002; Huggett et al., 2003). Thus, the concentrations of phenanthrene-equivalent FACs in Beaufort Sea fish are near the natural background concentrations.

The concentration of BaP-equivalent bile FACs in English sole from a PAH-contaminated site in Eagle Harbor, WA (Puget Sound) ranged from 0.25 to 1.30 µg/mg protein (Myers et al., 2008), higher than that observed in fish from the Beaufort Sea (Table 3-7). Thus, Beaufort Sea fish were being exposed in 2004 and 2005 low concentrations of low molecular weight, 2- and 3-ring PAH (probably mostly petrogenic) and high molecular weight 4- through 6-ring PAH (probably mostly pyrogenic).

3.1.1.7 Comparison to FAC Concentration in Fish from ANIMIDA (2001)

Phenanthrene-equivalent and BaP-equivalent bile FACs were measured in 57 samples of bile from 5 species of fish collected in 2001 at 5 locations in the Beaufort Sea as part of Task 8 of the ANIMIDA Program (Spies et al., 2003). FAC concentrations were not normalized to bile protein. Mean concentrations of phenanthrene-equivalent and BaP-equivalent FACs ranged from 0.05 to 12 µg/g and 0.02 to 0.32 µg/g, respectively, in the different fish species. The highest mean phenanthrene-equivalent FAC concentration was in arctic cod from Liberty; the highest mean BaP-equivalent FAC concentration was in arctic cisco from Stump Island (between Northstar and West Dock). Both phenanthrene-equivalent and BaP-equivalent bile FAC concentrations if Beaufort Sea fish were lower in 2001 than in 2004 and 2005. The bile FAC data for the 3 years indicate that fish in coastal waters of the Beaufort Sea development area are being exposed to low concentrations of low and high molecular weight PAH.

3.1.2 Hydrocarbons in Bivalve Mollusks and Crustaceans

3.1.2.1 Hydrocarbons in Clams, Amphipods, Isopods, and Mysids

Soft tissues of clams (*Astarte montagui* and *Cyrtodaria kurriana*) and amphipods (*Anonyx nugax*) collected at different locations in the Beaufort Sea during cANIMIDA were analyzed for PAH, saturated hydrocarbons (SHC), and sterane/triterpane biomarkers (StTr). Isopods (*Saduria sabini*) and mysids (*Mysis* sp) were collected opportunistically and were analyzed for PAH.

Summer of 2004

The mean concentrations of TPAH were similar in amphipods (68.2 ± 33.8 ng/g dry wt) and clams (95.8 ± 52.6 ng/g) collected in 2004 (Table 3-9). TPAH concentrations were quite low in the amphipod and clam tissues (39.6 to 168 ng/g dry wt), comparable to background concentrations in the same or similar species from clean marine environments.

Total resolved SHC concentrations in amphipod and clam tissues collected in 2004 were much higher than TPAH concentrations, ranging from 1446 to 35,350 ng/g dry wt (Table 3-9). Concentrations were higher in amphipods (mean 29,110 ± 4546 ng/g) than in clams (1866 ± 494 ng/g). Much of the interspecies difference was due to high concentrations of pristane in amphipod tissues (Table 3-8). Pristane represented more than 90 percent of the resolved SHC in amphipod tissues and less than 10 percent in clams.

Table 3-9. Concentrations of total PAH, total resolved SHC, pristane, and total sterane/triterpane (StTr) biomarkers in amphipods and clams collected at Northstar, Liberty, and the BSMP in 2004. Concentrations are ng/g dry wt.

Hydrocarbon	Mean	SD	Median	Minimum	Maximum
Amphipods (*Annonyx nugax*)					
Total PAH	68.2	33.8	64.2	39.6	143
Total SHC	29,110	4546	27,900	23,620	35,350
Pristane	27,516	3,991	26,295	22,875	32,955
Total StTr	8.16	4.92	8.10	1.30	17.7
Clams (*Astarte montagui* and *Cyrtoderia kurriana*)					
Total PAH	95.8	52.6	86.22	42.7	168.1
Total SHC	1866	494	1644	1446	2799
Pristane	134	54	124	80	209
Total StTr	8.54	6.21	7.10	2.92	17.06

Concentrations of total StTr were low in amphipod and clam tissues in 2004 (Table 3-9). The concentrations were about the same in the two species and the low concentrations probably reflect the low solubility and bioavailability of these refractory organic compounds. StTr occur at low concentrations in crude oils and heavy refined and residual products. Each crude oil has a unique profile of StTr. Natural nonpolar organic matter in soils and sediments also contains StTr derived from the decay of plant material. The modern StTr usually are different from the fossil StTr. These properties make StTr good biomarkers to identify possible sources of hydrocarbon mixtures in sediments and biological samples (Wang et al., 2006).

Summer of 2005

Concentrations of TPAH were lower in soft tissues of amphipods (25.4 ± 13.5 ng/g dry wt) and clams (38.4 ± 15.1 ng/g) collected in 2005 than in soft tissues of the same species collected in 2004 (68.2 ± 33.8 ng/g and 95.8 ± 52.6 ng/g, respectively) (Tables 3-9, 3-10). The interannual differences were statistically significant for amphipods but not for clams (Table 3-11). TPAH concentrations in isopods (70.2 ± 7.6 ng/g) collected in 2005 were higher than concentrations of TPAH in amphipods and clams in 2005 (Table 3-10) and lower than concentrations of TPAH in amphipods and clams collected in 2004 (Table 3-9). Again, all concentrations were quite low, in the range observed in the same or similar species from clean marine environments.

Mean total resolved SHC concentrations in amphipods (29,550 ± 17,524 ng/g) collected in 2005 were higher than concentrations in clams (1,510 ± 517 ng/g) collected in 2005 (Table 3-10). Concentrations were similar to those in amphipods and clams collected in 2004 (Table 3-9). As observed during 2004, more than 90 percent of the total resolved SHC in the amphipod tissues was pristane. Pristane represented less than 30 percent of the total SHC in clam tissues. Pristane concentrations were similar in amphipods in 2004 and 2005, but higher in clams collected in 2005 (434 ± 629 ng/g) than in those collected in 2004 (134 ± 54 ng/g) (Tables 3-9, 3-10).

Concentrations of total StTr biomarkers were similar in amphipods and clams collected in 2005 (1.84 ± 2.37 ng/g and 1.72 ± 2.98 ng/g, respectively) (Table 3-10). These concentrations were about one-quarter the total StTr concentrations measured in amphipods and clams collected in 2004 (8.16 ± 4.92 ng/g and 8.54 ± 6/21 ng/g, respectively) (Table 3-9).

Table 3-10. Concentrations of total PAH, total resolved SHC, pristane, and total (StTr) in amphipods, isopods, and clams collected at four locations in the Alaskan Beaufort Sea in 2005. Concentrations are ng/g dry wt.

Hydrocarbon	Mean	SD	Median	Minimum	Maximum
Amphipods (*Anonyx nugax*)					
Total PAH	25.4	13.5	26.4	8.3	50.0
Total SHC	29,550	17,524	24,510	7364	61,510
Pristane	27,483	16,485	22,561	6,515	57,414
Total StTr	1.84	2.37	0.79	0.00	7.41
Isopods (*Saduria sabini*)					
Total PAH	70.2	7.6	71.3	55.7	78.4
Clams (*Astarte montagui* and *Cyrtoderia kurriana*)					
Total PAH	38.4	15.1	42.3	21.8	51.2
Total SHC	1,510	517	1,273	1,029	2,228
Pristane	434	629	86	56	1,159
Total StTr	1.72	2.98	0.00	0.00	5.16

Concentrations of TPAH were significantly higher in tissues of amphipods collected from Northstar and the BSMP in 2004 than in those collected in 2005 (Table 3-11). The differences were greater for Northstar than for the BSMP. Although the mean TPAH concentration in clams collected in the BSMP in 2004 where much higher than the mean TPAH concentration in clams collected in the BSMP in 2005, the difference was not statistically significant because of the high variance in TPAH concentrations in 2004.

Table 3-11. Results of statistical analysis of differences in concentrations of TPAH in amphipods from BSMP and Northstar and in clams from BSMP in 2004 and 2005. Significant between-year differences ($p < 0.05$) are highlighted.

Location	2004			2005			p-Value
	N	Mean	S.D	N	Mean	S.D.	
Amphipods							
BSMP	3	49.5	16.7	4	23.6	9.24	0.04
Northstar	3	83.3	52.7	3	13.8	8.96	0.02
Clams							
BSMP	3	97.1	64.3	3	38.4	15.1	0.15

Summer of 2006

TPAH concentrations were similar in amphipods, isopods, and mysids, and higher in the two clam samples from Liberty in 2006 (Table 3-12). Total PAH concentrations in amphipods collected during 2006 (59.6 ± 29.8 ng/g) were similar to those measured in amphipods collected during 2004 (68.2 ± 33.8 ng/g) and higher than concentrations measured in amphipods collected in 2005 (25.4 ± 13.5 ng/g) (Tables 3-9, 3-10, 3-12). Amphipods were collected at 23 stations in 5 areas of the development area in 2006. Highest mean total PAH concentrations were in amphipods from West Dock (103 ± 19.6 ng/g) and Liberty (92.2 ± 26.6 ng/g) and lowest in amphipods from Northstar (41.3 ± 27.4 ng/g) (Table 3-13). The mean TPAH concentration in amphipods was significantly lower at Northstar than at the other 4 locations where samples were collected in 2006 ($p = 0.007$). TPAH concentrations were similar in amphipods collected at the same stations in 2004 and 2006, but higher than concentrations measured in 2005 (Tables 3-9, 3-10, 3-12; Figure 3-10). The differences in TPAH concentrations in amphipods collected at BSMP, Liberty, and Northstar in 2004, 2005, and 2006 were statistically significant at $p<0.001$, because of the lower concentration of TPAH in amphipods collected at BSMP and Northstar in 2005.

Table 3-12. Concentrations of total PAH, total resolved SHC, pristane, and total StTr in amphipods and clams, and total PAH in isopods and mysids collected in 2006 at 1 to 5 areas in the development area. Clams were collected at just Liberty and mysids were collected at just Northstar. Concentrations are ng/g dry wt.

Hydrocarbon	Mean	SD	Median	Minimum	Maximum
Amphipods (*Anonyx nugax*)					
Total PAH	59.6	29.8	55.0	19.7	123
Total SHC	41,300	37,300	24,800	2,675	147,000
Pristane	38,400	35,000	22,600	1,971	137,000
Total StTr	6.01	11.03	2.48	0.0	52.63
Isopods (*Saduria sabini*)					
Total PAH	98.9	48.1	85.4	59.2	188

Table 3–12. Concentrations of total PAH, total resolved SHC, pristane, and total StTr in amphipods and clams, and total PAH in isopods and mysids collected in 2006 at 1 to 5 areas in the development area. Clams were collected at just Liberty and mysids were collected at just Northstar. Concentrations are ng/g dry wt, continued.

Hydrocarbon	Mean	SD	Median	Minimum	Maximum
Clams (*Astarte montagui* and *Cyrtoderia kurriana*)					
Total PAH	141	57.8	141	100	182
Total SHC	5,280	666	5,280	4,610	5940
Pristane	97	36	97	71	122
Total StTr	0	0	0	0	0
Mysids (*Mysis* sp)					
Total PAH	89.3	24.4	89.1	65.0	114

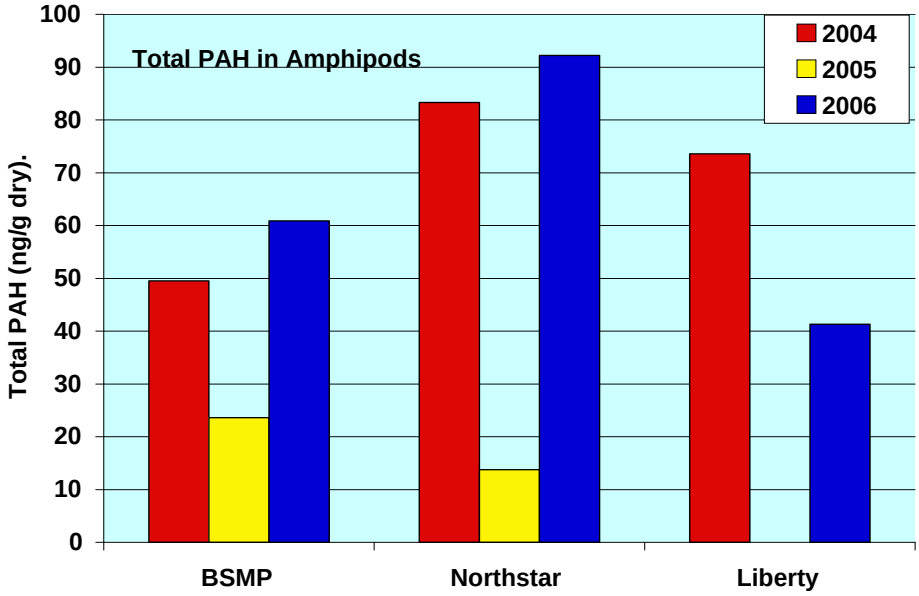


Figure 3-10. Mean concentrations of TPAH in amphipods collected in 2004, 2005, and 2006 from BSMP, Northstar, and Liberty.

The total resolved SHC concentrations were quite variable in amphipods collected in 2006, ranging from 2675 to 147,000 ng/g dry weight with a mean of 41,300 ± 37,3000 (Table 3-12). This is due largely to variability in pristane concentrations, which ranged from 1,971 to 137,000 ng/g dry weight (Table 3-12). The two amphipod samples containing the highest concentrations of pristane were from the Boulder Patch (Station BP01 north of Liberty) (Table 3-13). These two samples also contained 137,000 and 154,000 ng/g total SHC. The amphipod sample from BSMP Station 7E in Harrison Bay contained the lowest concentration of pristane (1970 ng/g) and the highest concentration of total SHC (249,000 ng/g) (Table 3-13). Thus, the contribution of pristane to total SHC in amphipods is highly variable and probably varies with the diet of these carnivorous amphipods.

Table 3-13. Concentrations of total PAH, total SHC, and total St/Tr in amphipods (*Anonyx nugax*) sampled at 23 sites in 5 monitoring areas in 2006. n = number of samples. Concentrations are ng/g dry wt.

Hydrocarbon	n	Location	Mean	SD	Range
Total PAH	2	Liberty	92.2	26.6	73.4 – 111
	10	Northstar	41.3	27.4	19.7 – 114
	2	West Dock	103	19.6	83.6 – 123
	2	Boulder Patch	70.9	16.5	54.4 – 87.4
	7	BSMP	60.9	14.4	34.8 – 80.1
Total SHC	2	Liberty	66,400	37,700	39,700 – 93,100
	10	Northstar	33,800	20,100	13,700 – 71,700
	2	West Dock	25,900	732	25,200 – 26,700
	2	Boulder Patch	145,500	8,300	137,000 – 154,000
	7	BSMP	90,800	77,600	26,100 – 249,000
Pristane	2	Liberty	48,100	36,600	22,300 – 74,000
	10	Northstar	24,700	18,800	6,400 – 61,000
	2	West Dock	15,600	1,020	14,600 – 16,600
	2	Boulder Patch	114,000	22,600	91,400 – 136,600
	7	BSMP	34,000	34,000	1,970 – 88,500
Total StTr	2	Liberty	6.70	3.74	2.96 – 10.45
	10	Northstar	1.09	2.28	0 – 7.48
	2	West Dock	0	---	0
	2	Boulder Patch	10.5	6.66	3.39 – 16.7
	7	BSMP	13.42	16.45	0 – 52.6

Total StTr concentrations in amphipods ranged from 0 to 52.63 ng/g, with a mean concentration of 6.01 ± 11.0 ng/g (Table 3-12). The amphipods containing the highest concentration of total StTr (52.6 ng/g) were from BSMP Station 7E in Harrison Bay (Table 3-13), the same amphipods that contained the highest total SHC and lowest pristane concentrations.

TPAH concentrations in mysids and isopods were similar in 2006, ranging from 59.2 to 188 ng/g, and higher than in amphipods; the mean TPAH concentrations in isopods and mysids collected in 2006 were 98.9 ± 48.1 ng/g and 89.3 ± 24/4 ng/g, respectively (Table 3-12). Isopods were collected in 2006 at 6 locations, 1 each at Liberty, BSMP, and West Dock, and 3 at Northstar. The highest TPAH concentration was in isopods from West Dock (188 ng/g) and lowest was in isopods from Northstar (67.6 ± 12.8 ng/g) (Table 3-14). The TPAH concentration was significantly higher in isopods from West Dock and BSMP ($p = 0.04$) than in those from Liberty and Northstar in 2006.

The two clam samples collected at Liberty in 2006 contained a higher mean TPAH concentration (141.4 ± 57.8 ng/g) than the three crustaceans did (Table 3-12). Total SCH concentrations ranged from 4,610 to 5,942 ng/g dry weight, with pristane contributing less than 2% to the total SHC. Most of the total SHC was n-alkanes ranging from C_{20} to C_{37}. StTr biomarkers were not detected in clams collected in 2006.

Table 3-14. Mean, standard deviation, minimum, and maximum concentrations of TPAH in tissues of isopods collected in 2006 from BSMP, Liberty, and Northsar.

Species	Location (n)	Mean	SD	Range
Isopod	BSMP (1)	114	---	114
	Liberty (1)	88.5	---	88.5
	Northstar (3)	67.6	12.8	59.2 – 82.4
	West Dock (1)	188	---	188

The PAH composition (PAH profile) was similar in tissues of amphipods, isopods, mysids, and clams collected in the BSMP, Liberty, and Northstar areas in 2006 (Figures 3-11 through 3-14). Alkylnaphthalenes were the most abundant PAH in tissues of all four species, followed in most cases by alkylphenanthrenes. Concentrations of 4- through 6-ring PAH (fluoranthene through benzo(ghi)perylene) usually were low.

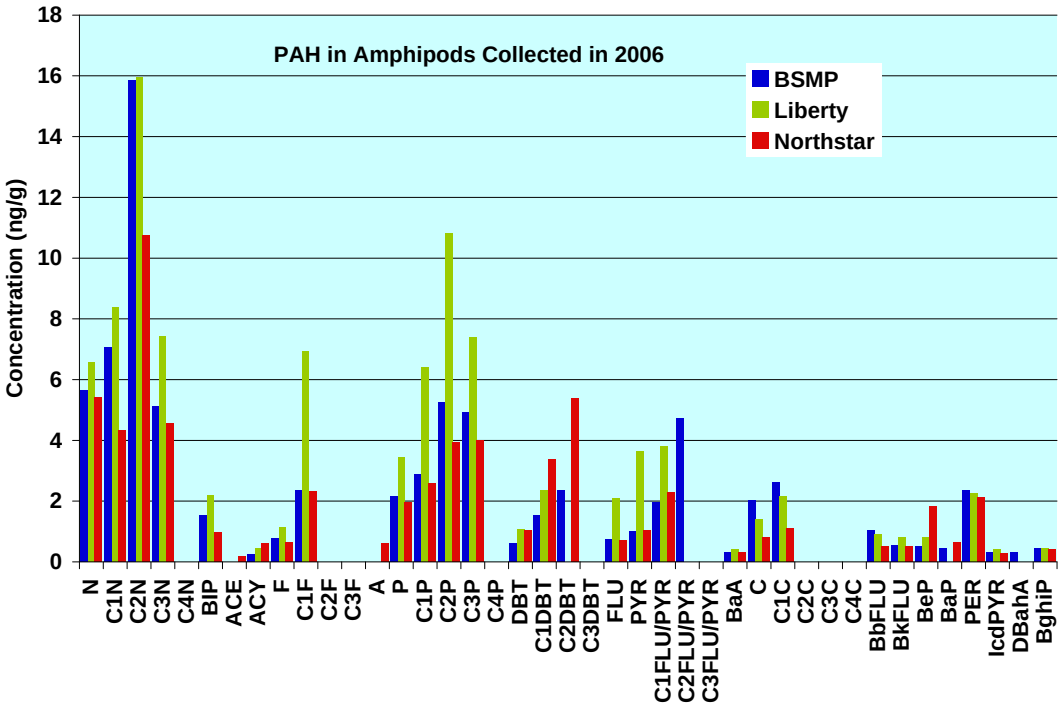


Figure 3-11. PAH composition in tissues of amphipods collected at BSMP, Liberty, and Northstar in 2006.

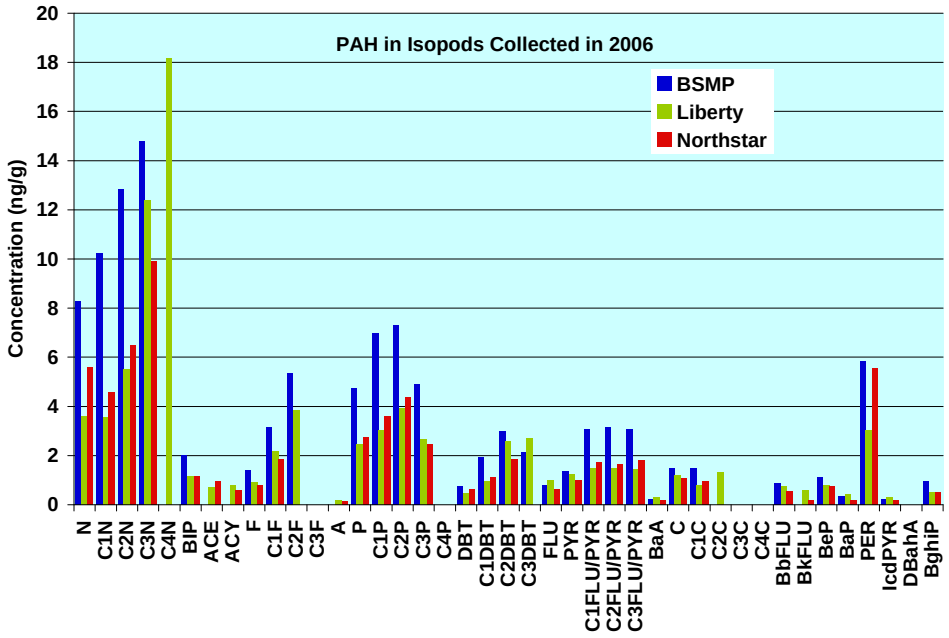


Figure 3-12. PAH composition in tissues of isopods collected at BSMP, Liberty and Northstar in 2006.

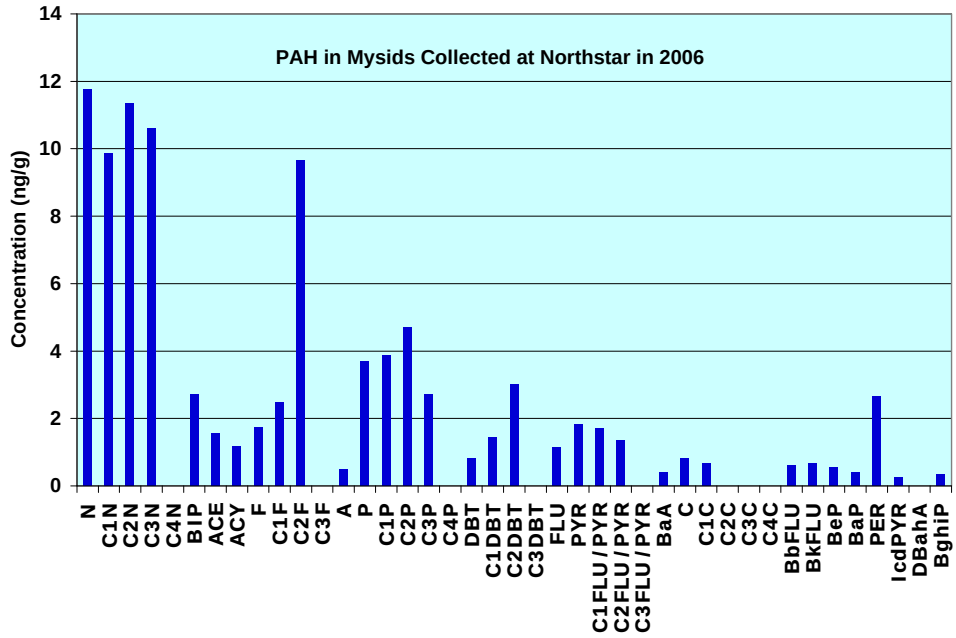


Figure 3-13. PAH composition in tissues of mysids collected at Northstar in 2006.

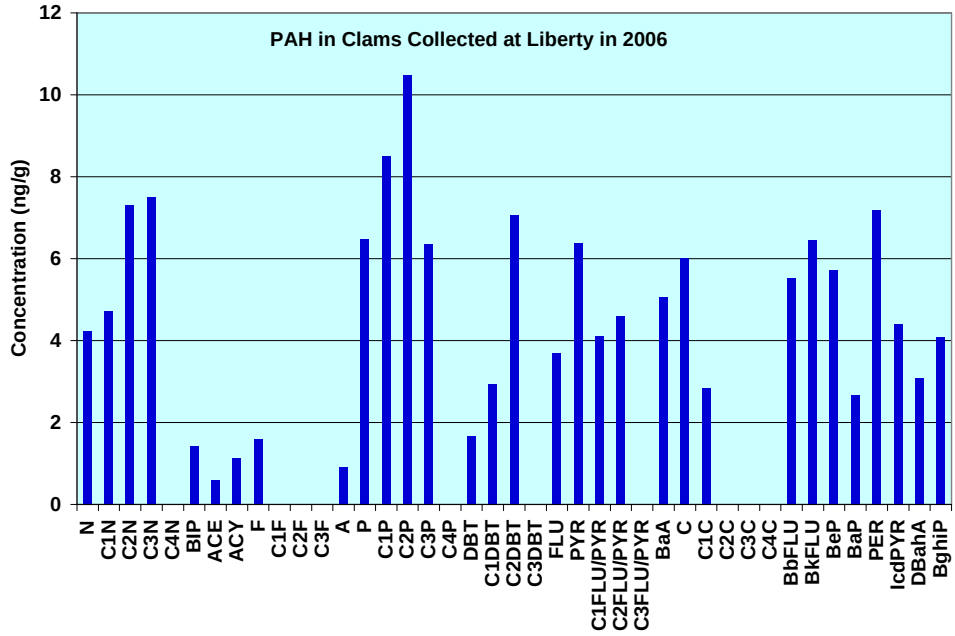


Figure 3-14. PAH composition in tissues of clams collected at Liberty in 2006.

In most cases, perylene was the most abundant high molecular weight PAH in invertebrate tissues. Most of the perylene in environmental samples is biogenic, derived from the diagenesis of organic plant remains under anaerobic conditions (Venkatesan, 1988). Peat often contains a high concentration of perylene. A sample of Colville River peat collected in 2006 contained 737 ng/g TPAH, including 43.7 ng/g perylene (Figure 3-15). Peat samples collected in 2006 from the Kuparuk, Sagavanirktok, and Pingok Rivers contained lower concentrations of TPAH (12.8 to 291 ng/g) and perylene (0.19 to 28.9 ng/g). The abundance of alkylnaphthalenes and alkylphenanthrenes also was high in the river peat samples. Thus, peat may have been a source of PAH in the clam and crustacean tissues. However, StTr biomarker profiles in peat samples were somewhat different from those in amphipods and clams. StTr were not measured in isopod and mysid samples. The most abundant biomarker in Colville River peat and amphipod tissues was 17a(H),21b(H)-hopane (Figure 3-16). 22R-17a(H),21b(H)-30-homohopane was abundant in Colville River and Sagavanirktok River peat and amphipods from the BSMP area, but non-detectable in amphipods from Liberty and Northstar (Figure 3-17). Clams collected at Liberty in 2006 did not contain detectable levels of any StTr biomarkers.

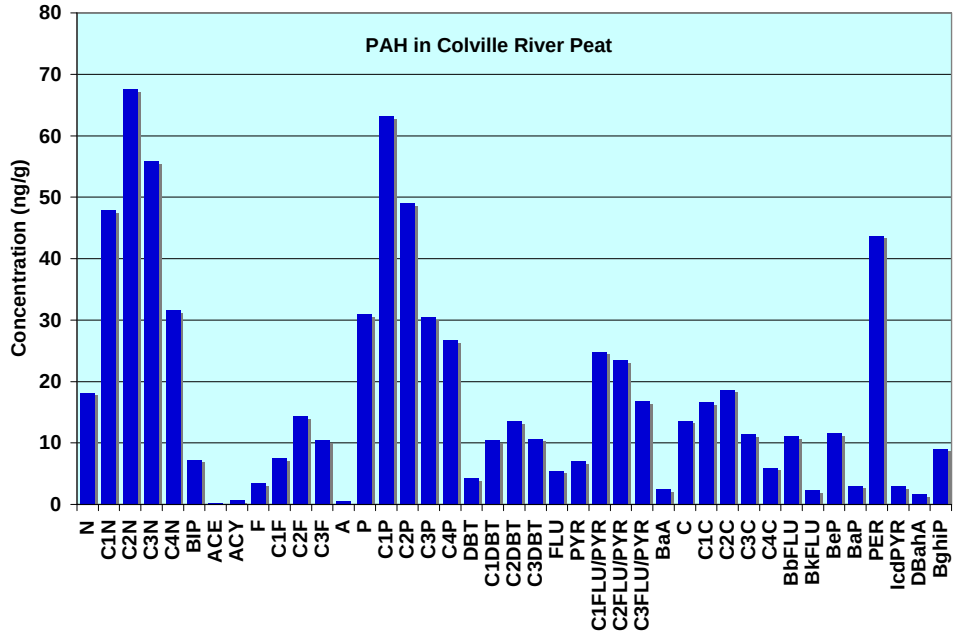


Figure 3-15. PAH composition of Colville River peat collected in 2006.

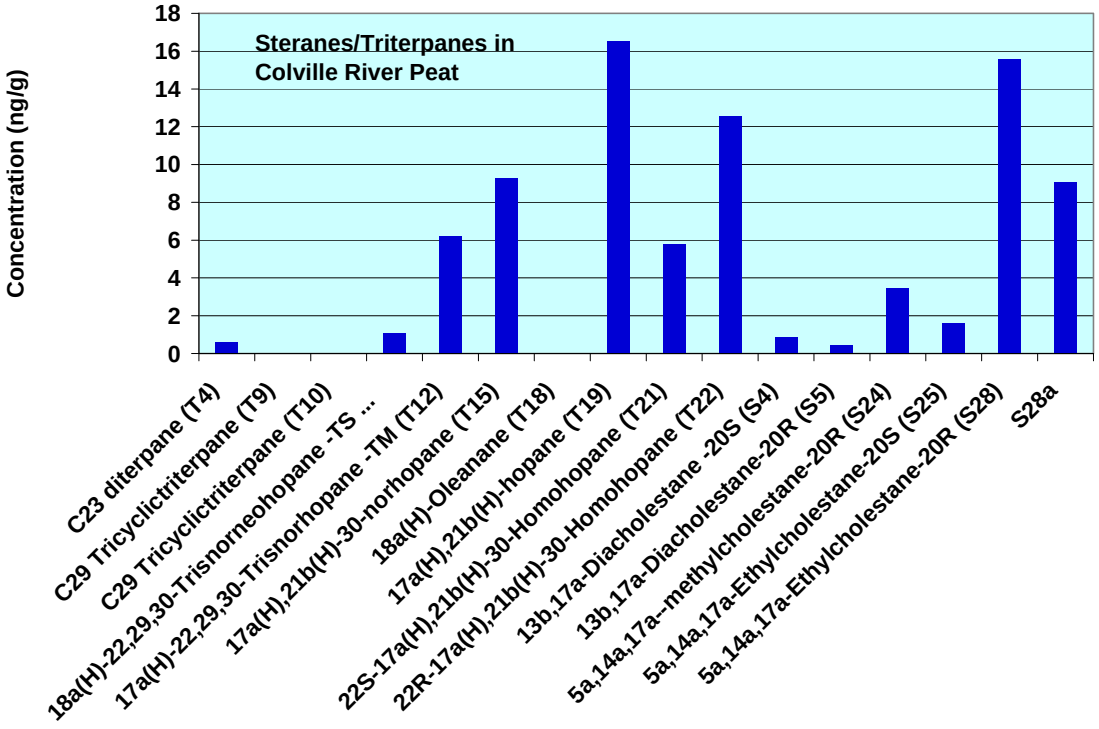


Figure 3-16. Sterane/triterpane (StTr) biomarker composition of Coleville River peat collected in 2006.

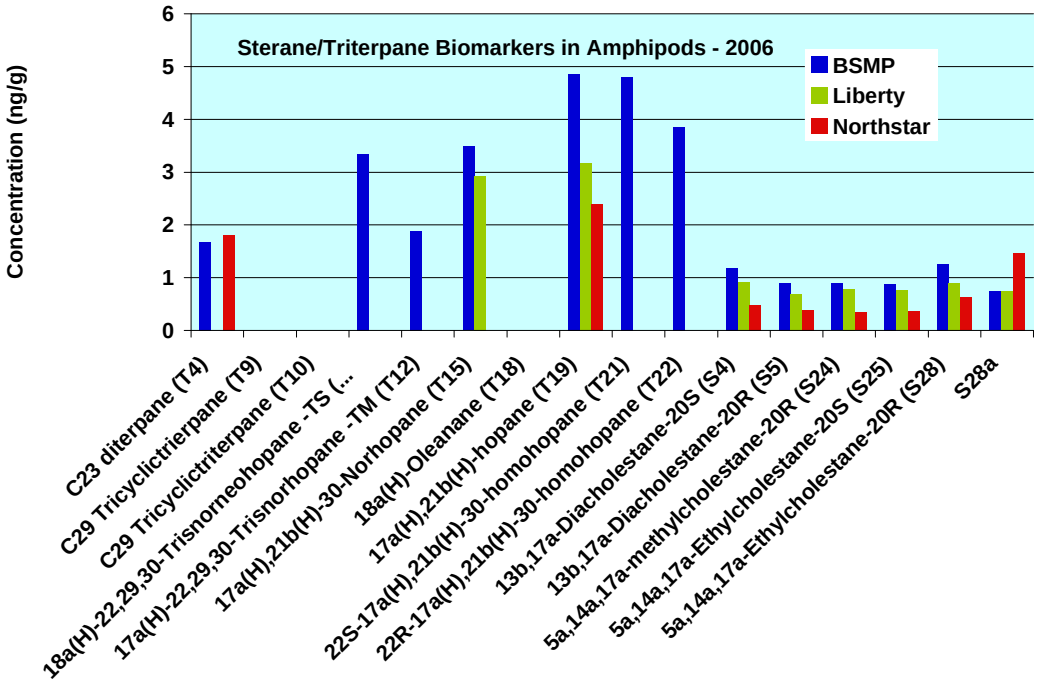


Figure 3-17. Sterane/triterpane (StTr) biomarker composition in tissues of amphipods collected in 2006 from the BSMP, Liberty, and Northstar areas.

The PAH and StTr compositions of Northstar crude oil were different from those in tissues of clams and crustaceans collected in 2006 from the BSMP, Liberty, and Northstar. Northstar is a light crude oil (°API = 40.8). The most abundant PAH are the 2-ring naphthalene and alkyl naphthalenes (Figure 3-18). Alkyl phenanthrenes are the most abundant 3-ring PAH. Most high molecular weight 4- through 6-ring PAH, except the chrysenes, are present at concentrations near or below the method detection limit. This PAH profile is typical for a light crude oil.

Northstar crude oil contains a full suite of StTr biomarkers, except oleanane and S28a (Figure 3-19). The most abundant biomarkers are 17a(H),21b(H)-hopane (which is abundant in amphipod tissues: Figure 3-17) and 13b,17a-diacholestane-20S (which is present at a low concentration in amphipod tissue: Figure 3-17). Northstar crude oil does not contain a detectable concentration of the S28a sterane, which is present at low concentrations in amphipod tissues (Figure 3-17) and Coleville River peat (Figure 3-16). Northstar crude oil probably contributes little if at all to the PAH and StTr biomarker residues in tissues of indigenous clams and crustaceans in the study area.

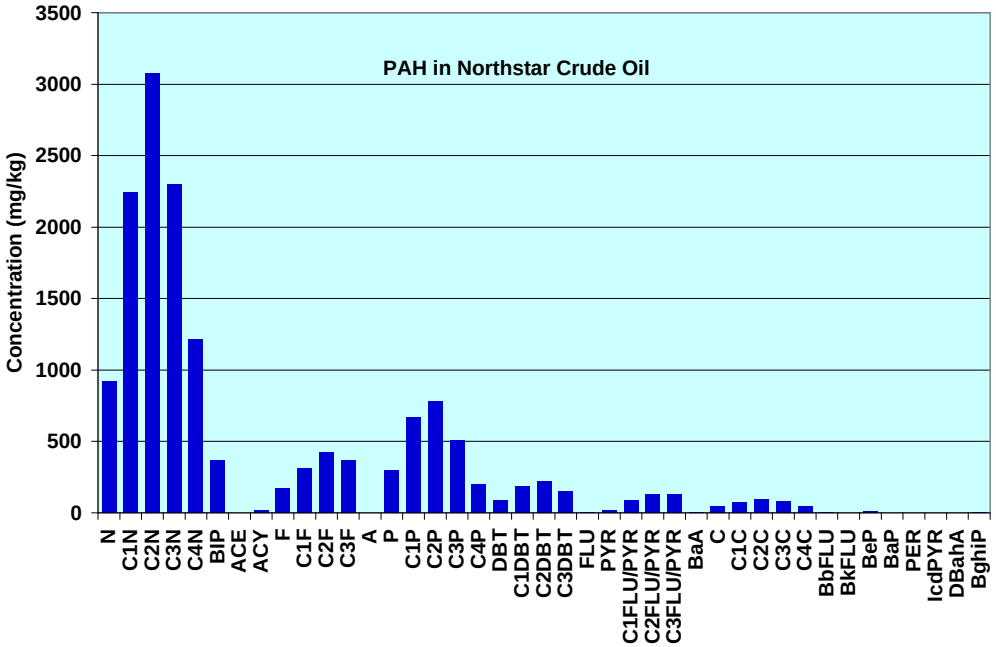


Figure 3-18. The PAH composition of Northstar crude oil.

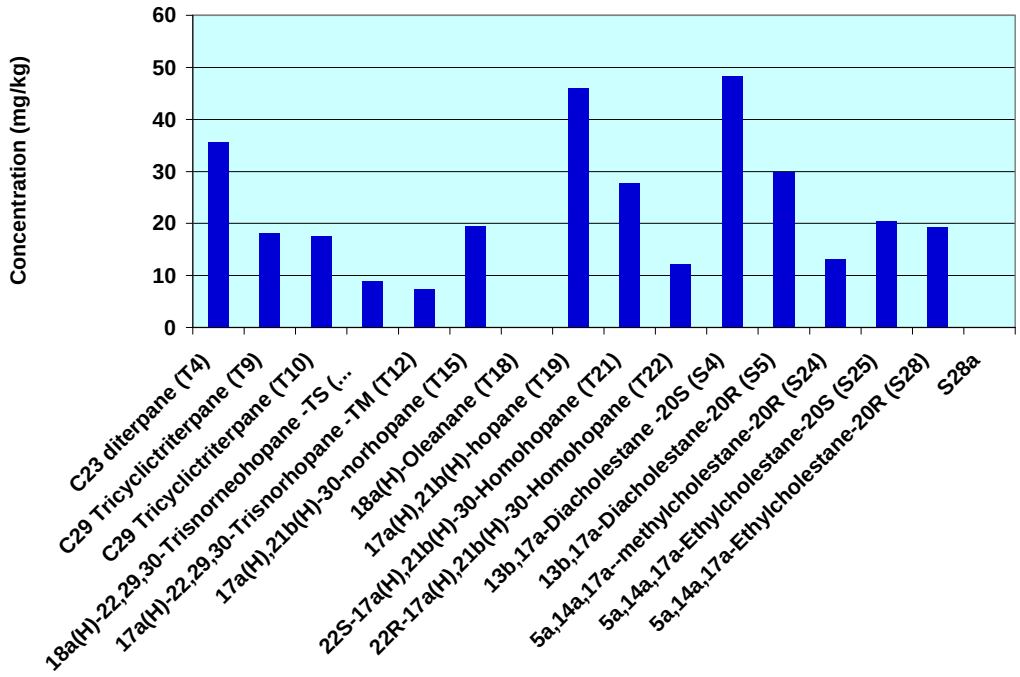


Figure 3-19. The sterane/triterpane (StTr) biomarker composition of Northstar crude oil.

3.1.2.2 Comparison to Hydrocarbons in Amphipods and Clams from ANIMIDA (2000 and 2002)

PAH, SHC, and StTr were measured in soft tissues of amphipods (*Anonyx nugax*) and clams *(Astarte montagui* and *Cyrtodaria kurriana*) in 2000 and 2002 as part of ANIMIDA Task 2 (Brown et al., 2005). Sampling locations were similar to those used in this investigation and were primarily near the Northstar Development, the Liberty Prospect, and at a few BSMP locations. The results of these analyses are compared to those for 2004, 2005, and 2006 in Table 3-15.

Table 3-15. Mean, SD, and range of concentrations of TPAH, TSHC, and StTr in soft tissues of amphipods (*Anonyx nugax*) and clams (*Astarte montagui* and *Cyrtodaria kurriana*) collected from the Beaufort Sea in 2000 and 2002 (ANIMIDA Task 2) and 2004, 2005, and 2006 (cANIMIDA Task 5). Concentrations are ng/g dry wt (data for 2000 and 2002 were converted from wet wt. by multiplying by 5, based on ~89% moisture).

Hydrocarbon	Year	Amphipods		Clams	
		Mean ±SD	Range	Mean (SD)	Range
TPAH	2000	85.8 ± 18.4	60.0 – 115	90.4 ± 54.4	37.0 – 195
	2002	95.6 ± 41.6	55.0 – 175	80.6 ± 47.8	48.0 – 175
	2004	68.2 ± 33.8	39.6 – 143	95.8 ± 52.6	42.7 – 168
	2005	25.4 ± 13.5	8.25 – 50.0	38.4 ± 15.1	21.8 – 51.2
	2006	59.6 ± 29.8	19.7 – 123	141 ± 40.9	100 – 182
TSHC	2000	55,800 ± 45,300	0 – 130,000	6000 ± 8580	0 – 22,000
	2002	113,000 ± 71,800	23,500 – 260,000	14,100 ± 1390	12,500 – 16,000
	2004	444000 ± 8540	29800 - 54100	26,800 ± 14,400	678 – 39,700
	2005	31,600 ± 18,400	5171 – 67,100	1510 ± 517	1030 - 2230
	2006	63,000 ± 57,100	13,700 – 249,000	12,600 ± 2410	10,200 – 15,000
StTr	2000	18.8 ± 10.2	10.0 – 40.5	14.8 ± 3.67	10 - 20
	2002	10.8 ± 2.46	7.5 – 16.0	8.40 ± 3.97	5.00 – 15.5
	2004	12.2 ± 3.45	8.36 – 17.8	8.54 ± 5.37	2.92 – 17.1
	2005	6.22 ±1.89	4.53 – 8.02	1.72 ± 2.43	0 – 5.16
	2006	29.0 ± 11.8	20.7 – 55.0	0	0

Mean concentrations of TPAH were similar in all years except 2005 in tissues of amphipods and clams from the Beaufort Sea development area (Table 3-15). TPAH concentrations in both taxa were lower in 2005 than in other years. The mean TPAH concentration in amphipods declined each year between 2000 and 2006; this trend was not observed in clams. Clams (*Cyrtodaria kurriana*) collected off the Kuparuk River in Gwydyr Bay (station 5F) in 2000 and 2002 contained higher TPAH concentrations (195 and 175 ng/g dry wt) than the *Astarte montagui* collected in 2000 and 2002 further offshore.

Mean concentrations of TSHC were high and extremely variable, both within a year and from one year to another, in tissues of both amphipods and clams (Table 3-15). Highest TSHC concentrations in both taxa were in animals collected in 2004 and lowest concentrations were in animals collected in 2005. In any year, TSHC concentrations were higher in amphipod tissues than in clam tissues. Most of this difference was due to much higher pristane concentrations in amphipods than in clams. Mean StTr biomarker concentrations were roughly similar in all years

except 2005 in amphipods (Table 3-15). Mean biomarker concentrations tended to decline with time in clam tissues and were not detected in 2006.

3.1.2.3 Hydrocarbons in Deployed Mussels

Summer 2004

Three composite samples of mussels deployed near Northstar, two samples deployed in the BSMP area, and one sample deployed at Liberty were recovered in 2004 for analysis (Table 2-1; Figures 2-1 through 2-3). The mussels used in the Beaufort Sea deployments were collected from Port Chatham on the southwest coast of the Kenai Peninsula at the mouth of Cook Inlet. Three replicate composite mussel samples, collected at the same time and place, were not deployed but were used as zero-time reference samples.

Polycyclic Aromatic Hydrocarbons (PAH)

The reference mussels contained 195 to 275 ng/g dry wt TPAH (mean 227 ± 34.9 ng/g dry wt). All mussel samples, following deployment at Northstar, Liberty, and the BSMP area, contained lower TPAH concentrations than the mean for the three reference mussel samples (Table 3-16; Figure 3-20), indicating that they released some of the accumulated PAH during the deployment in the Beaufort Sea. TPAH concentrations in individual composite samples of deployed mussels ranged from 91.5 to 204 ng/g. The Port Chatham mussels, although collected from a commercial mussel harvest area that was considered uncontaminated, contained low concentrations of PAH. This indicates that the Beaufort Sea environment contains lower concentrations of bioavailable PAH than the Port Chatham environment.

Between 46 and 62 percent of the TPAH in zero-time mussels and mussels that had been deployed at Northstar and BSMP sites were naphthalene and alkylnaphthalenes (Figure 3-21). Naphthalenes represented 22 percent of the TPAH in mussels that had been deployed at Liberty. Biphenyl was present at higher than expected concentrations. The laboratory blanks contained traces of naphthalene, C1-C2-naphthalenes, and biphenyl, indicating that some of the low levels of naphthalenes and biphenyl in these samples may have been contributed by laboratory contamination.

Alkylphenanthrenes also were abundant in all the mussels, indicating that most of the PAH in the mussels probably were from petroleum sources or from peat (Figure 3-15). Mean naphthalene concentration was higher in mussels deployed at BSMP and lower in mussels from Liberty than in zero-time control mussels. There were differences in concentrations of several other PAH in deployed and in reference mussels, indicating that there was some exchange of individual PAH during deployment. Mussels from Liberty and Northstar contained a higher concentration of perylene than zero-time controls did, indicating that the mussels were bioaccumulating perylene during deployment at these sites, probably from a natural, biogenic source, probably peat.

Table 3-16. Mean concentrations of total PAH, total resolved SHC, and total StTr in reference mussels (*Mytilus trossulus*) and mussels that had been deployed at different locations in the Beaufort Sea study area for 14 to 17 days in 2004, 2005, and 2006. Standard deviations are included if there was more than one replicate at a location and time. Concentrations are ng/g dry wt.

Location	Year	Total PAH	Total Resolved SHC	Total StTr
Reference (Port Chatham)	2004	227 ± 34.9	6051 ± 522	12.2 ± 3.79
	2005	32.8 ± 27.0	3632 ± 151	5.87 ± 5.87
	2006	164 ± 36.2	23,159 ± 2294	ND
BSMP	2004	157 ± 46.8	6246 ± 123	14.1 ± 6.43
	2005	31.5 ± 1.46	3137 ± 380	ND
	2006	52.6	16,033	ND
Liberty	2004	92.8	5725	13.5
	2005	24.8 ±14.7	2689 ± 384	ND
	2006	134 ± 6.74	16,040 ± 3145	ND
Northstar	2004	148 ± 45.8	7624 ± 2494	27.2 ± 16.2
	2005	13.0	3381	6.9
	2006	91.6 ± 5.18	21,024 ± 3599	ND
Prudhoe Bay	2005	64.2	4820	ND
SDI (Endicott)	2006	121	20,147	101
West Dock	2006	150	29,134	25.3

ND, not detected.

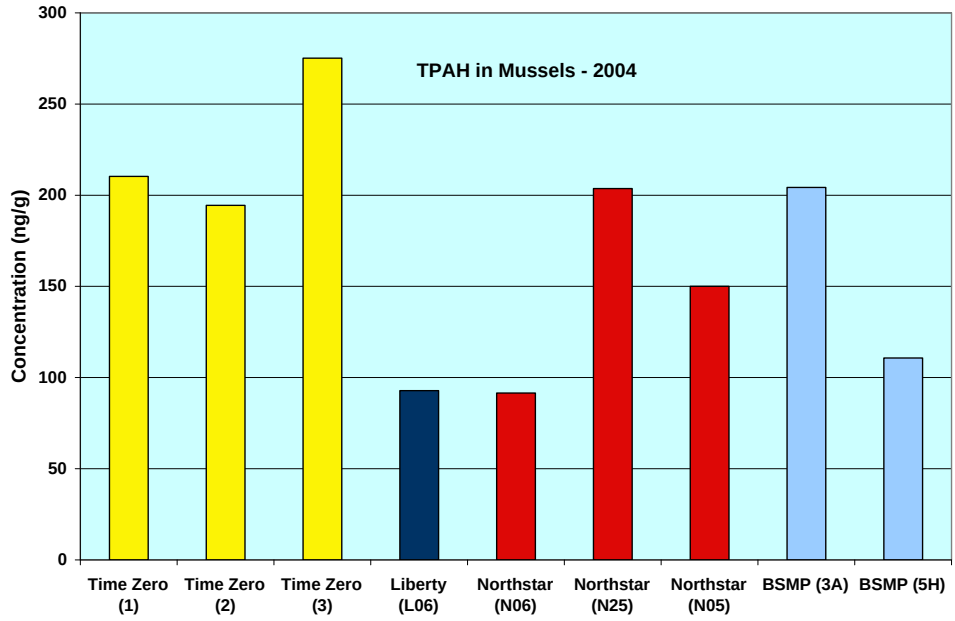


Figure 3-20. Concentrations of TPAH in tissues of zero-time reference mussels (Mytilus trossulus) (from the collection site at Port Chatham, AK) and mussels deployed for 14 to 17 days at three Beaufort Sea locations in 2004. Concentrations are ng/g dry wt.

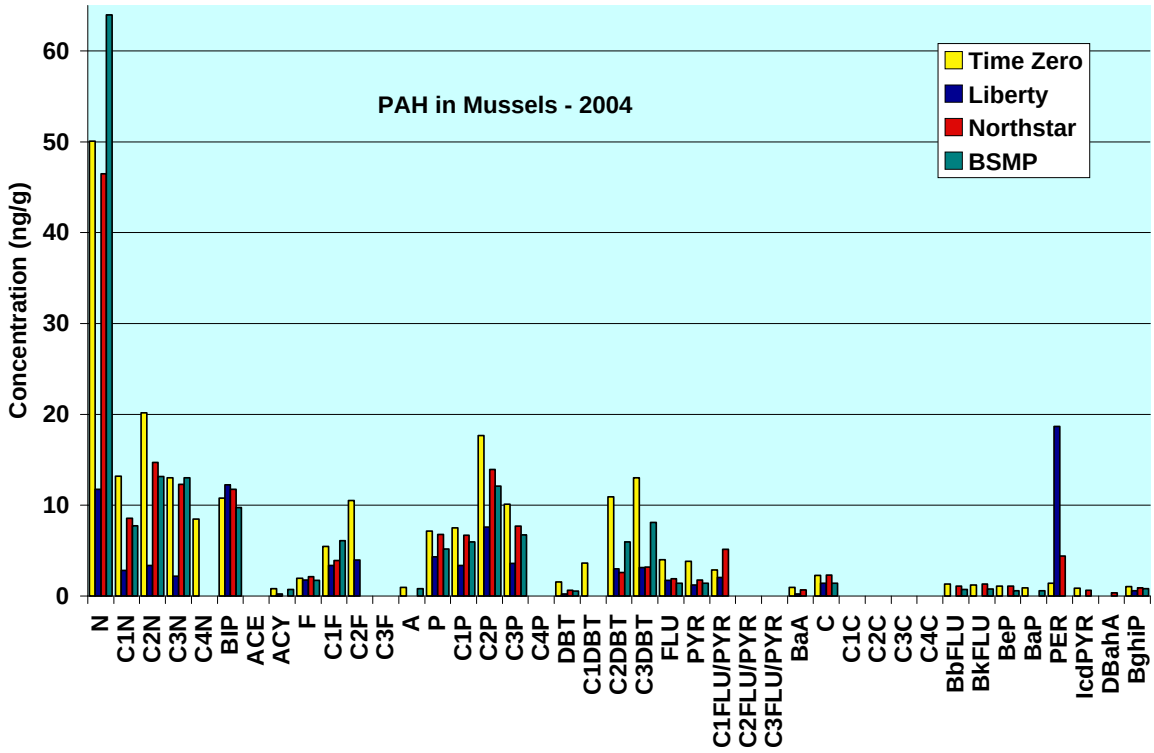


Figure 3-21. Concentrations of individual PAH in zero-time mussels (Mytilus trossulus) and mussels that had been deployed at three locations in the Beaufort Sea in 2004.

Saturated Hydrocarbons (SHC)

Mussels also were analyzed by GC/FID for total and resolved saturated hydrocarbons (SHC) and steranes/triterpanes (StTr). The mean concentration of total resolved SHC in reference mussels was 6051 ± 522 ng/g dry wt. Mean concentrations in mussels following two-week deployments in three areas of the Beaufort Sea ranged from 5725 ng/g in the single sample from Liberty to 7624 ± 2494 ng/g at Northstar (Table 3-16).

The concentrations of individual resolved SHC in the mussel tissues are summarized in Figure 3-22. The most abundant SHC in most mussel samples were *n*-C_{15}, *n*-C_{16}, *n*-C_{17}, and *n*-C_{25} *n*-paraffins. Concentrations of pristane and phytane were similar to those of n-paraffins with similar molecular weights.

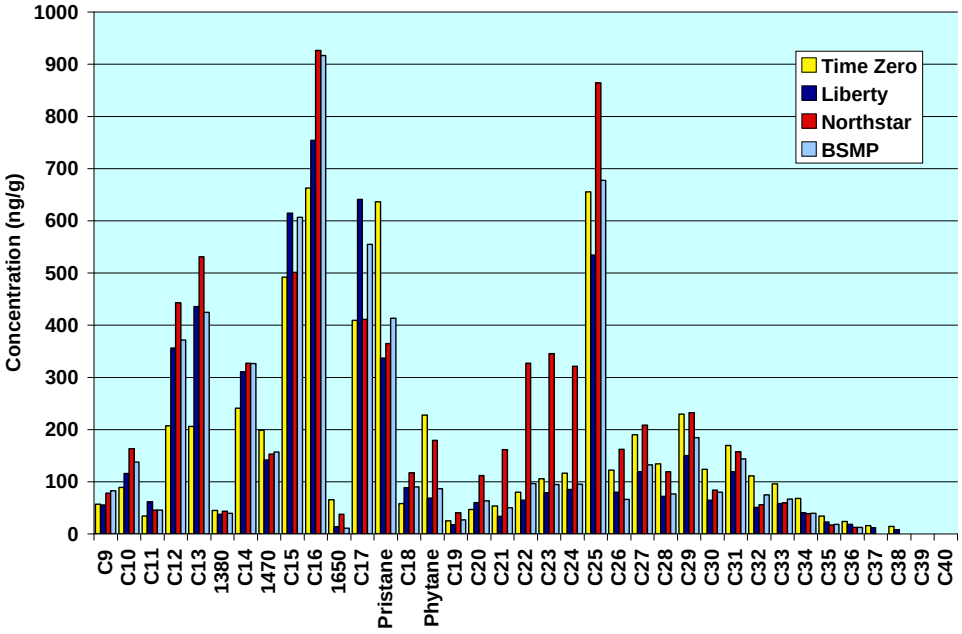


Figure 3-22. Concentrations of individual resolved saturated hydrocarbons (SHC), mostly n-paraffins, in soft tissues of mussels that had been deployed in the Beaufort Sea for approximately two weeks in 2004. Data for reference mussels are included.

Sterane/Triterpane (StTr) Biomarkers

StTr biomarker concentrations were similar in time zero mussels and in mussels following deployment in the BSMP, Liberty, and Northstar areas. The mean concentration in reference mussels in 2004 was 12.2 ± 3.79 ng/g and concentrations in deployed mussels ranged from 13.5 at Liberty to 27.2 ng/g at Northstar (Table 3-16). The higher mean concentration in deployed than in reference mussels was due mainly to S28a (an ethylcholestane) and 17a(H)-22,29,30-trisnorhopane, which were not detected in reference mussels in 2004 (Figure 3-23). S28a also was not detected in Northstar crude oil (Figure 3-19), suggesting that mussels were not bioaccumulating hydrocarbons from this crude oil. These two biomarkers were present in tissues of mussels deployed at BSMP and Northstar, but not Liberty. The StTr profiles in tissues of reference and deployed mussels were almost identical, with the exception of the two biomarkers that were detected in just the deployed mussels. These results indicate that there was not much exchange between hydrocarbons in reference mussels and hydrocarbons in the Beaufort Sea environment during the two-week deployment.

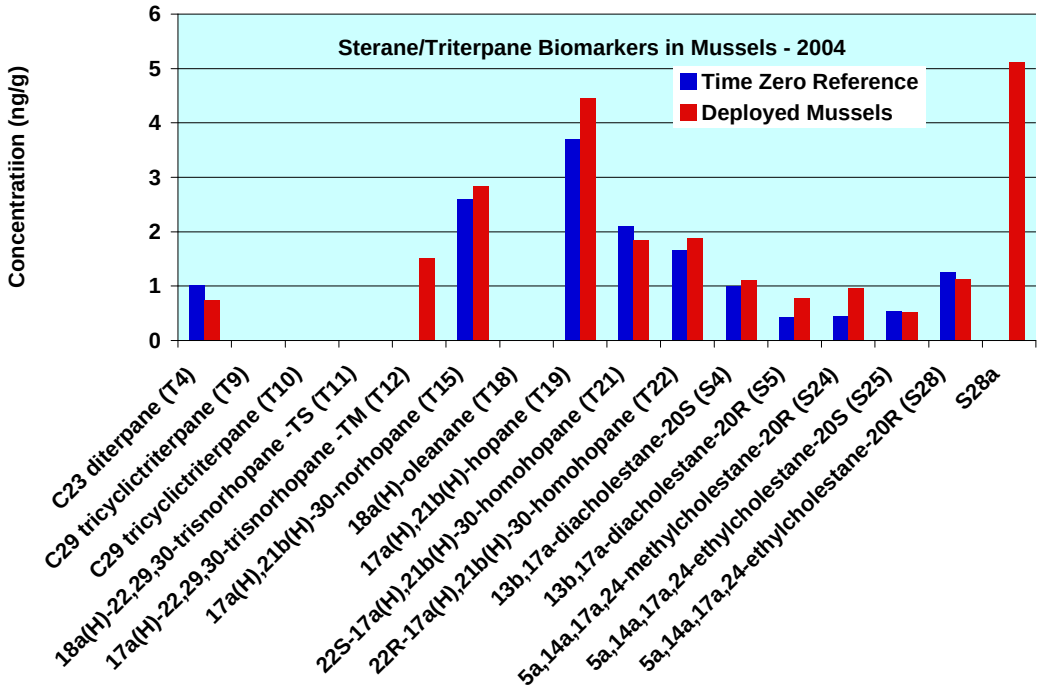


Figure 3-23. Mean concentrations of StTr in reference mussels (3 samples) and mussels that had been deployed in the BSMP, Liberty, and Northstar areas (6 samples) in 2004.

Summer 2005

Seven of the eight subsurface moorings containing caged mussels that were deployed in 2005 were recovered, including one each from Liberty, Northstar, Prudhoe Bay, near Endicott, and the Boulder Patch, and two in the BSMP area (Table 2-2). Two reference samples also were collected, one was frozen immediately upon collection in Port Chatham; the other was transported to the North Slope and frozen when the other samples were deployed. Mussels were deployed in 2005 for 13 to 14 days.

Polycyclic Aromatic Hydrocarbons (PAH).

TPAH concentrations ranged from 10.5 to 64.2 ng/g dry weight in the reference and deployed mussels in 2005 (Figure 3-24). The average TPAH concentration was highest in mussels deployed in Prudhoe Bay (64.2 ng/g dry wt) and lowest in mussels deployed near Northstar (13.0 ng/g). The reference mussels contained an average of 32.8 ± 27.0 ng/g TPAH (Table 3-16). Both reference mussels and mussels that had been deployed for 13 to 14 days at six Beaufort Sea locations in 2005 contained significantly lower concentrations of TPAH than the mussels deployed in 2004.

The PAH composition in the reference mussels was quite different from that in the mussels following deployment at the different Beaufort Sea Sites. For example, the deployed mussels contained relatively high concentrations of C2- and C3-phenanthrenes and smaller amounts of chrysene, both absent from tissues of reference mussels (Figure 3-25). Reference mussels contained higher concentrations of naphthalene, C1- through C3-naphthalenes, and biphenyl than deployed mussels did.

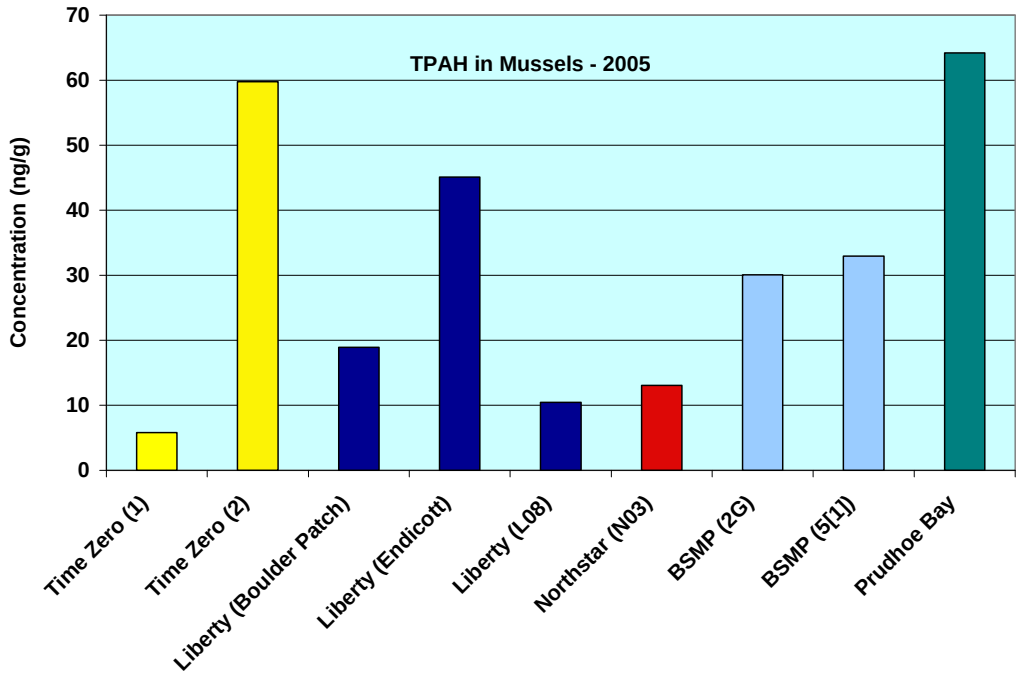


Figure 3-24. Concentrations of total PAH in tissues of reference mussels (Mytilus trossulus) (from the collection site at Port Chatham, AK) and mussels that had been deployed for 13 to 14 days at six Beaufort Sea locations in 2005. Concentrations are ng/g dry wt.

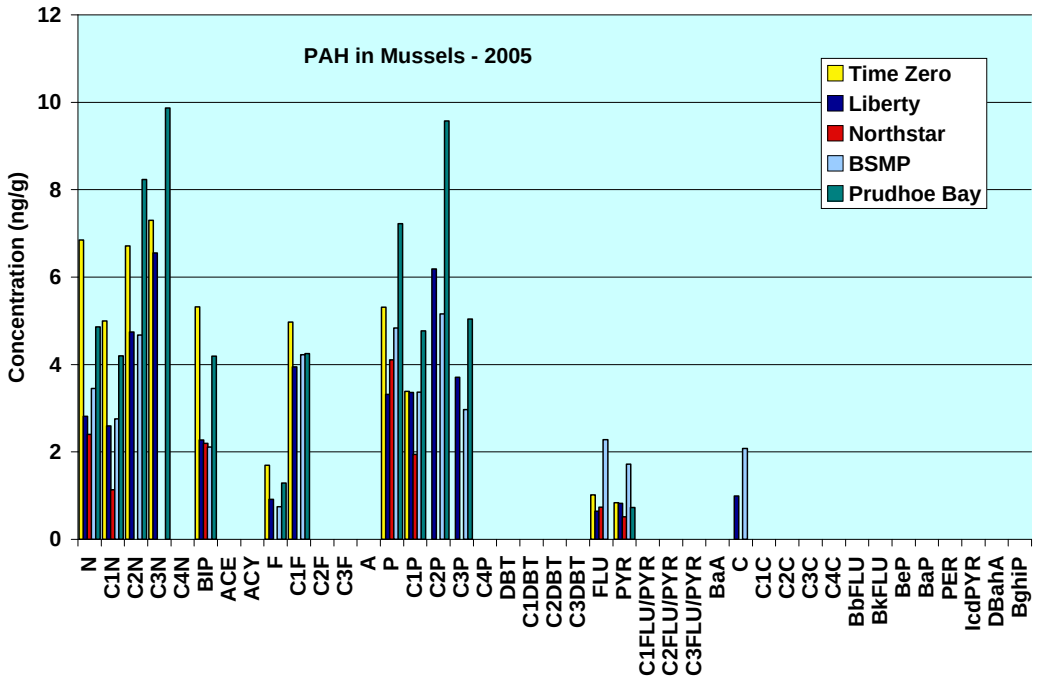


Figure 3-25. Concentrations of individual PAH in reference mussels (Mytilus trossulus) and mussels that had been deployed at four locations in the Beaufort Sea in 2005.

Resolved Saturated Hydrocarbons (SHC).
The total resolved SHC in the mussels collected during the 2005 field season ranged from 2370 ng/g dry wt in a sample deployed near Liberty to 4820 ng/g in the mussels deployed in Prudhoe Bay (Table 3-16). The reference mussels contained an average concentration of 3632 ng/g total resolved SHC; mussels deployed near Liberty, Northstar, and at historical BSMP stations contained a lower mean SHC concentration than the reference mussels.

The reference mussels contained appreciable amounts of n-C_{11}, Isoprenoid 1470, n-C_{15}, and pristane (Figure 3-26). The distribution of the mid-molecular weight alkanes (n-C_{20} to n-C_{30}) in all samples tended to exhibit a bias toward the odd-numbered alkanes; this is particularly evident in the mussels deployed in Prudhoe Bay.

Sterane/Triterpane (StTr) Biomarkers
One of two reference mussel samples collected in 2005 contained 11.74 ng/g dry wt total StTr. The other reference mussel sample did not contain any StTr above the method detection limit. The single deployed mussel sample from Northstar contained 6.9 ng/g total StTr; all other mussels did not contain StTr at detectable concentrations (Table 3-16). The reference sample contained just S28a and the sample from Northstar contained just 17a(H),21b(H)-30-norhoane.

Summer 2006
Eight subsurface moorings containing caged mussels were deployed for 13 to 14 days and recovered in 2006. There were two deployments near Northstar, and one deployment each in the BSMP area, the Boulder Patch, near Endicott, at Liberty, at SDI, and at West Dock (Table 2-3; Figures 2-7 through 2-9). Three 'time zero' (reference) mussel samples were collected and used as trip blanks. One reference samplewas frozen immediately after collection in Port Chatham. The other two reference mussels samples were transported to the North Slop with the mussels destined for deployment and were frozen for shipment when mussels were deployed at the Boulder Patch and at West Dock locations.

Polycyclic Aromatic Hydrocarbons (PAH)
Individual samples of both reference mussels and mussels that had been deployed at seven Beaufort Sea locations in 2006 contained between 50 ng/g dry wt and 200 ng/g TPAH (Figure 3-27). Mean TPAH concentrations ranged from 52.6 ng/g at BSMP to 164 ± 36.2 ng/g in the time zero reference mussels (Table 3-16). Mussels were deployed for the first time in this study at the Endicott SDI and at West Dock. These mussels contained 121 ng/g and 150 ng/g TPAH, respectively. Mussels deployed at West Dock contained the highest TPAH concentrations in mussels deployed in the Beaufort Sea in 2006.

The PAH composition was similar in the reference mussels and the mussels that had been deployed at five sites in the Beaufort Sea in 2006 (Figure 3-28). Naphthalene, phenanthrene, C1- and C2-phenanthrenes, dibenzothiophene, C1- and C2-dibenzothiophenes, fluoranthene, and pyrene were more abundant in reference mussels than in deployed mussels. The deployed mussels contained higher concentrations of perylene, a biogenic PAH, than the reference mussels did.

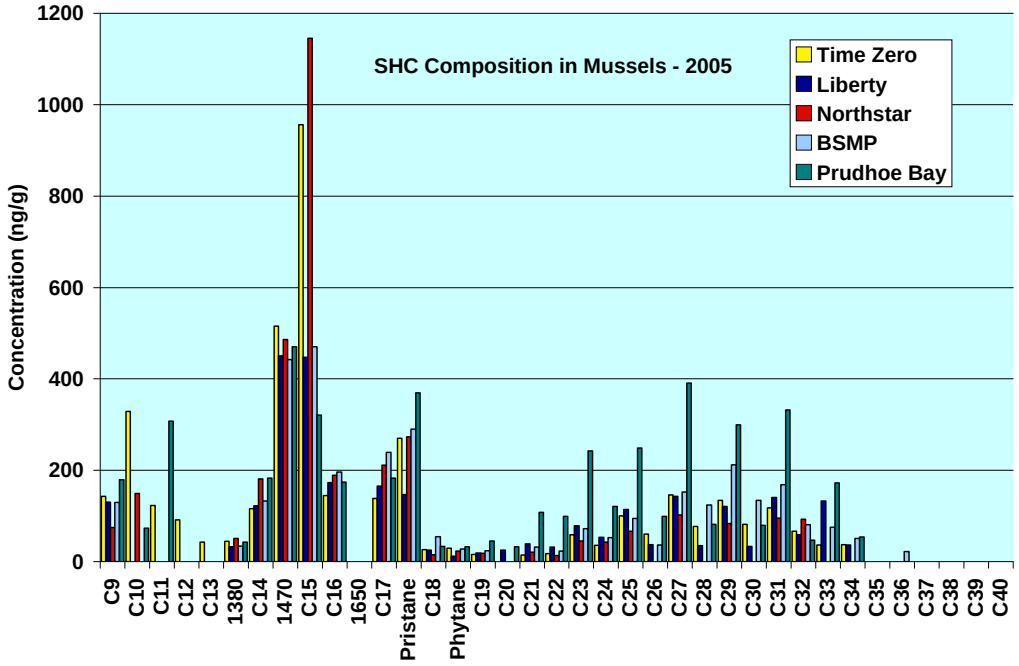


Figure 3-26. Concentrations of individual resolved saturated hydrocarbons (SHC) in soft tissues of mussels that had been deployed in the Beaufort Sea for approximately two weeks in 2005. Data for zero-time reference mussels are included.

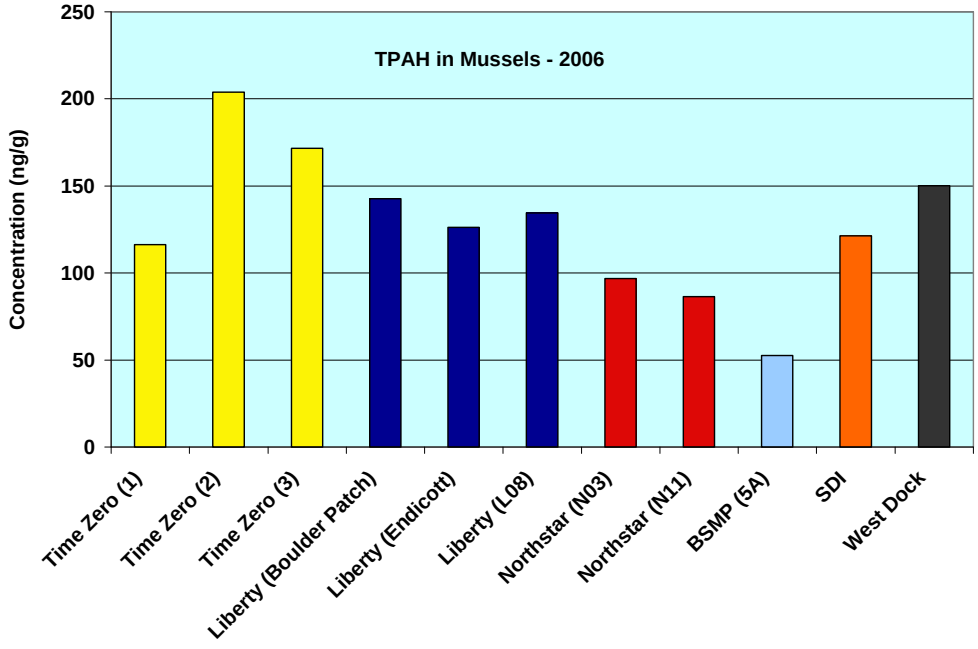


Figure 3-27. Concentrations of total PAH in tissues of reference mussels (Mytilus trossulus) (from the collection site at Port Chatham, AK) and mussels that had been deployed for 13 to 14 days at six Beaufort Sea locations in 2006. Concentrations are ng/g.

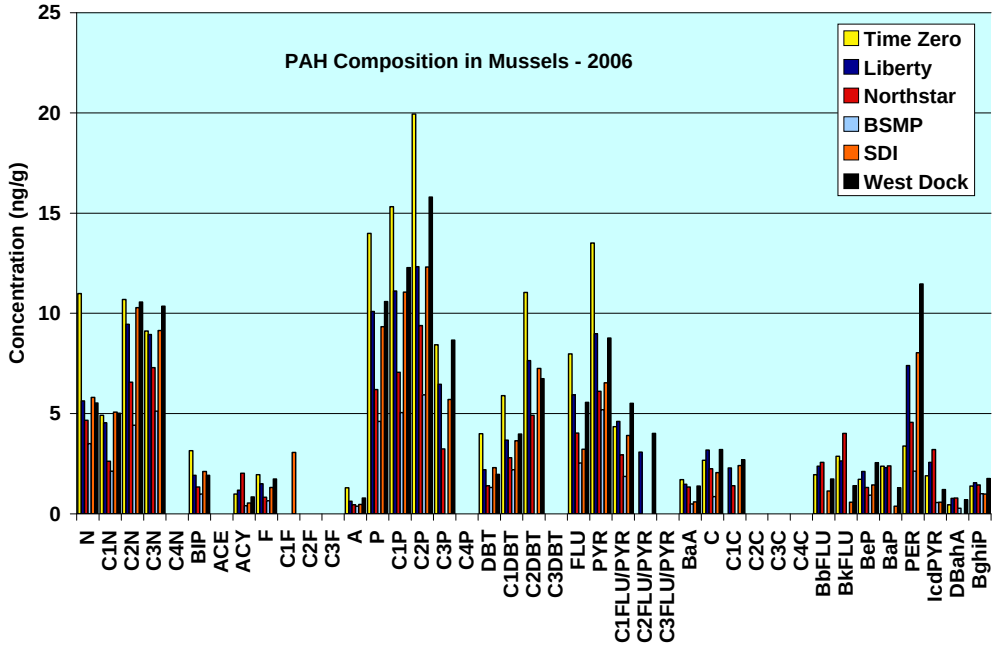


Figure 3-28. Concentrations of individual PAH in reference mussels (Mytilus trossulus) and mussels that had been deployed at five locations in the Beaufort Sea in 2006.

Resolved Saturated Hydrocarbons (SHC)

In 2006, the deployed and reference mussels contained mean resolved total SHC concentrations ranging from 16,033 ng/g dry weight (single sample from BSMP) to 29,134 ng/g in a single sample from West Dock (Table 3-16). The mean concentration of SHC in the reference mussels was 23,159 ± 2294 ng/g, higher than in any of the deployed mussels except those deployed at West Dock. A substantial portion of the SHC in all mussel samples was *n*-paraffins ranging from *n*-C_{23} to *n*-C_{37}. Pristane also was abundant. The *n*-alkanes exhibited a bell-shaped distribution with the highest individual concentrations at n-C_{27} through n-C_{30} (Figure 3-29). The SHC profile was similar in reference and deployed mussels in 2006.

Sterane/Triterpane (StTr) Biomarkers

Sterane/triterpane (StTr) biomarkers were detected in 2006 in only the mussel samples deployed at SDI and West Dock. Concentrations were 101 ng/g and 25.3 ng/g, respectively (Table 3-16). The three most abundant compounds in both samples were the sterane, S28a (an ethylcholestane), and the terpanes, 17a(H),21b(H)-hopane, and 17a(H),21b(H)-30-norhopane, (Figure 3-30). The concentration of S28a in the mussels deployed at SDI and West Dock in 2006 was 101 ng/g and 25.3 ng/g, respectively, the highest concentration of a single StTr detected in mussels in all three years.

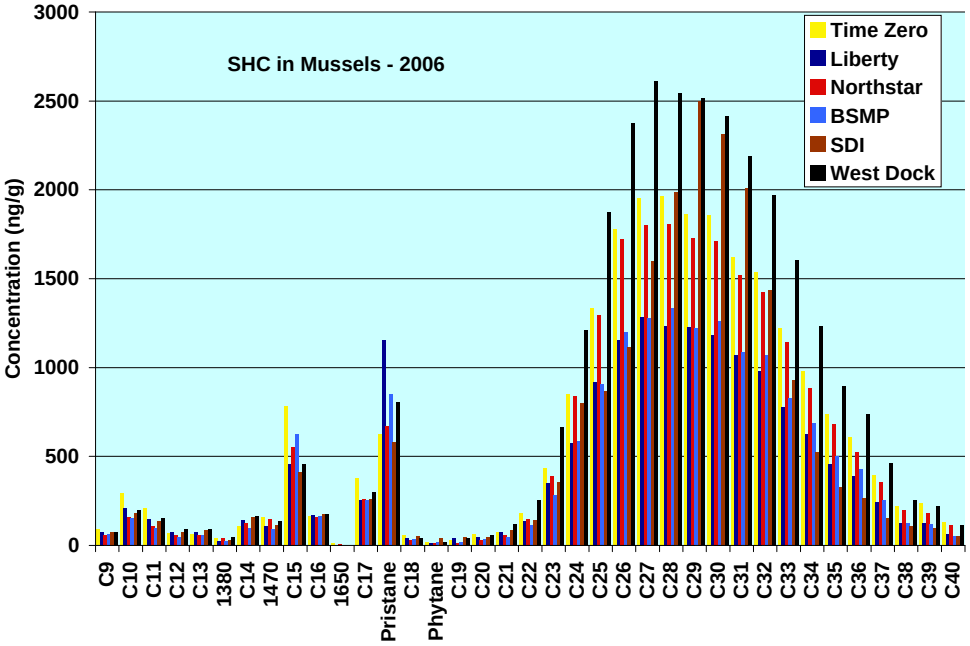


Figure 3-29. Concentrations of individual resolved saturated hydrocarbons (SHC) in soft tissues of mussels that had been deployed in the Beaufort Sea for approximately two weeks in 2006.

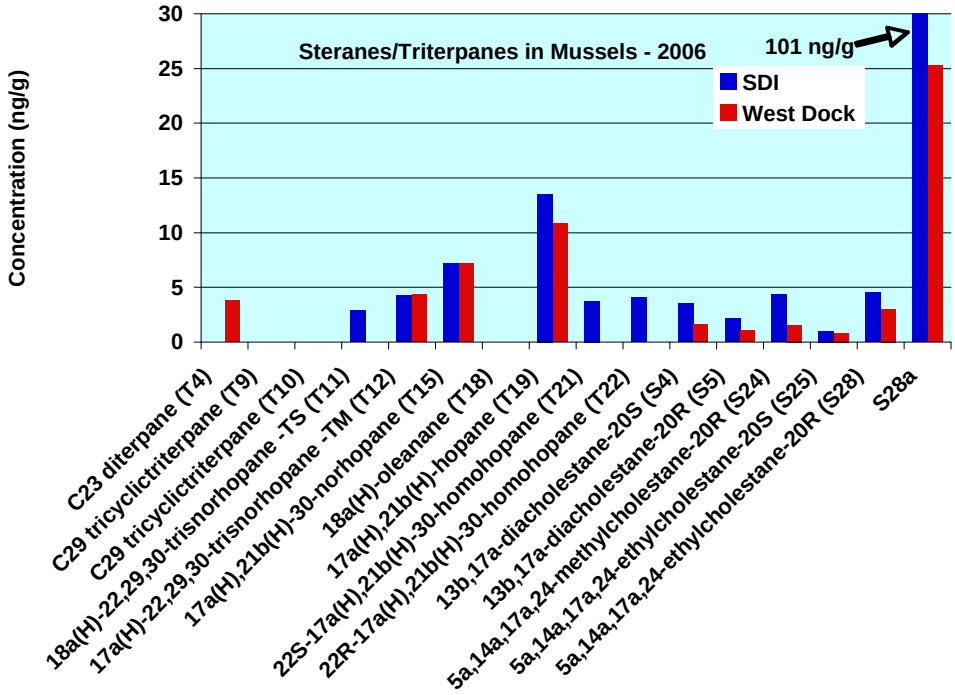


Figure 3-30. Concentrations of individual StTr biomarkers in tissues of mussels (Mytilus trossulus) deployed at SDI and West Dock in 2006. No other mussels collected in 2006 contained detectable concentrations of StTr. Concentrations are ng/g dry wt.

.

3.1.2.4 Inter-annual Variation in Hydrocarbon Concentrations in Mussel Tissues and Comparison to 2002 ANIMIDA Data

Reference Mussels

Time zero reference mussels collected from Port Chatham in lower Cook Inlet contained highly variable concentrations of PAH, resolved SHC, and StTr biomarkers in the 3 years of this monitoring program (Table 3-16), reflecting exposure differences. Three replicate reference samples were collected in 2004 and frozen immediately for analysis. These three replicates contained 195 to 275 ng/g TPAH (mean 227 ± 34.9 ng/g) (Figure 3-31). A single sample of pre-deployment mussels collected in 2002 contained 39.0 ng/g TPAH. Two types of time zero reference samples were collected in 2005. One mussel sample was frozen for analysis immediately after collection at Port Chatham. The other sample was transported to the North Slope with the mussels destined for deployment and then frozen for analysis when the other mussels were deployed. The two samples contained 5.81 ng/g and 59.8 ng/g TPAH, respectively (mean 32.8 ± 27.0 ng/g).

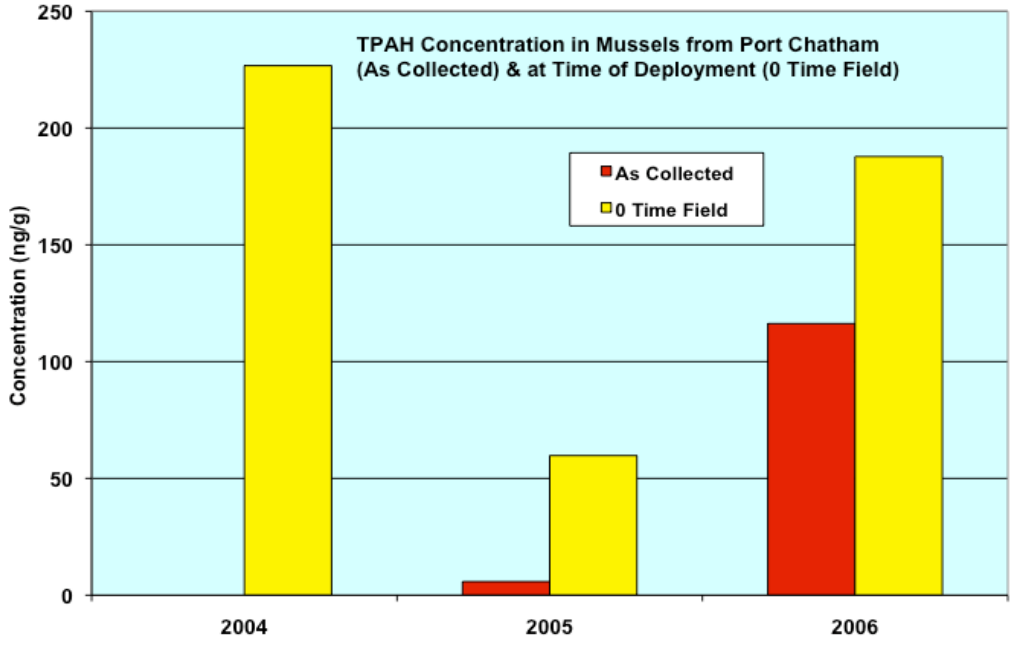


Figure 3-31. Concentrations of TPAH in time zero reference mussels (Mytilus trossulus) collected in 2004, 2005, and 2006 from Port Chatham and either frozen for analysis immediately (as collected) or after transport to the North Slope (time zero field). Concentrations are ng/g dry wt.

Three reference samples were collected in 2006. One sample was frozen for analysis immediately after collection; the other two samples were transported to the North Slope and frozen when the first mussel samples were deployed in the Beaufort Sea in 2006. The first reference sample contained 116 ng/g TPAH; the other two contained 204 and 172 ng/g. Thus,

reference mussels that were transported to the North Slope and held there for a short period of time before freezing contained higher TPAH concentrations than reference mussels that were frozen immediately after collection. Apparently, the mussels became exposed to low level PAH or starved, decreasing tissue mass, during transport and holding.

Mussels also were deployed in the Beaufort Sea in 2002 as part of Task 2 of the ANIMIDA program (Brown et al., 2005). The mussels were from Port Chatham, the same location were mussels used in the cANIMIDA Program were obtained. One reference sample was transported to the North Slope and frozen for analysis when the other mussels were deployed. This reference mussel sample contained 39 ng/g TPAH, a lower concentration than the time zero reference mussel samples from 2004, 2005, and 2006 in the cANIMIDA Program, suggesting changes in the amount of PAH the mussels were exposed to at Port Chatham.

Much smaller differences were observed in TSHC and StTr concentrations between reference mussels that were frozen immediately or transported to the North Slope before freezing. However, SHC concentrations in reference mussels were significantly higher in 2006 (23,159 ± 2294 ng/g) than in 2004 (6051 ± 522 ng/g) or in 2005 (3632 ± 151 ng/g) (Table 3-16). As with TPAH, mean TSHC concentrations in all mussels were lower in 2005 than in 2004 and 2006. Zero-time reference mussels from 2002 contained 19,000 ng/g TSHC, a concentration similar to that of reference mussels collected in 2006, which was lower than concentrations in mussels collected in 2004 and 2005.

StTr biomarkers were detected in reference mussels collected in 2004 and 2005 (12.2 ± 3.79 and 5.87 ± 5.87 ng/g, respectively: Table 3-16), but not in 2006. Reference mussels from 2002 contained 5.0 ng/g StTr. Thus, reference mussels collected at Port Chatham in 2002 as part of the ANIMIDA Program contained lower concentrations of TPAH and similar concentrations of TSHC and StTr compared to zero-time reference mussels collected in 2004, 2005, and 2006 as part of the cANIMIDA Program.

The composition of the PAH and SHC assemblages in reference mussels also varied among the three years of this monitoring program. The inter-annual differences in compositions of the TPAH and TSHC assemblages are partially obscured by the large differences among the three years in TPAH and TSHC concentrations. If concentrations of individual analytes are normalized to the TPAH or TSHC concentration in the sample, to produce profiles of fraction TPAH or TSHC, the differences become more apparent. The PAH profile of the 2004 reference mussels was dominated by naphthalene, which represented more than 20 percent of TPAH (Figure 3-32). The 2005 reference mussels were enriched in C1-C3-naphthalenes, biphenyl, and phenanthrene compared to reference mussels from 2004 and 2006. The 2006 reference mussels were enriched in C1-C2-phenanthrenes and pyrene relative to reference mussels from 2004 and 2005. The PAH profile of the 2002 zero-time reference mussels resembled that of the reference mussels from 2005.

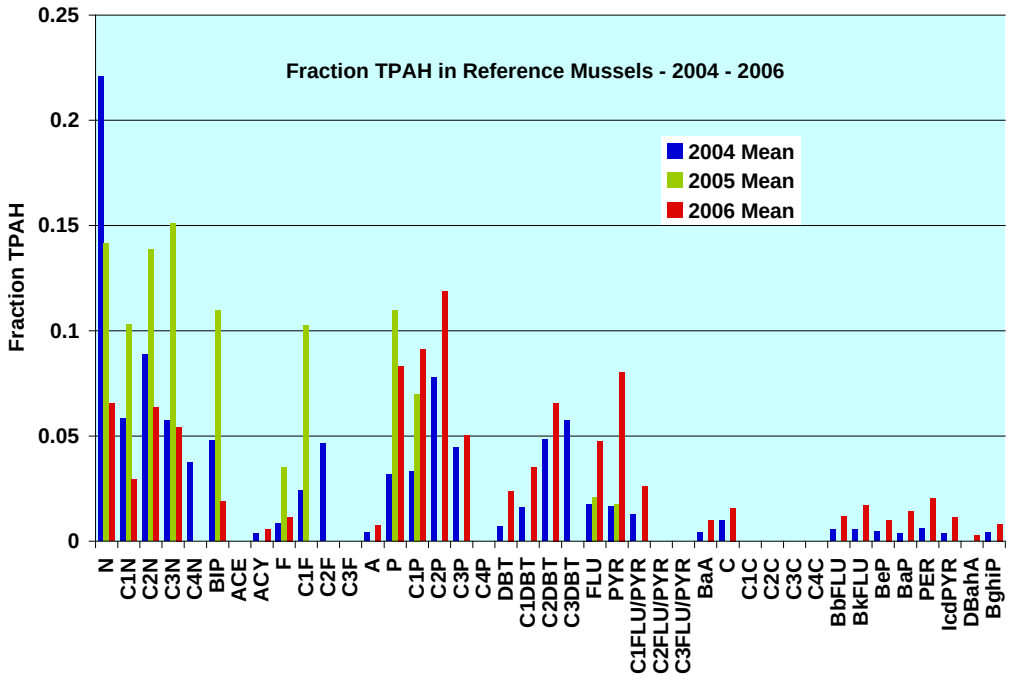


Figure 3-32. Mean fraction TPAH in reference mussels in 2004, 2005, and 2006. Fraction TPAH is the ratio of analyte concentration to TPAH concentration.

Reference mussels collected in 2004 were enriched in the C_{16}, C_{17}, and C_{25} *n*-alkanes, pristane, and phytane, compared to reference mussels from 2005 and 2006 (Figure 3-33). The 2005 reference mussels were enriched in the C_{10} and C_{15} *n*-alkanes and the 1470 isoprenoid alkane. The 2006 reference mussels were enriched in the C_{24} through C_{40} *n*-alkanes, compared to the 2004 and 2005 reference mussels. The SHC profile of the 2002 zero-time reference mussels resembled that of the reference mussels from 2006.

Concentrations and compositions of StTr biomarkers in reference mussels varied widely from year to year. The dominant StTr in reference mussels collected in 2002 and 2004 were the T19-hopane and the T15-norhopane. Only the S28a- ethylcholestane was detected in reference mussels collected in 2005. These large inter-annual variations in the concentrations and compositions of the PAH, SHC, and StTr biomarker assemblages in the reference mussel tissues from the original Port Chatham source makes it difficult, if not impossible to identify bioaccumulation of individual or total PAH, SHC, and StTr biomarkers in mussels that have been deployed in the Beaufort Sea. Longer deployment periods would likely have reduced the impact of residual Port Chatham originating hydrocarbons, and improved the ability to interpret the information from the deployed mussels, but this was unfortunately not logistically possible.

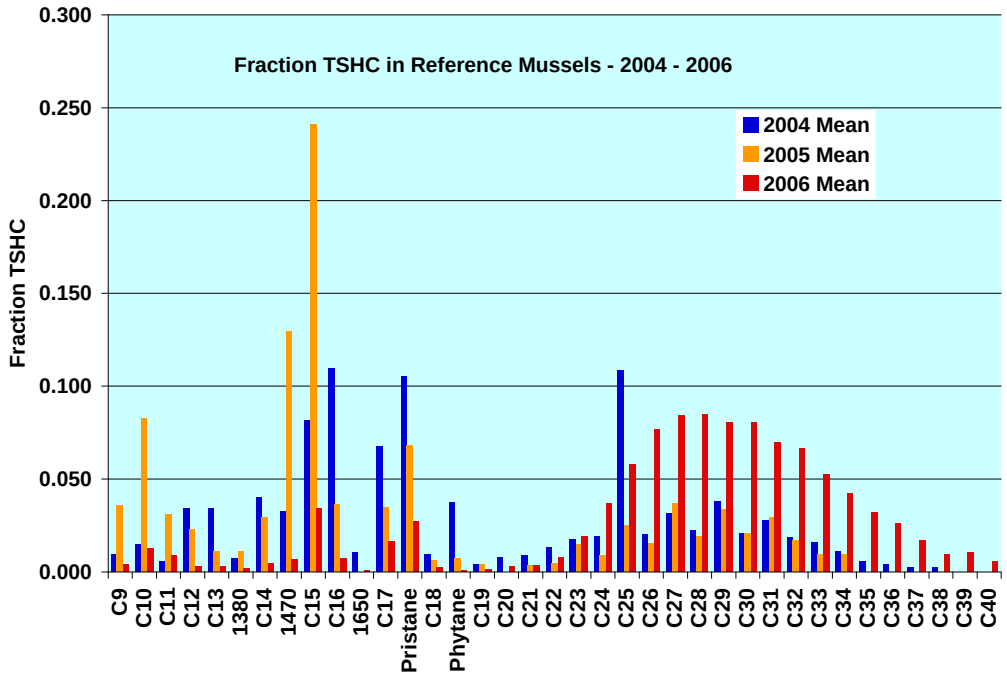


Figure 3-33. Mean fraction TSHC in time zero (reference) mussels in 2004, 2005, and 2006. Fraction TSHC is the ratio of analyte concentration to TSHC concentration.

Deployed Mussels

Concentrations of TPAH, TSHC, and StTr varied widely in mussel samples that were deployed in the Beaufort Sea in 2004, 2005, and 2006 (Table 3-16; Figure 3-34). Concentrations of all three hydrocarbon types in mussel tissues were lower in 2005 than in 2004 and 2006. An ANOVA analysis of the TPAH concentrations in mussels from reference, BSMP, Liberty, and Northstar in 2004, 2005, and 2006 revealed that differences were significant by year ($p<0.001$) and location ($p = 0.03$) but the interaction of year and location was not significant. Duncan's multiple range test revealed that TPAH concentrations in all mussel groups in 2004, 2005, and 2006 were statistically significantly different. Reference mussels contained a significantly different mean TPAH concentration than mussels from all deployment sites in 2004 and 2005, but not in 2006. Mean TPAH concentrations in mussels deployed for 21 days in 2002 approximately 1.5 km west of the Northstar Development and at a reference site approximately 4 km southwest of Pole Island (west of Bullen Point) contained 80 ± 6.0 ng/g and 65 ± 6.5 ng/g TPAH, respectively (Brown et al., 2005), similar to the TPAH concentrations in Northstar mussels in 2006.

Statistical comparisons were not performed for the TSHC and StTr concentrations in mussel tissues. However, the temporal and location differences followed the same trends as the TPAH data. Mussels deployed near the oil production site at Northstar had similar interannual fluctuations and roughly similar concentrations of TPAH, TSHC, and biomarker concentrations as mussels deployed at BSMP and Liberty stations. TSHC and StTr concentrations also were similar in mussels deployed in 2002 at Northstar and a reference site.

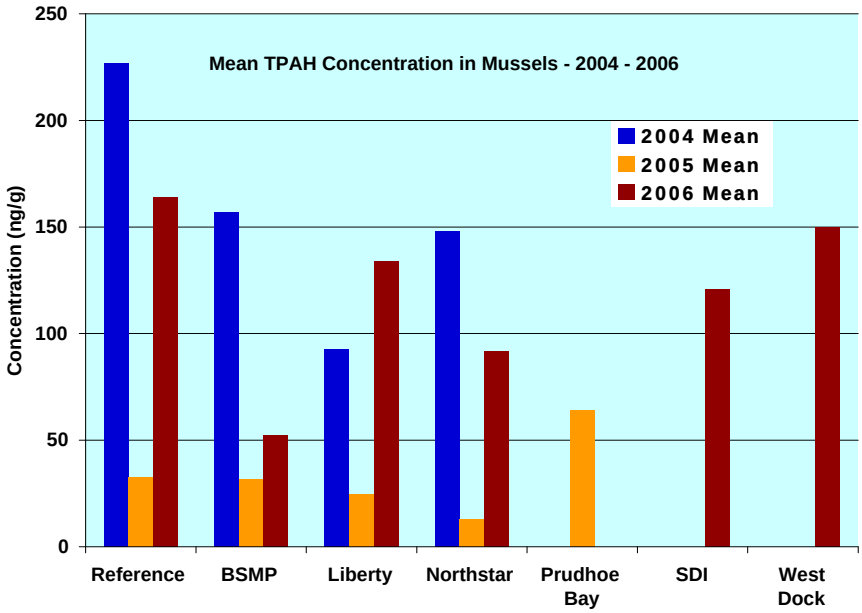


Figure 3-34. Mean TPAH concentrations in reference and deployed mussels in 2004, 2005, and 2006. TPAH concentrations in reference and deployed mussels were significantly different in 2005 from concentrations in 2004 and 2006. Concentrations are ng/g dry wt.

3.1.2.5 PAH in Semipermeable Membrane Devices (SPMDs)

Semipermeable membrane devices (SPMDs) were deployed with mussels at Northstar, Liberty, and the BSMP in 2004. Background concentrations of PAH were determined by analysis of an unused SPMD (unused blank) and an SPMD taken to the field but not deployed (trip blank). The unused blank SPMD contained 754 ng TPAH/SPMD; the trip blank contained 643 ng TPAH/SPMD. The mean TPAH concentration in blank SPMDs was 699 ± 55.0 ng/SPMD (Table 3-17). Mean concentrations of TPAH in the SPMDs that were deployed in the Beaufort Sea in 2004 ranged from 606 ± 156 ng/SPMD at BSMP stations to 945 ng/SPMD at Liberty stations. The overall mean concentration of TPAH in all SPMDs deployed in the Beaufort Sea was 737 ± 170 ng/SPMD, slightly, but not significantly higher than the mean concentration in blank SPMDs. The SPMDs clearly accumulated little PAH from the surrounding waters, and the baseline hydrocarbon concentrations in the SPMDs further confounded the data interpretation.

SPMDs also were deployed near the Northstar Development and at a reference site west of Pole Island in 2002 as part of the ANIMIDA Program. The blank SPMD contained 1604 ng/SPMD TPAH (Table 3-17) more than twice the concentration in the 2004 blanks. SPMDs that had been deployed for 21 days near Northstar and at a reference site west of Pole Island contained similar concentrations, slightly higher than 1000 ng/SPMD, about 30% higher than the concentration in SPMDs that had been deployed at Northstar in 2004.

Table 3-17. Mean and range of concentrations of TPAH in SPMDs blank samples and SPMDs deployed at BSMP stations and near the Liberty Prospect and the Northstar Development in 2004 in the cANIMIDA Program and at near Northstar and a reference site in 2002 in the ANIMIDA Program. Concentrations are ng TPAH/SPMD.

SPMD Sample	No. Samples	Mean ± Standard Deviation	Range
2002 (ANIMIDA)			
Blank	1	1604	1604
Northstar	3	1001 ± 60.4	942 – 1084
Reference	3	1013 ± 55.9	949 – 1085
2004 (cANIMIDA)			
Blanks	2	699 ± 55.0	643 – 754
BSMP	2	606 ± 156	496 – 716
Liberty	1	945	945
Northstar	4	750 ± 182	633 – 1022

The PAH assemblage in the blank SPMDs used in 2004 was dominated by naphthalene and alkyl naphthalenes (Figure 3-35). Naphthalenes also were the most abundant PAH in the SPMDs that had been deployed at Liberty, Northstar, and BSMP and amounts of most three- and four-ring PAH were higher in the deployed than in the blank SPMDs. Amounts of most three- and four-ring PAH ranged from about 10 to 30 ng/SPMD. SPMDs from Liberty and Northstar also contained trace amounts of perylene, a biogenic PAH. Alkyl naphthalenes, fluorenes, phenanthrenes, and dibenzothiophenes usually were more abundant than the parent PAH in both blank and deployed SPMDs. These observations indicate that the SPMDs were accumulating small amounts of petrogenic PAH during deployment. The relatively high and variable background PAH concentrations in blank SPMDs, both SPMDs that had been transported to the field (i.e., field blanks) and those that had not (base SPMD blanks), preclude use of the SPMD data to estimate PAH concentrations in Beaufort Sea water or even to estimate the composition of the PAH assemblage in the water because of the very low concentrations in the water and the minimal accumulation in the deployed SPMDs.

The PAH assemblage in the blank SPMD that was used in 2002 was quite different from that in the 2004 blank SPMDs (Figure 3-36). The blank 2002 blank SPMD contained 1604 ng/SPMD TPAH, 1290 ng/SPMD (80%) of which was naphthalene and C1- and C2-naphthalenes. The blank sample also contained small amounts of C3- and C4-naphthalenes, biphenyl, and phenanthrene. The PAH assemblage was similar in SPMDs that had been deployed near Northstar and at a reference station in 2002. Alkyl naphthalenes, fluorenes, and dibenzothiophenes were the most abundant PAH present in the SPMDs. Phenanthrene and C1- and C2-phenanthrenes also were abundant. The PAH assemblage in the SPMDs deployed at Northstar and the reference site resembled that in a light refined petroleum product.

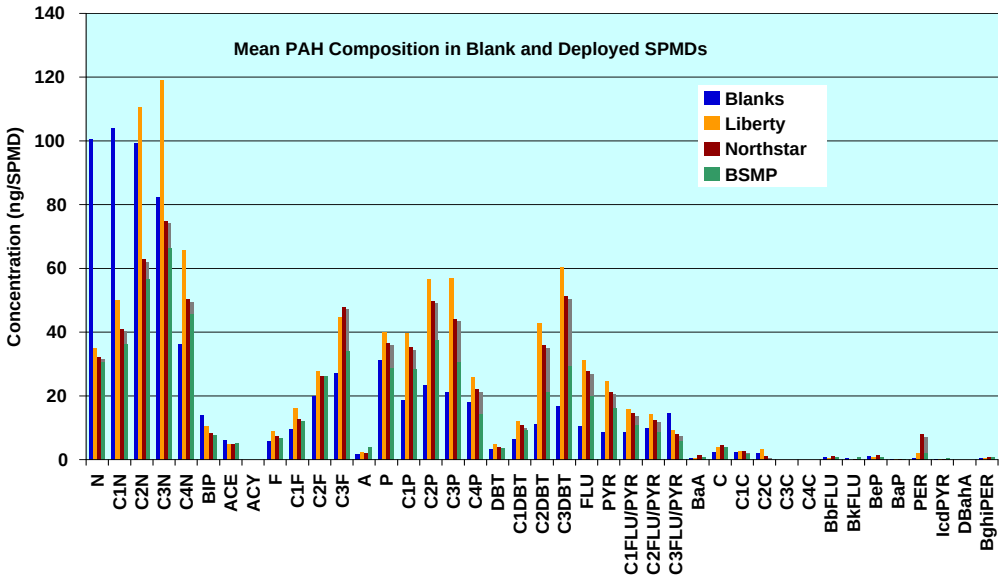


Figure 3-35. The mean composition of the PAH assemblage in blank SPMDs and SPMDs that had been deployed for approximately two weeks in 2004 at three locations in the Beaufort Sea. Concentrations of individual PAH are ng/SPMD.

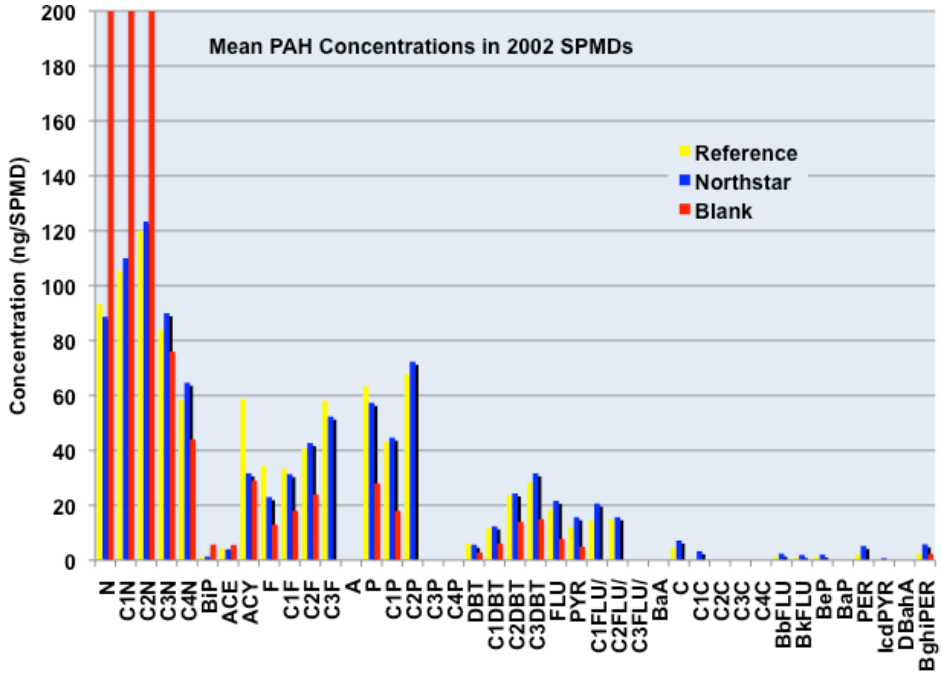


Figure 3-36. The mean composition of the PAH assemblage in a blank SPMD and SPMDs that had been deployed for 21 days in 2002 at Northstar and a reference site in the Beaufort Sea as part of the ANIMIDA Program. Concentrations of N, C1N, and C2N in the blank SPMD are 510, 550, and 230 ng/SPMD, respectively. Concentrations of individual PAH are ng/SPMD.

3.2 Metals in Tissues of Marine Animals

3.2.1 Metals in Fish

3.2.1.1 Summer 2004

Concentrations of 13 metals were measured in seven species of fish collected from locations near the Northstar Development and the Liberty Prospect and at a reference site near Tigvariak Island. Metal concentrations in fish tissue are summarized by species in Table 3-18. Mean concentrations of each metal varied by a factor of up to about ten among the seven species of fish. Variance within a species usually was much lower. The most variable mean concentrations were the lowest ones. Arctic cod and four-horn sculpin contained the highest concentrations of most metals. Arctic flounder also contained elevated concentrations of some metals. These three species are demersal and probably accumulated metals by ingestion of metal-rich sediment particles or benthic fauna. Concentrations of As and Ni were significantly higher in tissues of least cisco from Northstar than in those from Liberty; concentrations of Ba, Hg, Se, and Zn were significantly higher in tissues of Arctic cisco from Liberty than in those from Northstar (Table 3-18).

Table 3-18. Concentrations of 13 metals in whole samples of seven species of fish collected from the Beaufort Sea study area during the summer of 2004. n = number of replicates. Concentrations are μg/g dry wt. Metals concentrations that were significantly different in a fish species at different sampling locations are highlighted.

Metal	Parameter	Arctic Char (n= 2-3)	Arctic Cisco (n= 4-5)	Arctic Cod (n= 4-5)	Arctic Flounder (n= 2)	Broad White-fish (n=2)	Four Horn Sculpin (n=5-6)	Least Cisco (n= 6)
Ag	Mean±SD	0.04±0.01	0.02±0.01	0.10±0.03	0.03±0.01	0.07±0.06	0.19±0.10	0.02±0.01
	Range	0.03-0.05	0.01-0.03	0.06-0.14	0.02-0.03	0.02-0.11	0.06-0.35	0.01-0.04
As	Mean±SD	2.51±0.80	3.05±0.52	9.45±5.33	5.74±0.88	1.43±0.44	4.87±2.32	**2.98±0.91**[a]
	Range	1.59-3.03	2.57-3.79	2.07-16.2	5.12-6.36	1.12-1.74	3.40-9.47	**1.56-3.94**
Ba	Mean±SD	1.9±0.3	**2.5±4.3**[b]	4.0±2.3	5.6±2.1	2.7±0	9.3±4.1	1.3±1.0
	Range	1.7-2.1	**0.3-8.9**	1.7-6.9	4.1-7.0	2.7-2.7	3.0-14.2	0.5-3.0
Cd	Mean±SD	0.10±0.04	0.07±0.04	0.17±0.11	0.04±0.02	0.05±0.03	0.18±0.09	0.06±0.06
	Range	0.05-0.13	0.03-0.14	0.05-0.27	0.03-0.06	0.03-0.07	0.05-0.32	0.02-0.18
Cr	Mean±SD	0.12±0.04	0.19±0.20	0.53±0.40	0.57±0.12	0.32±0.07	0.34±0.54	0.11±0.03
	Range	0.09-0.17	0.05-0.54	1.14±0.12	0.48-0.65	0.27-0.37	0.22-1.17	0.16±0.07
Cu	Mean±SD	4.5±0.3	3.0±0.5	4.5±2.2	3.4±0.9	3.7±0.7	11.1±4.1	2.3±0.5
	Range	4.2-4.7	2.4-3.6	2.2-7.9	2.7-4.0	3.2-4.2	6.1-18.2	1.5-2.9
Fe	Mean±SD	54.5±17.3	63.0±30.1	172±165	90.4±68.7	162±26.2	252±161	56.7±15.5
	Range	37.7-72.2	40.0-112	37.2-424	41.8-139	143-180	114-541	44.5-87.1
Hg	Mean±SD	0.06±0.02	**0.06±0.02**[b]	0.03±0.02	0.17±0.09	0.07±0.04	0.21±0.13	0.14±0.06
	Range	0.05-0.09	**0.04-0.08**	0.02-0.06	0.11-0.23	0.05-0.10	0.10-0.45	0.05-0.20
Ni	Mean±SD	0.06±0.02	0.19±0.10	0.60±0.24	0.36±0.16	0.20±0.03	0.48±0.33	**0.19±0.10**[a]
	Range	0.05-0.08	0.11±0.37	0.30-0.91	0.24-0.47	0.18-0.22	0.17-0.92	**0.09-0.35**

[a] Northstar > Liberty; [b] Liberty > Northstar.

Table 3–18. Concentrations of 13 metals in whole samples of seven species of fish collected from the Beaufort Sea study area during the summer of 2004. n = number of replicates. Concentrations are μg/g dry wt. Metals concentrations that were significantly different in a fish species at different sampling locations are highlighted, continued.

Metal	Parameter	Arctic Char (n= 2-3)	Arctic Cisco (n= 4-5)	Arctic Cod (n= 4-5)	Arctic Flounder (n= 2)	Broad White-fish (n=2)	Four Horn Sculpin (n=5-6)	Least Cisco (n= 6)
Pb	Mean±SD	0.19±0.13	0.07±0.04	0.30±0.19	0.11±0.06	0.11±0.10	0.22±0.09	0.10±0.05
	Range	0.11-0.33	0.02-0.12	0.09-0.57	0.07-0.15	0.03-0.18	0.07-0.32	0.04-0.18
Se	Mean±SD	3.69±1.23	**1.94±0.46**[b]	3.34±0.99	3.19±0.27	2.64±0.45	4.10±1.03	2.30±0.23
	Range	2.71-5.07	**1.57-2.53**	1.73-4.12	3.0-3.38	2.32-2.96	2.49-5.66	1.89-2.59
V	Mean±SD	0.14±0.05	0.15±0.15	1.12±1.18	0.55±0.14	0.57±0.09	1.32±1.08	0.13±0.06
	Range	0.10-0.19	0.05-0.40	0.19-3.02	0.45-0.65	0.50-0.63	0.34-3.38	0.05-0.19
Zn	Mean±SD	72.0±22.3	**58.8±21.2**[b]	82.9±22.1	91.6±16.1	50.5±12.3	94.0±13.1	66.5±26.1
	Range	53.1-96.6	**36.0-82.5**	44.5-97.8	103-80.2	41.8-59.2	70.8-106	38.7-109

[a] Northstar > Liberty; [b] Liberty > Northstar.

The data from different species also were evaluated together, based on location, to summarize overall mean concentrations in each area (Figures 3-37 and 3-38). There was little variation in concentrations of the 13 metals in tissues of the seven species of fish at the three sampling areas, Northstar, Liberty, and Tigvariak. There were no significant differences among the three sampling areas in the concentration of any metal in all fish species combined, because of the large standard deviations around the means. Concentrations of Cu and Fe were slightly, but not significantly, higher in fish from Northstar (the production site) than in fish from Liberty and Tigvariak. The mean Ba concentration was slightly, but not significantly, higher in fish from Liberty. Metals present at highest concentrations in all fish tissues combined were Fe (range 37.2 to 541 μg/g) and Zn (range 36.0 to 109 μg/g) (Table 3-18). Concentrations of Ag, Cd, and Hg usually were below 0.1 μg/g (range 0.01 to 0.45 μg/g) (Table 3-18).

3.2.1.2 Summer 2005

Mean metals concentrations in tissues of five species of fish collected near Northstar and Liberty in 2005 were similar to those in the same species of fish collected in 2004 (Table 3-19). However, mean concentrations of six metals (Ba, Cd, Cr, Cu, Fe, and Ni) were higher in four-horn sculpin collected in 2005 than in sculpin collected in 2004. These metals may have been associated with ingested sediment particles. Concentrations of these metals were similar in the other fish species collected in both 2004 and 2005.

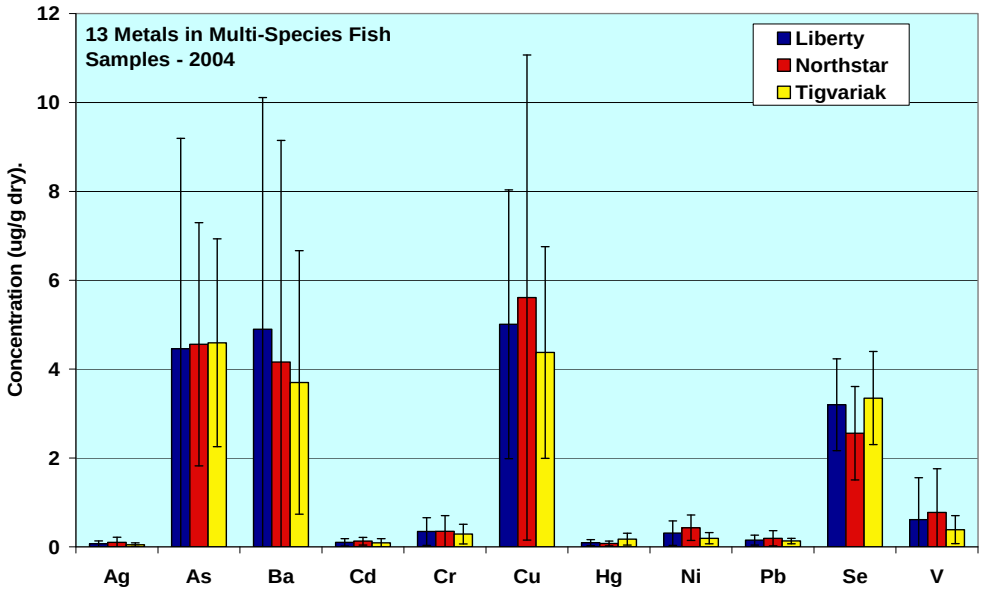


Figure 3-37. Mean and standard deviations concentrations of 11 metals in tissues of seven species of fish collected during the summer of 2004 from Liberty and Northstar prospects and from Tigvariak, east of Liberty. Concentrations are µg/g dry wt.

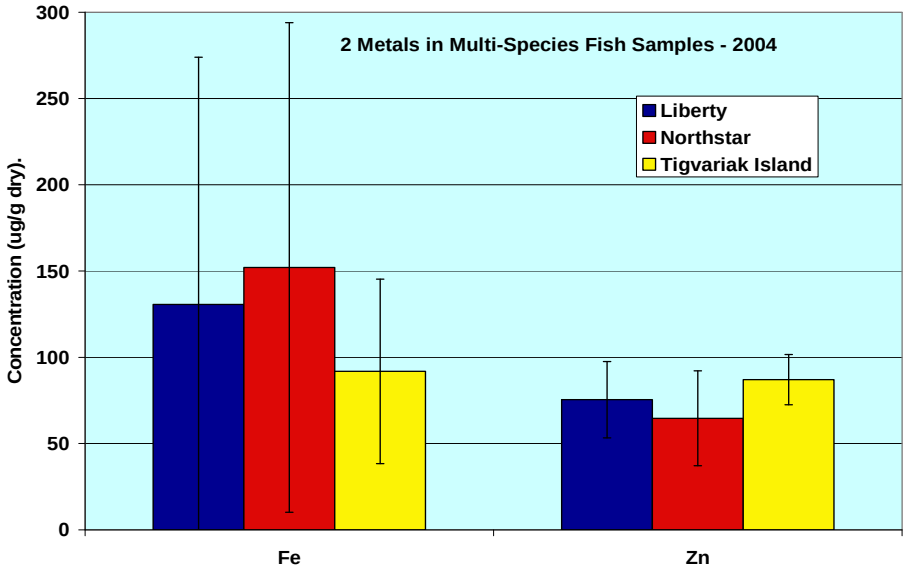


Figure 3-38. Mean and standard deviations concentrations of iron and zinc in tissues of five species of fish collected during the summer of 2004 from Liberty and Northstar prospects. Concentrations are µg/g dry wt.

Table 3-19. Concentrations of 13 metals in whole fish samples of seven species collected from the Beaufort Sea study area during the summer of 2005. n = number of replicates. Concentrations are μg/g dry wt.

Metal	Parameter	Arctic Char (n=3)	Arctic Cisco (n= 3)	Arctic Flounder (n= 2)	Four Horn Sculpin (n= 2)	Humpback Broad Whitefish (Liberty) (n= 5)	Humpback Broad Whitefish (Northstar) (n= 5)
Ag	Mean±SD	0.03±0.01	0.01±0.00	0.03±0.02	0.22±0.19	0.02±0.00	0.02±0.01
	Range	0.03-0.04	0.01-0.01	0.02-0.04	0.08-0.35	0.01-0.02	0.01-0.04
As	Mean±SD	4.39±2.64	3.23±0.58	3.24±0.23	3.38±0.66	1.61±0.51	1.29±0.18
	Range	2.28-7.35	2.59-3.72	3.08-3.40	2.91-3.85	0.81-2.12	1.00-1.45
Ba	Mean±SD	1.1±1.0	2.7±1.3	10.1±7.8	32.7±19.7	5.7±2.8	3.5±3.4
	Range	0.5-2.2	1.2-3.5	4.5-15.6	18.7±46.6	3.4-10.2	0.7-9.3
Cd	Mean±SD	0.04±0.02	0.02±0.01	0.03±0.01	0.29±0.11	0.03±0.02	0.05±0.02
	Range	0.02-0.06	0.01-0.03	0.03±0.04	0.21-0.37	0.02-0.06	0.03-0.08
Cr	Mean±SD	0.29±0.40	0.07±0.06	0.47±0.45	2.21±2.27	0.18±0.14	0.28±0.36
	Range	0.04-0.75	0.03-0.13	0.15-0.78	0.60-3.81	0.07-0.38	0.05-0.92
Cu	Mean±SD	4.2±0.6	2.2±1.0	3.1±1.4	13.0±11.7	2.3±0.7	2.9±1.5
	Range	3.7-4.8	1.1-2.8	2.1-4.1	4.7-21.2	1.8-3.3	2.1-5.6
Fe	Mean±SD	53.6±2.1	43.5±22.1	121±127	656±841	86.1±64.1	147±202
	Range	51.3-55.2	19.4-62.9	31.9-211	61.3-1250	36.9-193	30.8±507
Hg	Mean±SD	0.06±0.02	0.06±0.00	0.08±0.01	0.12±0.13	0.15±0.08	0.12±0.05
	Range	0.05-0.07	0.06±0.06	0.07-0.08	0.03-0.21	0.07-0.27	0.07-0.20
Ni	Mean±SD	0.05±0.03	0.11±0.04	0.33±0.21	1.12±1.0	0.21±0.06	0.27±0.16
	Range	0.03-0.08	0.07-0.15	0.18-0.47	0.41-1.82	0.16-0.32	0.14-0.53
Pb	Mean±SD	0.11±0.02	0.06±0.03	0.10±0.01	0.46±0.48	0.07±0.03	0.11±0.09
	Range	0.09-0.13	0.03-0.10	0.10-0.11	0.12-0.79	0.03-0.11	0.03-0.26
Se	Mean±SD	2.89±0.36	1.47±0.47	3.08±0.33	3.50±0.21	2.96±0.50	2.40±0.31
	Range	2.49-3.18	0.94-1.83	2.84-3.31	3.35-3.64	2.46-3.52	2.21-2.95
V	Mean±SD	0.14±0.02	0.10±0.04	0.32±0.32	2.42±2.12	0.34±0.20	0.28±0.26
	Range	0.12-0.15	0.08-0.15	0.09-0.54	0.92-3.92	0.16-0.56	0.12±0.74
Zn	Mean±SD	93.8±30.8	69.2±25.5	106±10.2	91.8±2.7	75.6±24.4	51.4±14.8
	Range	59.5-119	40.9-90.5	98.6-113	89.9-93.7	51.2-115	42.4-77.7

Humpback broad whitefish was the only species for which more than one sample was collected at both Northstar and Liberty in 2005. Five replicate whitefish samples were collected at each location. Mean concentrations of all 13 metals were similar in humpback broad whitefish from Northstar and Liberty (Table 3-19).

When metals data for all five fish species are combined, mean concentrations of each metal are roughly similar in fish from Northstar and Liberty (Figures 3-39 and 3-40). There were no significant differences among sampling areas in 2005 because of the large standard deviations around the means. Mean concentrations of Ba, Cu, and Fe were higher though not significantly

so, in fish from Northstar than in those from Liberty, due mainly to the high concentrations of these metals in the two samples of four horn sculpin collected at Northstar. The excess Ba and Fe in four horn sculpin probably are from ingestion of sediments; Cu concentration was higher in all three years in four horn sculpin than in the other fish species, perhaps due to consumption of benthic crustaceans, often rich in Cu.

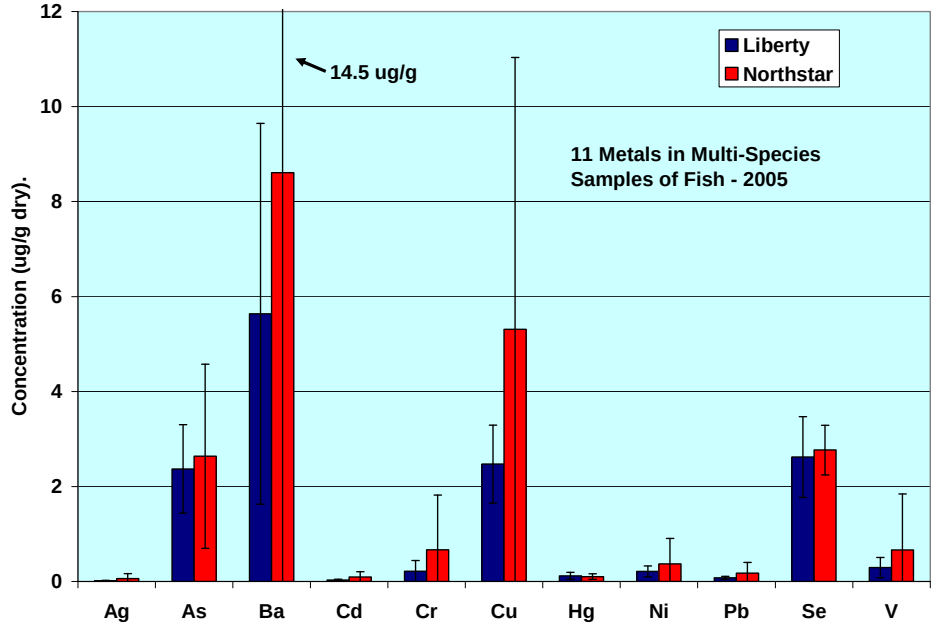


Figure 3-39. Mean and standard deviations of concentrations of 11 metals in tissues of five species of fish collected during the summer of 2005 from Liberty and Northstar prospects. Concentrations are µg/g dry wt.

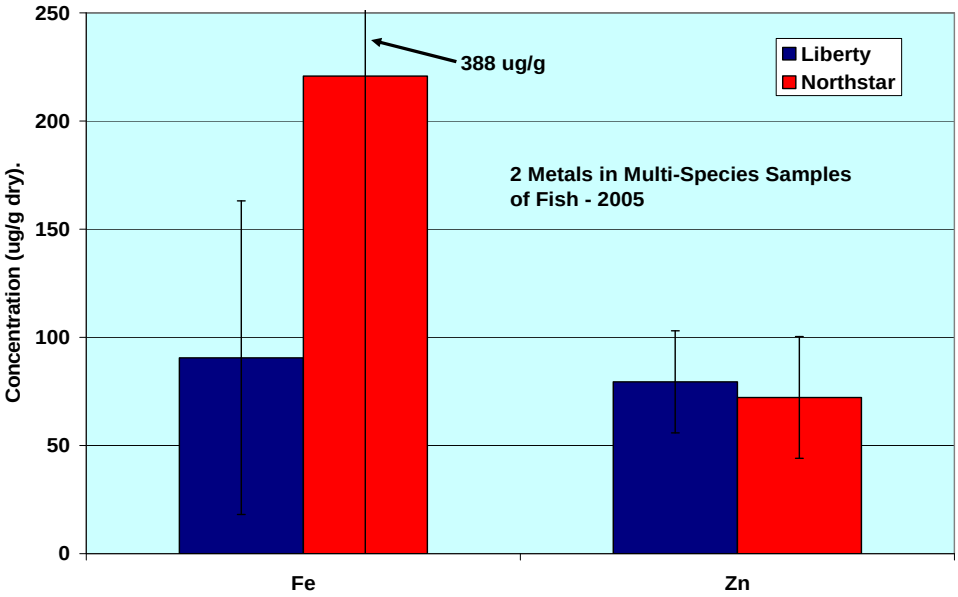


Figure 3-40. Mean and standard deviations of concentrations of iron and zinc in tissues of five species of fish collected during the summer of 2005 from Liberty and Northstar prospects. Concentrations are µg/g dry wt.

There were significant differences in concentrations of Ag, Cd, and Cu in combined fish samples collected in 2004 and 2005 at Liberty and of As in combined fish samples collected in 2004 and 2005 at Northstar (Table 3-20). In all cases, metals concentrations were higher in fish collected in 2004 than in 2005. More four horn sculpin (containing high concentrations of Ag, Cd, and Cu) were sampled in 2004 than in 2005, explaining the higher concentration of these metals in combined fish samples from Liberty in 2004 than in 2005; arctic cod (contain a high concentration of As, probably from consumption of oceanic copepods) were collected in 2004 but not 2005, explaining the higher concentration of As in combined fish samples from Northstar in 2004 than in 2005.

Table 3-20. Results of statistical analysis of differences in concentrations of 13 metals in combined fish samples collected from Liberty and Northstar in 2004 and 2005. Significant between-year differences ($p < 0.05$) are highlighted.

Liberty							
Parameter	**2004**			**2005**			**p-Value**
	N	**Mean**	**S.D.**	**N**	**Mean**	**S.D.**	
Ag	11	0.069	0.065	10	0.017	0.010	**0.02**
As	11	4.46	4.73	10	2.42	0.962	0.27
Ba	11	3.93	3.19	10	5.64	4.23	0.37
Cd	11	0.103	0.083	10	0.029	0.013	**0.004**
Cr	11	0.345	0.311	10	0.202	0.231	0.08
Cu	11	5.01	3.02	10	2.45	0.857	**0.004**
Fe	11	131	143	10	80.4	67.5	0.20
Hg	11	0.095	0.068	10	0.110	0.071	0.41
Ni	11	0.309	0.275	10	0.204	0.115	0.53
Pb	11	0.153	0.112	10	0.075	0.031	0.19
Se	11	3.20	1.03	10	2.54	0.845	0.12
V	11	0.615	0.942	10	0.265	0.204	0.34
Zn	11	75.4	22.1	10	79.7	24.9	0.73
Northstar							
Parameter	**2004**			**2005**			p-Value
	N	**Mean**	**S.D.**	**N**	**Mean**	**S.D.**	
Ag	10	0.102	0.114	10	0.062	0.104	0.18
As	10	4.56	2.74	10	2.64	1.94	**0.03**
Ba	10	4.39	5.23	10	8.61	14.5	0.48
Cd	10	0.128	0.086	10	0.093	0.112	0.20
Cr	10	0.35	0.355	10	0.67	1.15	0.85
Cu	10	5.61	5.46	10	5.31	5.73	0.95
Fe	10	152	142	10	221	389	0.89
Hg	10	0.077	0.053	10	0.103	0.061	0.26
Ni	10	0.43	0.287	10	0.373	0.533	0.21
Pb	10	0.194	0.171	10	0.178	0.224	0.67
Se	10	2.56	1.05	10	2.768	0.523	0.34
V	10	0.775	0.982	10	0.665	1.18	0.95
Zn	10	64.6	27.5	10	72.2	28.1	0.51

3.2.1.1 Summer 2006

The mean metal concentrations in fish collected in 2006 were similar to those observed during previous years. The elevated mean metal concentrations in four horn sculpin were not as apparent in 2006 and the data were similar to those observed during 2004. Multiple samples of both least cisco and four horn sculpin were collected from both Liberty and Northstar during 2006; there usually was little difference observed between the locations for either species (Table 3-21). However, the concentration of Ni was significantly higher ($p = 0.035$) in least cisco from Northstar than in least cisco from Liberty. The differences are small and may be related to contact with stainless steel in lab processing.

Table 3-21. Concentrations of 13 metals in whole samples of species of fish collected from the Beaufort Sea study area during the summer of 2006. n = number of replicates. Concentrations are μg/g dry wt. Metals concentrations that were significantly different in tissues of a species from Liberty and Northstar are highlighted

Metal	Parameter	Arctic Flounder (n= 2)	Four Horn Sculpin (Liberty) (n= 3)	Four Horn Sculpin (Northstar) (n= 2)	Humpback Broad Whitefish (Liberty) (n=3)	Least Cisco (Liberty) (n= 3)	Least Cisco (Northstar) (n= 4)
Ag	Mean±SD	0.009±0.007	0.07±0.02	0.10±0.02	0.006±0.003	0.005±0.001	0.02±0.01
	Range	0.004-0.014	0.06-0.09	0.09-0.11	0.004-0.009	0.004-0.006	0.01-0.03
As	Mean±SD	4.79±2.09	3.49±0.56	2.36±1.03	1.84±0.42	1.98±0.65	2.12±0.16
	Range	3.31-6.27	2.92-4.04	1.63-3.08	1.54-2.33	1.55-2.73	1.96-2.32
Ba	Mean±SD	8.64±1.28	6.84±2.21	7.98±1.00	2.70±1.01	1.67±0.55	2.36±0.88
	Range	7.73-9.54	4.28-8.17	7.27-8.68	1.68-3.69	1.13-2.22	1.09-3.06
Cd	Mean±SD	0.07±0.02	0.18±0.08	0.08±0.003	0.03±0.02	0.029±0.001	0.03±0.01
	Range	0.06-0.09	0.11-0.27	0.076-0.08	0.02-0.05	0.028-0.03	0.03-0.04
Cr	Mean±SD	0.51±0.13	0.29±0.12	0.43±0.21	0.17±0.06	0.15±0.05	0.14±0.09
	Range	0.41-0.6	0.21-0.43	0.28-0.58	0.11-0.22	0.1-0.18	0.08-0.26
Cu	Mean±SD	2.9±0.4	6.6±0.3	7.5±0.4	2.5±0.7	2.2±0.2	3.3±1.0
	Range	2.6-3.1	6.3-6.9	7.2-7.8	1.8-3.2	2.0-2.3	2.4-4.7
Fe	Mean±SD	149±141	111±19.3	123±40.7	81.4±44.1	43.9±4.9	54.1±12.9
	Range	49-249	90.6-129	94.5-152	51.3-132	38.7-48.4	40.8-65.8
Hg	Mean±SD	0.15±0.07	0.29±0.11	0.12±0.001	0.09±0.01	0.09±0.02	0.09±0.03
	Range	0.098-0.198	0.20-0.41	0.121-0.123	0.08-0.11	0.07-0.10	0.046-0.116
Ni	Mean±SD	0.75±0.02	0.55±0.11	0.67±0.09	0.24±0.06	**0.25±0.03**	**0.37±0.07**
	Range	0.73-0.76	0.43-0.63	0.6-0.73	0.19-0.31	**0.23-0.28**	**0.28-0.44**
Pb	Mean±SD	0.09±0.05	0.08±0.02	0.1±0.01	0.04±0.02	0.008±0.002	0.01±0.01
	Range	0.06-0.13	0.06-0.09	0.09-0.11	0.01-0.05	0.006-0.01	0.01-0.03
Se	Mean±SD	4.42±0.16	4.32±0.86	3.44±0.09	3.00±0.23	2.6±0.53	2.59±0.22
	Range	4.31-4.53	3.54-5.24	3.37-3.5	2.75-3.21	2.13-3.17	2.39-2.79
V	Mean±SD	2.92±2.97	2.52±0.43	2.88±1.15	0.85±0.29	0.46±0.15	0.36±0.08
	Range	0.82-5.02	2.05-2.88	2.06-3.69	0.52-1.08	0.33-0.63	0.25-0.43
Zn	Mean±SD	80.0±10.9	92.3±9.80	94.7±14.6	56.8±12.8	80.3±5.6	73.4±7.0
	Range	72.3-87.7	83.8-103	84.3-105	47.2-71.3	74.1-85	63-78.2

When metals concentrations in all four fish species collected in 2006 were combined, the concentration of each metal was similar in fish from Liberty and Northstar (Figures 3-41 and 3-42). Mean concentrations of Ba, Cu, and Fe were slightly, but not significantly, higher in combined fish samples from Northstar than in those from Liberty. As in 2004 and 2005, concentrations of Ag, Cd, Hg, and Pb in combined fish tissues were very low, ranging from 0.001 to 0.41 µg/g (Table 3-21).

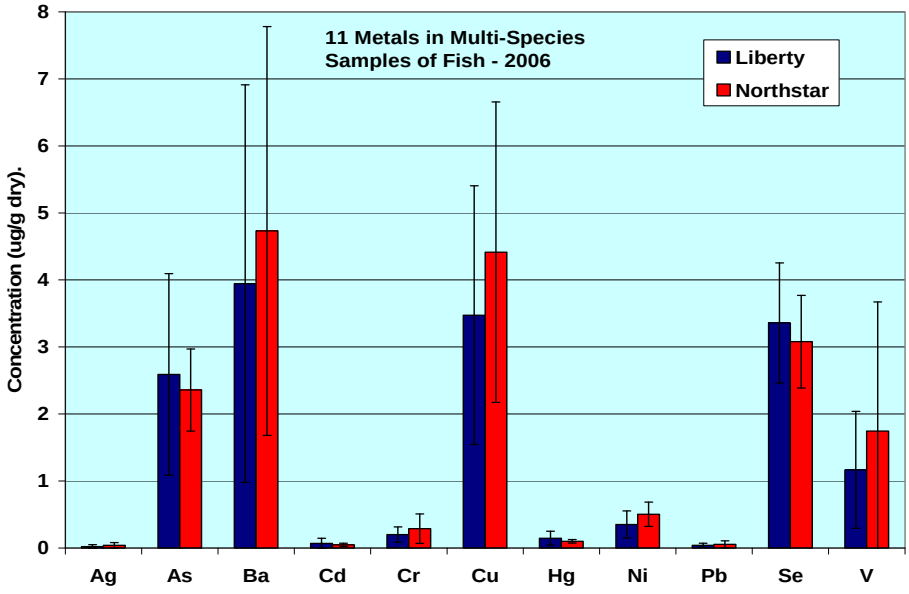


Figure 3-41. Mean and standard deviations of concentrations of 11 metals in tissues of five species of fish collected during the summer of 2006 from Liberty and Northstar prospects. Concentrations are µg/g dry wt.

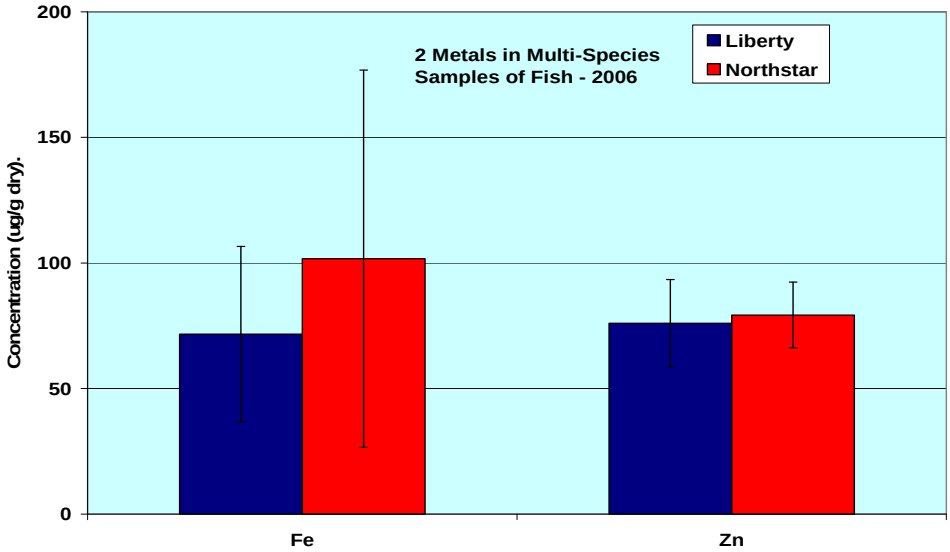


Figure 3-42. Mean and standard deviations of concentrations of iron and zinc in tissues of five species of fish collected during the summer of 2006 from Liberty and Northstar. Concentrations are µg/g dry wt.

Concentrations of all metals were similar in all species collected at Liberty and Northstar in 2004, 2005, and 2006. When combined fish tissue data for 2005 and 2006 were combined, concentrations of Ag and Cu were significantly higher in fish from Northstar than in those from Liberty. The concentration of Se is significantly higher in fish collected in 2006 than in those collected in 2005. However, when combined fish tissue data for 2004, 2005, and 2006 were compared, there were no significant differences by year or location.

3.2.2 Metals in Amphipods, Isopods, and Clams

3.2.2.1 Summer of 2004

Amphipods

Amphipods (*Anonyx nugax*) were collected in 2004 at three stations near the Northstar Development, two stations near the Liberty Prospect, and at three stations in the BSMP area (Appendix A, Table A-5). Concentrations of most metals, except Cd, were lower in amphipods collected at the BSMP stations than in amphipods collected at Northstar and Liberty in 2004 (Table 3-22). Concentrations of several metals were highest in amphipods from Northstar. The

Table 3-22. Concentrations of 18 metals in amphipods (*Anonyx nugax*) collected from three locations in the Beaufort Sea in 2004. n= number of samples. Concentrations are μg/g dry wt (ppm).

Metal	Parameter	Northstar (n= 3)	Liberty (n= 2)	BSMP (n= 3)
Ag	Mean±SD	4.05±1.69	2.98±0.07	2.41±0.51
	Range	2.75 - 5.96	2.91 - 3.04	1.85 - 2.84
Al	Mean±SD	444±257	531±33	278±101
	Range	271 - 740	494 - 555	164 - 358
As	Mean±SD	12.79±2.98	7.24±0.57	7.44±1.58
	Range	9.97 - 15.9	6.59 - 7.57	6.39 - 9.26
Ba	Mean±SD	34.8±14.1	23.9±1.1	20.1±13.5
	Range	22.8 - 50.4	23.0 - 25.1	10.7 - 35.6
Be	Mean±SD	0.011±0.007	0.011±0.002	0.008±0.003
	Range	0.007 - 0.019	0.010 - 0.013	0.005 - 0.01
Cd	Mean±SD	0.805±0.460	0.590±0.019	1.124±0.808
	Range	0.434 - 1.32	0.569 - 0.605	0.559 - 2.05
Co	Mean±SD	2.62±0.74	2.36±0.13	1.82±0.78
	Range	2.16 - 3.48	2.21 - 2.44	0.95 - 2.44
Cr	Mean±SD	0.62±0.21	0.86±0.17	0.46±0.13
	Range	0.43 - 0.85	0.66 - 0.96	0.35 - 0.6
Cu	Mean±SD	238±83	156±8	135±25
	Range	176 - 333	147 - 160	108 - 158
Fe	Mean±SD	299±122	326±26	184±71
	Range	218 - 439	297 - 345	103 - 237
Hg	Mean±SD	0.052±0.007	0.071±0.009	0.104±0.076
	Range	0.045 - 0.059	0.063 - 0.08	0.053 - 0.191
Mn	Mean±SD	61.1±33.2	43.0±2.3	35.5±23.3
	Range	39.7 - 99.4	40.9 - 45.5	10.3 - 56.2

Table 3–22. Concentrations of 18 metals in amphipods (Anonyx nugax) collected from three locations in the Beaufort Sea in 2004. n= number of samples. Concentrations are μg/g dry wt (ppm), continued.

Metal	Parameter	Northstar (n= 3)	Liberty (n= 2)	BSMP (n= 3)
Ni	Mean±SD	3.11±2.09	2.56±0.17	1.81±0.66
	Range	1.7 - 5.51	2.37 - 2.69	1.05 - 2.2
Pb	Mean±SD	2.01±2.11	0.294±0.010	0.221±0.144
	Range	0.371 - 4.39	0.285 - 0.305	0.097 - 0.379
Sb	Mean±SD	0.025±0.008	0.015±0.002	0.014±0.004
	Range	0.02 - 0.034	0.013 - 0.016	0.009 - 0.017
Tl	Mean±SD	0.013±0.005	0.011±0.001	0.016±0.010
	Range	0.008 - 0.018	0.01 - 0.012	0.009 - 0.028
V	Mean±SD	2.13±1.18	2.58±0.21	1.70±0.47
	Range	1.3 - 3.48	2.34 - 2.73	1.16 - 2.02
Zn	Mean±SD	145±60	103±2	122.6±41.3
	Range	103 - 214	101 - 105	94.8 - 170

biggest differences were for Pb, which was present in Northstar amphipods at a mean concentration of 2.01± 2.11 μg/g dry wt, compared to mean concentrations of 0.29 ± 0.01 in Liberty amphipods and 0.22 ± 0.14 μg/g in BSMP amphipods. The unusually high concentration of Pb in amphipods from Northstar was not observed in samples collected in 2005 and 2006 and may be anomalous.

Clams

Clams (*Astarte montagui* and *Cyrtodaria kurriana*) were collected in 2004 at three stations in the BSMP area and at one location in the Liberty Prospect (Appendix A, Table A5). Concentrations of most metals were similar in clams collected at Liberty and BSMP in 2004 (Table 3-23). Because there was only a single clam sample from Liberty, meaningful statistical comparisons of metals concentrations in clams from Liberty and BSMP could not be made. However, the concentration of several metals in the single Liberty clams sample was higher than the highest concentration of that metal in the three BSMP clam samples. These metals include Al, Co, Cr, Fe, Hg, Mn, Ni, Pb, V, and Zn. This may indicate that clams from Liberty are retaining sediment particles in the gut or on the gills.

Inter-species Comparisons

Ag, Cu, and Zn concentrations were higher in amphipods than in clams. Al, Cr, Fe, Mn, and V concentrations were higher in clams than in the mussels (Tables 3-22 and 3-23). Cu and Zn are essential trace nutrients; crustaceans (amphipods) may require more of these metals than mollusks (clams) do. Al, Fe, and Mn are strongly associated with sediment particles; their high relative concentrations in clams may indicate ingestion of sediment particles by these filter-feeders. Concentrations of all 18 metals in both amphipods and clams collected in 2004 from different regions of the Beaufort Sea were low and in the range expected for these and related species from clean marine environments.

Table 3-23. Concentrations of 18 metals in indigenous clams (*Astarte montagui* and *Cyrtodaria kurriana*) collected from two locations in the Beaufort Sea in 2004. n= number of samples. Concentrations are μg/g dry wt (ppm).

Metal	Parameter	Liberty (n= 1)	BSMP (n= 3)
Ag	Mean±SD	0.123	0.104±0.026
	Range		0.087 - 0.134
Al	Mean±SD	2150	1184±563
	Range		721 – 1810
As	Mean±SD	14.3	11.4±3.53
	Range		8.22 - 15.2
Ba	Mean±SD	20.2	17.3±5.9
	Range		10.8 - 22.5
Be	Mean±SD	0.042	0.045±0.003
	Range		0.043 - 0.049
Cd	Mean±SD	5.24	3.81±2.87
	Range		0.53 - 5.85
Co	Mean±SD	3.92	1.98±0.96
	Range		1.11 - 3.01
Cr	Mean±SD	4.41	2.42±0.15
	Range		2.28 - 2.58
Cu	Mean±SD	13.2	13.8±4.1
	Range		11.2 - 18.5
Fe	Mean±SD	3640	1727±866
	Range		1040 - 2700
Hg	Mean±SD	0.075	0.067±0.005
	Range		0.062 - 0.072
Mn	Mean±SD	637	189±144
	Range		68.3 - 348
Ni	Mean±SD	5.28	3.41±1.31
	Range		1.92 - 4.35
Pb	Mean±SD	1.16	0.657±0.063
	Range		0.607 - 0.728
Sb	Mean±SD	0.026	0.020±0.002
	Range		0.018 - 0.021
Tl	Mean±SD	0.015	0.022±0.005
	Range		0.016 - 0.026
V	Mean±SD	6.91	4.19±1.01
	Range		3.34 - 5.31
Zn	Mean±SD	88.5	76.1±6.9
	Range		68.7 - 82.4

3.2.2.2 Summer of 2005

Amphipods

Amphipods were collected in 2005 at four stations near the Northstar Development, five stations in the BSMP area, and three stations near the Liberty Prospect. Concentrations of most metals were similar in amphipods collected in 2004 and 2005 (Tables 3-22 and 3-24). Concentrations of Cu and Pb were lower, but not significantly so, in amphipods collected from Northstar in 2005 than in 2004. Concentrations of Al, Co, Fe, Mn, Ni, and V were lower, but not significantly so, in 2004 than in 2005 in amphipods from the Liberty Prospect (Tables 3-22 and 3-24). All metals, except Ba and Sb, were present at similar concentrations in amphipods collected in 2004 and 2005 at the five BSMP stations. Concentrations of Ba and Sb were slightly, but significantly, higher in amphipods collected BSMP in 2005 than those collected in 2004.

Table 3-24. Concentrations of 18 metals in amphipods (*Anonyx nugax*) from three locations in the Beaufort Sea in 2005. n= number of samples. Concentrations are μg/g dry wt (ppm).

Parameter		Northstar (n= 4)	Liberty (n=3)	BSMP (n= 5)
Ag	Mean±SD	2.94±0.06	3.37±1.05	2.65±0.72
	Range	2.85 - 2.99	2.16 - 4.04	1.61 - 3.39
Al	Mean±SD	227±68	166±68.7	270±96.5
	Range	168 - 305	95.8 - 233	145 - 379
As	Mean±SD	11.8±2.7	11.8±6.27	10.7±2.42
	Range	7.9 - 14.2	5.01 - 17.4	8.12 - 13.3
Ba	Mean±SD	25.0±0.3	23.1±15.4	31.1±7.47
	Range	24.6 - 25.4	11.8 - 40.7	20.1 - 39.6
Be	Mean±SD	0.024±0.002	0.018±0.001	0.018±0.005
	Range	0.021 - 0.027	0.017 - 0.019	0.014 - 0.024
Cd	Mean±SD	0.78±0.12	1.64±0.91	0.989±0.307
	Range	0.68 - 0.938	0.615 - 2.37	0.681 - 1.39
Co	Mean±SD	1.85±0.19	1.41±0.03	2.14±0.50
	Range	1.69 - 2.02	1.38 - 1.44	1.59 - 2.88
Cr	Mean±SD	0.48±0.10	0.61±0.01	0.64±0.08
	Range	0.41 - 0.55	0.6 - 0.62	0.56 - 0.73
Cu	Mean±SD	190±12	167±30	151±35
	Range	173 - 200	132 - 185	100 - 196
Fe	Mean±SD	208±24	189±5.3	231±43
	Range	177 - 235	183 - 193	168 - 280
Hg	Mean±SD	0.061±0.023	0.082±0.033	0.068±0.046
	Range	0.045 - 0.095	0.049 - 0.115	0.001 - 0.117
Mn	Mean±SD	42.7±9.3	26.0±16.6	34.9±8.0
	Range	28.8 - 47.7	15.3 - 45.1	26.6 - 44.9
Ni	Mean±SD	2.14±1.05	1.39±0.46	2.9±1.5
	Range	0.84 - 3.03	1.07 - 1.92	1.4 - 5.2

Table.3–24. Concentrations of 18 metals in amphipods (Anonyx nugax) from three locations in the Beaufort Sea in 2005. n= number of samples. Concentrations are μg/g dry wt (ppm), continued.

Parameter		Northstar (n= 4)	Liberty (n=3)	BSMP (n= 5)
Pb	Mean±SD	0.256±0.042	0.161±0.122	0.094±0.045
	Range	0.217 - 0.308	0.057 - 0.296	0.047 - 0.159
Sb	Mean±SD	0.026±0.003	0.031±0.008	0.030±0.005
	Range	0.022 - 0.027	0.024 - 0.039	0.025 - 0.038
Tl	Mean±SD	0.009±0.001	0.014±0.004	0.012±0.008
	Range	0.009 - 0.01	0.01 - 0.017	0.005 - 0.026
V	Mean±SD	1.12±0.08	0.79±0.27	1.24±0.45
	Range	1.02 - 1.21	0.52 - 1.06	0.66 - 1.74
Zn	Mean±SD	109±7	129±42.9	103±15.6
	Range	102 - 119	79.4 - 156	85.7 - 125

Isopods

A sufficient biomass of isopods (*Saduria sabini*) was collected at four BSMP stations and three Liberty stations in 2005 to permit metals analysis (Table 3-25). There was insufficient biomass for analysis of the isopods collected in 2004 and 2006. There was not a statistically significant difference in the concentration of any metal in isopods collected at BSMP and Liberty in 2005.

Table 3-25. Concentrations of 18 metals in isopods (Saduria sabini) collected from two locations in the Beaufort Sea in 2005. n= number of samples. Concentrations are μg/g dry wt (ppm).

Metal	Parameter	Liberty (n=3)	BSMP (n=4)
Ag	Mean±SD	2.68±0.22	2.23±0.40
	Range	2.47 - 2.90	1.88 - 2.77
Al	Mean±SD	2133±611	3413±1213
	Range	1650 - 2820	2250 - 4480
As	Mean±SD	22.7±1.4	21.6±9.3
	Range	21.4 - 24.1	13.9 - 34.4
Ba	Mean±SD	58.2±7.3	63.9±8.7
	Range	51.8 - 66.1	53.6 - 73
Be	Mean±SD	0.082±0.013	0.136±0.039
	Range	0.071 - 0.096	0.093 - 0.170
Cd	Mean±SD	0.924±0.124	1.46±0.48
	Range	0.782 - 1.01	0.833 - 1.95
Co	Mean±SD	3.84±0.25	4.73±0.86
	Range	3.67 - 4.13	3.56 - 5.42
Cr	Mean±SD	3.85±1.61	5.17±1.36
	Range	2.71 - 5.69	3.98 - 6.72
Cu	Mean±SD	124±19	124±25
	Range	104 - 141	104 - 160

Table 3–25. Concentrations of 18 metals in isopods (Saduria sabini) collected from two locations in the Beaufort Sea in 2005. n= number of samples. Concentrations are μg/g dry wt (ppm), continued.

Metal	Parameter	Liberty (n=3)	BSMP (n=4)
Fe	Mean±SD	1843±359	2788±900
	Range	1570 - 2250	1740 - 3810
Hg	Mean±SD	0.054±0.005	0.071±0.022
	Range	0.049 - 0.059	0.041 - 0.095
Mn	Mean±SD	411±159	384±159
	Range	316 - 594	234 - 608
Ni	Mean±SD	3.65±1.04	5.47±1.63
	Range	2.74 - 4.78	3.27 - 7.21
Pb	Mean±SD	0.804±0.190	1.46±0.46
	Range	0.635 - 1.01	1.07 - 2.01
Sb	Mean±SD	0.027±0.005	0.024±0.005
	Range	0.022 - 0.032	0.018 - 0.029
Tl	Mean±SD	0.031±0.009	0.027±0.011
	Range	0.021 - 0.037	0.014 - 0.038
V	Mean±SD	7.05±2.26	8.78±2.82
	Range	5.09 - 9.52	6.37 - 12.7
Zn	Mean±SD	84.2±6.6	82.2±5.8
	Range	80.1 - 91.8	77.8 - 90.7

Clams

Clams (*Astarte montagui* and *Cyrtodaria kurriana*) were collected in 2005 from one station near Liberty and 5 stations in the BSMP area. Concentrations of Al, Fe, Mn, and V were slightly higher in clams collected in 2004 than those collected in 2005 (Tables 3-23 and 3-26). However, these interannual differences were not statistically significant at the BSMP, the only location where sufficient replicate clam samples were collected to allow for statistical comparisons. Hg concentration was significantly higher and Sb concentration was significantly lower in clams collected in 2004 than in those collected in 2005 from the BSMP. Concentrations of the other metals were similar in clams collected from Liberty and the BSMP in 2004 and 2005.

Interspecies Comparisons

Concentrations of Al, Ba, Co, Cr, Mn, and V were higher in isopods than in amphipods and clams collected in 2005 (Tables 3-24, 3-25, and 3-26). Concentrations of Cr, Fe, and Pb were similar in isopods and clams, but lower in amphipods collected in 2005. These metals tend to be associated with nearly insoluble mineral phases of oxidized sediments. The higher concentrations in isopods may indicate that the isopods had more sediment particles in the gut than amphipods and clams did. Copper concentrations were lower in clams than in isopods and amphipods. This reflects the high levels of copper-containing respiratory pigments in most crustaceans.

Table 3-26. Concentrations of 18 metals in indigenous clams (*Astarte montagui* and *Cyrtodaria kurriana*) collected from two locations in the Beaufort Sea in 2005. n= number of samples. Concentrations are µg/g dry wt (ppm).

Metal	Parameter	Liberty (n= 1)	BSMP (n= 5)
Ag	Mean±SD	0.066	0.085±0.031
	Range		0.052 - 0.137
Al	Mean±SD	1220	623±495
	Range		98.5 - 1320
As	Mean±SD	16	13.0±2.6
	Range		11.3 - 17.4
Ba	Mean±SD	24.8	17.3±13.0
	Range		7.9 - 39.5
Be	Mean±SD	0.085	0.046±0.020
	Range		0.019 - 0.066
Cd	Mean±SD	3.88	4.34±3.45
	Range		1.13 - 9.55
Co	Mean±SD	1.67	1.16±0.35
	Range		0.75 - 1.68
Cr	Mean±SD	3.7	2.46±1.60
	Range		0.91 - 5.15
Cu	Mean±SD	11.1	14.0±2.5
	Range		12.0 - 17.1
Fe	Mean±SD	1910	1227±592
	Range		771 - 2110
Hg	Mean±SD	0.081	0.042±0.007
	Range		0.030 - 0.047
Mn	Mean±SD	65.5	128±57.4
	Range		78.4 - 205
Ni	Mean±SD	4.45	4.05±1.27
	Range		2.04 - 5.34
Pb	Mean±SD	0.707	0.5274±0.426
	Range		0.184 - 1.24
Sb	Mean±SD	0.045	0.046±0.011
	Range		0.031 - 0.059
Tl	Mean±SD	0.021	0.022±0.006
	Range		0.015 - 0.030
V	Mean±SD	3.37	2.41±1.49
	Range		0.97 - 4.72
Zn	Mean±SD	73.1	70.8±9.2
	Range		57.8 - 79.3

3.2.2.3 Summer 2006

Amphipods

Amphipods were collected from three locations in 2006: Northstar, BSMP, and near West Dock, an industrial area near Prudhoe Bay. Metals concentrations were similar in amphipods collected at the three locations in 2006 (Table 3-27). Concentrations of Ba, Mn, and Ni were significantly lower in amphipods from Northstar than in those from BSMP and West Dock. There were no significant differences in concentrations of the other 15 metals in amphipods from the three locations.

Table 3-27. Concentrations of 18 metals in amphipods (Anonyx nugax) collected from four locations in the Beaufort Sea in 2006. n= number of samples. Concentrations are μg/g dry wt (ppm).

Metal	Parameter	Northstar (n= 9)	West Dock (n= 2)	BSMP (n= 5)
Ag	Mean±SD	2.18±0.85	2.38±0.19	2.18±0.44
	Range	0.95 - 3.01	2.24 - 2.51	1.55 - 2.65
Al	Mean±SD	256±194	345±57	381±290
	Range	124 - 747	305 - 385	207 - 894
As	Mean±SD	12.8±4.42	5.49±0.21	9.88±3.27
	Range	4.97 - 16.9	5.34 - 5.64	6.77 - 13.7
Ba	Mean±SD	17.2±6.38	36.9±0.1	32.2±19.0
	Range	12.0 - 33.5	36.8 - 37.0	16.2 - 59.0
Be	Mean±SD	0.008±0.004	0.012±0	0.011±0.006
	Range	0.005 - 0.017	0.012 - 0.012	0.006 - 0.02
Cd	Mean±SD	1.01±0.45	0.519±0.021	0.677±0.228
	Range	0.386 - 1.74	0.504 - 0.534	0.401 - 0.909
Co	Mean±SD	1.55±0.20	1.77±0.04	1.62±0.13
	Range	1.29 - 1.81	1.74 - 1.80	1.40 - 1.73
Cr	Mean±SD	0.43±0.26	0.68±0.04	0.70±0.66
	Range	0.18 - 1.04	0.65 - 0.71	0.30 - 1.86
Cu	Mean±SD	167±18.0	144.5±3.5	156±42
	Range	124 - 189	142 - 147	114 - 203
Fe	Mean±SD	222±92	275±33	287±108
	Range	137 - 431	252 - 298	221 - 477
Hg	Mean±SD	0.088±0.044	0.081±0.007	0.053±0.012
	Range	0.046 - 0.158	0.076 - 0.086	0.037 - 0.066
Mn	Mean±SD	32.3±9.2	70.1±1.5	42.1±9.3
	Range	21.6 - 48.6	69.0 - 71.1	36 - 58
Ni	Mean±SD	2.79±0.33	3.64±0.064	3.31±0.52
	Range	2.31 - 3.32	3.59 - 3.68	2.78 - 3.92
Pb	Mean±SD	0.148±0.055	0.130±0.025	0.140±0.076
	Range	0.091 - 0.248	0.112 - 0.147	0.070 - 0.223
Sb	Mean±SD	0.014±0.005	0.018±0.001	0.019±0.013
	Range	0.007 - 0.019	0.017 - 0.018	0.007 - 0.040

Table 3–27. Concentrations of 18 metals in amphipods (Anonyx nugax) collected from four locations in the Beaufort Sea in 2006. n= number of samples. Concentrations are μg/g dry wt (ppm).

Metal	Parameter	Northstar (n= 9)	West Dock (n= 2)	BSMP (n= 5)
Tl	Mean±SD	0.008±0.001	0.012±0	0.011±0.006
	Range	0.007 - 0.011	0.012 - 0.012	0.006 - 0.019
V	Mean±SD	1.27±0.26	1.05±0	1.26±0.46
	Range	0.89 - 1.59	1.05 - 1.05	0.86 - 1.99
Zn	Mean±SD	116±22	86.9±1.5	92.6±12.6
	Range	83.6 - 145	85.8 - 87.9	74.4 - 104

Interannual Variation in Concentrations of Metals in Amphipods: 2004, 2005, 2006

The concentrations of the 18 metals in tissues of amphipods collected at Northstar, Liberty, and BSMP in 2004, 2005, and 2006 were compared statistically by a two-way General Linear Model (GLM) application of the analysis of variance (ANOVA), because the data were not normally distributed. There were statistically significant differences in concentrations of nine of the 18 metals among years, locations, or a combination of the two (Table 3-28). The statistically significant difference was among the three years of the program or year x location for eight of the nine metals. There was a statistically significant difference in copper concentration in amphipods from the three sampling locations. Mean copper concentration was higher in amphipods from Northstar than in those from Liberty and BSMP in all three years.

Concentrations of Ag, Al, Pb, Sb, and V in amphipod tissues were higher in 2004 than in 2005 or 2006. The concentration of Be was higher in amphipod tissues in 2005 than in 2004 and 2006. The concentration of Co in amphipod tissues was higher at Northstar and Liberty than at BSMP in 2004 and was higher at BSMP than at Northstar and Liberty in 2006. The concentration of Fe in amphipod tissues was higher in 2004 than in 2005 and 2006 at Northstar and Liberty and higher in amphipods from BSMP in 2006. The highest Pb concentration was in amphipods collected in 2004 from Northstar; the lowest concentration was in amphipods collected at BSMP in 2005. In most cases, these differences were small and there was no clear pattern of difference in metals concentrations in amphipod tissues with year or location.

Table 3-28. Statistical comparison of metals concentrations in amphipods (*Anonyx* sp) from BSMP, Liberty, and Northstar in 2004, 2005, and 2006. Only metals for which statistically significant differences were detected are included. Differences were considered significant at $p \leq 0.05$ and are highlighted.

Metal	Source	DF	Pr > F	Approach
Ag – Rank	Year	2	0.0217	Two-Way
	Location	2	0.0549	Two-Way
	Year x Location	3	0.6824	Two-Way
Al – Log	Year	2	0.0362	Two-Way
	Location	2	0.9723	Two-Way
	Year x Location	3	0.1378	Two-Way

Table 3–28. Statistical comparison of metals concentrations in amphipods (Anonyx sp) from BSMP, Liberty, and Northstar in 2004, 2005, and 2006. Only metals for which statistically significant differences were detected are included. Differences were considered significant at $p \leq 0.05$ and are highlighted, continued.

Metal	Source	DF	Pr > F	Approach
Be - Rank	Year	2	<0.001	Two-Way
	Location	2	0.6945	Two-Way
	Year x Location	3	0.0818	Two-Way
Co – Log	Year	2	0.0023	Two-Way
	Location	2	0.3564	Two-Way
	Year x Location	3	0.0204	Two-Way
	Location x Year	7	0.0041	One-Way
Cu – Rank	Year	2	0.5339	Two-Way
	Location	2	0.0039	Two-Way
	Year x Location	3	0.1146	Two-Way
Fe - Log	Year	2	0.2842	Two-Way
	Location	2	0.7292	Two-Way
	Year x Location	3	0.0442	Two-Way
Pb – Rank	Year	2	0.0030	Two-Way
	Location	2	0.0054	Two-Way
	Year x Location	3	0.1759	Two-Way
Sb	Year	2	0.0003	Two-Way
	Location	2	0.9385	Two-Way
	Year x Location	3	0.0847	Two-Way
V – Log	Year	2	0.0082	Two-Way
	Location	2	0.9022	Two-Way
	Year x Location	3	0.1561	Two-Way

Clams

Clams were collected during 2006 at only two stations near Liberty. Mean concentrations of most metals were slightly lower in clams collected at Liberty in 2006 than in those collected at Liberty in 2004 and 2005 (Table 3-29). Mean Cd concentration was higher (7.78 ± 1.29 µg/g dry wt) in clams collected at Liberty in 2006 than in clams collected at Liberty in 2004 (3.81 ± 2.87 µg/g) or 2005 (4.34 ± 3.45 µg/g).

Table 3-29. Concentrations of 18 metals in indigenous clams (*Astarte montagui*) collected from Liberty (two stations) in 2006. n= number of samples. Concentrations are µg/g dry wt (ppm).

Metal	Parameter	Liberty (n= 2)
Ag	Mean±SD	0.070±0.011
	Range	0.062 - 0.078
Al	Mean±SD	992±26
	Range	973 - 1010
As	Mean±SD	11.2±0.6
	Range	10.7 - 11.6
Ba	Mean±SD	13.1±0.9
	Range	12.4 - 13.7
Be	Mean±SD	0.056±0.026
	Range	0.037 - 0.074
Cd	Mean±SD	7.78±1.29
	Range	6.86 - 8.69
Co	Mean±SD	1.13±0.13
	Range	1.03 - 1.22
Cr	Mean±SD	2.66±0.23
	Range	2.49 - 2.82
Cu	Mean±SD	10.2±0
	Range	10.2 - 10.2
Fe	Mean±SD	1590±57
	Range	1550 - 1630
Hg	Mean±SD	0.068±0.002
	Range	0.066 - 0.069
Mn	Mean±SD	60.2±18.5
	Range	47.1 - 73.3
Ni	Mean±SD	3.84±0.25
	Range	3.66 - 4.02
Pb	Mean±SD	0.467±0.374
	Range	0.202 - 0.731
Sb	Mean±SD	0.029±0.026
	Range	0.010 - 0.047
Tl	Mean±SD	0.016±0.004
	Range	0.013 - 0.018
V	Mean±SD	4.48±0.35
	Range	4.23 - 4.73
Zn	Mean±SD	78.9±3.6
	Range	76.3 - 81.4

Interannual Variation in Concentrations of Metals in clams: 2004, 2005, 2006
The concentrations of the 18 metals in tissues of clams collected at Liberty in 2004, 2005, and 2006, and at BSMP stations in 2004, 2005 were compared statistically by a two-way General Linear Model (GLM) application of the analysis of variance (ANOVA), because most data were not normally distributed. There were significant differences in concentrations of four metals among years or between locations (Table 3-30). The concentrations of Co and Mn in clams from Liberty and BSMP were significantly higher in 2004 than in 2005 and 2006. The Mn concentration in the single clam sample collected at Liberty in 2004 (637 μg/g) was much higher than Mn concentrations in clams collected at other times or locations. The Fe concentration in the clam sample from Liberty collected in 2004 also was much higher (3640 μg/g) than the Fe concentrations in clams collected at all other times and locations, explaining the significant effect of location on Fe concentration in clams. The high concentrations of Mn and Fe in clams from Liberty in 2004 indicate that the clams may have ingested oxic sediments naturally high in these metals. The concentration of Hg was significantly different in clams collected at Liberty and BSMP, due primarily to a lower concentration in clams from BSMP in 2005 (0.042 ± 0.006 μg/g) than in clams from other years and locations (range of means, 0.067 – 0.081 μg/g). As with the amphipods, the differences in metals concentrations in clams among times and locations were relatively small and probably of no biological significance.

Table 3-30. Statistical comparison of metals concentrations in clams (*Astarte montagui* and *Cyrtodaria kurriana*) from Liberty in 2004, 2005, and 2006 and the BSMP stations in 2004 and 2005. Only metals for which statistically significant differences were detected are included. Differences were considered significant at $p \leq 0.05$ and are highlighted.

Metal	Source	DF	Pr > F	Approach
Co – Log	Year	2	0.0220	Two-Way
	Location	1	0.0577	Two-Way
	Year x Location	1	0.3827	Two-Way
Fe	Year	2	0.0678	Two-Way
	Location	1	0.0392	Two-Way
	Year x Location	1	0.2692	Two-Way
Hg	Year	2	0.0868	Two-Way
	Location	1	0.0015	Two-Way
	Year x Location	1	0.0139	Two-Way
	Location_Year	4	0.0009	One-Way
Mn – Log	Year	2	0.0247	Two-Way
	Location	1	0.3780	Two-Way
	Year x Location	1	0.0570	Two-Way

3.2.2.4 Time Series Analysis of Trace Metals in Amphipods and Clams: 1999 - 2006

Clams
As previously discussed, the ANIMIDA and cANIMIDA studies were designed to test the hypothesis that oil and gas industry activities in the development area will not result in an

increase in tissue concentrations of metals in sediments and tissues of marine animals. One challenge to testing this hypothesis is to establish a baseline of metal concentrations with a variance that is small enough to identify minor trends in concentrations, especially increases, over time or space. Data are available for 18 metals in samples of amphipods and clams collected during the ANIMIDA (1999, 2000, and 2002) and cANIMIDA (2004, 2005 and 2006) studies. In addition, data for Ba, Cd, Cu, Pb, V and Zn were obtained from previous studies in 1986 and 1989 (Boehm et al., 1990). The discussion that follows considers utility of the database with respect to identifying any future changes in metal concentrations.

Annual mean concentrations for each of 18 metals in amphipods (*Anonyx nugax*) from the coastal Beaufort Sea are plotted with their respective standard deviations (square root of the variance) in Figures 3-43 through 3-45. Table 3-31 shows the grand means and standard deviations for the 1999-2006 data along with the relative standard deviations [RSD = (mean/SD) x 100%], maximums and minimums. The 1999-2006 data sets were used to create Table 3-31 so that the same time interval would be used for all metals. Identification of any future changes in metal concentrations in amphipods relies on (1) a relatively low RSD or (2) low enough absolute metal values that changes can be distinguished before an adverse impact occurs.

Table 3-31. Means, standard deviations (SD), relative standard deviations [RSD = (mean/SD) x 100], maximums and minimums for metals in 54 amphipod (*Anonyx*) samples from the ANIMIDA and cANIMIDA study areas from 1999-2001, 2003, and 2004-2006.

Metal	Mean ± SD (µg/g, dry wt.)	RSD (%)	Maximum (µg/g, dry wt.)	Minimum (µg/g, dry wt.)	(Max/Mean)
Ag	2.5 ± 0.6	26	4.0	0.8	1.6
Al	391 ± 242	62	1220	96	3.1
As	9.4 ± 3.8	40	17.4	4.0	2.4
Ba	26.1 ± 10.6	41	59.0	7.4	3.5
Be	0.012 ± 0.007	57	0.031	0.002	2.6
Cd	0.79 ± 0.45	57	2.4	0.3	3.0
Co	1.7 ± 0.4	26	2.9	0.6	1.7
Cu	149 ± 37	25	206	41	1.4
Fe	293 ± 156	53	949	103	3.2
Hg	0.067 ± 0.033	49	0.19	0.02	2.8
Mn	40 ± 13	32	71	10	1.8
Ni	2.9 ± 1.2	40	6.7	0.8	2.3
Pb	0.23 ± 0.14	60	0.7	0.05	1.7
Sb	0.022 ± 0.008	37	0.04	0.01	1.8
Tl	0.011 ± 0.004	39	0.028	0.005	2.5
V	1.6 ± 0.7	43	3.4	0.5	2.1
Zn	105 ± 21	20	170	54	1.6
% H_2O	75.5 ± 2.3	3	80.0	71.0	1.06

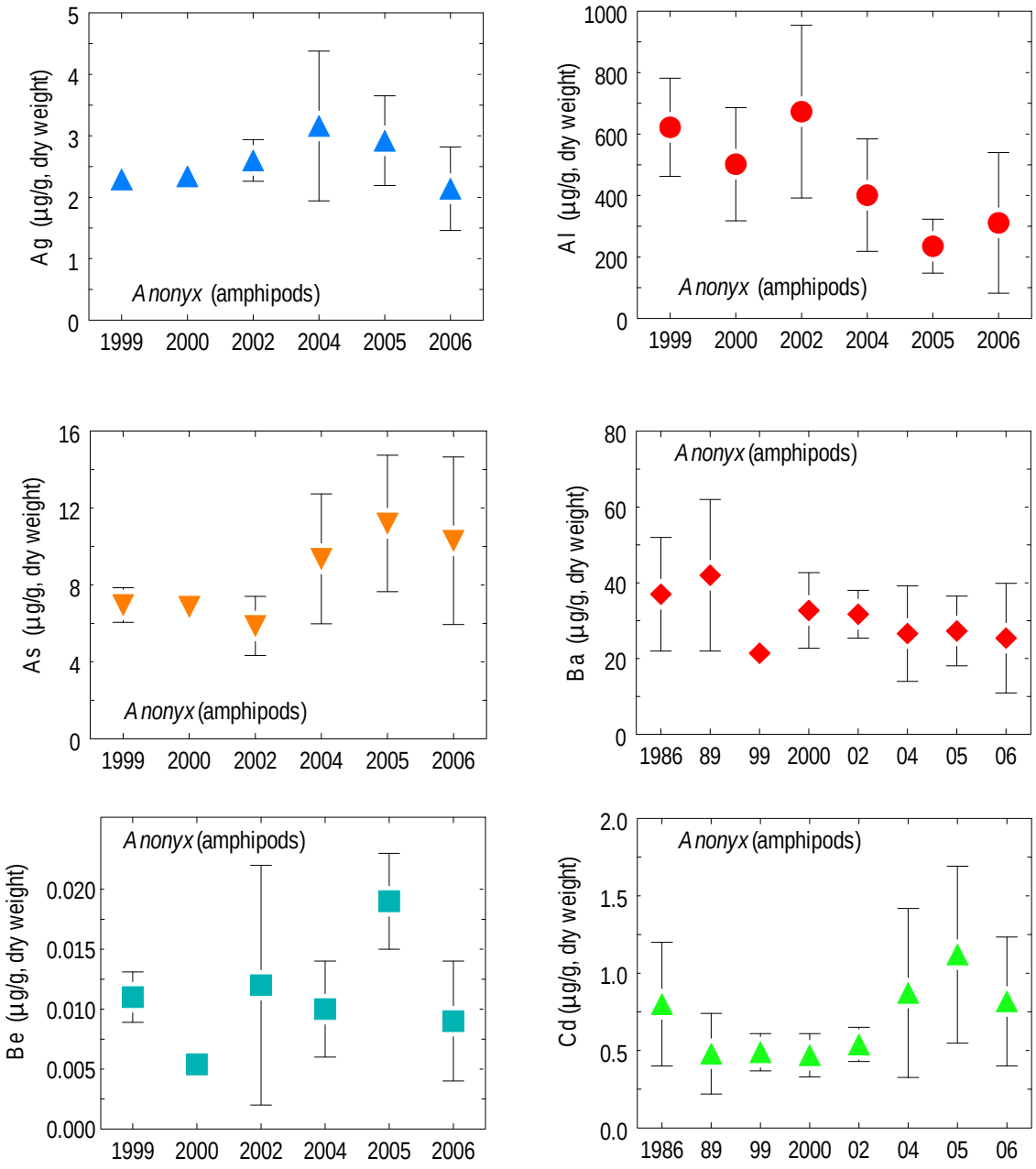


Figure 3-43. Concentrations of silver (Ag), aluminum (Al), arsenic (As), barium (Ba), beryllium (Be) and cadmium (Cd) in amphipods (Anonyx) from the coastal Beaufort Sea. Markers show the annual mean concentrations and lines show ±1 standard deviation (SD). Markers with no lines have an SD that is smaller than the marker.

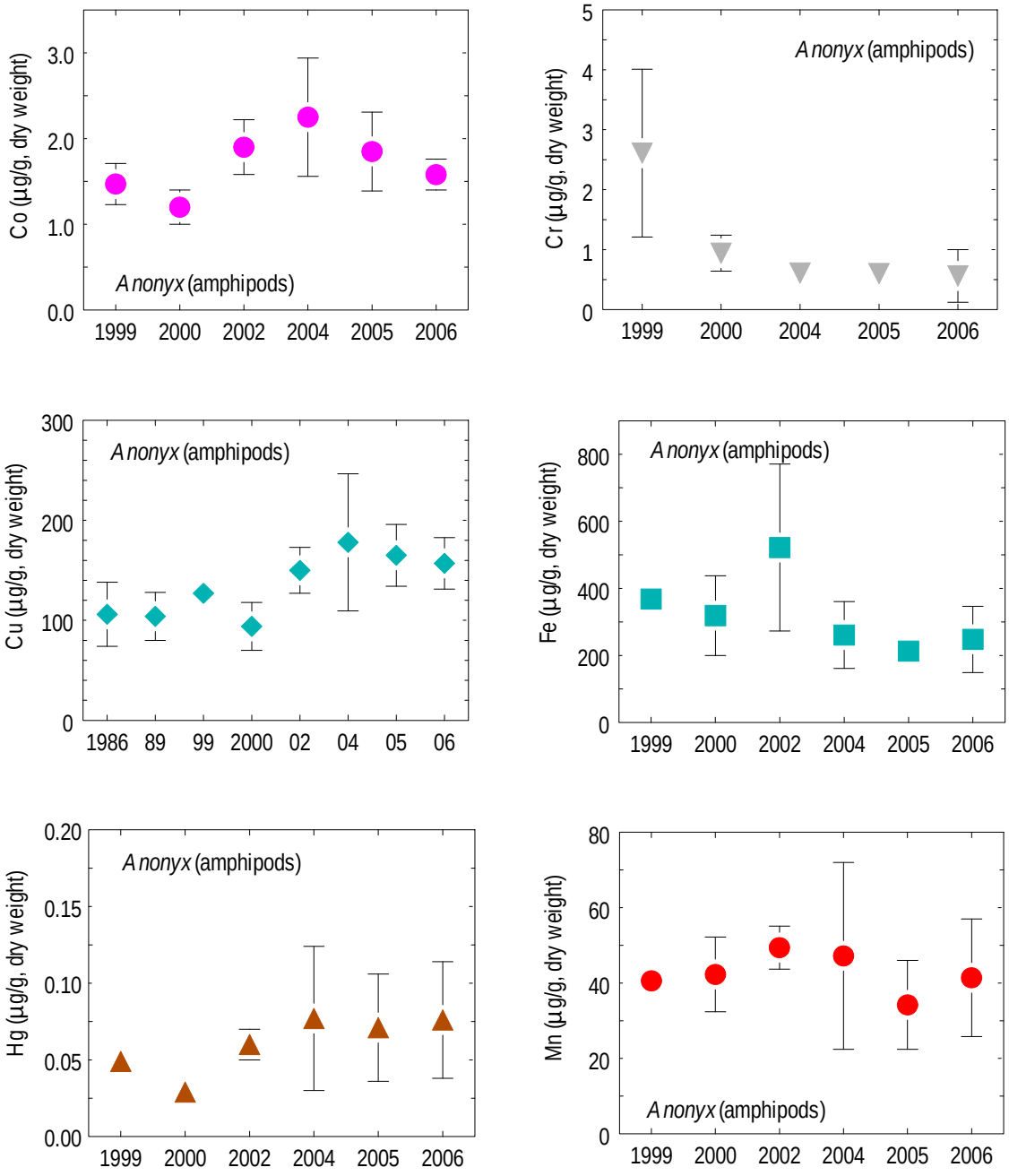


Figure 3-44. Concentrations of cobalt (Co), chromium (Cr), copper (Cu), iron (Fe), mercury (Hg) and manganese (Mn) in amphipods (Anonyx) from the coastal Beaufort Sea. Markers show the annual mean concentrations and lines show ±1 standard deviation (SD). Markers with no lines have an SD that is smaller than the marker.

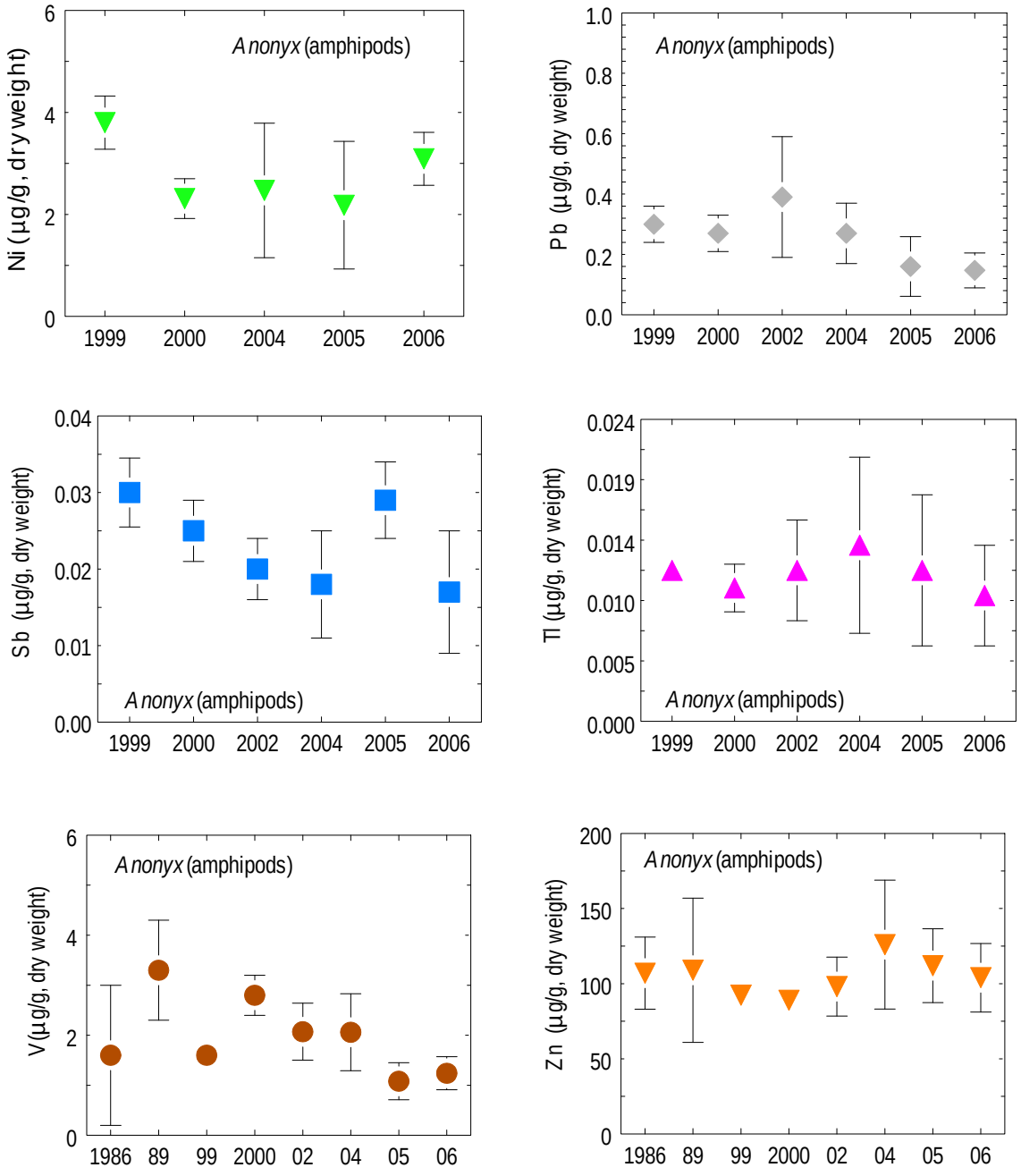


Figure 3-45. Concentrations of nickel (Ni), lead (Pb), antimony (Sb), thallium (Tl), vanadium (V) and zinc (Zn) in amphipods (Anonyx) from the coastal Beaufort Sea. Markers show the annual mean concentrations and lines show ±1 standard deviation (SD). Markers with no lines have an SD that is smaller than the marker.

Concentrations of Cu and Zn in amphipods were the most uniform in the database with RSDs of 25% and 20%, respectively, for the complete data set of 54 samples collected between 1999 and 2006 (Table 3-31 and Figures 3-44 and 3-45). This observation for Cu and Zn is most likely due ion regulation of these essential metals that helps control their concentrations in the amphipods. The other metals show a variety of values for the RSD that range from 26% for Ag and Co to 62% for Al (Table 3-31). Assuming that an increase of 2 standard deviations above the mean (for replicate samples) is required to identify changes in metal values, then a 40 to 52% increase in the absolute concentrations of Ag, Co, Cu and Zn would be needed to support a significant increase (Table 3-31). Similarly, increases in metal concentrations in amphipods of 64 to 70% (2 SDs) would be needed to identify changes in concentrations of Mn, Sb, Tl, As and Ni (Table 3-31).

Because average metal concentrations for several metals were <1 µg/g, the absolute magnitude of any increase in metal values could be small, yet easily detected. For example, concentrations of Be ranged from 0.002 to 0.031 µg/g (i.e., 2 to 31 parts per billion) with a mean of 0.012 ± 0.007 µg/g; thus, an increase of just 0.030 µg/g (30 parts per billion) would be equivalent to an increase of >4 standard deviations.

Clams

Annual mean concentrations for each of 18 metals in clams (*Astarte montagui* and *Cyrtodaria kurriana*) from the coastal Beaufort Sea are plotted with their respective standard deviations (square root of the variance) in Figures 3-46 through 3-48. Table 3-32 shows the grand means and standard deviations for the 1999-2006 data along with the relative standard deviations [RSD = (mean/SD) x 100], maximums and minimums. The 1999-2006 data sets were used to create Table 3-31 so that the same time interval would be used for all metals. As discussed above with the amphipod data, identification of any future changes in metal concentrations in amphipods relies on (1) a relatively low RSD or (2) low enough absolute metal values that changes can be distinguished before an adverse impact occurs.

Concentrations of As, Cu and Zn in clams were the most uniform in the data base with RSDs of 19%, 28% and 18%, respectively, for the complete data set of 22 samples collected between 1999 and 2006 (Table 3-32 and Figures 3-46, 3-47 and 3-48). The other metals show a variety of values for the RSD that range from 29% for Be and Co to 69% for Mn (Table 3-32). If an increase of 2 standard deviations above the mean (for replicate samples) is assumed to be required to support a change in metal values, then a 36 to 58% increase in the absolute concentrations of As, Be, Cu and Zn would be needed to identify a significant increase (Table 3-32). Similarly, increases in metal concentrations in amphipods of 64 to 70% would be needed to identify changes in concentrations of Ag, Hg, Pb and Tl (Table 3-32).

Because average metal concentrations for several metals are <1 µg/g, the absolute magnitude of any increase in metal values could be small, yet easily detected. For example, concentrations of Ag ranged from 0.04 to 0.13 µg/g with a mean of 0.07 ± 0.02 µg/g; thus, an increase of just 0.10 µg/g (100 parts per billion) would be equivalent to an increase of >5 standard deviations.

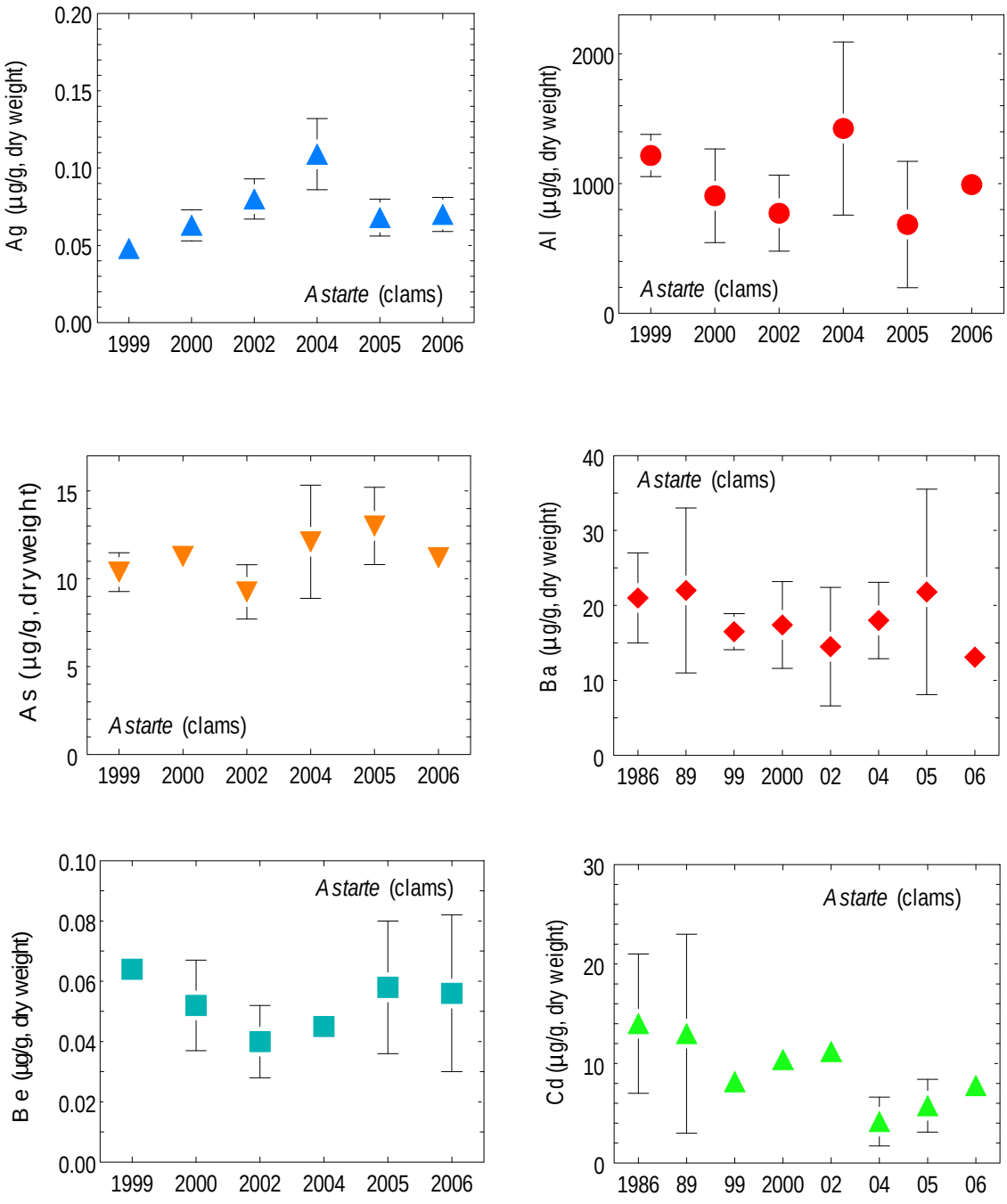


Figure 3-46. Concentrations of silver (Ag), aluminum (Al), arsenic (As), barium (Ba), beryllium (Be) and cadmium (Cd) in clams (Astarte) from the coastal Beaufort Sea. Markers show the annual mean concentrations and lines show ±1 standard deviation (SD). Markers with no lines have an SD that is smaller than the marker.

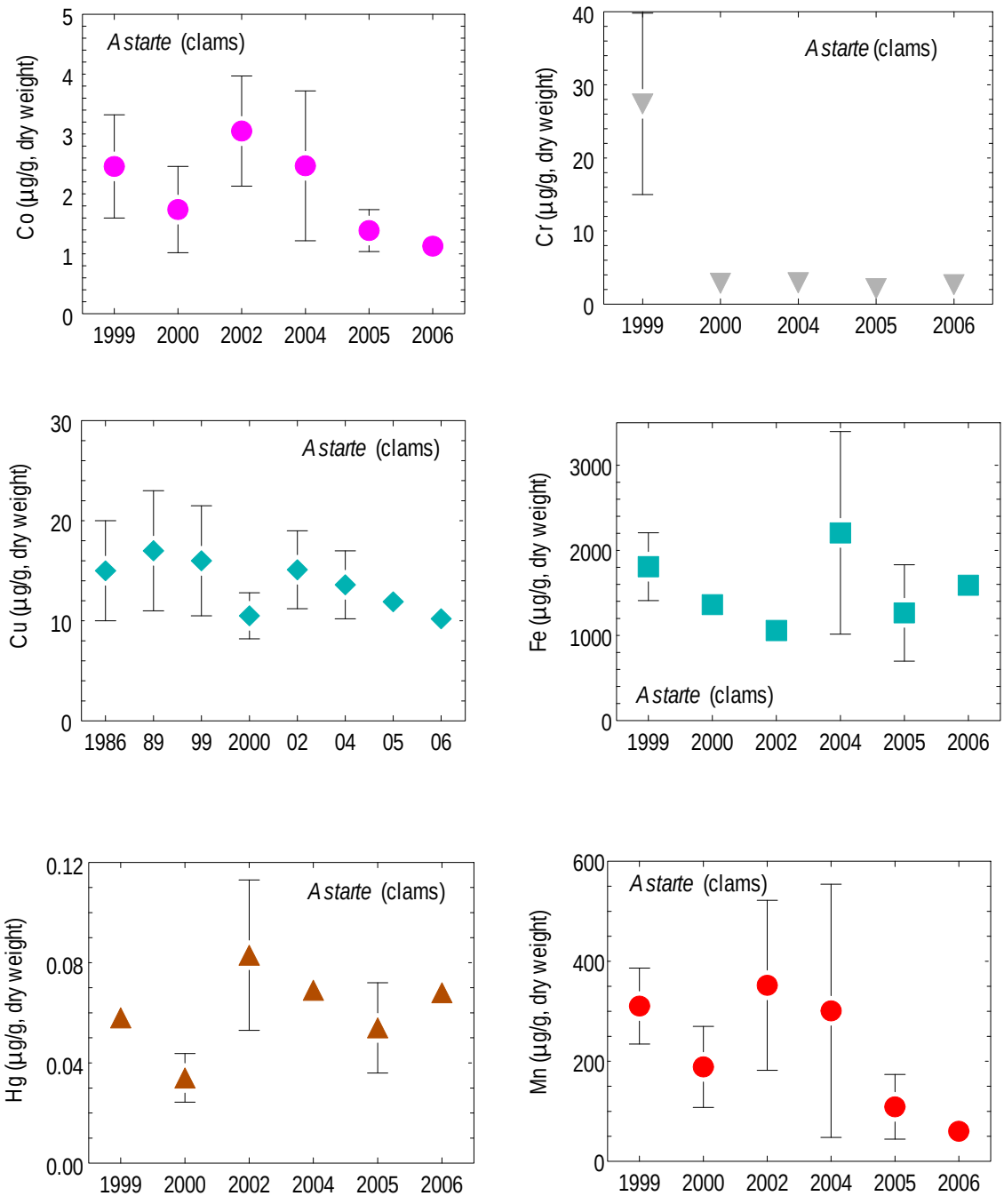


Figure 3-47. Concentrations of cobalt (Co), chromium (Cr), copper (Cu), iron (Fe), mercury (Hg) and manganese (Mn) in clams (Astarte) from the coastal Beaufort Sea. Markers show the annual mean concentrations and lines show ±1 standard deviation (SD). Markers with no lines have an SD that is smaller than the marker.

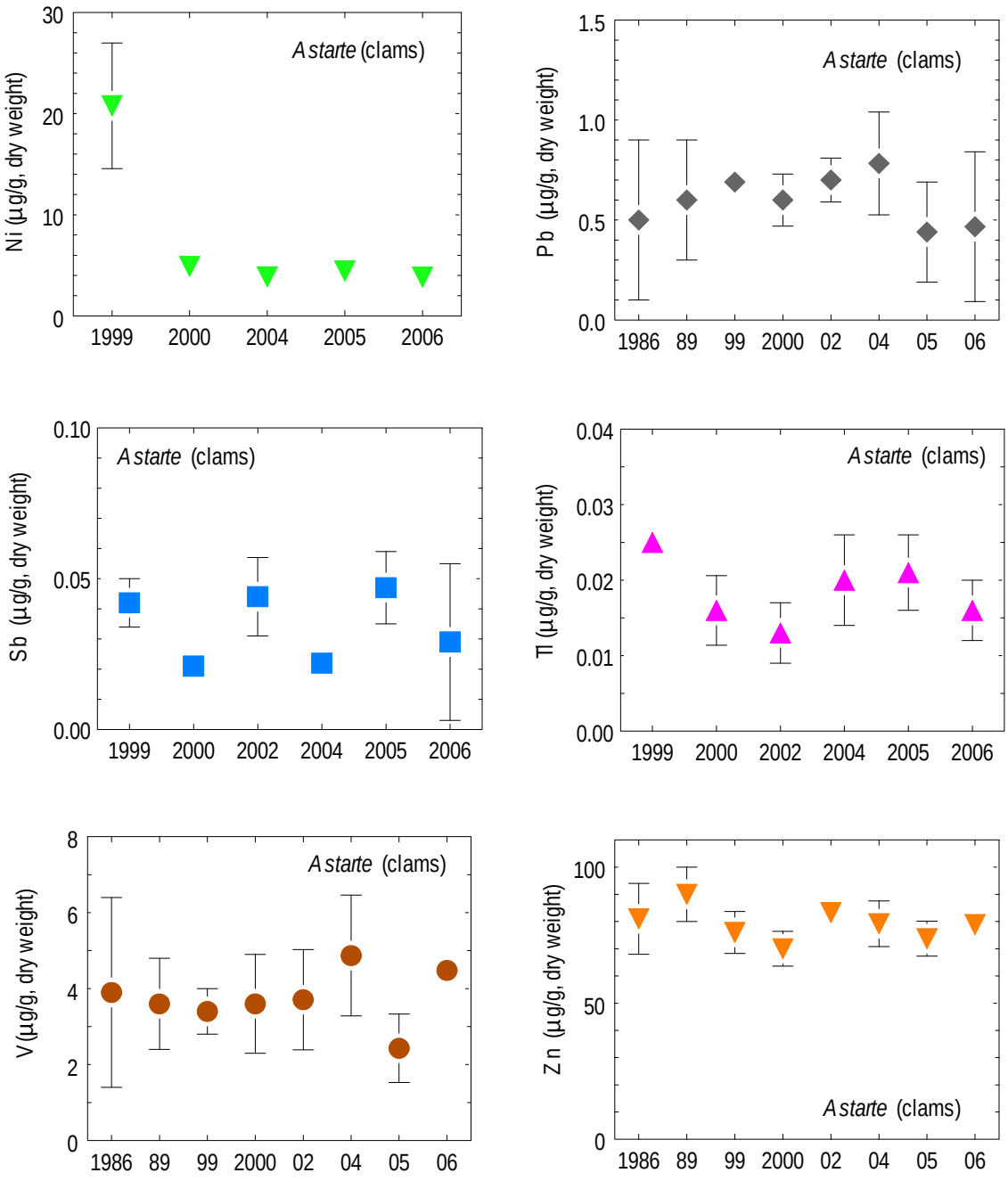


Figure 3-48. Concentrations of nickel (Ni), lead (Pb), antimony (Sb), thallium (Tl), vanadium (V) and zinc (Zn) in clams (Astarte) from the coastal Beaufort Sea. Markers show the annual mean concentrations and lines show ±1 standard deviation (SD). Markers with no lines have an SD that is smaller than the marker.

Table 3-32. Means, standard deviations (SD), relative standard deviations [RSD = (mean/SD) x 100], maximums and minimums for metals in 22 samples of clams (*Astarte montagui* and *Cyrtodaria kurriana*) from the ANIMIDA and cANIMIDA study areas for combined data from 1999-2001, 2003 and 2004-2006.

Metal	Mean ± SD (µg/g, dry wt.)	RSD (%)	Maximum (µg/g, dry wt.)	Minimum (µg/g, dry wt.)	(Mean/Min)
Ag	0.07 ± 0.02	32	0.13	0.04	1.8
Al	1000 ± 454	45	2150	98	2.2
As	11.2 ± 2.1	19	16	8	1.4
Ba	17.2 ± 7.2	42	40	7	2.3
Be	0.052 ± 0.015	29	0.085	0.029	1.6
Cd	7.9 ± 3.1	39	13.1	0.5	1.6
Co	2.1 ± 1.0	47	4.0	0.8	1.9
Cu	13.0 ± 3.6	28	24	7	1.8
Fe	1555 ± 672	43	3640	771	2.3
Hg	0.060 ± 0.021	35	0.127	0.025	2.1
Mn	235 ± 163	69	637	47	2.7
Pb	0.62 ± 0.22	35	1.2	0.2	1.9
Sb	0.034 ± 0.015	43	0.059	0.010	1.7
Tl	0.018 ± 0.006	33	0.028	0.008	1.6
V	3.4 ± 1.4	42	6.9	1.3	2.0
Zn	79.9 ± 14.1	18	125	62	1.6
% H_2O	84.4 ± 2.9	3.5	92	78	1.18

3.2.3 Metals in Deployed Mussels

3.2.3.1 Summer 2004

A total of 18 metals were measured in soft tissues of the zero-time reference mussels and mussels that had been deployed for approximately two weeks near the Northstar Development, the Liberty Prospect, and at twp BSMP stations in 2004 (Table 3-33). Concentrations of most of the metals were higher in mussels that had been deployed for about two weeks in the Beaufort Sea than in zero-time reference mussels. Only zinc concentration was slightly higher in zero-time reference mussels than in the mussels that had been deployed in the Beaufort Sea. Because of the small sample sizes and large standard deviations, none of the differences was statistically significant. The difference between Beaufort Sea and reference mussels was greatest for aluminum and iron, suggesting that the excess metals in the Beaufort Sea mussels was derived from sediment particles on the gills or in the gut. Concentrations of most metals were lower in the single sample of Liberty mussels than in mussels that had been deployed at Northstar or the BSMP.

Table 3-33. Concentrations of 18 metals in reference mussels (*Mytilus trossulus*) and mussels that had been deployed at three locations in the Beaufort Sea in 2004. n= number of samples. Concentrations are µg/g dry wt.

Metal	Parameter	Liberty (n= 1)	Northstar (n= 3)	BSMP (n= 2)	Reference (n= 3)
Ag	Mean±SD	0.095	0.117±0.016	0.097±0.011	0.086±0.007
	Range		0.096-0.133	0.089-0.104	0.080-0.094
Al	Mean±SD	479	1336±568	1164.5±842	447±481
	Range		854-2030	569-1760	130-1000
As	Mean±SD	8.52	10.2±1.11	8.90±0.05	8.36±1.35
	Range		8.92-11.5	8.86-8.93	6.91-9.59
Ba	Mean±SD	8.6	13.0±5.7	10.8±2.76	5.2±3.5
	Range		8.3-20	8.8-12.7	3.1-9.3
Be	Mean±SD	0.012	0.027±0.012	0.027±0.014	0.009±0.006
	Range		0.018-0.044	0.017-0.037	0.005-0.016
Cd	Mean±SD	2.58	2.32±0.56	2.15±0.39	2.38±0.71
	Range		1.51-2.71	1.87-2.42	1.58-2.92
Co	Mean±SD	0.59	0.95±0.08	0.76±0.15	0.70±0.08
	Range		0.87-1.04	0.65-0.86	0.62-0.77
Cr	Mean±SD	1.52	2.34±0.85	2.63±1.55	1.42±0.72
	Range		1.63-3.4	1.53-3.72	0.99-2.26
Cu	Mean±SD	8.1	8.3±0.97	7.95±0.35	7.0±1.3
	Range		7.5-9.4	7.7-8.2	5.5-8
Fe	Mean±SD	380	865±341	811±494	374±265
	Range		569-1230	461-1160	198-678
Hg	Mean±SD	0.069	0.090±0.014	0.093±0.021	0.102±0.012
	Range		0.074-0.109	0.078-0.108	0.092-0.116
Mn	Mean±SD	9	17.1±3.5	17.9±11.0	9.5±5.5
	Range		13.7-21.2	10.1-25.6	5.8-15.8
Ni	Mean±SD	1.44	1.96±0.30	1.76±0.66	1.35±0.31
	Range		1.74-2.39	1.29-2.22	1.07-1.68
Pb	Mean±SD	0.731	0.994±0.108	0.815±0.054	0.88±0.33
	Range		0.861-1.1	0.777-0.853	0.504-1.10
Sb	Mean±SD	0.013	0.013±0.005	0.014±0.002	0.013±0.002
	Range		0.009-0.02	0.012-0.015	0.012-0.015
Tl	Mean±SD	0.035	0.024±0.003	0.027±0.004	0.005±0.001
	Range		0.022-0.028	0.024-0.029	0.005-0.006
V	Mean±SD	1.9	3.65±0.94	3.21±1.56	1.67±1.15
	Range		2.79-4.65	2.11-4.31	0.98-2.99
Zn	Mean±SD	93.8	93.3±34.2	92.7±8.6	99.7±33.7
	Range		46.8-121	86.6-98.8	64.1-131

3.2.3.2 Summer 2005

Mussels were deployed at Prudhoe Bay, in addition to Liberty, Nortstar, and BSMP in 2005. Concentrations of 10 (Al, Co, Cr, Cu, Fe, Hg, Mn, Ni, Pb, and V) of the 18 metals analyzed were higher in mussels deployed at Prudhoe Bay than in those deployed at Liberty, Northstar, and BSMP or in zero time reference mussels (Table 3-34). The largest differences were for Al, Fe, and Mn, indicating that the mussels at Prudhoe Bay were retaining suspended sediment particles in the gut and gills. Most of the other metals probably also were associated with ingested sediment particles.

Table 3-34. Concentrations of 18 metals in Mussels (*Mytilus trossulus*) following deployment in four locations in the Beaufort Sea in 2005, compared to reference animals. n= number of samples. Concentrations are µg/g dry wt.

Metal	Parameter	Liberty (n=3)	Northstar (n= 1)	BSMP (n= 2)	Prudhoe Bay (n= 1)	Reference (n= 2)
Ag	Mean±SD	0.074±0.005	0.089	0.092±0.021	0.086	0.076±0.006
	Range	0.069-0.078		0.077-0.106		0.071-0.08
Al	Mean±SD	806±202	131	613±433	1470	371±40
	Range	606-1010		306-919		342-399
As	Mean±SD	11.0±0.98	11.3	11.2±1.1	12.1	12.4±1.1
	Range	10.2-12.1		10.4-12		11.6-13.1
Ba	Mean±SD	8.77±1.84	7.2	6.9±2.1	8.6	6.0±0.7
	Range	7.2-10.8		5.4-8.3		5.5-6.5
Be	Mean±SD	0.027±0.008	0.017	0.024±0.018	0.05	0.014±0.006
	Range	0.019-0.034		0.011-0.037		0.01-0.018
Cd	Mean±SD	3.99±0.11	4.77	3.98±0.52	4.25	4.69±1.05
	Range	3.89-4.11		3.61-4.34		3.94-5.43
Co	Mean±SD	0.70±0.14	0.76	0.53±0.09	1.01	0.64±0.05
	Range	0.57-0.85		0.46-0.59		0.6-0.67
Cr	Mean±SD	1.84±0.35	1.12	1.91±0.15	3.31	2.21±0.21
	Range	1.46-2.14		1.8-2.01		2.06-2.35
Cu	Mean±SD	7.5±1.1	7	7.3±0.3	9.2	7.9±0.4
	Range	6.5-8.6		7.1-7.5		7.6-8.1
Fe	Mean±SD	685±164	265	606±267	1180	460.5±0.7
	Range	520-847		417-794		460-461
Hg	Mean±SD	0.066±0.003	0.253	0.073±0.013	0.451	0.052±0.001
	Range	0.063-0.068		0.064-0.082		0.051-0.052
Mn	Mean±SD	16.5±1.5	8.5	17.0±5.9	29.1	14.6±0.2
	Range	14.8-17.7		12.8-21.1		14.4-14.7
Ni	Mean±SD	1.76±0.19	1.27	1.57±0.13	2.86	1.81±0.13
	Range	1.56-1.94		1.47-1.66		1.72-1.9
Pb	Mean±SD	0.66±0.10	0.524	0.606±0.097	0.899	0.610±0.015
	Range	0.602-0.78		0.537-0.674		0.599-0.62
Sb	Mean±SD	0.015±0.007	0.008	0.013±0.008	0.018	0.022±0.001
	Range	0.007-0.02		0.007-0.018		0.021-0.022

Table 3–34. Concentrations of 18 metals in Mussels (Mytilus trossulus) following deployment in four locations in the Beaufort Sea in 2005, compared to reference animals. n= number of samples. Concentrations are µg/g dry wt., continued.

Metal	Parameter	Liberty (n=3)	Northstar (n= 1)	BSMP (n= 2)	Prudhoe Bay (n= 1)	Reference (n= 2)
Tl	Mean±SD	0.029±0.008	0.037	0.025±0.010	0.017	0.022±0.001
	Range	0.02-0.035		0.018-0.032		0.021-0.022
V	Mean±SD	1.65±0.54	0.99	0.98±0.09	3.23	1.11±0.06
	Range	1.08-2.15		0.91-1.04		1.07-1.15
Zn	Mean±SD	93±18.7	95.6	98.4±1.1	104	101±2
	Range	76-113		97.6-99.1		99-102

Concentrations of the 18 metals in mussels deployed at four locations in the Beaufort Sea in 2005 were similar to concentrations in the mussels that had been deployed there in 2004. As in 2004, metals concentrations in reference mussels usually were lower than concentrations in the mussels that had been deployed at the stations in the Beaufort Sea. Again, greatest differences were for aluminum and iron, suggesting that the differences were caused primarily by ingestion by the deployed mussels of suspended fine sediment particles.

3.2.3.3 Summer 2006

Mussels were deployed at SDI, and West Dock, in addition to Liberty, Northstar, and BSMP in 2006. Concentrations of most metals were similar in mussels from SDI and West Dock and higher than concentrations in time zero reference mussels and mussels that had been deployed at Northstar (Table 3-35). Largest differences among metals concentrations in mussels deployed at SDI and West Dock and reference mussels from Port Chatham were for Al, Ba, and Fe, possibly indicating a higher concentration of suspended clay-sized sediment particles at SDI and West Dock than at the mussel collection site. Largest differences between metals concentrations in mussels deployed at SDI and West Dock and those deployed at Northstar were for Ba, Cr, and Fe, all associated with drilling discharges and fine-grained sediments.

Table 3-35. Concentrations of 18 metals in Mussels (*Mytilus trossulus*) following deployment in five regions of the Beaufort Sea, compared to the time zero reference animals in 2006. n= number of samples. Concentrations are µg/g dry wt.

Metal	Parameter	Liberty (n= 3)	Northstar (n= 2)	BSMP (n= 1)	SDI (n= 1)	West Dock (n= 1)	Time Zero (n= 1)
Ag	Mean±SD	0.075±0.010	0.065±0.004	0.103	0.069	0.05	0.07
	Range	0.064-0.081	0.062-0.067				
Al	Mean±SD	1182±448	328±74.2	1910	1860	1690	474
	Range	665-1450	275-380				
As	Mean±SD	10.9±0.2	8.97±0.47	11.7	10.8	10.1	10.8
	Range	10.8-11.1	8.63-9.30				

Table 3–35. Concentrations of 18 metals in Mussels (Mytilus trossulus) following deployment in five regions of the Beaufort Sea, compared to the time zero reference animals in 2006. n= number of samples. Concentrations are µg/g dry wt., continued.

Metal	Parameter	Liberty (n= 3)	Northstar (n= 2)	BSMP (n= 1)	SDI (n= 1)	West Dock (n= 1)	Time Zero (n= 1)
Ba	Mean±SD	10.4±2.86	3.34±0.59	8.72	17.9	14.4	3.75
	Range	7.09-12.1	2.92-3.76				
Be	Mean±SD	0.028±0.005	0.012±0.004	0.021	0.038	0.038	0.011
	Range	0.022-0.032	0.009-0.014				
Cd	Mean±SD	4.23±0.22	3.14±0.37	5.27	4.16	3.72	4.07
	Range	3.98-4.39	2.88-3.4				
Co	Mean±SD	0.87±0.08	0.556±0.062	1.56	1.2	1.08	0.803
	Range	0.77-0.93	0.512-0.599				
Cr	Mean±SD	2.32±0.49	1.19±0.11	7.32	4.17	3.24	2.09
	Range	1.75-2.64	1.11-1.27				
Cu	Mean±SD	7.3±0.6	5.75±0.49	7.4	7.9	8.2	6
	Range	6.9-8.0	5.4-6.1				
Fe	Mean±SD	976±273	424±81	2010	1790	1470	659
	Range	669-1190	366-481				
Hg	Mean±SD	0.095±0.006	0.062±0.003	0.104	0.112	0.121	0.086
	Range	0.091-0.102	0.06-0.064				
Mn	Mean±SD	18.0±3.9	13±2.8	61.5	29.8	29.6	21.3
	Range	13.8-21.4	11-15				
Ni	Mean±SD	2.08±0.33	1.63±0.08	4.94	3.4	3.12	2.1
	Range	1.70-2.33	1.57-1.68				
Pb	Mean±SD	0.697±0.170	0.457±0.030	0.795	0.933	0.631	0.587
	Range	0.513-0.849	0.435-0.478				
Sb	Mean±SD	0.007±0	0.011±0.030	0.011	0.011	0.013	0.005
	Range	0.007-0.007	0.009-0.013				
Tl	Mean±SD	0.016±0.004	0.009±0.002	0.019	0.024	0.011	0.012
	Range	0.012-0.019	0.007-0.01				
V	Mean±SD	4.06±1.39	1.88±0.24	6.74	5.27	4.48	2.98
	Range	2.68-5.46	1.71-2.05				
Zn	Mean±SD	103.6±9.05	91.9±6.0	95	110	105	85.9
	Range	97.9-114	87.6-96.1				

3.2.3.4 Interannual Variation in Metals Concentrations in Zero-Time Reference Mussels and Mussels That Had Been Deployed in the Beaufort Sea: 2004, 2005, 2006

Concentrations of most metals in zero time reference mussels were similar in 2004, 2005, and 2006 (Figure 3-49). Interannual differences rarely were more than two-fold. Largest differences were for abundant metals, such as Fe and Mn. Thus, the zero time mussels serve as a good reference for the mussels that are deployed at locations in the Beaufort Sea.

Mussels were deployed at Liberty, Northstar, and BSMP in 2004, 2005, and 2006, so statistical comparisons can be made for all 18 metals in mussels deployed in three years and in three locations. There were statistically significant differences in concentrations of nine metals in the three years of the study: Ag, As, Cd, Co, Mn, Ni, Pb, and V (Table 3-36). Significant differences also were observed for Mn and Tl for location and year x location and for Ni for year x location. None of the differences in metal concentrations in mussels deployed in 2004, 2005, and 2006 at BSMP, Liberty, and Northstar were large.

Ag and Pb concentrations were significantly higher in deployed mussels in 2004 than in 2005 and 2006; As and Cd concentrations were lower in 2004 than in 2005 and 2006. V concentration was higher in 2005 than in 2004 and 2006. Co, Mn, and Ni concentrations were significantly higher in mussels deployed at BSMP in 2006 than in mussels deployed at other locations and years. Hg and Tl concentrations were very low in all mussels, but concentrations varied among years and deployment locations.

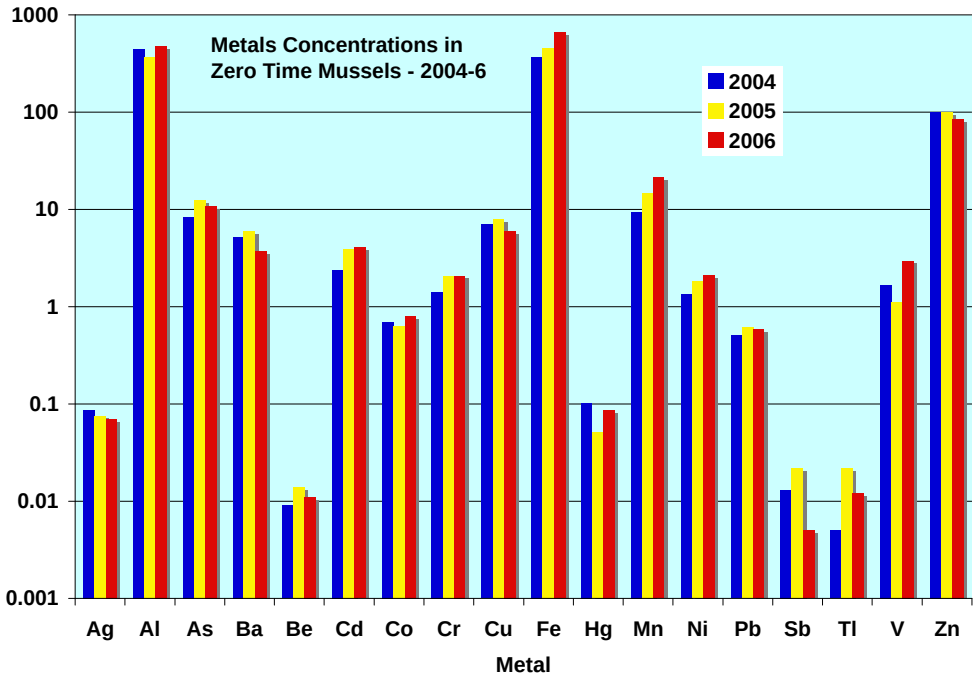


Figure 3-49. Mean metals concentrations in reference mussels collected at Port Chatham in 2004, 2005, and 2006. Note that the y-axis is log scale.

Table 3-36. Statistical comparison of metals concentrations in mussels (*Mytilus trossulus*) that were deployed at BSMP, Liberty, and Northstar in 2004, 2005, and 2006. Only metals for which statistically significant differences were detected are included. Differences were considered significant at $p \leq 0.05$ and are highlighted.

Metal	Source	DF	F Value	Pr > F
Ag	Year	2	5.65	0.0187
	Location	3	2.93	0.0770
	Year x Location	6	2.00	0.1451
As	Year	2	13.20	0.0009
	Location	3	0.37	0.7758
	Year x Location	6	2.46	0.0871
Cd	Year	2	30.10	<0.0001
	Location	3	0.56	0.6540
	Year x Location	6	2.12	0.1266
Co – Rank	Year	2	4.55	0.0339
	Location	3	0.28	0.8406
	Year x Location	6	6.88	0.0024
Hg	Year	2	2.56	0.1183
	Location	3	1.46	0.2755
	Year x Location	6	6.84	0.0024
Mn – Log	Year	2	6.76	0.0108
	Location	3	4.47	0.0251
	Year x Location	6	3.25	0.0392
Ni – Rank	Year	2	4.00	0.0466
	Location	3	0.61	0.6241
	Year x Location	6	3.15	0.0428
Pb	Year	2	4.60	0.0329
	Location	3	0.23	0.8755
	Year x Location	6	095	0.4971
Tl	Year	2	13.29	0.0009
	Location	3	7.50	0.0044
	Year x Location	6	3.87	0.0220
V	Year	2	13.72	0.0008
	Location	3	1.66	0.2285
	Year x Location	6	2.94	0.0528

3.2.3.5 Temporal Trends in Metals Concentrations in Zero-Time Mussels and Mussels Deployed at Northstar: 2002 through 2006

Twelve metals (Ag, As, Ba, Be, Cd, Cu, Fe, Hg, Pb, Tl, V, and Zn) were measured in the soft tissues of reference mussels and mussels deployed near the Northstar Development as part of Task 2 of the ANIMIDA Program (Brown et al., 2005). These 12 metals and an additional eight metals were analyzed in mussels in 2004 through 2006 as part of the cANIMIDA Program.

Concentrations of Ba, Be, Cu, Fe, and Zn were higher in reference mussels and mussels that had been deployed at the Northstar Development in 2002 than in 2004, 2005, and 2006 (Table 3-37). Concentrations of Ag and As were slightly lower in mussels deployed at the Northstar Development in 2002 than in those deployed there in 2004, 2005, and 2006. In all cases, the differences were small and the number of replicates in each year was small, indicating that the differences were not ecologically significant. In each year, the concentration of each metal was similar in reference mussels and mussels that were deployed at the Northstar Development, indicating the metals were accumulated primarily at the reference site, Port Chatham.

Table 3-37. Mean (± Sd) concentrations of 12 metals in tissues of reference mussels from Port Chatham, AK, and mussels from Port Chatham that had been deployed for 2 to 3 weeks near the Northstar Development in 2002 (ANIMIDA Program), 2004, 2005, and 2006 (cANIMIDA Program). . n= number of samples. Concentrations are µg/g dry wt.

Metal	**Reference Mussels**				**Mussels Deployed at Northstar**			
	2002 (n = 2)	2004 (n – 3)	2005 (n = 2)	2006 (n = 1)	2002 (n = 2)	2004 (n – 3)	2005 (n = 1)	2006 (n – 2)
Ag	0.08±0.01	0.09±0.01	0.08±0.01	0.07	0.08±0.01	0.12±0.02	0.09	0.06±0.00
As	6.9±0.6	8.4±1.4	12.4±1.1	10.8	6.7±0.10	10.2±1.11	11.3	8.97±0.47
Ba	18±3	5.2±3.5	6.0±0.70	3.75	17±2	13.0±5.7	7.2	3.34±0.59
Be	0.04±0.01	0.01±0.01	0.01±0.01	0.01	0.04±0.00	0.03±0.01	0.02	0.01±0.00
Cd	3.9±0.4	2.4±0.71	4.69±1.05	4.07	3.9±0.30	2.32±1.51	4.77	3.14±0.37
Cu	9.9±1.4	7.0±1.3	7.9±0.40	6.0	9.1±0.10	8.3±0.97	7.0	5.75±0.49
Fe	1300±80	374±265	460±0.7	659	1370±130	865±341	265	424±81
Hg	0.11±0.01	0.10±0.01	0.05±0.00	0.09	0.09±0.01	0.09±0.01	0.25	0.06±0.00
Pb	0.75±0.10	0.88±0.33	0.61±0.02	0.59	0.71±0.05	0.99±0.11	0.52	0.46±0.03
Tl	0.02±0.004	0.01±0.00	0.02±0.00	0.01	0.02±0.00	0.02±0.00	0.04	0.01±0.00
V	4.2±0.01	1.67±1.15	1.11±0.06	2.98	4.0±0.50	3.65±0.94	0.99	1.88±0.24
Zn	114±4	99.7±33.7	101±2.0	85.9	106±2.0	93.3±34.2	95.6	91.9±6.0

This page intentionally left blank

4.0 DISCUSSION

4.1 Hydrocarbons in Indigenous Fish and Invertebrates, Deployed Mussels, and SPMDs in the Oil and Gas Development Area of the Alaskan Beaufort Sea

4.1.1 Data Quality

A summary of of the quality control results from the invertebrate and fish tissue analyses generated in Task 5 of the cANIMIDA Program is included in Appendix C. The discussion below deals with the apparent interannual differences in hydrocarbon concentrations in fish and invertebrate tissues.

A review of the Summer 2005 hydrocarbon data for fish, indigenous invertebrates, and the deployed mussels indicated lower PAH and SHC concentrations in 2005, compared to 2004 and 2006. Data for selected 2004 and 2005 fish and mussel samples (i.e., four horn sculpin from the Northstar area and reference mussels) were reviewed in 2006; chromatograms indicated that the 2005 sample extracts appeared "cleaner" than the 2004 samples. Some additional chromatographic contribution could be seen in the 2004 samples suggesting there could some overestimation of PAH (from fluorene to alkyl phenanthrenes/anthracenes) in the 2004 samples. However, another possibility is that "cleaner" than typical chromatograms for the 2005 samples resulted in a small underestimation of the sample concentrations in that year, or that the measured concentrations indeed represent the field sample concentrations well despite some differences in the appearance of the chromatograms, and the year-to-year variations.

The method for calculating total saturated hydrocarbon (total SHC) concentrations in 2005 was changed slightly from the method used in 2004. This affects the sediment and tissue FID analysis data generated for summer 2004 samples. However, the difference is generally ~50-100 ng/g and the impact would be minimal and would, if anything, have resulted in a minor *negative* bias in the 2004 data, which was the year for which the highest hydrocarbon concentrations were generally observed.

The decline in hydrocarbon concentration that was observed for many biota samples between 2004 and 2005 was also observed for the sediment samples. In order to better understand this difference, selected fish and amphipod samples collected and analyzed in 2004 and 2005 were re-extracted and re-analyzed along with the 2006 samples, to ensure that all processing and analyses were identical; observed differences would therefore be due to actual field sample concentration differences. Samples for this exercise were selected from stations that were sampled during all three field seasons and the results were compared to the previous analyses to assess if any observed differences could be attributed to analytical differences between different years, or if the changes observed are more likely due changing field sample concentrations.

Generally, the reanalysis of the 2004 fish sample exhibited quite close agreement with the original analysis; the discrepancy were significantly smaller than had been observed between in the 2004 and 2005 field sample data for the same fish species and location. The reanalysis of the 2005 fish sample yielded somewhat higher PAH concentrations than the original analysis but was less useful for interpretation purposes, as the original 2005 analysis had poor sensitivity and limited individual compound data. The data from the re-extracted and re-analyzed 2004 and

2005 amphipod samples provided more useful information for data evaluation purposes, and included PAH, SHC, and S/T results (not only PAH). The reanalysis agreed well with the original analysis for all three organic parameter classes, despite the large differences in the results between 2004 and 2005.

The original results were, for the most part, confirmed in the re-analyses, and surprisingly large year-to-year variability in the field sample concentrations of PAH, SHC, and S/T was demonstrated. The results from the analysis of the samples collected in 2006 demonstrate continued large fluctuations in field sample concentrations, with much of the 2006 data falling somewhere between that observed for 2004 and 2005. The data from the re-analysis of the 2004 and 2005 samples, together with the analysis of the 2006 samples, demonstrated that the observed fluctuations could *not* be attributed to laboratory artifacts or any analytical factors, but indeed appear to reflect actual fluctuations in the organic compound concentrations in the field.

The differences in the analytical observations, and the significant variability, should be kept in mind in comparing the analytical results for hydrocarbons in tissues for 2004, 2005, and 2006. All mean hydrocarbon concentrations were higher in marine animals collected in 2004 and 2006 (and indeed in prior ANIMIDA years) than in 2005 (Tables 4-1 and 4-2), and it is unclear if 2005 is an anomalously low year, historically, and what caused the variations. However, much of the interannual variation in concentrations of PAH in tissues of marine animals in the coastal Beaufort Sea probably was caused by natural large interannual variations in the flux of hydrocarbon-containing suspended particulate matter from the rivers entering the Beaufort Sea. Brown et al. (2005) reported that a summer storm in 2004 may have resuspended surface fine-grained sediments from the study area and transported them offshore. Because most PAH in sediments are associated with the silt/clay fraction, such winnowing would have reduced PAH concentrations in sediments in 2005.

Table 4-1. Mean Concentrations of total polycyclic aromatic hydrocarbons (TPAH), total saturated hydrocarbons (TSHC), pristane, and steranes/triterpanes (StTr) in tissues of indigenous fish, clams, amphipods, isopods, and mysids collected at several locations in the Alaskan Beaufort Sea in 2004 through 2006. Significant within-year differences in PAH concentrations among locations are highlighted.

Taxon	Analytes	Location	Mean (SD) TPAH Concentration (ng/g dry wt)		
			2004	2005	2006
Fish (8 species)	TPAH	Northstar	38.8 ± 20.7	9.44 ± 6.85	**52.4 ± 12.8**
		Liberty	47.8 ± 25.3	10.1 ± 6.45	**24.8 ± 6.4**
		Tigvariak Island	30.4 ± 7.7	No data	No data
Amphipods	TPAH	Northstar	83.3 ± 52.7	13.8 ± 8.96	41.3 ± 27.4
		Liberty	73.6 ± 10.3	39.5 ± 10.1	81.5 ± 23.9
		BSMP	49.5 ± 16.7	23.6 ± 9.24	60.9 ± 14.4
	TSHC	Northstar	31,458 ± 2784	18,003 ± 1746	26,681 ± 17,987
		Liberty	26,203 ± 2010	44,625 ± 14,523	85,152 ± 41,029
		BSMP	28,704 ± 3948	26,914 ± 9980	43,679 ± 31,008
	Pristane	Northstar	26,968 ± 3237	16,598 ± 8736	24,749 ± 18,785
		Liberty	24,634 ± 2488	42,127 ± 17,377	81,071 ± 47,234
		BSMP	27,254 ± 5160	24,644 ± 14,816	39,986 ± 3397

Table 4–1. Mean Concentrations of total polycyclic aromatic hydrocarbons (TPAH), total saturated hydrocarbons (TSHC), pristane, and steranes/triterpanes (StTr) in tissues of indigenous fish, clams, amphipods, isopods, and mysids collected at several locations in the Alaskan Beaufort Sea in 2004 through 2006. Significant within-year differences in PAH concentrations among locations are highlighted, continued.

Taxon	Analytes	Location	Mean (SD) TPAH Concentration (ng/g dry wt)		
			2004	2005	2006
	StTr	Northstar	12.7 ± 4.37	0.57 ± 0.25	1.09 ± 2.40
		Liberty	8.10 ± 0.40	4.88 ± 2.27	8.38 ± 6.53
		BSMP	3.70 ± 1.93	0.52 ± 0.32	13.4 ± 17.8
Isopods	TPAH	Northstar	No data	No data	67.6 ± 12.8
		Liberty	No data	67.0 ± 9.83	88.5
		BSMP	No data	73.37 ± 4.36	114
Mysids	TPAH	Northstar	No data	No data	89.3 ± 24.4
Clams	TPAH	Liberty	91.85	No data	141 ± 57.8
		BSMP	97.1 ± 52.5	38.4 ± 12.3	No data
	TSHC	Liberty	1644	No data	5276 ± 666
		BSMP	1922 ± 621	1510 ± 634	No Data
	Pristane	Liberty	80.2	No data	96.6 ± 36,0
		BSMP	152 ± 40.6	434 ± 629	No data
	StTr	Liberty	8.99	No data	0
		BSMP	8.40 ± 6.20	1.72 ± 2.98	No data

Table 4-2. Mean Concentrations of total polycyclic aromatic hydrocarbons (TPAH), total saturated hydrocarbons (TSHC), pristane, and steranes/triterpanes (StTr) in tissues of reference and deployed mussels and of TPAH in blank and deployed SPMDs used to monitor hydrocarbon concentrations in the water column of the Alaskan Beaufort Sea in 2004 through 2006.

Matrix	Analytes	Location	Mean (SD) TPAH Concentration (ng/g dry wt or ng/SPMD)		
			2004	2005	2006
Deployed Mussels	TPAH	Reference	227 ± 34.9	32.8 ± 27.0	164 ± 36.2
		Northstar	148 ± 45.8	13.0	91.6 ± 5.18
		Liberty	92.8	24.8 ± 14.7	134 ± 6.74
		BSMP	157 ± 46.8	31.5 ± 1.46	52.7
	TSHC	Reference	6051 ± 522	3632 ± 151	23,159 ± 2294
		Northstar	7624 ± 2494	3381	21,024 ± 3599
		Liberty	5725	2689 ± 384	16,040 ± 3145
		BSMP	6246 ± 123	3137 ± 380	16,033
	Pristane	Reference	637 ± 63.4	270 ± 10.9	627 ± 86.0
		Northstar	365 ± 79.5	273	671 ± 180
		Liberty	337	146 ± 29.1	1153 ± 214
		BSMP	413 ± 162	290 ± 106	850

Table 4–2. Mean Concentrations of total polycyclic aromatic hydrocarbons (TPAH), total saturated hydrocarbons (TSHC), pristane, and steranes/triterpanes (StTr) in tissues of reference and deployed mussels and of TPAH in blank and deployed SPMDs used to monitor hydrocarbon concentrations in the water column of the Alaskan Beaufort Sea in 2004 through 2006, continued.

Matrix	Analytes	Location	Mean (SD) TPAH Concentration (ng/g dry wt or ng/SPMD)		
			2004	2005	2006
Deployed Mussels	StTr	Reference	12.2 ± 3.79	5.87 ± 5.87	ND
		Northstar	27.2 ± 16.2	6.9	ND
		Liberty	13.5	ND	ND
		BSMP	14.1 ± 6.43	ND	ND
SPMDs	TPAH	Blank	699 ± 55.0	No data	No data
		Northstar	750 ± 182	No data	No data
		Liberty	945	No data	No data
		BSMP	606 ± 155	No data	No data

ND, not detected.

4.1.2 Polycyclic Aromatic Hydrocarbons (PAH) in Marine Animals

4.1.2.1 Indigenous Marine Animals

Where data were available to make comparisons, PAH concentrations in indigenous marine animals collected near the Northstar Development (the site of oil development) usually were similar to those in the same species collected near the Liberty Prospect and at reference stations at Tigvariak Island and in the BSMP area (Table 4-1).

PAH in Fish Tissues

Eight species of fish were collected at one or more Beaufort Sea locations in one or more years of the program. TPAH concentrations and compositions were similar in all species. TPAH concentrations in all species combined were lower in 2005 than in 2004 and 2006. Mean TPAH concentration in all fish species combined was significantly higher at Northstar than at Liberty in 2006, but not in 2004 and 2005.

The PAH assemblage in all fish collected in the three years of the program was dominated by alkylnaphthalenes, with much lower concentrations of alkylphenanthrenes, particularly in 2006. This composition is consistent with a light petroleum source (e.g., middle distillate fuels or light crude oil). Traces of several pyrogenic PAH also were present in most fish tissues.

Exposure Biomarkers in Fish

Histochemical or enzymatic evidence of induction (increase in activity) of cytochrome P450 mixed function oxygenase activity (CYP1A) in fish tissues is used frequently as a biomarker of exposure to PAH and certain other inducers, such as PCBs and chlorinated pesticides (Collier et al., 1995; van der Oost et al., 2003). Concentrations of PAH metabolites, measured as fluorescent aromatic compounds (FAC) in bile, also are used frequently as biomarkers of exposure to PAH (Meador et al., 2008). CYP1A staining and bile FAC analysis were evaluated as biomarkers of

petroleum exposure in several species of fish collected in the Beaufort Sea in 2004 and 2005. CYP1A staining was very light in several tissues of five species of fish collected at several sampling sites in 2004 and 2005. Usually, CYP1A activity was lower in fish from Northstar than in those collected near Liberty or at other sampling sites. However, in 2005, CYP1A staining of hepatocytes and kidney tubules in humpback broad whitefish (a predominantly freshwater species that feeds in the summer in estuarine and nearshore marine waters) was stronger in fish from Northstar than in those from Point Brower (near Liberty). Bile FAC concentrations were similar in fish from Northstar and Liberty in both 2004 and 2005. These results are consistent with the tissue residue data in indicating very low-level exposure to PAH, possibly of petroleum origin, at all stations.

PAH in Indigenous Bivalve Mollusks and Crustaceans

Indigenous clams (*Astarte montagui* and *Cyrtodaria kurriana*) and three taxa of crustaceans (amphipods, isopods, and mysids) contained similar concentrations of TPAH in any one year of the program, with lowest concentrations in all species in 2005 (Table 4-1). As expected, mean TPAH concentrations in whole fish tissues were lower each year than in the four taxa of marine invertebrates, because of the greater ability of fish than mollusks and crustaceans to metabolize and excrete PAH (Neff, 2002a). The mean TPAH concentration in amphipods from Liberty was higher than in amphipods from Northstar in 2005; the mean TPAH concentration was significantly higher in amphipods from Liberty than in those from BSMP and Northstar in 2006 (Table 4-1).

The PAH assemblage in amphipods, isopods, mysids, and clams was dominated by alkylnapthalenes and alkylphenanthrenes, consistent with a predominantly petroleum source for the PAH in the invertebrate tissues. The most abundant five- through six-ring PAH in tissues of all four taxa was perylene, a predominantly biogenic PAH derived from diagenesis of organic matter of plant origin in anoxic sediments (Venkatesan, 1988). This may indicate that eroding coastal peat (high in perylene) may be important in the food web leading to these mollusks and crustaceans.

4.1.2.2 Deployed Mussels and SPMDs

Deployed Mussels

Mussels (*Mytilus trossulus*) collected each year from a mussel mariculture operation, certified by the state for mussel harvesting, in Port Chatham at the mouth of Lower Cook Inlet, were deployed for approximately two weeks at Northstar, Liberty, and BSMP, to monitor PAH in the water column. Unfortunately, reference mussels (collected at Port Chatham with mussels destined for deployment, but frozen for analysis immediately or after shipment to the North Slope), contained TPAH at concentrations similar to those in mussels following deployment in the Beaufort Sea (Table 4-2). In each year, the mean TPAH concentration in reference mussels was higher than that in mussels that were deployed at the three Beaufort Sea locations.

Concentrations of individual PAH in the PAH assemblage in the deployed mussels were different than that in the reference mussels, indicating that some exchange of PAH between Beaufort Sea water and suspended particles and the mussel tissues had occurred during the deployment.

The limited bioaccumulation of PAH by deployed mussels could have been due to a too short deployment period to allow for equilibration between Beaufort Sea water and mussel tissues or to stressful environmental conditions for Cook Inlet mussels in the Beaufort Sea. A deployment period of at least one month usually is recommended to allow tissue concentrations of non-polar organic chemicals, such as PAH, to equilibrate with concentrations in the ambient water and mussel food (Salazar and Salazar, 1995; Durell et al., 2006). The deployment period for mussels in the Beaufort Sea in 2004, 2005, and 2006 was limited to approximately two weeks because of logistic constraints. This may have been insufficient time for equilibration of some PAH, particularly three- through six-ring PAH that equilibrate slowly because of their low aqueous solubilities (Neff, 2002a). However, mussels were deployed near Northstar and at a reference site for three weeks in 2002 as part of the ANIMIDA program. TPAH concentrations in the mussels from the longer 2002 deployment were similar to those in mussels deployed for 2 weeks in 2004, 2005, and 2006. Thus, a short deployment period doesn't completely explain the lack of bioaccumulation of PAH by the deployed mussels.

The short deployment period probably combined with unsatisfactory environmental conditions to limit PAH equilibration between mussel tissues and Beaufort Sea water. Ambient water temperature and salinity in July and August within a few meters of the bottom at the locations where mussels were deployed usually was in the range of -1.5ºC to +2ºC and 20‰ to 32‰, respectively (Trefry 2006 personal communication). *Mytilus trossulus* is the most cold-tolerant of the mytilid bivalves; they can tolerate temperatures at least as low as -2ºC (Cusson et al., 2005). However, clearance rate, food absorption efficiency, and metabolic rate, measured as the scope for growth, decreases to low levels in *M. trossulus* at a temperature of - 1˚C; mussels transferred from + 4 or + 8˚C to - 1˚C decreased respiration and required more than 5 days to recover (Cusson et al., 2005). Feeding also is depressed markedly by high suspended sediment concentrations and low phytoplankton concentrations (Thompson, 1984). The salinity of the near-bottom water of the Beaufort Sea is in the optimum range for *M. trossulus* and similar to what it is at Port Chatham. Thus, since the Port Chatham mussels were not acclimated for more than a few days to Beaufort Sea conditions before deployment, low ambient temperature and high suspended sediment loads, probably coupled with a short deployment time may have limited PAH bioaccumulation (and depuration) in the deployed mussels. However, mussels from Port Chatham that were deployed near Northstar and at a reference area in 2002 as part of the ANIMIDA Program exhibited bysal thread growth, an indication that they were active during deployment. Activity probably was slowed enough to slow equilibration of hydrocarbons between Beaufort Sea water and food and tissues of deployed mussels. Future deployments should be for longer periods of time (preferably one month or more) with mussels that have been acclimated to low Beaufort Sea water temperatures for a few weeks.

There also may have been limited bioconcentration of PAH by mussels deployed in the Beaufort Sea because of the low concentrations of dissolved PAH in surface waters there. Trefry et al. (2004) reported that concentrations of dissolved total PAH in surface waters near the Northstar and Liberty prospects in 2001 and 2002 ranged from 12.7 to 18.5 ng/L (parts per trillion). By comparison, dissolved TPAH concentrations in areas of Prince William Sound, AK, not oiled by the 1989 *Exxon Valdez* oil spill ranged from 1 to 39 ng/L (Boehm et al., 2007) a concentration range considered typical for uncontaminated near-shore waters. The concentration of TPAH in waters of lower Cook Inlet has not been measured. However, it is possible to conclude that the

low TPAH concentrations in mussels following deployment for a few weeks in the Beaufort is due in part to the low ambient concentrations of dissolved PAH in the water column.

SPMDs

SPMDs were deployed at three locations in the Beaufort Sea in 2004 to measure PAH in the water column. They produced little useful data because blank (unused) SPMDs contained about as much TPAH as deployed ones (Table 4-2). As with the deployed mussels, there was some evidence of exchange of some PAH between the ambient Beaufort Sea water and the SPMDs. PAH exchange probably was slowed by the low ambient temperatures and short deployment period. As with mussels, SPMDs and other passive samplers require deployment times of a month or more to reach equilibrium with dissolved PAH, particularly the higher molecular weight PAH, in the water column (Cornelissen et al., 2008). The dominant PAH in both blank and deployed SPMDs were naphthalenes; very little high molecular weight PAH were present, indicating limited equilibration and low concentrations of high molecular weight PAH in Beaufort Sea sea water. Therefore, SPMDs showed no advantage over mussels and they were not used in subsequent years.

SPMDs were deployed in 2002 at locations near the Northstar Development and at a reference site to the east as part of the ANIMIDA Program. Blank SPMDs from 2002 contained more than twice the amount of PAH contained in blank SPMDs from 2004 (1604 ng/SPMD versus 699 ng/SPMD). As in 2004, naphthalenes were the dominant PAH in blank and deployed SPMDs in 2002. Concentrations of most PAH in the SPMDs deployed in 2002 were higher than the concentrations of the corresponding PAH in SPMDs deployed in 2004, confirming that the blank PAH were masking any PAH analytes accumulated during deployment.

The combined SPMD and deployed mussel PAH data indicate that PAH concentrations were very low (probably lower than in Port Chatham) in Beaufort Sea water and suspended fine particulate matter. Amounts of PAH in SPMDs following deployment in the Beaufort Sea were lower than amounts in most SPMDs that had been deployed in the vicinity of offshore production platforms in the North Sea (Durell et al., 2006). Estimated TPAH concentrations in North Sea water, estimated from amounts of individual PAH in SPMDs that had been deployed for a month in the North Sea, range from about 4 to 25 ng/L at locations where the SPMDS contained amounts of TPAH similar to those in the SPMDs deployed in the Beaufort Sea. The background concentration of TPAH in coastal estuarine and marine waters is about 5 to 10 ng/L (Neff, 2002a). Beaufort Sea water probably contained similar TPAH concentrations.

4.1.2.3 Integration

Mean TPAH concentrations in fish, deployed mussels, amphipods, isopods, mysids, and clams collected from all stations in 2004, 2005, and 2006, as well as fish collected in 2001 and amphipods, clams, and deployed mussels collected in 2000 and 2002 in the ANIMIDA Program ranged from 9.44 ng/g dry wt to 157 ng/g (parts per billion) (Tables 4-1 and 4-2). By comparison, the median concentration of TPAH in tissues of mussels and oysters collected in coastal waters of the United States in 2002/2003 and analyzed in the National Status and Trends Mussel Watch Program 220 ng/g dry wt (O'Connor and Lauenstein, 2006). The lowest concentration of TPAH measured in mussels and oysters in the Mussel Watch Program was 7.3

ng/g. Thus, invertebrates and fish contain low concentrations of TPAH, compared to mussels and oysters (the greatest bioaccumulators) from the other US states.

The concentrations of TPAH in the tissues of most of the bivalve mollusks, crustaceans, and fish sampled in this program were in the range of or lower than expected for the same or similar species in relatively unpolluted marine environments throughout the world (Table 4-3). PAH usually are more abundant in mollusks than in crustaceans and fish. Fish and crustaceans have a well developed, inducible cytochrome P450 mixed function oxygenase system that rapidly metabolizes bioaccumulated PAH, facilitating their rapid excretion. This pattern of distribution of TPAH was evident in bivalve mollusks, crustaceans, and fish in the Alaskan Beaufort Sea (Tables 4-1 and 4-2).

As discussed above, the PAH profiles in animal tissues and SPMDs from the Beaufort Sea are consistent with a primarily petrogenic source. Thus, marine biota from throughout the production area of the Beaufort Sea are being exposed to low concentrations of fossil hydrocarbon PAH. These PAH may be coming from aerial deposition (arctic haze and combustion plumes offshore and onshore oil and gas operations), small boat fuel leaks, small releases from production activities on and offshore, and runoff from land, mainly via rivers (Steinhauer and Boehm, 1992), particularly from erosion of peat from river banks and the shore. TPAH concentrations were not higher in marine animals collected near the Northstar Development than at other areas of the Beaufort Sea, indicating that the offshore operations were not a major source of PAH in the animal tissues. The TPAH concentrations in tissues of Beaufort Sea invertebrates and fish are well below concentrations that might pose a health risk to fish, wildlife, and humans that might consume marine foods from the Beaufort Sea (Neff et al., 2006).

Table 4-3. Concentrations of several metals and TPAH in whole soft tissues or muscle tissues (fish) of marine bivalve mollusks, crustaceans, and fish from unpolluted marine environments throughout the world. The data for bivalve mollusks include results of "mussel watch" programs from the U.S. and Europe. Concentrations are μg/g dry wt. From Neff (2002a).

Chemical	Bivalve Mollusks		Crustaceans		Fish	
	Geomean	Range	Geomean	Range	Geomean	Range
Arsenic	11	0.13 – 214	15	<0.1 – 270	6.1	0.05 – 450
Barium	4.4	0.09 – 179	3.4	0.02 – 202	0.13	0.007 – 49
Cadmium	1.18	0.05 – 26.1	1.85	0.14 – 117	0.10	0.001 – 5.8
Mercury	0.17	0.004 – 11.7	0.45	0.02 – 6.2	0.77	0.01 – 115
Chromium	3.5[a]	0.1 – 10.0	2.6[a]	0.12 – 10.1	2.1[a]	0.03 – 5.8
Copper	51.8[a]	6.4 – 150	75.4[a]	8.8 – 241	3.9[a]	0.6 – 26
Lead	4.5[a]	<0.1 – 21.4	4.5[a]	0.03 – 17.5	7.09[a]	0.02 – 55.9
Zinc	290[a]	40 – 1315	68.6[a]	23.9 – 96.5	28.8[a]	4.1 – 58.8
TPAH	0.65	0.003 – 1729	0.17	0.004 – 13.4	0.19	0.002 – 23.4

[a] arithmetic mean.

4.1.3 Saturated Hydrocarbons (SHC) and Steranes/Triterpanes (StTr) in Mollusks and Crustaceans

4.1.3.1 Saturated Hydrocarbons (SHC)

The saturated hydrocarbon data yielded some information that, with the TPAH and St/Tr data, was useful in the food chain conceptual model that was developed as part of this task. Total SHC concentrations were very high in tissues of amphipods, clams, and deployed mussels (Tables 4-1 and 4-2). Most of the SHC in amphipods, but not in clams or deployed mussels, was a single isprenoid hydrocarbon, pristane. Pristane is present in petroleum, but its main source in the marine environment is from zooplankton, particularly copepods of several genera, including *Neocalanus* and *Pseudocalanus*, the dominant zooplankton in Beaufort Sea waters in most seasons (Horner and Murphy, 1985).

Pristane is abundant in marine food webs, particularly at Arctic and sum-Arctic latitudes, that include large populations of large lipid-rich Calanoid copepods. Calanoid copepods bioaccumulate phytol, a monounsaturated diterpenyl alcohol that is esterified with chlorophyll, in their phytoplankton food and convert it to pristane; the pristane accumulates to high concentrations in lipid droplets in the copepods (Avigan and Blumer, 1968). Pristane is readily bioaccumulated by the many species of marine invertebrates, fish, birds, and mammals that rely on copepods or their predators for food (Short, 2005). Short (2005) showed that mussels in Prince William Sound, AK, bioaccumulate pristane from fecal material produced by juvenile pink salmon that feed primarily on *Neocalanus* and not from copepod feces or dissolved pristane in the ambient water. *Neocalanus* from Prince William Sound contain 4000 to 8000 μg/g pristane (Short, 2005). Pristane does not accumulate in marine sediments, but apparently is biodegraded rapidly at the sediment/water interface (Prahl and Carpenter, 1984). The amphipods sampled in this study (*Anonyx nugax*) are carnivores/scavengers that consume any carrion that settles to the sea floor and have been observed preying directly on pelagic copepods near the ice edge in the Barents Sea (Werner et al., 2004). Highest pristane concentrations were in amphipods collected in the Boulder Patch, a highly productive area north of the Liberty Prospect. Lowest concentrations were in amphipods from Harrison Bay, suggesting that the pristane in amphipods doesn't come primarily from peat in river runoff.

Amphipods also may bioaccumulate pristane from ingestion of detritus that accumulates at the sediment/water interface. Pristane is present at low concentrations in peat, which is eroding into coastal waters from shoreline sediments and upland soils (Steinhauer and Boehm, 1992). Peat collected from four rivers emptying into the Beaufort Sea contained 22 to 84 ng/g pristane. The amphipods probably accumulate pristane from ingestion of copepods and detritus, derived from dead copepods and zooplanktivorous fish, and from eroded peat. Clams may be feeding on a different fraction of the benthos. The indigenous clams, *Astarte* and *Cyrtodaria*, occupy sandy nearshore sediments and finer shallow bay and river mouth sediments, respectively, and may be feeding on organic particles in interstitial water and ingested sediments (Bernard, 1979). The deployed mussels are accumulating SHC, including pristane, from suspended organic matter (seston). Reference mussels contained similar or slightly higher concentrations of SHC and pristane as deployed mussels (Table 4-2), indicating that most of the SHC were accumulated from the waters of Port Chatham. Port Chatham may have a higher biomass of calanoid copepods than the nearshore Beaufort Sea. Much of the remaining SHC in the mussels is from

terrestrial plant material, which may enter lower Cook Inlet in larger amounts than in the coastal Beaufort Sea.

4.1.3.2 Steranes/Triterpanes (StTr)

Concentrations of St/Tr, often used as tracers of crude oil sources, were low in all benthic invertebrates analyzed; amphipods, clams, and deployed mussels. The small amounts present may be from terrestrial plant materials, detritus, peat, or petroleum. The StTr profiles in the invertebrate tissues had some resemblance to the StTr profiles in Colville River peat, but little resemblance to the StTr in Northstar crude oil. Both recent and fossil biomarkers were present in the tissues, indicating that they were bioaccumulating hydrocarbons from organic detritus, peat, and possibly petroleum. Amphipods, mysids and clams also contained high concentrations of the diagenic PAH, perylene, which is abundant in Coleville River peat (Figure 3-16). The tissue residue data for PAH, SHC, and StTr indicate that hydrocarbons in tissues of near-shore benthic animals from the development area are derived in part from organic matter entering the Beaufort Sea from nearby rivers and also from coastal erosion. Petrogenic St/Tr from the Northstar Development contributes little or none to the St/Tr in tussues of marine animals near Northstar.

4.2 Metals in Beaufort Sea Indigenous Marine Animals and Deployed Mussels

Concentrations of 18 metals in tissues of several species of fish, amphipods, isopods, and clams collected near the Northstar Development, in the Liberty Prospect area, and in other reference areas were similar in 2004, 2005, and 2006, and comparable to metals concentrations in the same species collected in the ANIMIDA Program. When statistically significant differences were detected among years or locations, the differences were small. Northstar is the only offshore area where a large amount of development drilling has occurred and where oil production currently is occurring. A few metals concentrations were higher in marine animals from Northstar than in the same species from the other sampling sites (Pb in amphipods and Ag and Cu in fish); however, there were no consistent differences by year or location.

Although there were interannual differences in concentrations for several metals, the differences were small and all concentrations were in the range for marine animals from clean marine environments (Table 4-3). Concentrations of most metals were higher in clams and mussels than in amphipods and fish. Aluminum and iron concentrations tended to be higher in clams than in crustaceans and fish, possibly indicating that the clams are retaining sediment particles in the gut and gills.

There were no consistent trends in concentrations of metals in fish tissues. Demersal species, such as four horn sculpin tended to contain higher concentrations of metals than the more pelagic species. The anadromous species, arctic char, tended to contain lower concentrations of metals and hydrocarbons than the other species did, perhaps because they spend much of the year in fresh water where dissolved concentrations of most metals are lower than in the ocean.

The concentrations of metals in the tissues of most of the bivalve mollusks, crustaceans, and fish sampled in this program were in the range expected for the same or similar species in relatively unpolluted marine environments throughout the world (Table 4-3). Concentrations of different metals vary widely in tissues of different taxa of marine animals. Zinc is particularly abundant in mollusks (particularly oysters); copper often is abundant in crustaceans and oysters. Mercury

tends to be more abundant in fish tissues, particularly muscle, than in tissues of marine invertebrates. Most of the mercury in fish muscle is methylmercury. This pattern of distribution of metals was evident in bivalve mollusks, crustaceans, and fish in the Alaskan Beaufort Sea.

The National Status and Trends median metals concentrations in mussels and oysters can be condidered normal values (Table 4-4). A few fish, mussel, amphipod, and clam samples from the Beaufort Sea contained higher than the NS&T median concentrations of the 8 metals for which median values are available. As expected, amphipods contained copper at concentrations that exceeded the median value for copper in mussels, but not oysters. Crustaceans often contain naturally high concentrations of copper. As discussed above fish muscle often contains slightly elevated concentrations of mercury, as methylmercury. Some fish collected in the Beaufort Sea contained greater than the mussel median value for mercury of 0.24 μg/g. The mercury concentration in Beaufort Sea fish is well below the 1 μg/g wet wt screening concentration for edible tissues of fishery products consumed by man set by the Food and Drug Administration (FDA, 2001). Most marine fish contain mercury concentrations similar to or higher than concentrations reported in this investigation for Beaufort Sea fish. These concentrations of metals in tissues probably are not harmful to the animals themselves or to consumers of these species, including man.

Table 4-4. Concentration ranges of several metals in fish, mussels, amphipods, and clams collected in the Beaufort Sea between 2000 and 2006 as part of the ANIMIDA and cANIMIDA Programs, compared to the National Status and Trends median concentration ranges for mussels or oysters collected in US coastal waters between 1986 and 2003 (From O'Connor and Lauenstein, 2006). Concentrations are μg/g dry wt (ppm).

Metal	Fish	Mussel	Amphipod	Clam	NS&T Medians
Silver (Ag)	0.01 – 0.35	0.05 – 2.5	0.8 – 4.0	0.04 – 0.13	---
Aluminum (Al)	---	131 - 2000	96 - 1200	98 - 2200	---
Arsenic (As)	0.55 – 16	6.2 - 13	4.0 – 17	8 - 16	8.1 – 9.6
Barium (Ba)	0.30 – 47	2.9 - 20	7.4 - 59	7 - 40	---
Berylium (Be)	---	0.01 – 0.06	0.01 – 0.03	0.03 – 0.08	---
Cadmium (Cd)	0.01 – 0.37	0.29 – 5.4	0.3 – 2.4	0.53 – 13	2.1 – 2.9
Cobalt (Co)	---	0.46 – 2.6	0.6 – 2.9	0.8 – 4.0	---
Chromium (Cr)	0.04 – 3.8	0.65 – 7.3	---	---	---
Copper (Cu)	1.1 – 21	5.4– 9.40	41 - 210	7.0 - 24	(8.0 – 10)[a] (91 – 140)[b]
Iron (Fe)	19 – 1200	200 - 2000	100 - 950	770 - 3600	---
Mercury (Hg)	0.02 – 0.5	0.01 – 0.45	0.02 – 0.19	0.0 – 0.13	0.09 – 0.11
Manganese (Mn)	---	5.8 - 260	10 - 71	47 - 640	---
Nickel (Ni)	0.03 – 4.4	1.1 – 4.9	0.8 – 6.7	1.92 – 5.34	1.6 – 2.2
Lead (Pb)	0.01 – 2.6	0.22 – 1.1	0.05 – 0.7	0.18 – 1.9	0.63 – 0.98
Antimony (Sb)	---	0.01 – 0.03	0.01 – 0.04	0.01 – 0.06	---
Selenium (Se)	0.94 – 5.7	---	---	---	2.3 – 3.0
Tellurium (Tl)	---	0.01 – 0.04	0.01 – 0.03	0.01 – 0.03	---
Vanadium (V)	0.05 – 5.1	0.91 – 6.7	0.5 – 3.4	1.3 – 6.9	---
Zinc (Zn)	36.0 – 120	47 – 130	54 - 170	62 - 130	(110 – 140)[a] (1600 – 2400)[b]

[a] Mussels; [b] Oysters

4.3 Conceptual Model of Bioaccumulation and Trophic Transfer in the Beaufort Sea Food Web

A conceptual food web model is useful as a tool for designing scientifically defensible studies of the sources and distribution of chemical contaminants in the coastal Alaskan Beaufort Sea. It provides the basis for development of testable null hypotheses about relationships among physical and chemical disturbance of marine ecosystems by offshore oil development and production operations, climate change, and ecological changes.

4.3.1 Contaminant Sources in the Beaufort Sea

Metals and hydrocarbons (SHC, PAH, St/Tr) are natural ingredients of the Alaskan Beaufort Sea environment. Large amounts of metals and hydrocarbons enter the Beaufort Sea each year

associated with suspended particles in river runoff (Steinhauer and Boehm, 1992; Trefry et al., 2003; Rember and Trefry, 2004). Additional metals and hydrocarbons enter coastal waters and sediments of the Beaufort Sea from dry and wet deposition from the atmosphere. The arctic aerosol over northern Canada and Alaska contains relatively high concentrations of metals, PAH, SHC, and other persistent organic pollutants (POPs) (Matsumoto et al., 1998; Muir et al., 1999; Cheng and Schroeder, 2000). Climate change in the arctic is changing wind patterns, changing the patterns and masses of contaminants deposited to the Arctic seas from aerosols (Macdonald, et al., 2005).

Metals also may enter coastal and offshore waters of the Beaufort Sea from coastal and offshore oil and gas operations (Steinhauer and Boehm, 1992; Trefry et al., 2003). The major sources of metals in Beaufort Sea sediments from oil development activities are causeway and drilling island construction and drilling mud and cuttings discharges (Northern Technical Services, 1982; Naidu et al., 2001; Neff, 2002b, 2005). Island construction material usually has a metal concentration similar to that of natural soils and sediments, but sometimes may contain elevated concentrations of some metals (e.g., Crippen et al., 1980).

The metals frequently found in drilling muds and cuttings that are of greatest concern because of their potential toxicity and/or abundance in drilling muds include arsenic, barium, chromium, cadmium, copper, iron, lead, mercury, nickel, and zinc. Only barium, chromium, iron, lead, and zinc, are present frequently in drilling mud/cuttings solids at concentrations significantly higher (> 100-fold) than concentrations in clean marine sediments (Neff, 2005). However, most of these metals are present as insoluble sulfide inclusions in drilling barite and are not bioavailable to marine animals (Neff, 2007).

PAH also have natural and anthropogenic sources (Neff, 2002a). The major sources of the complex PAH assemblages found in most soils and sediments are combustion soot and fossil hydrocarbon mixtures (peat, coal, and petroleum). PAH from these sources enter the Beaufort Sea in petroleum spills and in river runoff and aerial deposition.

Oil development and production activities on the North Slope also may contribute PAH to coastal waters of the Beaufort Sea. In the past, the major sources of PAH inputs to the environment from oil and gas activities were permitted discharges of drilling mud/cuttings and produced water (Neff, 2002a, 2005). However, produced water is not discharged to coastal waters of the Alaskan Beaufort Sea; it is reinjected. Currently, water based drilling muds are the only ones used offshore in the Beaufort Sea. The current NPDES permit for Alaska allows discharge of water based drilling muds and cuttings to federal waters of the Beaufort Sea if they meet effluent limitation guidelines. Drilling mud discharges usually are limited to winter discharges onto the ice in deep water. Water based drilling muds, if used and discharged in accordance with regulations; contribute only small amounts of metals and hydrocarbons to the local marine environment. Completion fluids that usually are used to drill the hydrocarbon-bearing formation are not permitted for discharge because they often contain free oil. They may be reinjected if a suitable geologic formation is available or transported to shore for upland disposal.

PAH also are emitted to the atmosphere by flaring waste gases at production platforms or gas-treatment facilities. Approximately 162,000 million standard cubic feet (mscf) of waste gas was flared at Northstar in 2004 (Alaska Oil and Gas Conservation Commission). The PAH concentration in the flare exhaust is not known. Hydrocarbons, including PAH, may also be emitted in diesel exhaust and fugitive emissions from petroleum production, treatment, storage, and transportation facilities. Accidental drilling mud/cuttings, petroleum, and wastewater releases also are a potential source of PAH to the Beaufort Sea.

4.3.2 Bioaccumulation and Trophic Transfer of Metals and Hydrocarbons in the Beaufort Sea Food Web

There is concern that offshore oil and gas development may harm the Beaufort Sea marine environment by habitat alteration and by contamination of the local food web with harmful chemicals from accidental or intentional discharges from development and production activities. Marine invertebrates, fish, birds, and mammals living in the Beaufort Sea are able to bioaccumulate metals and PAH from the ambient seawater, from contaminated sediments, and from their food (Neff, 2002a). The concentrations of metals and PAH in tissues of marine animals are assumed to be at equilibrium with concentrations in the ambient water and food. Benthic invertebrates bioaccumulate contaminants primarily from solution in sediment pore water, overlying bottom water, and ingestion of sediment particles; benthic carnivores may also bioaccumulate contaminants from their food. Demersal and pelagic invertebrates, fish, birds, and mammals bioaccumulate the contaminants primarily from their food. Thus, a local increase in concentrations of metals and PAH in water or sediments due to oil development and production discharges may lead to an increase in the concentration of the contaminants in tissues of marine organisms in the local food web.

None of the target PAH or metals is known to biomagnify in marine food webs (with the possible exception of methylmercury) (Neff, 2002a,b; Dehn et al., 2006a,b). Some metals, such as cadmium, bioaccumulate continuously in certain tissues, such as kidneys, during the life of marine animals (Dehn et al., 2006a,b). The accumulated metals usually are in solid, non-bioavailable concretions, such as mercuric selenide in cetacean liver; this is not considered biomagnification (Neff, 2002b). This means that the concentrations of the metals and hydrocarbons do not increase “up the food chain” with highest concentrations in the top consumer, usually a marine bird or mammal. However, all the target contaminants are transferred through marine food webs; measured concentrations are lower in the whole predator tissues than in the whole prey tissues. Arctic marine animals are about as sensitive as temperate and tropical species to petroleum and metals toxicity (Neff, 2002a; Perkins et al., 2003). Thus, if chemicals from offshore oil development activities are bioaccumulated to high enough concentrations, they may cause adverse effects in the Beaufort Sea food web.

This conceptual model of bioaccumulation, trophic transfer, and effects was used as a basis for developing the objectives and study design for this task. A major focus of this task is to develop the information needed to protect indigenous people and the subsistence resources upon which they rely from injury due chemical contamination from offshore oil and gas operations. This conceptual model is being used to evaluate the possible bioaccumulation and food web transfer of metals and PAH derived from offshore oil and gas development activities in the Beaufort Sea. Sediments, benthic invertebrates (lower trophic level), and fish (higher trophic level) from the

Beaufort Sea development area have been analyzed in the ANIMIDA and cANIMIDA Programs for several metals and PAH that are associated with oil development activities. Marine mammals and birds (the apex consumers: Figure 4-1) have not been analyzed for the target chemicals. However, there is a large amount of data available on concentrations of several metals and organochlorine compounds, but not hydrocarbons, in several species of marine birds and mammals from the Beaufort Sea (e.g., Honda et al., 1990; Zeisler et al., 1993; Mackey et al., 1995, 1996; Krone et al., 1999; Woshner et al., 2001, 2002; Hoekstra et al., 2002, 2003, 2005; Kucklick et al., 2002; Borga et al., 2004; Franson et al., 2004; Dehn et al., 2005, 2006a,b; Riget et al., 2005; O'hara et al., 1999, 2006).

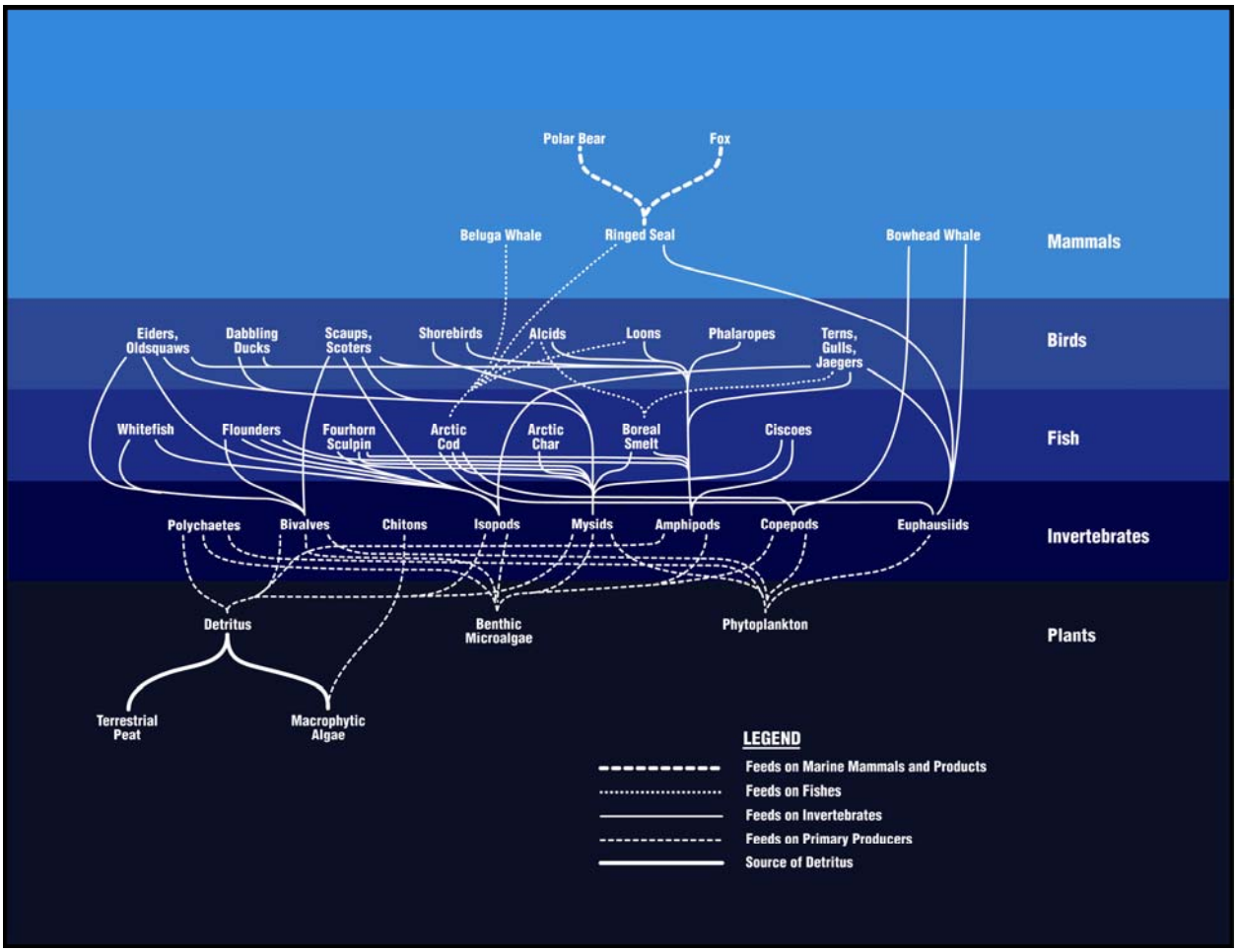


Figure 4-1. A diagram of the Beaufort/Chukchi Sea food web model, showing carbon pathways from terrestrial peat and primary production to top keystone species of marine birds and mammals. Modified from MMS (1990).

4.3.3 The Beaufort Sea Food Web

The harsh marine environment of the Beaufort Sea, with wide seasonal variations in light intensity, ice cover, freshwater input from rivers, and nutrient concentrations results in wide seasonal cycles of primary production, and complex, seasonally variable food webs (Walsh et al., 2005). Food web structure and carbon flow on the inner continental shelf of the Alaskan

Beaufort Sea are different in the nearshore (inside the barrier islands) and offshore environments and are weakly linked (Figure 4-2). For example, although terrestrially derived peat makes a substantial contribution to available particulate organic carbon (POC) in the nearshore marine environment (left side of Figure 4-2), it contributes much less to offshore organic carbon fluxes (right side of Figure 4-2) (Schell, 1983; Dunton et al., 2006). For example, arctic cod in Beaufort Sea lagoons may derive up to 70 percent of their carbon from terrestrial carbon (mostly peat); offshore populations derive most of their carbon from zooplankton (Dunton et al., 2006).

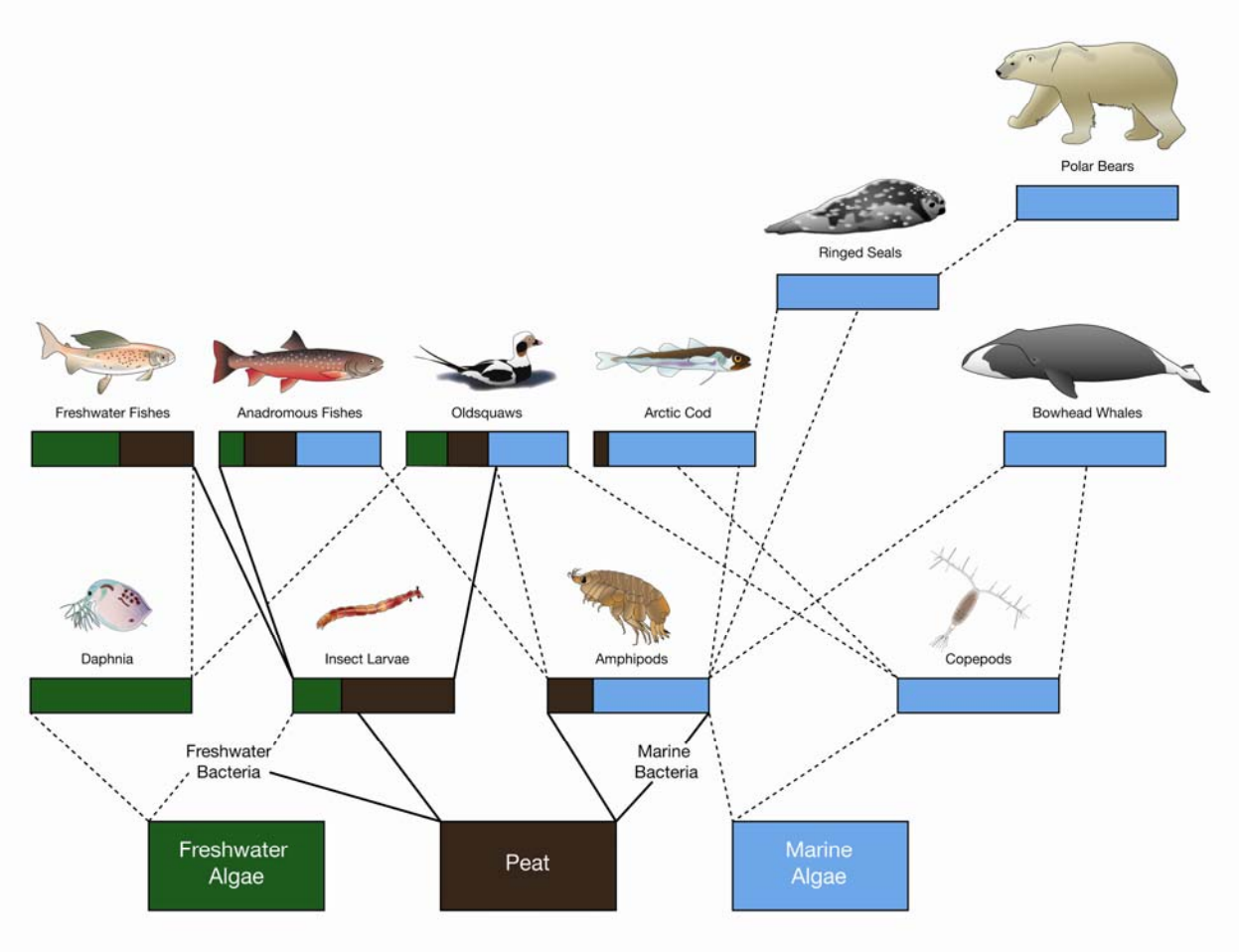


Figure 4-2. A generalized food web structure showing the sources of organic carbon, including peat, in the Alaskan Beaufort Sea coastal ecosystems. The shadings of the rectangles below each taxon shows the relative contribution and pathway of carbon from freshwater algae, terrestrial peat, and marine algae in the food web. Food chain structure grades from near-shore, freshwater/estuarine on the left to offshore marine on the right. From Schell et al. (1982) as presented in Norton and Weller (1984).

The Beaufort Sea food web is a relatively simple one with relatively few abundant taxa at each trophic level (Figure 4-1). Nutrients to support primary production in the Alaskan Beaufort Sea are derived primarily from inflows from the Bering Sea, Anadyr Water (from the Russian Chukchi Sea), the Canadian Beaufort Sea, and upwelling from the Arctic Basin (Dunton et al., 2003a,b, 2004). Primary production is pelagic (phytoplankton), eponic (living on the underside

of sea ice), and benthic (micro- and macro-algae living on the sea floor). Primary production, measured as chlorophyll a concentrations in surface waters, is lower in the Beaufort than in the Chukchi Sea (Dunton et al., 2003a,b, 2004). Near-shore areas east of Barrow and off the Coleville River have the highest primary production in coastal and continental shelf waters of the Beaufort Sea.

Particulate organic carbon (POC) produced by primary producers (phytoplankton) or in runoff from land may be consumed by primary consumers (mostly zooplankton), or exported to sediments or off the shelf (Wasserman et al., 2003; Moran et al., 2005). The POC supports blooms of zooplankton communities, dominated by calanoid copepods and euphausiids, which are consumed by bowhead whales.

In the nutrient rich northeastern Chukchi Sea, the fraction of primary production that is exported from the water column to the underlying sediments increases from about 15 percent in the spring to about 32 percent in the summer (Moran et al., 2005). POC in sediments supports development of a rich benthic fauna that supports benthic feeders, such as some seals and many species of demersal fish, and results in strong benthic/pelagic coupling of nutrients (Figure 4-2). This organic matter tends to sequester metals and hydrocarbons and is ingested by benthic fauna, facilitating bioaccumulation and trophic transfer of contaminants in the Chukchi Sea food web.

POC flux to the benthos probably is lower in the nutrient-depleted Beaufort Sea (Dunton et al., 2003a,b, 2004, 2006). Because of the lower POC flux, benthic biomass in the Beaufort Sea (average 33 g/m^2) is much lower than that in the Chukchi Sea (167 g/m^2) (Dunton et al., 2003a,b, 2004). Sediments in the Beaufort Sea contain organic matter from both marine and terrigenous sources (Yunker et al., 2005). As discussed above, much of the terrigenous organic matter is delivered to the Beaufort Sea in outflows from large Canadian and Alaskan rivers and from coastal erosion and often is enriched in metals and hydrocarbons.

Global warming may dramatically change ecosystem dynamics, including the distribution of marine organisms and chemical contaminants in the Chukchi Sea. Environmental changes may include: 1) increased pelagic primary and secondary production during extended open-water conditions in the summer (based on surplus nutrients, currently underutilized); 2) reduced benthic and pelagic biomass in coastal/shelf areas (due to increased river runoff and resulting changes in salinity and turbidity; and 3) increased pelagic grazing and recycling in open-water conditions at the expense of the current benthic/pelagic coupling in part of the ice-covered shelf regions (due to increased pelagic consumption versus vertical flux) (Bluhm and Gradinger, 2008). These changes may benefit pelagic feeders, such as bowhead whales and some pinnipeds and marine birds, and harm benthic feeders such benthic-feeding pinnipeds and fish. It will also affect the trophic transfer and cycling of chemical contaminants in the Chukchi Sea food web.

Grainger (1965) identified two zooplankton communities in waters less than 100 m deep in the Alaskan Beaufort Sea. The community occupying Arctic surface water is dominated by coelenterates, ctenophores, and copepods. Dominant copepods include *Calanus hyperboreus, Calanus glacialis, Pseudocalanus minutus, Metridia longa,* and *Oithoma similes.* The second zooplankton community is associated in summer with warmer, less saline nearshore waters, primarily inside the barrier islands. Dominant copepods include *Limnocalanus macrouris, Acartia clausi, Eurytemora herdmani,* and *Derjugina tolli.* Calanoid copepods, primarily

Pseudocalanus, are abundant under the sea ice during winter and early spring; they are replaced in early summer by cyclopoid and harpacticoid copepods, hydrozoans, amphipods, larvaceans and larval stages of planktonic and benthic invertebrates (Horner and Murphy, 1985). Griffiths and Thompson (2002) identified 3 zooplankton communities in waters out to 200 m off the eastern Alaskan Beaufort Sea (Camden Bay and eastward to the Canadian border). Copepods are dominant in all 3 communities, accounting for 50 to 75 percent of the total zooplankton biomass. Three species represent more than 85 percent of the total copepod biomass: *Calanus hyperboreus, C. glacialis,* and *Limnocalanus macrourus.*

Euphausiids (*Thysanoessa inermis* and *T. raschii*) are much less abundant than calanoid copepods in the eastern Beaufort Sea. However, they are seasonally abundant in the Chukchi Sea and western Beaufort Sea. Euphausiids are transported by deep-water currents from the Bering Sea, through the Chukchi Sea to waters off Barrow, and then into the western Beaufort Sea (Berline et al., 2008). They are the main prey of bowhead whales that aggregate off Barrow during the fall migration.

The abundance and distribution of zooplankton is extremely variable in different seasons and years. Total zooplankton wet biomass in waters of the eastern Alaskan Beaufort Sea ranges from 170 to more than 383 mg/m^3 in different years; biomass is greater at depths of more than 10 m than at the surface (Griffiths and Thomson (2002). Wet biomass of zooplankton can be as high as 3,500 mg/m^3 in dense patches. The feeding threshold for bowhead whales may be as high as 800 mg zooplankton wet biomass/m^3 of seawater (Griffiths et al., 2002; MMS, 2006), highlighting the importance of dense zooplankton patches to bowhead whales.

These primary consumers are preyed on by a variety of secondary consumers (carnivores), including several species of anadromous and marine fish, marine birds, and marine mammals, including some ice seals (e.g., ringed, ribbon, and bearded seals) and baleen whales (bowhead and gray whales). Other marine fish, birds, and mammals consume primarily fish or other marine mammals (polar bears). Crustaceans, such as copepods, euphausiids, amphipods, isopods, and mysids, are abundant in coastal, estuarine waters where they are preyed upon heavily by several species of fish (Table 4-5). Many of the coastal fish in the Beaufort Sea feed primarily on small fish, particularly arctic cod (Frost and Lowry, 1984).

Arctic cod (*Boreogadus saida*) is considered a key species in the Beaufort Sea marine ecosystem (Lowry and Frost, 1981; Bradstreet et al., 1986). They are a major link between lower trophic level benthic and pelagic primary consumers and upper trophic level apex consumers, such as several species of marine birds, seals, and mammals (Figure 4-2 and 4-3; Tables 4-6, 4-7, and 4-8). They consume primarily gammarid amphipods, copepods, and mysids, and are consumed by many birds and mammals, including black kittiwakes, arctic terns, thick-billed murres, black guillemots, and gloucus gulls.

Table 4-5. Anadromous and marine fish collected for Task 5 of the cANIMIDA program from coastal waters of the Beaufort Sea and their food preferences. Data from Thorsteinson and Wilson (2006) and Craig et al. (1984).

Common Name	Scientific Name	Food Preferences
Marine Fish		
Arctic cod	*Boreogadus saida*	Mysids, amphipods, copepods
Four horn sculpin	*Myxocephalus quadricornis*	Amphipods, isopods, polychaetes
Arctic flounder	*Liopseta glacialis*	Demersal & benthic crustacea, polychaetes
Anadromous Fish		
Arctic char	*Salvelinus alpinus*	Mysids, small fish
Arctic cisco	*Coregonus autumnalis*	Mysids, larval & juvenile fish
Least cisco	*C. sardinella*	Mysids, amphipods
Broad whitefish	*C. nasus*	Chironomids, amphipods
Humpback broad whitefish	*C. pidschian*	Chironomids, amphipods

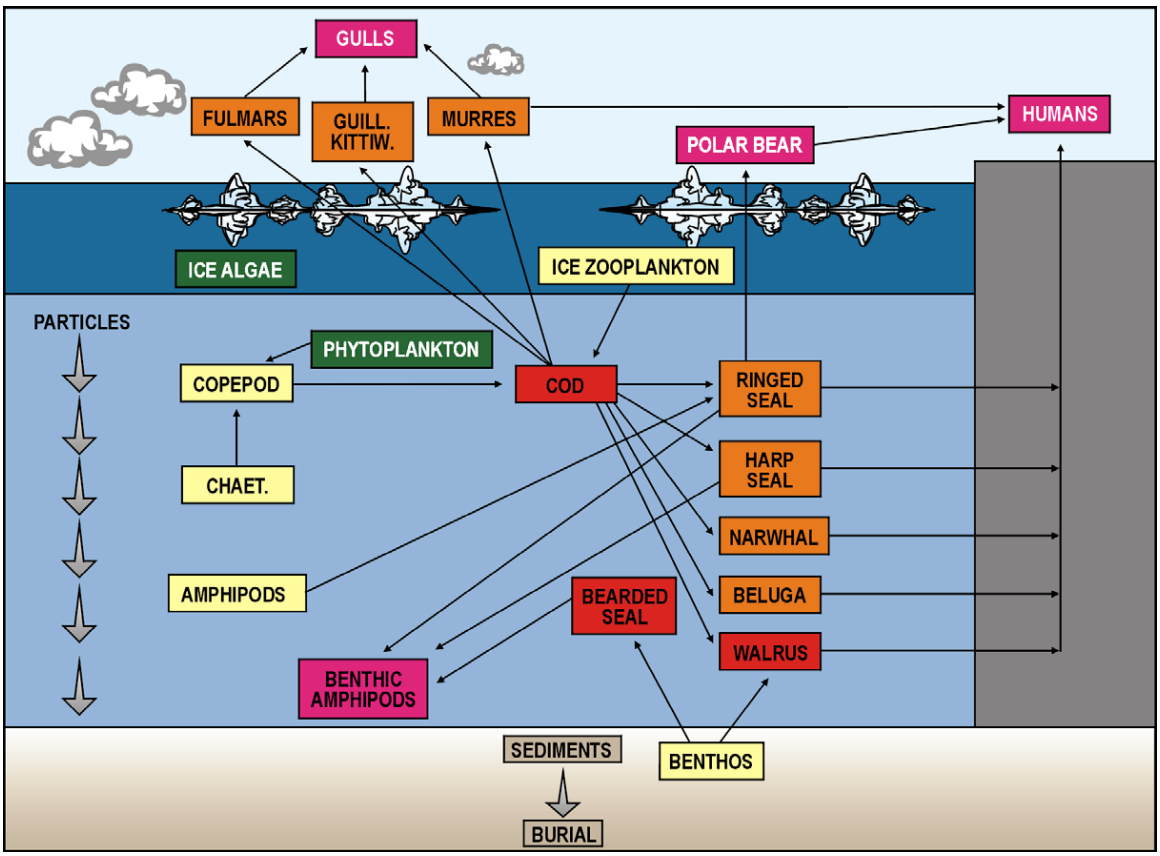


Figure 4-3. The importance of arctic cod (cod) in the Arctic food web. From Macdonald et al. (2005).

Table 4-6. Diet composition (% of stomach contents) in arctic cod, bowhead whales, beluga whales, and ringed seals in the Alaskan Beaufort Sea. From Frost and Lowry (1984) and Lowry and Sheffield (2002).

Prey	Arctic Cod	Bowhead Whales		Beluga Whale	Ringed Seal		
		E. Beaufort	W. Beaufort		Nov-Mar	Apr-Jun	Aug-Sep
Isopod						16	
Hyperiid Amphipod	1				0 – 17		44
Gammarid Amphipod	12					32	
Copepod	50	60	8				
Euphausiid	3	37	92			20	21
Mysid	20	1				19	
Arctic Cod	5			80	75 - 90	6	30
Other Fish		1		10			
Misc.	9	1		10		7	5

Table 4-7. Diet composition in black kittiwakes, red phalaropes, sabine's gulls, and arctic terns feeding in nearshore and offshore waters of the Alaskan Beaufort Sea. From Frost and Lowry (1984).

Prey	Black Kittiwake		Red Phalarope	Sabine's Gull		Arctic Tern	
	Offshore	Nearshore	Offshore	Offshore	Nearshore	Offshore	Nearshore
Isopod							
Hyperiid Amphipod	1	67	1	54	6	1	
Gammarid Amphiopd		14	49		49		31
Copepod			11				
Euphausiid			5	13	4	35	23
Mysid		11	13		24		
Shrimp	1			3			
Mollusks							
Arctic Cod	95	5		13	4	64	20
Other Fish			1				12
Misc.	3	3	20	17	13		14

Bowhead whales and some marine birds and seals also rely on marine crustaceans, particularly copepods and euphausiids, for food. (Tables 4-6 and 4-7). The amphipods, mysids, and isopods that are such important foods for many fish, birds and mammals, consume primarily smaller benthic crustaceans and polychaetes, diatoms, and peat (Craig et al., 1984).

Table 4-8. Diet composition in long-tailed ducks, common and king eiders, thick billed murres, black guillemots, and gloucus gulls feeding in nearshore and offshore waters of the Alaskan Beaufort Sea. From Frost and Lowry (1984).

Prey	Long-tail Duck	Comm. Eider	King Eider	Thick-bill Murre	Black Guillemot	Glaucous Gull	
	Nearshore	Nearshore	Nearshore	Offshore	Offshore	Offshore	Nearshore
Isopod		83	89				12
Hyperiid Amphipod				1		1	1
Gammarid Amphipod	23	1	2				
Copepod							
Euphausiid	17						13
Mysid	20	15					
Shrimp							
Mollusks	22	1	2				
Arctic Cod				99	100	17	60
Other Fish							
Misc.						75	

Bowhead whales feed on pelagic euphausids, copepods, mysids, hyperiid amphipods, and occasionally small fish (Lowry and Frost, 1984; Richardson et al., 1995; Lowry et al., 2004). Bowhead whales feed throughout the Alaskan Beaufort Sea during both the spring (eastward) and fall (westward) migrations between the Bering/Chukchi Seas and the Canadian Beaufort Sea. They feed less during the spring than the fall migration (Lowry et al., 2004). Carroll et al. (1987) reported large numbers of bowheads feeding near Point Barrow during the 1985 spring migration. The dominant prey in the whale stomachs were calanoid copepods and euphausiids. They also feed extensively during their fall, nearshore migration through the Alaskan Beaufort Sea to northeastern Chukchi Sea (Lowry et al., 2004). (Landino et al., 1994) reported a large aggregation of bowhead whales feeding near Point Barrow in late October 1992. Calanoid copepods were the dominant prey in bowhead whales collected off Kaktovik (eastern Beaufort Sea) and euphausiids were the dominant prey in whales collected off Point Barrow (western Beaufort Sea) (Table 4-6).

Lee and Schell (2002) compared carbon isotopes in zooplankton to those in bowhead whale muscle and estimated that 10 to 26% of annual bowhead feeding activity was in the eastern and central Beaufort Sea. Thompson et al. (2002) estimated that they may obtain approximately 2.4% of their annual energy requirements feeding in the eastern Alaskan Beaufort Sea (Flaxman Island to the Canadian border). Although bowheads feed extensively in the Alaskan Beaufort Sea, the times, extent, and locations of feeding vary widely from year to year. The variation is due in large part to variations in the temporal and spatial distribution of dense patches of zooplankton upon which the whales depend, which are controlled by ocean conditions (Asjian et al., 2009).

Beluga whales (*Delphinapterus leucas*) migrate annually through the Alaskan Beaufort Sea, usually further offshore than bowhead whales, between summer feeding areas in the Canadian Beaufort Sea and wintering areas in the Bering Sea and feed heavily on pelagic and demersal fish (Treacy, 2002). The dominant prey of beluga whales, because of its abundance in offshore waters, is the arctic cod (Table 4-6).

Some ice-associated seals, such as the bearded seal (*Erignathus barbatus*) and ribbon seal (*Phoca faciata*), which feed in coastal waters of the Beaufort Sea in some seasons, feed heavily on benthic invertebrates and demersal fish, such as shrimp, crabs, arctic cod, and sculpins (Wynne, 1997). Ringed seals (*Phoca hispida*), the principal food of polar bears, are common in all seasons in the Beaufort Sea. Their diet varies seasonally from primarily isopods, amphipods, euphausiids, and mysids in spring during retreat of the sea ice, to amphipods, euphausiids, and arctic cod during the summer open-water season, to primarily arctic cod during the winter when the seals occupy the sea ice (Table 4-6).

The protected spectacled eider (*Somateria fischeri*) and king eider (*Somateria spectabilis*) which breed along the arctic coastal plain from Barrow to the Canadian border, feed primarily on benthic isopods and mysids, and small numbers of mollusks that they gather in shallow coastal waters (<30 m) (Dau and Kistchinski, 1977; Frost and Lowry, 1984) (Table 4-8). Several species of marine birds in the Alaskan Beaufort Sea, including black kittiwakes, arctic terns, thick-billed murres, black guillemots, and gloucus gulls, feed heavily on arctic cod, which, in turn, feed mainly on mysids, copepods, and amphipods (Frost and Lowry, 1984) (Tables 4-6, 4-7, and 4-8). The copepods that are the principal foods of bowhead whales also are consumed by red phalaropes, Sabine's gulls, arctic terns, long-tailed ducks, and gloucus gulls (Tables 4-7 and 4-8).

Contaminants introduced into the marine environment from offshore oil and gas operations are likely to accumulate in this Beaufort Sea food web. Although none of the contaminants measured in tissues of Beaufort Sea animals in Task 5 are known to biomagnify, they all can be spread through trophic transfer through the food web. The tissue residue data collected to date in Task 5 of the cANIMIDA Program has not identified any metal or PAH contaminants from offshore oil and gas operations that are present at elevated concentrations in selected representatives of the Beaufort Sea food web. Some of the petroleum PAH in the tissues of invertebrates and fish could have been derived from offshore operations; however, concentrations are very low, mostly in the range of natural background, indicating a lack of significant risk to the local ecosystem.

An example of the distribution of total mercury in selected components of the Beaufort Sea food web is shown in Figure 4-4. The mercury data, collected from several sources, including cANIMIDA (this report, Semmler, 2003), show that there is a large increase in total mercury concentrations from the ambient water, to phytoplankton, to zooplankton. The apparent biomagnifications factor (BF: concentration in consumer/concentration in prey) for the transfer from phytoplankton to zooplankton is 2.8. Mercury concentration then decreases from zooplankton to arctic cod (BF = 0.54). Highest concentrations of total mercury are in liver of the three species of marine mammals in Figure 4-4. The apparent BFs for total mercury concentration from whole soft tissues of prey to liver tissue of bowhead whales, beluga whales, and ringed seals are 3.6, 2000, and 586, respectively. Thus, apparent biomagnifications of mercury from zooplankton to bowhead whales is much less than biomagnification from arctic

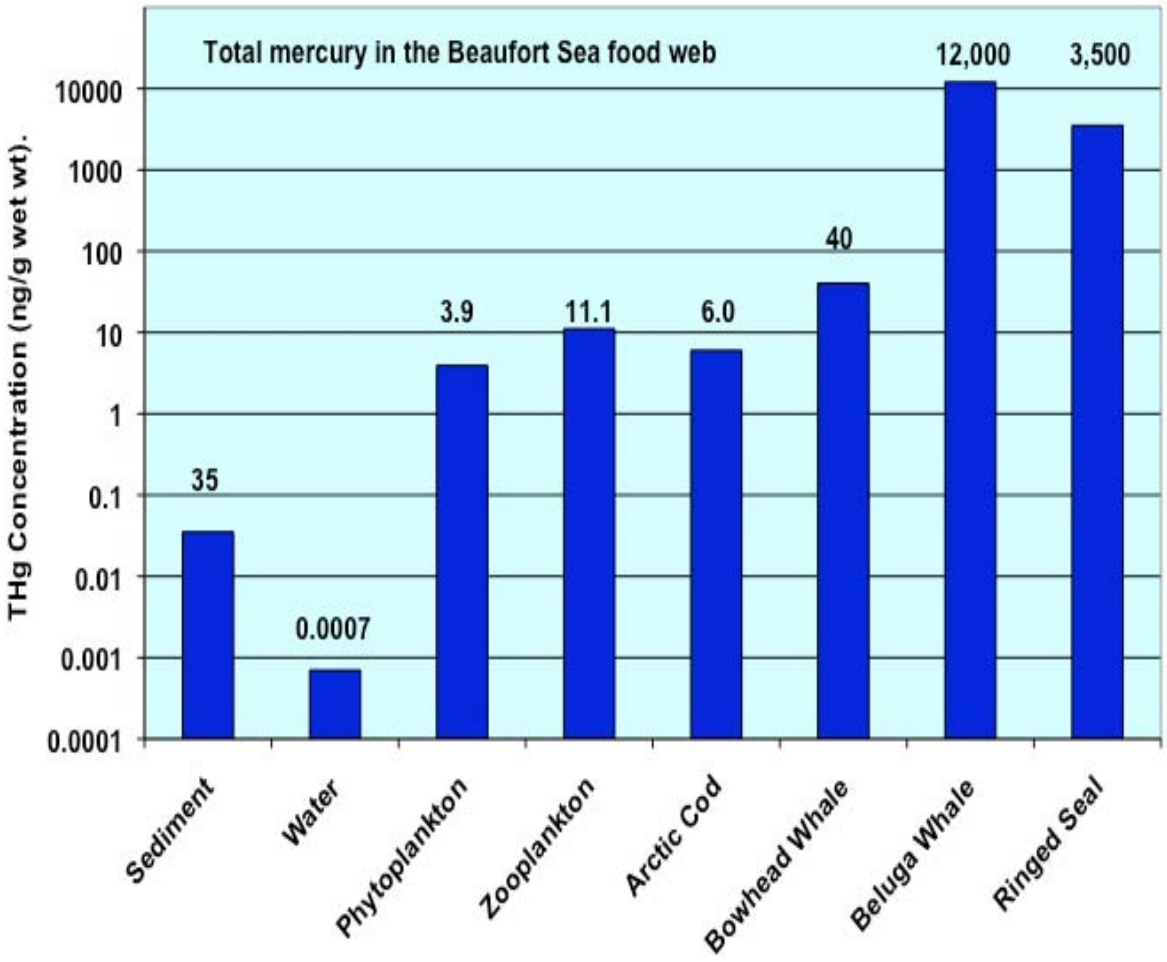


Figure 4-4. Mean concentrations of total mercury in several components of the Beaufort Sea food web. Mean mercury concentrations are for whole phytoplankton, zooplankton, and arctic cod, and for liver of bowhead whales, beluga whales, and ringed seals. Data for sediment, water, phytoplankton, and zooplankton are from Semmler (2003) as reported by Trefry et al. (2009); data for arctic cod are from this report; data for bowhead and beluga whales are from Dehn et al. (2006a); data for ringed seals are from Woshner et al. (2001).

cod to beluga whales or ringed seals. Polar bears (not included in Figure 4-4) from the Beaufort Sea contain a mean of 14,220 ng/g wet wt total mercury (Woshner et al., 2001); the BF for total mercury from prey (ringed seal) liver to polar bear liver is 4.0.

Biomagnification of total mercury in marine food chains usually is attributed to selective accumulation and retention of methylmercury. Mason et al. (1995, 1996) showed that the preferential bioaccumulation of methylmercury in pelagic marine food webs occurs primarily at the level of phytoplankton. Phytoplankton are able to bioaccumulate both inorganic mercury and methylmercury from water. The inorganic mercury binds to cell membranes of the plants in a relatively non-bioavailable form, whereas the methylmercury accumulates in the cytoplasm. Zooplankton that feed on phytoplankton assimilate more methylmercury than inorganic mercury.

Marine animals that feed on the zooplankton also assimilate more methylmercury than inorganic mercury; they also are able to eliminated inorganic mercury more rapidly than methylmercury from their tissues, facilitating the greater accumulation of methylmercury, particularly at higher trophic levels in marine food webs.

Mercury concentrations in marine waters of much of the Arctic are higher than concentrations in temperate and tropical waters due in large part to deposition of metallic and inorganic mercury from long-range transport and deposition from the atmosphere (Skov et al., 2004; Macdonald et al., 2005). There is no evidence that significant amounts of mercury are coming from oil operations around Prudhoe Bay (Snyder-Conn et al., 1997).

Less than 3.5 percent of the total mercury in Beaufort Sea coastal water is methylmercury (Semmler, 2003). Methylmercury in the water column and in tissues of marine organisms is derived from microbial methylation of inorganic mercury in suboxic layers in the water column and sediments (Rolfhus and Fitzgerald, 1995; Gagnon et al., 1996). The methylmercury in Beaufort Sea coastal water probably is from upwelling of offshore water near the edge of the pack ice (Semmler, 2003) and from runoff from melting of permafrost soils containing a large inventory of mercury and methylmercury from aerial deposition and methylation in suboxic layers of frozen soil (Macdonald et al., 2005). Naidu et al. (2001) suggested that methylmercury in Beaufort Sea sediments could be mobilized into the overlying water column during sediment reworking by ice gouging, storm-induced sediment erosion and resuspension, and bioturbation. This is possible.

Much of the mercury in marine sediments is complexed with dissolved and particulate organic matter in the sediments (Neff, 2002b). The remaining inorganic mercury forms strong, stable complexes with iron oxyhydroxides in oxidized surface layers. In oxidized layers of marine sediments, where most biological activity occurs, concentrations of solid iron and manganese oxides are very high (usually several percent) and most of the mercury not complexed with sediment organic matter is adsorbed to the solid oxides. However, if the sediments become anoxic, the iron oxyhydroxides dissolve, releasing adsorbed metals, including mercury. The inorganic mercury released from oxyhydroxides at the redox potential discontinuity (the location in the sediment column where Eh is about 0 mV) in sediments is bioavailable for bioaccumulation and methylation by sulfate reducing bacteria. However, as redox potential decreases further, particularly in marine sediments rich in sulfate, the mercury precipitates as insoluble, nonbioavailable mercuric sulfide. Thus, mercury methylation seems to occur at highest rates and methylmercury concentrations are highest in the narrow band of the redox potential discontinuity in the sediments where the mercury can form slightly soluble mercury-sulfide complexes. Any disturbance that moves the RPD toward the sediment-water interface tends to increase the rate of flux of methylmercury from sediment to the water column. Thus, physical disturbance of the sediment surface or a reduction in oxygen concentration at the sediment-water interface caused, for instance, by a decrease in photosynthesis in the water column during low-light conditions in winter or a seasonal increased flux of organic carbon to surface sediments from the water column (Macdonald et al., 2005), can lead to an increased flux of methylmercury into the overlying bottom water.

The percent of total mercury that is methylmercury increases from 3 percent in phytoplankton, to 7.6 percent in zooplankton, and more than 90 percent in arctic cod (Semmler, 2003), indicating that methylmercury is preferentially biomagnifying in this part of the Beaufort Sea food chain. Methylmercury was not measured in bowhead whale liver; because of the high selenium/total mercury ratio, most of the mercury in bowhead liver probably is present as an insoluble mercury-selenium complex (Dehn et al., 2006a). Three to 10 percent of the total mercury in ringed seal and polar bear liver is methylmercury; the remainder probably is present as mercury-selenium complex (Woshner et al., 2001). Mercury concentrations in liver are much higher than those in other tissues of marine mammals. Thus, much of the methyl mercury accumulated from zooplankton by bowhead whales and from arctic cod by beluga whales and ringed seals is demethylated and stored as a mercury-selenium complex, probably solid mercuric selenide, primarily in the liver (Khan and Wang, 2009). Thus, the apparent biomagnifications of mercury in the trophic step from water-breathing prey to air-breathing predator is the result of assimilation of bioavailable methylmercury and chemical transformation of methylmercury and sequestration as solid mercuric selenide.

This page intentionally left blank

5.0 REFERENCES

Ashjian, C.J., R.G. Campbell, J.C. George, S.E. Moore, S.R. Okkonen, B.F. Sherr, and E.B. Sherr. 2009. Impact of inter-annual variability in ocean conditions on bowhead feeding near Barrow, Alaska. Poster in the Alaska Marine Science Symposium, Anchorage, AK.

Avigan, J. and M. Blumer. 1968. On the origin of pristane in marine organisms. J. Lipid Res. 9:350-352.

Bard, S.M. 1999. Global transport of anthropogenic contaminants and the consequences for the Arctic marine ecosystem. Mar. Pollut. Bull. 38:356-379.

Beliaeff, B., O'Connor, T. P., and Claisse, D. 1998. Comparison of chemical concentrations in mussels and oysters from the United States and France. 1998; Environ. Monitor. Assess. 49:87-95.

Berline, L., Y.H. Spitz, C.J. Ashjian, R.G. Campbell, W. Maslowski, and S.E. Moore. 2008. Euphausiid transport in the western Arctic Ocean. Mar. Ecol. Prog. Ser. 360:163-178.

Bernard, F.R. 1979. Bivalve mollusks of the western Beaufort Sea. Contrib. Sci. Nat. Hist. Mus. Los Angeles County. 313:1-80.

Bluhm, B.A. and R. Gradinger. 2008. Regional variability in food availability for Arctic marine mammals. Ecol. Applic. 18:S77-S96.

Boehm, P.D., L. Le Blanc, J. Trefry, P. Marajh-Whittemore, J. Brown, A. Schutzberg and A. Kick. 1990. Monitoring hydrocarbons and trace metals in Beaufort Sea sediments and organisms. Final report to U.S. Department of the Interior Minerals Management Service Alaska OCS Office, Anchorage, Alaska. 199 pp. + 36 Appendices.

Boehm, P.D., J.M. Neff, and D.S. Page. 2007. Assessment of hydrocarbon exposure in the waters of Prince William Sound after the *Exxon Valdez* oil spill: 1989-2005. Mar. Pollut. Bull. 54:339-367.

Borgå, K., A.T. Fisk, P.F. Hoekstra, and D.C.G. Muir. 2004. Biological and chemical factors of importance in the bioaccumulation and trophic transfer of persistent organochlorine contaminants in Arctic marine food webs. Environ. Toxicol. Chem. 23:2367-2385.

BP Exploration (Alaska), Inc. 2007. Liberty Development Project. Development and Production Plan. Report submitted to U.S Dept. of the Interior, Minerals Management Service, Alaska OCS Region, Anchorage, AK. BP Exploration (Alaska) Inc., Anchorage, AK.

Bradstreet, M.S.W., K.J. Finley, A.D. Sekerak, W.B. Griffiths, C.R. Evans, M.F. Fabijan, et al. 1986. Aspects of the biology of Arctic cod (*Boreogadus saida*) and its importance in Arctic marine food chains. Can Tech Rep Fish Aquat Sci 1986:1-193.

Brown, J., P.D. Boehm, L. Cook, J. Trefry, and W. Smith. 2005. ANIMIDA Task 2. Hydrocarbon and Metal Characterization of Sediments, Bivalves, and Amphipods in the ANIMIDA Study Area. OCS Study MMS 2004-024. Final Report to U.S. Dept. of the Interior, Minerals Management Service, Anchorage, Alaska. Contract No. 143501-99-CT-30998, to 10902. Battelle, Waltham, Ma.

Carroll, G.M., J.C. George, L.F. Lowry, and K.O. Coyle. 1987. Bowhead whale (*Balaena mysticetus*) feeding near Point Barrow, Alaska, during the 1985 spring migration. Arctic 40:105-110.

Cheng, M.D., and W.H. Schroeder. 2000. Potential atmospheric transport pathways for mercury measured in the Canadian high Arctic. J. Atmos. Chem. 35:101-107.

Collier, T.K., B.F. Anulacion, J.E. Stein, and A. Goksøyr. 1995. A field evaluation of cytochrome PF501A as a biomarker of contaminant exposure in three species of flatfish. Environ. Toxicol. Chem. 14:143-152.

Cornelissen, G., A. Pettersen, D. Broman, P. Mayer, and G.D. Breedveld. 2008. Field testing of equilibrium passive samplers to determine freely dissolved native polycyclic aromatic hydrocarbon concentrations. Environ. Toxicol. Chem. 27:499-508.

Craig, P.C., W.B. Griffiths, and S.R. Johnson. 1984. Trophic dynamics in an arctic lagoon. Pages 347-380 In: P.W. Barnes, D.M. Schell, and E. Reimitz, Eds., The Alaskan Beaufort Sea: Ecosystems and Environments. Academic Press, NY.

Crippen, R.W., S.L. Hood, and G. Greene. 1980. Metal levels in sediment and benthos resulting from drilling fluid discharge into the Beaufort Sea. Pages 636-669 In: Symposium on Research on Environmental Fate and Effects of Drilling Fluids and Cuttings. American Petroleum Institute, Washington, DC.

Cusson, M., R. Tremblay, G. Daigle, and M. Roussy. 2005. Modeling the depuration potential of blue mussels (*Mytilus* spp.) in response to thermal shock. Aquacult. 250:183-193.

Daskalakis, K.D. and T.P. O'Connor. 1995. Distribution of chemical concentrations in US coastal and estuarine sediment. Mar. Environ. Res. 40:381-398.

Dau, C.P. and S.A. Kistchinski. 1977. Seasonal movements and distribution of the spectacled eider. Waterfowl 28:65-75.

Dehn, L.-A., E.H. Follmann, C. Rosa, L.K. Duffy, D.L. Thomas, G.R. Bratton, R.J. Taylor, and T.M. O'Hara. 2006a. Stable isotope and trace element status of subsistence-hunted bowhead and beluga whales in Alaska and gray whales in Chukotka. Mar. Pollut. Bull. 52:301-319.

Dehn, L.-A., E.H. Follmann, D.L. Thomas, G.G. Sheffield, C. Rosa, L.K. Duffy, and T.M. O'Hara. 2006b. Trophic relationships in an Arctic food web and implications for trace metal transfer. Sci. Tot. Environ. 362:103-123.

Dehn, L.-A., G.G. Sheffield, E.H. Follmann, L.K. Duffy, D.L. Thomas, G.R. Bratton, R.J. Taylor, and T.M. O'Hara. 2005. Trace elements in tissues of phocid seals harvested in the Alaskan and Canadian Arctic: influence of age and feeding ecology. Can. J. Zool. 83:726-746.

Duncan, D. B. 1975. t-test and intervals for comparisons suggested by the data. Biometrics, 31: 339-359.

Dunton, K., A. Burd, D. Funk, and R. Maffione. 2003a. Linking Water Turbidity and TSS Loading to Kelp Productivity within the Stefansson Sound Boulder Patch. ANIMIDA Task 6 (Boulder Patch). Report to US Dept. of the Interior, Minerals Management Service, Alaska Outer Continental Shelf Office, Anchorage, AK.

Dunton, K., J.M Grebmeier, D.R. Maidment, and S.V. Schonberg. 2003b. SBI I Final Report. Benthic Community Structure and Biomass in the Western Arctic: Linkage to Biological and Physical Properties. Final Report to NSF, Washington, DC.

Dunton, K., A. Burd, D. Funk, and R. Maffione. 2004. Linking Water Turbidity and Total Suspended Solids Loading to Kelp Productivity within the Stefansson Sound Boulder Patch. ANIMIDA Task 6 (Boulder Patch). OCS Report MMS 2005-011 to US Dept. of the Interior, Minerals Management Service, Alaska Outer Continental Shelf Office, Anchorage, AK.

Dunton, K., T. Weingartner, and E. Carmack. 2006. The nearshore western Beaufort Sea ecosystem: circulation and importance of terrestrial carbon in arctic coastal food webs. Prog. Oceanogr. 71:362-378.

Durell, G., T. Røe Utvik, S. Johnsen, T. Frost, and J.M. Neff. 2006. Oil Well Produced Water Discharges to the North Sea. Part I: Comparison of Deployed Mussels (*Mytilus edulis*), Semi Permeable Membrane Devices, and the DREAM Model Predictions to Estimate the Dispersion of Polycyclic Aromatic Hydrocarbons. Mar. Environ. Res. 62:194-223.

Fisk, A.T., K.A. Hobson, and R.J. Norstrom. 2001. Influence of chemical and biological factors on trophic transfer of persistent organic pollutants in the Northwater Polynya marine food web. Environ. Sci. Technol. 35:732-738.

Food and Drug Administration. 2001. Mercury levels in seafood species. U.S. Food and Drug Administration, Center for Food Safety and Applied Nutrition, Office of Seafood, Washington, DC.

Franson, J.C., T.E. Hollmén, P.L. Flint, J.B. Grand, and R.B. Lanctot. 2004. Contaminants in molting long-tailed ducks and nesting common eiders in the Beaufort Sea. Mar. Pollut. Bull. 48:504-513.

Frost, K.J. and L.F. Lowry. 1984. Trophic relationships of vertebrate consumers in the Alaskan Beaufort Sea. Pages 381-401 In: P.W. Barnes, D.M. Schell, and E. Reimitz, Eds., The Alaskan Beaufort Sea: Ecosystems and Environments. Academic Press, NY.

Gagnon, C., É. Pelletier, A. Mucci, and W.F. Fitzgerald. 1996. Diagenetic behavior of methylmercury in organic-rich coastal sediments. Limnol. Oceanogr. 41:428-434.

Grainger, E. 1965. Zooplankton from the Arctic Ocean and adjacent Canadian waters. J. Fish. Res. Bd. Canada. 22:543-564.

Griffiths, W. and D. Thomson. 2002. Species composition, biomass, and local distribution of zooplankton relative to water masses in the eastern Alaskan Beaufort Sea. Chapter 5 In: J. Richardson and D. Thomson, Eds., Bowhead Whale Feeding in the Eastern Alaskan Beaufort Sea: Update of Scientific and Traditional Knowledge. OCS Study MMS 2002-012. LGL Report TA2996-7. US. Dept. of the Interior, Minerals Management Service, Alaska OCS Region, Anchorage, AK 277 pp.

Griffiths, W.B., D.H. Thomson, and M.S.W. Bradstreet. 2002. Zooplankton and water masses at bowhead whale feeding locations in the eastern Beaufort Sea. Pages 1-44 In: W.J. Richardson and D.H. Thomson, Eds., Bowhead Whale Feeding in the Eastern Alaskan Beaufort Sea: Update of Scientific and Traditional Knowledge. LGL Report TA196-7. US. Dept. of the Interior, Minerals Management Service, Alaska OCS Region, Anchorage, AK 277 pp.

Halsall, C.J., L.A. Barrie, P. Fellin, D.C.G. Muir, B.N. Billeck, L. Lockhart, F.Y. Rovinsky, E.Y. Kononov, and B. Pastukhov. 1997. Spatial and temporal variation of polycyclic aromatic hydrocarbons in the arctic atmosphere. Environ. Sci. Technol. 31:3593-3599.

Heidam, N.Z. The components of the Arctic aerosol. 1984. Atmos. Environ. 18: 329-343.

Hoekstra, P.F., L.A. Dehn, J.C. George, K.R. Solomon, D.C.G. Muir, and T.M. O'Hara. 2002. Trophic ecology of bowhead whales (*Balaena mysticetus*) compared with that of other arctic marine biota as interpreted from carbon-, nitrogen-, and sulfur-isotope signatures. Can. J. Zool. 80:223-231.

Hoekstra, P.F., T.M. O'Hara, S.M. Backus, C. Hanns, and D.C.G. Muir. 2005. Concentrations of persistent organochlorine contaminants in bowhead whale tissues and other biota from northern Alaska: Implications for human exposure from a subsistence diet. Environ. Res. 98:329-340.

Hoekstra, P.F., T.M. O'Hara, A.T. Fisk, K. Borgå, K.R. Solomon, and D.C.G. Muir. 2003. Trophic transfer of persistent organochlorine contaminants (OCs) within an Arctic marine food web from the southern Beaufort-Chukchi Seas. Environ. Pollut. 124:509-522.

Hoekstra, P.F., T.M. O'Hara, S.J. Pallant, K.R. Solomon, and D.C.G. Muir. 2002. Bioaccumulation of organochlorine contaminants in bowhead whales (*Balaena mysticetus*) from Barrow, Alaska. Arch. Environ. Contam. Toxicol. 42:497-507.

Hom, T., U. Varanasi, J.E. Stein, C.A. Sloan, K.L. Tilbury, and S.L. Chan. 1996. Assessment of the exposure of subsistence fish to aromatic compounds after the *Exxon Valdez* oil spill. Amer. Fish. Soc. Symp. 18:856-866.

Honda, K., J.E. Marcovecchio, S. Kan, R. Tatsukawa, and H. Ogi. 1990. Metal concentrations in pelagic birds from the North Pacific Ocean. Arch. Environ. Contam. Toxicol. 19:704-711.

Horner, R. And D. Murphy. 1985. Species composition and abundance of zooplankton in the nearshore Beaufort Sea in winter-spring. Arctic 36:201-209.

Huggett, R.J., J.J. Stegeman, D.S. Page, K.R. Parker, B. Woodin, and J.S. Brown. 2003. Biomarkers in fish from Prince William Sound and the Gulf of Alaska; 1999-2000. Environ. Sci. Technol. 37:4043-4051.

Jewett, S.C., T.A. Dean, B.R. Woodin, M.K. Hoberg, and J.J. Stegeman. 2002. Exposure to hydrocarbons 10 years after the *Exxon Valdez* oil spill: evidence from cytochrome P4501A expression and biliary FACs in nearshore demersal fishes. Mar. Environ. Res. 54:21-48.

Khan, M.A.K. and F. Wang. 2009. Mercury-selenium compounds and their toxicological significance: toward a molecular understanding of the mercury-selenium antagonism. Environ. Toxicol. Chem. (in press).

Kopponen, P., J. Tarhanen, J. Ruuskanen, R. Torronen, and S. Karenlampi. 1993. Peat induces cytochrome P451A1 in hepa-1- cell line. Comparison with fly ashes from combustion of peat, coal, heavy fuel oil and hazardous waste. Chemosphere 26:1499-1506.

Krone, C.A., P.A. Robisch, K.L. Tilbury, J.E. Stein, E.A. Mackey, P.R. Becker, T.M. O'Hara, and L.M. Philo. 1999. Elements in liver tissues of bowhead whales (*Balaena mysticetus*). Mar. Mamm. Sci. 15:123-142.

Kucklick, J.R., W.D.J. Struntz, P.R. Becker, G.W. York, T.M. O'Hara, and J.E. Bohonowych. 2002. Persistent organochlorine pollutants in ringed seals and polar bears collected from northern Alaska. Sci. Tot. Environ. 287:45-59.

Landino, S.W., S.D. Treacy, S.A. Zerwick, and J.B. Dunlap. 1994. A large aggregation of bowhead whales (*Balaena mysticetus*) feeding near Point Barrow, Alaska, in late October 1992. Arctic 47:232-235.

Lee, S. and D. Schell. 2002. Regional and seasonal feeding by bowhead whales as indicated by stable isotope ratios. Pages 20-1 to 20-25 In: J. Richardson and D. Thomson, Eds., Bowhead Whale Feeding in the Eastern Alaskan Beaufort Sea: Update of Scientific and Traditional Knowledge. OCS Study MMS 2002-012. LGL Report TA2996-7. US. Dept. of the Interior, Minerals Management Service, Alaska OCS Region, Anchorage, AK 277 pp.

Lowry, L.F. and K.J. Frost. 1981. Distribution, growth, and foods of Arctic cod (*Boreogadus saida*) in the Bering, Chukchi, and Beaufort Seas. Can. Field Natur. 95:186-191.

Lowry, L.F. and K.J. Frost. 1984. Foods and feeding of bowhead whales in western and northern Alaska. Sci. Rep. Whale Res. Instit. 35:1-16.

Lowry, L. and G. Sheffield. 2002. Stomach contents of bowhead whales harvested in the Alaskan Beaufort Sea. Pages 18-1 to 18-20 In: J. Richardson and D. Thomson, Eds., Bowhead Whale Feeding in the Eastern Alaskan Beaufort Sea: Update of Scientific and Traditional Knowledge. OCS Study MMS 2002-012. LGL Report TA2996-7. US. Dept. of the Interior, Minerals Management Service, Alaska OCS Region, Anchorage, AK 277 pp.

Lowry, L.F., G. Sheffield, and J.C. George. 2004. Bowhead whale feeding in the Alaskan Beaufort Sea, based on stomach contents analysis. J. Cetacean Res. Manage. 6:215-223.

MacDonald RW, L.A. Barrie, T.F. Bidleman, M.L. Diamond, D.J. Gregor, R.G. Semkin, et al. 2000. Contaminants in the Canadian Arctic: 5 years of progress in understanding sources, occurrence and pathways. Sci. Tot. Environ 254:93–234.

Macdonald, R.W., T. Harner, and J. Fyfe. 2005. Recent climate change in the arctic and its impact on contaminant pathways and interpretation of temporal trend data. Sci. Tot. Environ. 342:5-86.

Mackey, E.A., P.R. Becker, R. Demiralp, R.R. Greenberg, B.J. Koster, and S.A. Wise. 1996. Bioaccumulation of vanadium and other trace metals in livers of Alaskan cetaceans and pinnipeds. Arch. Environ. Contam. Toxicol. 30:503-512.

Mackey, E.A., R. Demiralp, P.R. Becker, R.R. Greenberg, B.J. Koster, and W.A. Wise. 1995. Trace element concentrations in cetacean liver tissues archived in the National Marine Mammal Tissue Bank. Sci. Tot. Environ. 175:25-41.

Mason, R.P., J.R. Reinfelder, and F.M.M. Morel. 1995. Bioaccumulation of mercury and methylmercury. Water Air Soil Pollut. 80:915-921.

Mason, R.P., J.R. Reinfelder, and F.M.M. Morel. 1996. Uptake, toxicity, and trophic transfer of mercury in a coastal diatom. Environ. Sci. Technol. 30:1835-1845.

Matsumoto, K., I. Nagao, H. Tanaka, H. Miyaji, T. Iida, and Y. Ikebe. 1998. Seasonal characteristics of organic and inorganic species and their size distributions in atmospheric aerosols over the northwest Pacific Ocean. Atmos. Environ. 32:1931-1946.

Meador, J.P., J. Buzitis, and C.F. Bravo. 2008. Using fluorescent aromatic compounds in bile from juvenile salmonids to predict exposure to polycyclic aromatic hydrocarbons. Environ. Toxicol. Chem. 27:845-853.

Miller, R. G. 1981. Simultaneous Statistical Inference. New York, Springer-Verlag.

Minerals Management Service. 1984. Proposed Diapir Field Lease Offering (June 1984) Sale 87, Final Environmental Impact Statement (2 Vols.). OCS EIS/EA MMS 84-0009. U.S. Dept. of the Interior, Minerals Management Service, Alaska OCS Region, Anchorage, AK.

Minerals Management Service. 1987. Alaska Outer Continental Shelf Beaufort Sea Sale 97 Final Environmental Impact Statement. 2 Vols. OCS EIS/EA, MMS 87-0069. U.S. Dept. of the Interior, Minerals Management Service, Alaska OCS Region, Anchorage, AK.

Minerals Management Service. 1990. Beaufort Sea Planning Area Oil and Gas Lease Sale 124. Draft Environmental Impact Statement. Volume 1. OCS EIS/EA MMS 90-0006. U.S. Dept. of the Interior, Minerals Management Service. Alaska OCS Region, Anchorage, AK.

Minerals Management Service. 2006. Biological Evaluation of the Potential Effects of Oil and Gas Leasing and Exploration in the Alaska OCS Beaufort Sea and Chukchi Sea Planning Areas on Endangered Bowhead Whales (*Balaena mysticitus*), Fin Whales (*Balaenoptera physalus*), and Humpback Whales (*Megaptera movaeangliae*). Biological Report prepared in accordance with Section 7 of the Endangered Species Act of the United States of 1973, as amended. U.S. Dept. of the Interior, Minerals Management Service, Alaska OCS Region, Anchorage, AK.

Minerals Management Service. 2007. Liberty Development and Production Plan. Ultra Extended Reach Drilling From Endicott – Satellite Drilling Island (SDI). Environmental Assessment. OCS EIS/EA MMS 2007-054. U.S. Dept. of the Interior, Minerals Management Service, Alaska OCS Region, Anchorage, AK.

Moran, S.B., R.P. Kelly, K. Hagstrom, J.N. Smith, J.M. Grebmeier, L.W. Cooper, G.F. Costa, J.J. Walsh, N.R. Bates, D.A. Hansell, W. Maslowski, R.P. Nelson, and S. Mulsow. 2005. Seasonal changes in POC export flux in the Chukchi Sea and implications for water column-benthic coupling in Arctic shelves. Deep-Sea Res II 52:3427-3451.

Muir, D., B. Braune, B. DeMarch, R. Norstrom, R. Wagemann, L. Lockhart, B. Hargrave, D. Bright, R. Addison, J. Payne, and K. Reimer. 1999. Spatial and temporal trends and effects of contaminants in the Canadian Arctic marine ecosystem: a review. Sci. Tot. Environ. 230:83-144.

Myers, M.S., B.F. Anulacion, B.L. French, W.L. Reichert, C.A. Laetz, J. Buzitis O.P. Olson, S. Sol, and T.K. Collier. 2008. Improved flatfish health following remediation of a PAH-contaminated site in Eagle Harbor, Washington. Aquat. Toxicol. 88:277-288.

Naidu, A.S., J.J. Goering, J.J. Kelly, and M.I. Venkatesan. 2001. Historical Changes in Trace Metals and Hydrocarbons in the Inner Shelf Sediments, Beaufort Sea: Prior and Subsequent to Petroleum-Related Industrial Developments. OCS Study MMS 2001-061. Institute of Marine Science, University of Alaska, Fairbanks, AK. 80 pp.

National Research Council. 2003. Cumulative Environmental Effects of Oil and Gas Activities on Alaska's North Slope. National Academy Press, Washington, DC. 304 pp.

Neff, J.M. 1997. Ecotoxicology of arsenic in the marine environment: a review. Environ. Toxicol.Chem. 16:917-927.

Neff, J.M. 2002a. Bioaccumulation in Marine Organisms. Effect of Contaminants from Oil Well Produced Water. Elsevier Science Publishers, Amsterdam. 452 pp.

Neff, J.M. 2002b. Fates and Effects of Mercury from Oil and Gas Exploration and Production Operations in the Marine Environment. Report to the American Petroleum Institute, Washington, DC. 164 pp.

Neff, J.M. 2005. Composition, Environmental Fates, and Biological Effect of Water Based Drilling Muds and Cuttings Discharged to the Marine Environment: A Synthesis and Annotated Bibliography. Report prepared for the Petroleum Environmental Research Forum (PERF). Available from American Petroleum Institute, Washington, DC. 73 pp.

Neff, J.M. 2007. Estimation of the bioavailability of metals from drilling mud barite. Integr. Environ. Assess. Manage. 4(2):184-193.

Neff, J.M., A.E. Bence, K.R. Parker, D.S. Page, J.S. Brown, and P.D. Boehm. 2006. Bioavailability of PAH from buried shoreline oil residues 13 years after the *Exxon Valdez* oil spill: a multispecies assessment. Environ. Toxicol. Chem. 25(4):947-961.

Northern Technical Services. 1982. Above-ice drilling effluent disposal tests. Sag Delta No. 7, Sag Delta No. 8, and Challenge Island No. 1 wells, Beaufort Sea, Alaska. Report prepared for SOHIO Alaska Petroleum Company, Anchorage, AK. 185 pp.

Norton, D. and G. Weller. 1984. The Beaufort Sea: Background, History, and Perspective. Pages 3-19 In: P.W. Barnes, D.M. Schell, and E. Reimitz, Eds., The Alaskan Beaufort Sea: Ecosystems and Environments. Academic Press, NY.

O'Connor, T.P. 2002. National distribution of chemical concentrations in mussels and oysters in the USA. Mar. Environ. Res. 53:117-143.

O'Connor, T.P. and G.G. Lauenstein. 2006. Trends in chemical concentrations in mussels and oysters collected along the US coast: update to 2003. Mar. Environ. Res. 62:261-285.

O'Hara, T.M., C. Hanns, G. Bratton, R. Taylor, and V.M. Woshner. 2006. Essential and non-essential elements in eight tissue types from subsistence-hunted bowhead whale: nutritional and toxicological assessment. Internat. J. Circumpolar Hlth. 65:228-242.

O'Hara, T.M., M.M. Krahn, D. Boyd, P.R. Becker, and L.M. Philo. 1999. Organochlorine contaminant levels in Eskimo harvested bowhead whales of Arctic Alaska. J. Wildlife Dis. 35:741-752.

Oliver, J.S. and P.N. Slattery. 1985. Destruction and opportunity on the sea floor: effects of gray whale feeding. Ecol. 66:1965-1975.

Percy, J.A. 1983. Distribution of Arctic marine isopods of the *Mesodotea* (= *Saduria*) complex in relation to depth, temperature, and salinity in the southern Beaufort Sea. Arctic 36:341-349.

Perkins, R.A., S. Rhoton, and C. Behr-Andres. 2003. Toxicity of dispersed and undispersed, fresh and weathered oil to larvae of a cold-water species, Tanner crab (*C. bairdi*), and standard warm-water test species. Cold Reg. Sci. Technol. 36:129-140.

Prahl, F. G., and R. Carpenter. 1984. Hydrocarbons in Washington coastal sediments. Estuar. Coast. Shelf Sci. 18:703-720.

Rember, R.D. and J.H. Trefry. 2004. Increased concentrations of dissolved trace metals and organic carbon during snowmelt in rivers of the Alaskan Arctic. Geochim. Cosmochim. Acta 68:477-489.

Richardson, W.J., K.J. Finley, G.W. Miller, R.A. Davis, and W.R. Koski. 1995. Feeding, social and migration behavior of bowhead whales, *Balaena mysticetus*, in Baffin Bay vs the Beaufort Sea - Regions with different amounts of human activity. Marine Mammal Science 11(1):1-45.

Riget, F., D. Muir, M. Kwan, T. Savinova, M. Nyman, V. Woshner, and T. O'Hara. 2005. Circumpolar pattern of mercury and cadmium in ringed seals. Sci. Tot. Environ. 351-352:312-322.

Rolfhus, K.R. and W.F Fitzgerald. 1995. Linkages between atmospheric mercury deposition and the methylmercury content of marine fish. Wat. Air Soil Pollut. 80:291-297.

Salazar, M.H., and S.M. Salazar. 1995. In-Situ bioassays using transplanted mussels: I. Estimating chemical exposure and bioeffects with bioaccumulation and growth. Pages 216-241 In: J. S. Hughes, G. R. Biddinger and E. Mones, Eds., Environmental Toxicology and Risk Assessment, ASTM STP 1218. American Society for Testing and Materials, Philadelphia.

Schell, D.J., P.J. Ziemann, D.M. Parrish, and E.J. Brown. 1982. OCSEAP Cumulative Summary Report, Research Unit #537. NOAA, OCSEAP, Anchorage, AK. From the University of Alaska, Fairbanks.

Schell, D.M. 1983. Carbon-13 and C-14 abundances in Alaskan aquatic organisms: delayed production from peat in Arctic food webs. Science 219:1068-1071.

Searle, S.R. 1971. Linear Models. John Wiley & Sons, Inc. New York.

Semmler, C.M. 2006. Sources, Cycling, and Fate of Arsenic and Mercury in the Coastal Beaufort Sea, Alaska. Master of Science Thesis, Florida Institute of Technology, Melbourne, FL.

Shapiro, S.S. and M.B. Wilk. 1965. An analysis of variance test for normality (complete samples). Biometrika 52: 591-611.

Short, J.W. 2005. Seasonal Variability of Pristane in Mussels (*Mytilus trossulus*) in Prince William Sound, Alaska. Ph.D. Thesis, University of Alaska, Fairbanks, AK.

Skov, H., J. Christensen, G. Asmund, S. Rusgaard, T.G. Nielsen, R. Dietz, and F. Riget. 2004. Fate of mercury in the Arctic (FOMA). National Environmental Research Institute, Denmark. NERI Technical Report No. 511. 56 pp. http\\technicalreport.dmu.dk.

Smolowitz, R.M., M.E. Hahn, J.J. Stegeman. 1991. Immunochemical localization of cytochrome P450IA1 induced by 3,3',4,4-tetrachlorobiphenyl and by 2,3,7,8-tetrachlorodibenzofuran in liver and extrahepatic tissues of *Stenotomus chrysops* (scup). Drug. Metab. Dispos. 19:113-123.

Snedecor, G.W., and W. G. Cochran. 1978. Statistical Methods. 6th ed. Iowa State Univ. Press, Ames, IA.

Snyder-Conn, E., J.R. Garbarino, G.L. Hoffman, and A. Oelkers. 1997. Soluble trace elements and total mercury in Arctic Alaskan snow. Arctic 50:201-215.

Spies, R., D. Hardin, J. Gold, and D. Bell. 2003. Baseline characterization of anthropogenic contaminants in biota associated with the Alaska OCS Liberty Prospect and Northstar Oil Production Unit in the Nearshore Beaufort Sea. Task 8 of the Arctic Nearshore Impact Monitoring in the Development Area Project (ANIMIDA). OCS Study MMS 2003-071. Final Report to the U.S. Department of the Interior, Minerals Management Service, Anchorage, AK. Contract No. 01-99-CT-30998, To 10908. Applied Marine Sciences, Inc., Livermore, CA.

Stegeman, J.J., J.J. Schlezinger, J.E. Craddock, and D.E. Tillett. 2001. Cytochrome P450 1A in midwater fishes: potential effects of chemical contaminants in remote oceanic zones. Environ. Sci. Technol. 35:54-62.

Steinhauer, M.S. and P.D. Boehm. 1992. The composition and distribution of saturated and aromatic hydrocarbons in nearshore sediments, river sediments, and coastal peat of the Alaskan Beaufort Sea: implications for detecting anthropogenic inputs. Mar. Environ. Res. 33:223-253.

Thompson, R.J. 1984. The reproductive cycle and physiological ecology of the mussel *Mytilus edulis* in a subarctic, non-estuarine environment. Mar. Biol. 79:277-288.

Thomson, D.H., W.R. Koski, and W.J. Richardson. 2002. Integration and conclusions. Pages 1-35 In: W.J. Richardson and D.H. Thomson, Eds., Bowhead Whale Feeding in the Eastern Alaskan Beaufort Sea. Update of Scientific and Traditional Knowledge. LGL Report TA2196-7. US. Dept. of the Interior, Minerals Management Service, Alaska OCS Region, Anchorage, AK 277 pp.

Thorsteinson, L.K. and W.J. Wilson. 2006. Anadromous fish of the Central Alaska Beaufort Sea. In: Anadromous Fish of the Central Alaska Beaufort Sea. U.S. Geological Survey, National Biological Service, Anchorage, AK. http://biology.usgs/s+t/no frame/s059.htm.

Treacy, S.D. 2002. Aerial Surveys of Endangered Whales in the Beaufort Sea, Fall 2000. OCS Study MMS 2002-014. U.S. Dept. of the Interior, Minerals Management Service, Alaska OCS Region, Anchorage, AK. 106 pp.

Trefry, J.H., R.D. Rember, R.P. Trocine, and J.S. Brown. 2003. Trace metals in sediments near offshore oil exploration and production sites in the Alaskan Arctic. Environ. Geol. 45:149-160.

Trefry, J.H., R.D. Rember, R.P. Trocine, and J.S. Brown. 2004. ANIMIDA Task 7. Partitioning of Potential Anthropogenic Chemicals between Dissolved and Particulate Phases in Arctic Rivers and the Coastal Beaufort Sea. OCS Study MMS 2004-031. Final report prepared by Battelle for the U.S. Dept. of the Interior, Minerals Management Sercice, Alaska OCS Region, Anchorage, AK.

Trefry, J.H., R.P. Trocine, M.B. Alkire, C.M. Semmler, M. Savoie, and R.D. Rember. 2009. Sources, Concentrations, Composition, Partitioning, and Dispersion Pathways for Suspended Sediment, Water, and Potential Metal Contaminants in the cANIMIDA Study Area, Coastal Beaufort Sea. Contract Nos. MP04PC00035 and MP04PC00036. Final Report submitted to U.S. Dept. of the Interior, Minerals Management Service, Alaska OCS Region, Anchorage, AK.

U.S. Environmental Protection Agency. 1991. Methods for the Determination of Metals in Environmental Samples. Office of Research & Development, U.S. EPA, Cincinnati, Ohio.

Van der Oost, R., J. Beyer, and N.P.E. Vermeulen. 2003. Fish bioaccumulation and biomarkers in environmental risk assessment: a review. Environ. Toxicol. Pharmacol. 13:57-149.

Venkatesan, M.I. 1988. Occurrence and possible sources of perylene in marine sediments - A review. Mar. Chem. 25:1-27.

Walsh, J.J., D.W. Dieterle, W. Maslowski, J.M. Gremeier, T.E. Whitledge, M. Flint, I.N. Sukhanova, N. Bates, G.F. Cota, D. Stockwell, S.B. Moran, D. A Hansell, and C.P. McRoy. 2005. A numerical model of seasonal primary production within the Chukchi/Beaufort Seas. Deep-Sea Res. II. 52:3541-3576.

Wang, Z., S.A. Stout, and M. Fingas. 2006. Forensic fingerprinting of biomarkers for oil spill characterization and source identification. Environ. Forens. 7:105-146.

Wasserman, P., E. Bauerfeind, M. Fourtier, M. Fukuchi, B. Hargrave, B. Moran, T. Naji, E.M. Nöthig, K. Olli, R. Reinert, H. Sasaki, and V.P. Shevchenko. 2003. Particulate organic carbon flux to the Arctic Ocean floor. In: R. Stein and R. Macdonald, Eds., The Organic Carbon Cycle of the Arctic Ocean. Springer, New York. 382 pp.

Weingartner, T. (2006) Circulation, thermohaline structure, and cross-shelf transport in the Alaskan Beaufort Sea. Final Report to the Minerals Management Service, OCS Study MMS 2006-031. 58 pp.

Werner, I., A.Holger, and K. Rainer. 2004. Occurrence of *Anonyx sari* (Amphipoda: Lysianassoidea) below Arctic pack ice: an example of cryo-benthic coupling? Polar Biol. 27:474-481.

Woodin, B.R., R.M. Smolowitz, and J.J. Stegeman. 1997. Induction of cytochrome P4501A in the intertidal fish *Anoplarchus purpurescens* by Prudhoe Bay crude oil and environmental induction in fish from Prince William Sound. Environ. Sci. Technol. 31:1198-1205.

Woshner, V.M., T.M. O'Hara, G.R. Bratton, R.S. Suydam, and V.R. Beasley. 2001. Concentrations and interactions of selected essential and non-essential elements in bowhead and beluga whales of Arctic Alaska. J. Wildlife Dis. 37:693-710.

Woshner, V.M., T.M. O'Hara, J.A. Eurell, M.A. Wallig, G.R. Bratton, R.S. Suydam, and V.R. Beasley. 2002. Distribution of inorganic mercury in liver and kidney of beluga and bowhead whales through automettalographic development of light microscsopic tissue sections. Toxicol. Pathol. 30:209-215.

Wynne, K. 1997. Guide to Marine Mammals of Alaska. Alaska Sea Grant College Program, University of Alaska, Fairbanks, AK. 75 pp.

Yunker, M.B., L.L. Belicka, H.R. Harvey, and R.W. Macdonald. 2005. Tracing the inputs and fate of marine and terrigenous organic matter in Arctic Ocean sediments: a multivariate analysis of lipid biomarkers. Deep-Sea Res. II. 52:3478-3508.

Zeisler, R., R. Demiralp, B.J. Koster, P.R. Becker, M. Burrow, P. Ostapuzuk, and S.A. Wise. 1993. Determination of inorganic constituents in marine mammal tissues. Sci. Tot. Environ. 139/140:365-386.