# University of Alaska Coastal Marine Institute

# **Annual Report No. 2**

Fiscal Year 1995 OCS Study MMS 95-0057

## **OCS Study MMS 95-0057**

## **University of Alaska Coastal Marine Institute**

In Cooperation: Minerals Management Service University of Alaska Fairbanks State of Alaska

## **Annual Report**

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## Contents

Introduction
Microbial Degradation of Aromatic Hydrocarbons in Marine Sediments Joan F. Braddock
Intertidal and Subtidal Effects of Pollution: Assessment of Top-trophic Level Predators as Bioindicators Lawrence K. Duffy, R. Terry Bowyer, James B. Faro, and Daniel D. Roby
A Study of the Adsorption and Biodegradation of Aromatic Hydrocarbons by Marine Sediments Susan M. Henrichs and Michelle Luoma
Kachemak Bay Experimental and Monitoring Studies Raymond C. Highsmith, Tama L. Rucker, and Susan M. Saupe
North Slope Amphidromy Assessment Thomas C. Kline and John J. Goering
Recruitment of Juvenile Flatfishes in Alaska: Habitat Preference at Kodiak Island Brenda L. Norcross
Juvenile Flatfish Habitat in Kachemak Bay: A Pilot Study Brenda L. Norcross
Testing Conceptual Models of Marine Mammal Trophic Dynamics Using Carbon and Nitrogen Stable Isotope Ratios Donald M. Schell
Circulation on the North Central Chukchi Sea Shelf Thomas J. Weingartner
Winter Circulation Processes in the Northeast Chukchi Sea Thomas J. Weingartner
New Projects
Funding Summary

## Introduction

The University of Alaska Coastal Marine Institute (CMI) produced its first Annual Report in 1994 to introduce the new organization. Since most of the research supported in fiscal year 1994 began in the spring of that year, and since no scientific results were available, the first Annual Report outlined the proposed work with a series of abstracts, one for each project being funded. In this 1995 Annual Report the CMI is pleased to present the preliminary scientific findings of the ten Alaskan CMI-funded projects as well as abstracts for two additional projects that received funding in 1995. This report demonstrates that the University of Alaska CMI has been active in addressing many of the marine science related research needs of the State of Alaska and the Minerals Management Service (MMS).

### Background

MMS administers the Outer Continental Shelf (OCS) natural gas, oil, and marine minerals program, which oversees the safe and environmentally sound leasing, exploration, and production of these resources within our nation's offshore areas. The Environmental Studies Program (ESP) was formally directed in 1978, under Section 20 of the OCS Lands Act Amendments, to provide information for decisions involving the planning, leasing, and management of exploration, development, and production activities. The research agenda is driven by the identification of specific issues, concerns, or data gaps by federal decision makers and state and local governments who participate in the process. ESP research focuses on the following broad issues associated with the development of OCS gas, oil, and minerals:

- What are the fates and effects of potential OCS-related pollutants (e.g., oil, noise, drilling muds and cuttings, products of fuel combustion) in the marine and coastal environment and the atmosphere?
- What biological resources (such as fish populations) exist, and what resources are at risk? What is the nature and extent of the risk? What measures must be taken to allow extraction to take place?
- How do the OCS activities affect people in terms of jobs and the economy? What are the direct and indirect effects on local culture? What are the psychological effects of the proposed OCS activities?

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Because MMS and individual states have distinct but complementary roles in the process that leads to decisions, scientific information is needed by MMS, the states, and localities potentially affected by CMI operations. In light of this, MMS has developed a locally managed CMI program. Under this program, MMS will take advantage of highly qualified, scientific expertise at local levels in order to:

- Collect and disseminate environmental information needed for OCS oil and gas and marine minerals decisions;
- Address local and regional OCS-related environmental and resource issues of mutual interest; and
- Strengthen the partnership between MMS and the states by addressing OCS oil and gas and marine minerals information needs.

MMS and the State of Alaska have joined in a five-year (1993–97) renewable Cooperative Agreement to establish a CMI to be administered by the University of Alaska Fairbanks School of Fisheries and Ocean Sciences, to address some of these concerns and share the cost of research of mutual interest. Alaska was selected as the location for this CMI because it contains some of the major potential offshore oil and gas producing areas in the United States. The University of Alaska Fairbanks is uniquely suited to participate in this venture by virtue of its flagship status within the state and its nationally recognized scope and depth of marine and coastal expertise relevant to the broad range of OCS program information needs. In addition, MMS and the University of Alaska have worked cooperatively on ESP studies for many years. Research projects funded by the CMI are required to have at least one active University of Alaska investigator. However, cooperative research between the University of Alaska scientists and various state agencies are encouraged.

A series of "Framework Issues" was developed during the formation of the CMI. This list of issues serves to identify and bracket the concerns to be addressed by the CMI. The Framework Issues have been identified as:

- 1) Studies to improve understanding of the affected marine, coastal, or human environment;
- 2) Modeling studies of environmental, social, and economic processes to improve predictive capabilities and to define information needs;
- 3) Experimental studies to improve understanding of environmental processes or the causes and effects of OCS activities;
- 4) Projects which design or establish mechanisms or protocols for the sharing of data or information regarding marine or coastal resources or human activities to support prudent management of oil and gas and marine mineral resources;
- 5) Synthesis studies of background information; and
- 6) Descriptive studies of offshore mining technologies.

Projects funded through the CMI will concentrate on providing information which can be used by the MMS and the state for management decisions specifically relevant to MMS mission responsibilities. That is, projects will be pertinent to either the OCS oil and gas program or the marine minerals mining program and should provide useful information for one of the phases of program management, or for the scientific understanding of the potential environmental effects of these resource development activities in arctic and subarctic environments.

Additional guidelines given to prospective researchers presently identify the Cook Inlet and Shelikof Strait areas as well as the Beaufort and Chukchi Seas as areas of primary concern to MMS and the state.

The proposal process is initiated each year with a request for letters of intent to address one or more of the Framework Issues stated above. This request is sent to researchers at the University of Alaska and to various state agencies. A CMI technical committee then decides which of the proposed letters of intent should be developed into proposals to be evaluated for possible funding.

Successful investigators are strongly encouraged to publish their results in peer-reviewed journals and to present them at national meetings. In addition, some investigators will also present information directly to the public in seminars.

Alaskans will benefit from the examination and increased understanding of the processes unique to Alaskan OCS and coastal waters because this enhanced understanding can be applied to other problems besides oil, gas, and mineral extraction. Spin-offs will likely benefit such activities as subsistence fisheries and northern shipping.

### **Project Reports**

Many of the CMI-funded projects address some combination of issues related to fisheries, biomonitoring, physical oceanography and the fates of oil. In the following section, the CMI project reports have been placed in alphabetic order by author. For the convenience of the reader, an abstract is provided at the beginning of each contribution. The conclusions presented here should be considered preliminary since most projects are ongoing.

## **Microbial Degradation of Aromatic Hydrocarbons in Marine Sediments**

#### Joan F. Braddock

Institute of Arctic Biology University of Alaska Fairbanks Fairbanks, AK 99775-7000

## Abstract

It has been estimated that 2-9 million tons of oil pollutants enter the marine system yearly from various sources worldwide. A number of physical-chemical processes affect the fate of these petroleum inputs, including adsorption of oil to particulate material. Microbial degradation of petroleum hydrocarbons is a major mechanism for the removal of oil from the environment, yet there are conflicting data regarding how adsorption affects both the rate and extent of biodegradation of hydrocarbon fractions. Polycyclic aromatic hydrocarbons are of significant concern in the environment due to their relative toxicity. Therefore, the initial focus has been on the aromatic hydrocarbon phenanthrene. It was found that populations of phenanthrene degraders are present in sediments collected in Cook Inlet. Their presence is likely related to naturally-occurring carbon substrates chemically similar to phenanthrene. It was also found that these populations could actively mineralize phenanthrene in sediment slurry microcosms. Phenanthrene adsorbs to these sediments and is not readily extracted by organic solvents. Radiolabeled phenanthrene is now being used to begin to determine the biodegradability of adsorbed hydrocarbon fractions. Two approaches are being used to study the interactions between microorganisms and hydrocarbons in sediment slurries. The first is a kinetic approach and the second is a bioassay. An understanding of the interactions between sorbed hydrocarbons and microbial communities is critical to assessing the fates and effects of both chronic and acute oil inputs to the marine environment.

### Introduction

Crude oil or refined petroleum products released at sea are subject to a number of physical-chemical and biological processes. These include "... evaporation, dissolution of specific components, dispersion, ingestion of oil droplets by pelagic organisms, oil adsorption onto suspended particulate material, photochemical oxidation of surface films, microbial degradation and removal by advective processes" (Payne et al., 1987). Some of these processes are well understood and can be incorporated into contaminant fate models. Other processes, such as the rates and extent of biodegradation, have been less well quantified. Nevertheless, the microbial degradation of petroleum hydrocarbons is known to be a major mechanism for removal of these compounds from the environment (Leahy and

Colwell, 1990). A number of studies have examined the potentials for microbial degradation of hydrocarbons in sediments either contaminated by acute inputs, such as from major oil spills (Colwell et al., 1978; Ward et al., 1980; Karl, 1992), or by additions of relatively high concentrations of hydrocarbons to sediments in laboratory studies (Bauer and Capone, 1985; 1988) or to outdoor wave tanks (Payne et al., 1984). Hundreds of marine sediment samples collected following the *Exxon Valdez* oil spill were assayed for microbial populations and activity (Braddock et al., 1993; Braddock et al., 1995). Much of the research to date has focused on microbial degradation and mineralization of relatively high concentrations of hydrocarbons. Chronic inputs may be of the greatest concern to the environment. However, the effects of these low-level additions are harder to quantify (Boesch et al., 1987).

Inputs of aromatic hydrocarbons have been of particular concern because many of these compounds are toxic, and some are potent mutagens and carcinogens. Generally, aromatic hydrocarbons are characterized as being relatively insoluble in water, and many of these compounds are also strongly adsorbed by soils and sediments (Means et al., 1980). In addition, polycyclic aromatic hydrocarbons (PAH) can also bind to microorganisms. By this mechanism, mobile microorganisms may enhance transport of PAHs in some systems (Jenkins and Lions, 1993). Microbial transformations in aquatic systems appear to be very important in influencing the transport of two- and three-ring PAHs (Herbes and Schall, 1978). For these reasons, this study focuses on the bioavailability of the PAH, phenanthrene, in marine sediment.

A number of studies have reported a positive correlation between the numbers of hydrocarbon-degrading microorganisms and oil pollution patterns in marine systems polluted by hydrocarbons (Colwell et al., 1978; Roubal and Atlas, 1978; Ward et al., 1980; Braddock et al., 1993; Braddock et al., 1995). Less often, hydrocarbon degraders may be present as a result of exposure to other chemically similar compounds in the environment. Seawater samples from Resurrection Bay, Alaska, were found to have high levels of hydrocarbon oxidization activity, which is, at least partly, a result of the presence of naturally occurring terpenes in those waters (Button, 1984; Button and Juttner, 1989). The composition of the microbial population has implications for the likely effects of acute or chronic oil inputs in ecosystems. For example, Heitkamp and Cerniglia (1987) found that PAH residues persisted two to four times longer in pristine ecosystems than in oil polluted ecosystems.

Another study which is relevant to the work described here was conducted in the early 1980s by Payne et al. (1984) on sediments collected from Kasitsna Bay, Alaska. The main objective of these studies was to investigate the rates and extent of physical and chemical changes which occur after a major petroleum spill in a subarctic marine system. While not the major focus of their study, Payne et al. did conduct outdoor experiments in flow-through wave tanks containing sea water at

5 -----

Kasitsna Bay to assess the impact of biodegradation on oil transformations for the oil weathering model they developed. High concentrations of oil were used to mimic the short-term effects of a large oil spill. The Payne et al. study did not specifically address the effects of adsorption on degradation of hydrocarbons, but it did look at overall short-term biodegradation. They found that the degradation was greatest in the summer (due to increased temperature) and occurred primarily on oiled surfaces. Payne et al. concluded that dissolution removed more oil than biodegradation, a result they attributed to the short residence time in the wave tanks. However, they conceded that microbial degradation is likely to be a more important component for long-term oil transformations in marine systems.

It is generally thought that sorption of hydrocarbons decreases the availability of PAH for microbial degradation (Manilal and Alexander, 1991; Weissenfels et al., 1992), but a few studies have provided evidence that sorbed compounds may be utilizable by some microorganisms (Fu and Alexander, 1992; Guerin and Boyd, 1992; Knaebel et al., 1994). Guerin and Boyd found large differences between two naphthalene-degrading bacteria in utilization of sorbed naphthalene. Sorption limited the rate and extent of biodegradation for the first microorganism, but enhanced total degradation for the second. Adsorption effects on bioavailability are not yet well understood. The effects depend upon the chemical nature of the sorbant, the mechanisms of sorption, the time allowed for equilibrium, and the properties of the associated microbial community. Adsorption of phenanthrene by marine sediments has recently been examined in another Coastal Marine Institute (CMI) study by Henrichs, and is presented elsewhere in this Annual Report. Henrichs found that phenanthrene is strongly adsorbed by Jakolof Bay sediments, and that the process is rapid with much of the adsorption occurring in less than fifteen minutes. The overall objective of this present study is to begin to understand the response of marine sediment microbial communities to low-level inputs of aromatic hydrocarbons. This report discusses the results of the first ten months of this project. This project has also funded the training of one M.S. student.

### Methods

#### **Sample Collection**

Sediments were collected on July 22, 1994 from the north shore of the eastern end (mouth) of Jakolof Bay. This bay is located near the Kasitsna Bay Research Laboratory on Lower Cook Inlet. The sediments are from the mudflats and are reasonably typical of intertidal sediments located throughout the Lower Cook Inlet region. Sediments were selected for their relative homogeneity, lack of macro-biological organisms, and for the presence of an easily collectible oxic layer. The sediments have not been particle-analyzed, but are fine in texture with a large silt component. Sediment samples were collected both for use in this

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project and for the CMI project of Henrichs. Sediments were kept on ice until they were transported to Fairbanks for processing.

#### **Microbial Populations**

Microbial populations of total heterotrophic microorganisms, hydrocarbon emulsifiers, and phenanthrene degraders were measured in freshly (within 48 hours of collection) collected sediment. Total heterotrophs were measured using a marine broth medium (Difco) in 24-well cell well plates (Corning) with a most probable number method (MPN) (Lindstrom et al., 1991). Sheen screen MPNs (Brown and Braddock, 1990) were used to assay the proportion of the population able to emulsify crude oil. Phenanthrene degraders were enumerated by a fivetube MPN procedure based on addition of <sup>14</sup>C-labeled phenanthrene as a sole carbon source to a series of MPN vials. Sediment was first diluted 1:10 in a Bushnell-Haas medium (Difco) amended with 2.5% NaCl. A 10-fold dilution series was set up with 5 replicate 40 mL septa vials (I-Chem Research) prepared for each dilution. Each vial was inoculated with 50  $\mu$ l of phenanthrene (2 g/L in acetone; final concentration 10 µg/mL slurry; containing about 50,000 dpm 9-14C phenanthrene). Abiotic controls were prepared both by autoclaving some vials and by adding 1 mL 10 N NaOH at time zero to other vials. The vials were incubated for two weeks then "killed" with the NaOH. The <sup>14</sup>CO<sub>2</sub> was recovered by acidifying the vials with 1.5 mL concentrated HCl and purging the CO<sub>2</sub> with a stream of nitrogen into a CO<sub>2</sub>-sorbing cocktail (see Brown et al., 1991). Vials containing CO<sub>2</sub> levels two times that of those measured in the abiotic controls were scored as positive for activity (Roubal and Atlas, 1978). This method was selected because it provided enough sensitivity to measure the populations.

#### **Microbial Activity Measurements/Kinetics Studies**

Microbial activity measurements were made using radiolabeled hydrocarbons (phenanthrene) in a manner similar to previous studies with sediment slurries (Braddock et al., 1993 after Brown et al., 1991). Duplicate vials of sediment slurries (concentrations specified for individual experiments) in a mineral salts medium (Bushnell-Haas medium amended with 2.5% NaCl; Atlas, 1994) were injected with phenanthrene in acetone (concentrations specified for individual experiments) with 9-14C labeled phenanthrene added (about 50,000 dpm per aliquot). Immediately after injecting the vials with radiolabled phenanthrene, 1 mL of 10 N NaOH was injected into a series of vials at time zero to stop microbial activity and trap CO<sub>2</sub>. The remaining vials were incubated for specified periods and then "killed" by adding the NaOH. Other chemical inhibitors of microbial activity were also used, including autoclaving and the addition of 200 mg/L (final concentration) sodium azide. The CO2 was then recovered by adding 1.5 mL HCl to each vial and purging the released CO<sub>2</sub> via a nitrogen stream into a CO2-sorbing cocktail. All final values were corrected for any label detected from abiotic controls.

7 —

The fate of phenanthrene in sediment added to Jakolof Bay sediment slurries was also examined by radiorespirometry. After incubation, <sup>14</sup>C-CO<sub>2</sub> was collected by removing 0.5 mL of headspace and 0.5 mL of supernatant with a gas-tight syringe (Hamilton). This aliquot was then added to a fresh vial containing 1 mL of concentrated HCl. The <sup>14</sup>C-CO<sub>2</sub> evolved was either collected in phenethylamine on filter paper in a center well, or by purging with nitrogen as previously described. Dissolved phenanthrene was recovered by first centrifuging the I-chem vial at 5000 x g for 5 minutes, then removing the supernatant and placing it in a fresh vial. The remaining sediment was then rinsed with 10 mL of mineral salts medium (Bushnell-Haas medium amended with 2.5% NaCl; Atlas, 1994), centrifuged again, and the supernatant removed. The supernatant was extracted by adding either 5 mL of benzene or 5 mL of methylene chloride, vortexing and removing 1 mL of the organic solvent for counting. The sediment remaining was extracted with three rinses of either benzene or methylene chloride, and then the aliquot was counted. Different solvent systems were used to optimize the extraction efficiency.

#### **Bioassay Development**

Phenanthrene degraders were isolated from enrichment cultures of freshly collected Jakolof Bay sediment. Four isolates were obtained using a spray plate protocol (Kiyohara, 1982). The isolates were then grown on a nutrient broth until dense. Standard curves of absorbance vs. cell number were made with standard plate count methods. The organisms were then grown to a standard optical density and the phenanthrene degradation activity measured as described earlier.

## **Results and Discussion**

Three groups of microbial populations were measured in Jakolof Bay sediments, including total heterotrophs, phenanthrene degraders, and hydrocarbon emulsifiers (Table 1). Total heterotrophs were about 2 x 10<sup>6</sup> per g sediment. For comparison, a direct count of total microorganisms in sediments from this same part of Cook Inlet found about 10<sup>8</sup> microorganisms/g sediment (Atlas et. al., 1983). Total

Sample	HC-emulsifiers	Phenanthrene Deg.	Heterotrophs	
		(cells/g sediment)		
1	<20	$1.7 \ge 10^3$	1.7 x 10 <sup>6</sup>	
2	<20	9.4 x 10 <sup>1</sup>	1.7 x 10 <sup>6</sup>	
3	<20	$3.4 \ge 10^{1}$	2.2 x 10 <sup>6</sup>	
Median	<20	$9.4 \ge 10^{1}$	<b>1.7 x 10</b> <sup>6</sup>	

Table 1. Microbial populations in Jakolof Bay sediments (three replicate samples) collected July 1994.

bacteria, as measured by direct count microscopy, also found about 10<sup>7</sup>-10<sup>8</sup> microorganisms/g sediment in Prince William Sound sediments (Braddock et al., 1993). Direct counts typically yield higher numbers than culture methods since culturing methods only measure active microorganisms that can grow under the conditions provided. Either approach can be used to monitor the relative magnitude of the heterotrophic population. Hydrocarbon degraders, as determined by the sheen screen MPN, were found to be below the detection limits of the assay (<20/g sediment). This is consistent with the use of this assay at other pristine marine sites (Braddock et al., 1995). However, using a radiorespirometric MPN procedure, Atlas et al. reported between 10<sup>2</sup> and 10<sup>3</sup> hydrocarbon utilizers in sediments from Lower Cook Inlet. The range measured by Atlas et al. more closely corresponds to the number of phenanthrene degraders enumerated in this study. Phenanthrene degraders were found to range from 10<sup>2</sup> to 10<sup>3</sup>. The populations of phenanthrene degraders may be related to the presence of naturally occurring aromatic compounds such as terpenes in the environment (Button, 1984). The presence of these organisms also has implications for the fate of hydrocarbons entering this environment. Phenanthrene activity was also measured in freshly collected Jakolof Bay sediments (Figure 1). Phenanthrene was mineralized at measurable rates after about a three-day lag phase. Phenanthrene utilization in the time course may be representative of the mineralization capacity of the naturally occurring population or may reflect an enrichment of the population of phenanthrene degraders to adapt to phenanthrene utilization under laboratory conditions. In previous experiments using a similar assay, it was found that PAH utilization after less than ten days incubation was reflective of the



Figure 1. Mineralization of <sup>14</sup>C-labeled phenanthrene (10  $\mu$ g/mL) with time for freshly collected Jakolof Bay sediment slurries (1:10 sediment to mineral salts medium). The values are means of duplicate points, and the range in values is shown with error bars.

9.

capacity of the naturally occurring population to utilize that substrate (Brown et al., 1991). A similar time course experiment run with sediment stored in a freezer for about six months showed no activity for phenanthrene utilization even after 30 days incubation, indicating that major population shifts occur in stored sediment (results not shown). Unfrozen sediment stored in a refrigerator began to go anaerobic shortly after collection. Therefore, to reflect processes occurring in the natural environment, laboratory work to determine microbial activity must be performed very soon after collecting fresh samples.

In related mineralization experiments, several methods were used to attempt to complete a mass balance (Table 2). At the concentrations of sediment (1:10 wet weight sediment to nutrient-amended seawater) and phenanthrene (10 µg/mL) used in these experiments, only about 50% of the initial labeled phenanthrene could be accounted for. The residual 50% was apparently sorbed to sediment and was not readily extracted using benzene or methylene chloride. Concentrations of sediment and phenanthrene and extraction protocols developed by Henrichs will be used in the next set of experiments. Two methods of collecting CO<sub>2</sub> were compared in this experiment: purging with nitrogen and collection in phenethylamine in a center well. Both methods recovered about the same amount of CO2. In nine-day incubations, about 10% of the label was evolved as CO2. No labelled CO2 was evolved after only one day of incubation. The dissolved concentration of phenanthrene measured (15% of the label added) corresponds to about 1.5 mg/L, slightly higher than would be expected from the aqueous solubility of phenanthrene (about 1 mg/L in pure water at 298 K; Shaw, 1989), and may reflect the presence of other products of phenanthrene degradation or contaminants in the phenanthrene. The commercially prepared <sup>14</sup>C-labeled phenanthrene used in these experiments and those of Henrichs may be contaminated by an unidenti-

Table 2. Fate of  $9^{-14}$ C phenanthrene (10 µg/mL) in sediment slurries (1:10 wet sediment to nutrient-amended seawater) after one-day and nine-day incubations. Abiotic controls ("killed" at time zero) were subtracted from each prior to calculation of percentages. Each value represents nine replicate vials ± 1 standard deviation. Method 1, CO<sub>2</sub> acidified then purged with a nitrogen stream into CO<sub>2</sub>-sorbing cocktail. Method 2, CO<sub>2</sub> collected on a phenethylamine-soaked filter paper in a center well.

Fraction	Method 1 9-day	Method 2 9-day	Method 2 1-day	
	% of ra	diolabeled phenanthren	e added	
CO <sub>2</sub>	$7\pm 6$	$10\pm4$	0	
dissolved	<sup>1</sup> 15 ± 2	$^{1}17 \pm 2$	$^{1}16 \pm 3$	
extracted from pellet	$^{2}29 \pm 1$	$^{2}23 \pm 3$	$^{3}20 \pm 7$	
total recovered	51	50	48	

<sup>1</sup>three seawater rinses

<sup>2</sup>benzene extraction

<sup>3</sup>methylene chloride ext.

fied hydrocarbon (Henrichs, pers. comm.). Mass balance experiments in the absence of microbial activity have recently been completed in Henrichs' laboratory. As soon as fresh sediment is collected in June 1995, the adsorption data collected from those experiments will be used to perform time course studies in the presence of the natural microbial populations. We have also begun to develop a bioassay for determining the bioavailability of phenanthrene in sediment slurries. Four phenanthrene degraders were isolated from fresh Jakolof Bay sediment collected in July 1994. These organisms have been characterized as Rhodococcus sp., Arthrobacter sp., and two Pseudomonas sp. Standard curves for growth, relating viable cell counts to absorbance, have been determined for each organism so that cultured organisms at specific population densities can be readily added to sediment slurries. These isolates all grow with phenanthrene as a sole carbon source on phenanthrene spray plates (Kiyohara et al., 1982). All of the isolates will also grow on a general heterotrophic medium. However, if they are grown for several generations on a general medium, they all lose their ability to mineralize phenanthrene. The activity of a constant biomass of these organisms is currently being determined in time course studies with <sup>14</sup>C phenanthrene. Once the phenanthrene utilization kinetics of each organism are established, this suite of organisms can be used to look at the effect on sediment and bioavailability of phenanthrene in slurry experiments. It is hoped that this second approach to measuring bioavailability will help in understanding the effect of sedimentsubstrate-microorganism interactions on availability of PAH carbon substrates.

### **Preliminary Conclusions**

Oil inputs to the marine environment have been studied intensively for the past several decades, and a number of processes have been identified that contribute to controlling the fate of these compounds in the marine environment. Some of these processes are fairly well understood, particularly for acute inputs of petroleum compounds. Other processes are less well understood and contradictory results have been reported, particularly where petroleum inputs are at low concentrations. Microbial transformations are often critical to the long-term fate of lower molecular weight aromatic hydrocarbons, but the degradability is likely to be affected by physical and chemical interactions of aromatics in sediments. The interactions between the microbial community and sorbed hydrocarbons may be very important in understanding the potential impacts of chronic petroleum additions to the marine environment. Further, microbial transformations may largely determine the persistence of compounds such as polycyclic aromatic hydrocarbons and thus affect their degree of hazard to the environment. The interactions between the extant microbial populations and the bioavailability of phenanthrene in sediment slurries from Lower Cook Inlet, Alaska, are beginning to be examined. These studies are important in predicting the fate of chronic and acute oil contamination to marine systems.

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## Intertidal and Subtidal Effects of Pollution: Assessment of Top-trophic Level Predators as Bioindicators

Lawrence K. Duffy R. Terry Bowyer Daniel D. Roby Institute of Arctic Biology University of Alaska Fairbanks Fairbanks, AK 99775-7000

James B. Faro Alaska Department of Fish and Game Sitka, AK 99835-7563

## Abstract

Pigeon guillemots (Cepphus columba) are well suited as bioindicators of contaminants in neritic food webs, because breeding pairs are widely dispersed and feed on nearshore demersal fishes. Average productivity of 62 nests monitored in Kachemak Bay was 0.4 fledglings per nest (Mayfield method). Low egg survival (50%) and hatching rates of surviving eggs (76%) were responsible for poor overall hatching success (38%); 57-69% of the chicks that hatched survived to fledgling. Predation was a major cause of nest failure. Diets of guillemot nestlings were dominated by sand lance (46%), with smaller proportions of blennies (29%) and sculpins (2%). Blood samples collected from adults and nestlings (20 d and 30 d post-hatch), showed haptoglobin values which ranged from 0 to 282 mg/dL (mean = 103, SD = 62.7). Mean levels differed among nests, but not between alpha- and beta-chicks from the same nest. However, haptoglobin levels did not differ among colonies, sampling dates, or nestling ages, nor were haptoglobin levels correlated with nestling growth performance. River otter latrine sites were documented and six animals were captured and sampled. These baseline biomarker values will aid future assessment of contaminant exposure.

## Introduction

Focusing on the biological responses in indicator species overcomes many of the limitations that plague chemical analysis of the environment (Payne et al., 1987). While measuring body burdens is an important aspect of using higher vertebrates as biomonitors, equally important is the measurement of biomarkers because they:

1) Provide evidence of exposure to compounds that do not bioaccumulate or are rapidly metabolized;

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- 2) Integrate the toxicological interactions resulting from exposure to complex mixtures of contaminants;
- 3) Present a biologically relevant measure of the cumulative adverse effect; and
- 4) Measure early responses of organisms to toxicant exposure and serve as short-term predictors of long-term adverse effects.

Development of biomonitors will lead to a better understanding of environmental and ecosystem processes related to outer continental shelf (OCS) activities. In this research our primary focus is to develop the pigeon guillemot and the river otter as new indicators of environmental stress in nearshore marine ecosystems.

Use of these bird and mammal models as upper trophic-level sentinels of bioavailable contaminants, such as oil, PCBs, and metals, will be developed to act as surrogates to estimate potential exposure and risk to humans living near sites of OCS activities (Fry and Lowenstine, 1985; Leighton 1985; Peakall et al., 1986). This research approach utilizes biomarkers (biochemical and cellular indicators of exposure), reproduction rates, and data on mortality to identify and quantify the present level of variability within the Lower Cook Inlet–Kachemak Bay ecosystem. This approach is necessary because evaluation of the potential for exposure to contaminants in the environment is extremely complex due to the differences in the biological availability of contaminants at different trophic levels and varying toxicological interactions within exposed organisms.

#### **Pigeon Guillemots**

Pigeon guillemots are one of the best-suited avian species for monitoring nearshore marine ecosystem health for several reasons:

- 1) they are a common and widespread seabird species breeding in coastal Alaska, specifically in Lower Cook Inlet and Shelikof Straits (Sowls et al., 1978);
- 2) they forage within 5 km of the nest site in the subtidal and nearshore zones (Drent, 1965);
- 3) unlike most seabird species, they do not breed in large, dense colonies that are sensitive to disturbance;
- 4) they raise their young almost entirely on fish, preying primarily on subtidal and nearshore bottomfish (blennies, sculpins), as well as a few schooling forage fish (e.g., sand lance; Drent, 1965);
- 5) the one- or two-chick broods are fed in the nest until the young reach adult body size; and
- 6) adults will breed in nest boxes, thus providing a readily accessible study population (Asbirk, 1979).

Guillemots begin breeding at two years of age and adults have high annual survivorship (85%; Asbirk, 1979). Young guillemots normally return to the natal

area to breed. Nest site fidelity of breeding pairs is high, and even in instances when pairs relocate nests, the distances involved are usually small (<30 m). Eggs are laid in a wide variety of natural crevices and holes, but most nest sites in the study area are located in cavities in cliffs or cracks in rock masses (G.C. West and D. Erikson, pers. comm.). Eggs are usually laid about 50 cm from the entrance of the nest crevice (Asbirk, 1979), thus the eggs, chicks and attending adults are normally accessible for data collection. Guillemots are unusual among alcids in that they normally lay two-egg clutches and raise two chicks per nesting attempt. Guillemots carry whole fish in their bills to the nest-site crevice to feed their young. Thus, individual prey items can be identified, weighed, measured, and if necessary, collected for contaminant analyses.

Guillemots have served as subjects in previous studies to assess the effects of ingested crude oil on marine birds (Peakall et al., 1980). Nestling black guillemots (*Cepphus grylle*), a very closely-related sibling species of the pigeon guillemot, were fed single doses of weathered South Louisiana crude oil (WSLC) and subsequently monitored in their natural nest site where they were cared for by their parents. These experiments demonstrated that single doses of as little as 0.1 mL WSLC resulted in declines in growth rates, increases in plasma sodium levels, and increases in nasal and adrenal gland masses. The effects of the single dose were not transient, as nestlings that were dosed at roughly two weeks posthatch were 20% lighter than controls at five weeks of age (just prior to fledging). Such persistent sublethal effects may have serious consequences for post-fledgling survival. Peakall et al.'s (1980) study clearly demonstrates that guillemot nestlings living normally in their chosen habitat are tolerant to the handling and disturbance associated with experiments designed for assessing pollutant toxicity.

Because of the repeated transfer of marine prey captured by adults to their young, chicks act as a potential sink for petroleum hydrocarbons in contaminated prey. After a large oil spill at high latitudes, chronic consumption of polycyclic aromatic hydrocarbons (PAH) by adult guillemots and the accumulation of PAH in chicks would be expected to result in reproductive effects. Thus guillemot chicks provide a good model for investigating the impact of petroleum hydrocarbon exposure on seabird reproduction. The high molecular weight aromatic fraction of crude oil is known to be the most persistent in the marine environment, and is also the causative agent in growth retardation and hypertrophy of adrenal and nasal glands in seabirds (Peakall et al., 1982). The assimilation, detoxification, and metabolism of PAH may interfere with the characteristic accumulation of lipid reserves in guillemot chicks. Presumably, chicks store assimilated aliphatic hydrocarbons from ingested crude oil in their large fat deposits. Much of these lipid reserves are metabolized by the chick prior to sea-going, at which time remobilized aliphatics may be pathogenic. In summary, guillemot chicks are well suited as models for monitoring sublethal effects of exposure to crude oil.

17 \_

#### **River Otters**

River otters inhabiting marine environments also serve as excellent monitors since they make extensive use of, and concentrate their activities in, intertidal and subtidal zones (Dubuc et al., 1990; Larsen, 1984; Woolington, 1984). These high trophic-level carnivores are long-lived ( $\geq 12$  years of age; Docktor et al., 1987), and occur at densities of 0.2–0.8 otters per km of shoreline throughout the Gulf of Alaska (Testa et al., 1994). River otters are extremely sensitive to aquatic pollutants (Table 1), yet continued to reside within the area of oil-contaminated shorelines in Prince William Sound, Alaska, following the *Exxon Valdez* oil spill (Testa et al., 1994). River otters commonly inhabit marine coastal environments, but have not been classified as marine mammals for purposes of the Marine Mammal Protection Act. Consequently, legal take permits are not required to conduct scientific research on this species. These characteristics make river otters an excellent model for assessing the effects of marine pollution on mammals, and provide an overall index to the health of the environment.

River otters consume a diet dominated by marine fishes, which they prey upon in intertidal and subtidal zones, although they also consume a wide variety of marine invertebrates. Such nearshore areas are the most often affected by pollution. For instance, oil spilled from the *Exxon Valdez* contaminated extensive areas of the intertidal and subtidal environments, which was reflected in a loss of dietary diversity for otters inhabiting oil-contaminated shorelines (Bowyer et al., 1995). Likewise, river otters living in oiled areas exhibited a significantly lower body mass (when controlled for sex and total body length) than did otters inhabiting oil-free areas (Duffy et al., 1993). Otters have extremely large home ranges (20–40 km of shoreline; Bowyer et al., 1995), and hence integrate the effects of pollution over wide areas.

Pesticides	Heavy Metals	Heavy Metals Cesium-137		Crude Oil	
Clark et al. 1981	Clark et al. 1981	Clark et al. 1981	Clark et al. 1981	Duffy et al. 1993	
Halbrook et al. 1981	O'Connor & Nielson 1981	Halbrook et al. 1981	Henney et al. 1981	Duffy et al. 1994	

Table 1. Published literature indicating river otters are especially sensitive to pollutants in aquatic systems.

A nonlethal method for evaluating the effects of marine pollutants (in this instance, crude oil) on the blood-enzyme chemistry of river otters has already been developed (Duffy et al., 1994). Blood haptoglobins (an acute-phase protein) were elevated in otters inhabiting areas where crude oil was prevalent one year following the oil spill (Duffy et al., 1993). Even two years after the oil spill and a major effort to clean oil-contaminated shores, a biostatistical model was successfully constructed, using logistic regression, in which > 85% of river otters were correctly classified as having been captured in oiled or non-oiled zones. This

highly sensitive model used only blood values for haptoglobin, interleukin-6 (a cytokine), and AST (a liver enzyme). The approach has already been extended for evaluating other marine mammals (Zenteno-Savin et al., 1993), and may be applicable to other vertebrates, especially marine birds. Coupling this knowledge of otters with similar physiological data for pigeon guillemots will provide a sensitive tool for examining the effects of pollution on the nearshore environment.

## Methods

Field studies were conducted during the 1994 breeding season in Kachemak Bay, Alaska (Figure 1). Approximately 500 pigeon guillemots nest along the shores of Kachemak Bay and the tributary bays on the south side of Kachemak Bay (Sowls et al., 1978). Active and accessible nests from Mallard Bay in the northeast to Seldovia Bay in the southwest were located and marked during the breeding season. These nests were closely monitored until the young fledged or the nesting attempt failed. From June to August, chicks that hatched in accessible nests were weighed regularly to determine individual growth rates throughout the nesting period. Alpha and beta nestlings were marked shortly after hatching for consistent identification throughout the nesting period. Blood samples (c. 1 mL) were collected in heparinized vials by brachial vein puncture from each nestling at ages 20 and 30 days post-hatch (guillemot chicks normally fledge at 30-40 days posthatch). The whole blood was centrifuged and the plasma frozen at  $-20^{\circ}$  for later analyses in the lab. Blood samples were analyzed for plasma protein, haptoglobins and IL-6. Twelve 30-day-old nestlings were collected for P450 assays. The taxonomic composition of guillemot diets was determined, with the aid of 15-60x spotting scopes, by observing adults transporting nestling meals in their bills.

River otters, which are moderately abundant in Kachemak Bay, were captured using Hancock live traps (Duffy et al., 1993) placed on trails at latrine sites. Initially, the otter was immobilized in the trap with a hand injection of ketamine hydrochloride (11 mg/kg estimated body weight, Sigma, St. Louis, Missouri, USA) and then placed in a drugging box. The blood biomarkers measured in this study were not induced for at least 24 hours after exposure to an inducer, so if ketamine hydrochloride was an inducer, the effect would not be measurable in blood samples. Total body mass and length measurements were taken and a blood sample drawn from the jugular vein. Sexes were distinguished by the relative position of urogenital openings and palpitation of the baculum (Larsen, 1984). Age estimates of river otters using age/length curves are inaccurate, so potential correlations between otter size and biomarker levels were controlled with the use of analysis of covariance.



Figure 1. Study area for pigeon guillemots in Kachemak Bay, Alaska, during the 1994 breeding season. All guillemot nests in the study were located along the south shore of Kachemak Bay from Seldovia to Bear Cove.

## **Results and Discussion**

#### Guillemots

Sixty-two active and accessible nests were found along the rocky shores of the south side of Kachemak Bay and on nearshore islets demonstrating that adequate numbers of accessible pigeon guillemot nests for the proposed 1995 dose-response experiment are present in Kachemak Bay. Most nests were located in crevices in vertical cliffs, presumably as a defense against nest predation by terrestrial mammals (e.g., mink), making access more time consuming than originally anticipated. Visual observation confirmed that marine fishes were by far the major food source of breeding pigeon guillemots in Kachemak Bay; guillemots were not observed to feed any invertebrates to their nestlings. Also, diet composition was different than that reported for other guillemot populations in the Pacific Northwest, where bennies and sculpins were a major part of the diet. In Kachemak Bay, 46% of prey items delivered to the nestling were sand lance (Ammodytes sp.), while sculpins comprised only 2%, and gunnels/pricklebacks, 29% of nestling diet (Figure 2). This diet composition is similar to that of guillemots breeding at Naked Island in Prince William Sound prior to the Exxon Valdez oil spill. Nestling diet composition in Kachemak Bay varied among sites (Figure 3). Sand lance

were much more prevalent and gunnels less prevalent at Moosehead Point (China Poot Bay) than at Halibut Cove or Kasitsna Bay to the east and west, respectively.

- a) Growth Rates. Growth rates in body mass of known-age chicks were measured and several other methods were used to incorporate data from chicks of unknown ages. Growth in mass is approximately linear from day 8 to 18 in guillemots. This rate is easy to calculate and can be used for comparisons, but confidence intervals for growth rates are large (Figure 4). Growth rates can also be estimated for chicks of unknown age by weighing many chicks at a constant time interval (e.g., three days). By determining the increase in mass over this interval for various initial weights (Figure 5), the change in mass with age can be reconstructed. Using measurements taken three days apart provided a growth curve very similar to that produced by using only values from known-age chicks (Figure 6). The growth rate from day 8 to 18 was 17.3 g per day.
- b) Productivity. Average clutch size was 1.68 eggs/nest (n=38 nests). Productivity and other parameters of reproductive success were estimated using the Mayfield Method. Productivity (number of fledglings produced per nest where at least one egg was laid) was low at 0.41 fledglings/nest, due mainly to a high predation rate on eggs (50%) and low hatching success (76%, Figure 7). Of those chicks that hatched, 57–69% survived to fledgling. There was considerable variation among locations within Kachemak Bay in the causes of nest failure (Figure 8). Guillemots breeding at Moosehead Point and China Poot Bay experienced much higher egg losses (69%) than birds nesting to the east (30%) or west (36%). However, guillemot nestlings suffered lower mortality rates at Moosehead Point/ China Poot Bay (2% of nests) than to the east (12%) or west (13%).
- c) Biomarkers. Extensive blood samples and liver tissue samples were collected from nestling guillemots. Many of these analyses are currently underway. Total plasma protein as well as classes were determined over time. These data are summarized in Tables 2-4. Haptoglobin levels were also measured in a timed series.

Haptoglobin levels varied significantly among nests but were not significantly different among ages, locations, dates, or between alpha and beta chicks. Overall serum protein levels were positively correlated with haptoglobin levels (R = 0.46) but did not vary among nests, locations, ages, dates, or alpha and beta chicks. The levels of haptoglobin and total proteins in serum were not correlated with growth performance (Figure 9) as indicated by the residuals of body mass as a function of wing length (Figure 10). The immune system was judged by IL-6 levels. All IL-6 levels were below detection, giving a relative zero as a baseline for the 1995 dosing experiment.

The variation in individual birds over time was interesting. For example, the chick in Nest 2 had an HP level of 52 on July 18, and an HP level of 99 on July 28, while the chick in Nest 43 had an HP level of 80 on August 4, and 0 on August 11.

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#### **River Otters**

Twenty active otter latrine sites were identified and scat samples collected for stable isotope analysis as well as metal and porphyrin analysis. Preliminary results indicated very low levels of total porphyrins in feces. Overall the baseline data collected in 1994 and the logistical experience gained in Kachemak Bay portend for a successful field season in 1995. River otter blood samples were not taken in 1995, but several whole river otter carcasses (including livers) were obtained for laboratory analysis.

#### **Comparative Implications of Bioindicator Studies**

Guillemot growth data from the study in Kachemak Bay was compared to similar data from oiled and unoiled areas of Prince William Sound in 1994 (D.L. Hayes, unpublished data). It was found that Kachemak Bay guillemot nestlings had significantly lower growth rates and fledged at significantly lower body mass than guillemot nestlings raised on Jackpot Island in an unoiled area within Prince William Sound. Growth rates and fledgling mass of guillemot chicks on Naked Island, in an oiled portion of Prince William Sound, were indistinguishable from those of Kachemak Bay guillemot chicks (Figure 11). This supports the concept that monitoring growth of guillemot chicks provides a sensitive model for studying the local effects of environmental pollution and prey availability. Kachemak Bay is not considered part of the Exxon Valdez Oil Spill Area, although shoreline at the mouth of the bay may have been lightly oiled. The differences in growth rate of guillemot nestlings between Kachemak Bay and Jackpot Island may be due to the differences in contaminant exposure and/or differences in the quantity or quality of forage fish. Studies designed to address these issues are being planned for 1995.





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Figure 3. Taxonomic composition of the diet for pigeon guillemot nestlings in three areas of Kachemak Bay, Alaska during 1994. Percentages represent the proportion of total observed prey items delivered by guillemot adults to nests containing chicks.



Linear Phase of the Growth Curve

Figure 4. Nestling body mass as a function of age (days post-hatch) for pigeon guillemots in Kachemak Bay, Alaska during 1994. The figure depicts the "linear phase" of nestling growth in body mass, about 8–18 days post-hatch. The line represents the least squares regression for the entire data set.

23 \_



Figure 5. Nestling body mass as a function of body mass three days earlier (initial mass) for pigeon guillemots breeding in Kachemak Bay, Alaska, during 1994. The figure shows the three-day mass increment in relation to the "no-growth" line (lower dashed line). These graphs can be used to construct sigmoidal growth curves from data obtained from nestlings of unknown age.













25 -

#	Date	Total Protein	γ2/1	β2	β1-2	β1	α2	α1	Alb/pre	HP
1	7/20	2.03	0.19	0.19	0.31	0.19			1.15	0
2	7/20	2.25	0.1/0.21	0.16	0.18	0.36			0.90/0.27	89
2	7/18	1.85	0.25		0.14	0.26			0.84/0.36	52
2	7/28	2.48	0.49	0.14	—	0.55	—	0.34	0.97	99
8	7/23	2.35	0.39	0.15	0.22	0.45		0.29	0.86	120
8	8/04	2.45	2.44	0.04	0.11	0.47	0.01	0.57	0.80	162
15	7/28	2.98	0.31	0.39		0.77			1.51	87
15	8/06	2.70	0.17	0.32		0.52			1.00	113
20	8/19	2.23	0.17	0.12		0.34		0.48	1.11	54
20	8/01	2.50	0.34	0.24		0.55		0.26	0.96	64
21	7/20	3.70	0.63		0.18	0.80	0.49	0.48	1.12	144
21	7/25	2.20	0.38		0.17	0.53		2.22	0.90	117
23	8/06	2.75	0.47	0.38		0.59	0.45	—	0.86	93
23	7/28	2.83	0.53	0.50	0.34	0.59			0.87	36
24	7/19	2.58	0.37	0.33		0.89			1.00	207
29	7/23	3.20	0.57	0.54		0.78			1.31	58
29	8/03	3.58	0.69	0.24	0.34	0.95		0.24	1.10	282
31	8/13	2.70	0.56	0.34		0.68		0.29	0.84	196
32	8/07	1.50	0.25	0.36	—	0.22			0.67	39
32	7/29	2.50	0.50	0.44	—	0.28	0.41	_	0.86	95
35	7/23	3.30	0.44	0.70		1.10	0.17	0.16	0.73	203
35	8/11	2.15	0.45	0.28	—	0.29		0.26	0.91	124
35	8/04	3.03	0.62	0.36		0.66	—	0.55	0.85	210
38	7/26	2.95	0.79			0.68		0.61	0.86	96
38	8/06	2.30	0.37	0.38		0.34	0.19	0.37	0.63	61 07
41	7/19	2.83	0.61	0.44		0.67		0.42	0.66	35
41	7/24	2.90	0.61	<u> </u>		0.89	0.28		1.12	57
42	7/19	2.60	0.11/0.29	00	00	0.79	0.18		1.04	50
42 43	7/31 8/11	2.30 2.03	0.45 0.28	0.05 0.26		0.43 0.30	0.10	0.46 0.11	0.80 1.08	108 0
43 43	8/11 8/04	2.03 2.15	0.28	0.26		0.30	0.11	0.11	0.84	80
43 44	7/23	2.13	0.52	0.11	0.16	0.55	0.41	0.33	0.84	228
44	7/23	2.30 3.13	0.05/0.57	0.21	0.13	0.52	0.41	0.17	0.55	228 151
46	7/18	1.88	0.05/0.57	0.20	0.25	0.30		0.18	0.55	169
46	7/23	2.30	0.13	0.20		0.57	_	0.33	0.57	113
47	7/20	2.50 2.50	Blank	lane	needs	to	be	run	0.57	113
47	7/31	2.50	0.43	0.23		0.35	0.20	0.58	0.70	139
49	8/15	2.50	0.48	0.29		0.39		0.30	0.90	47
49	8/06	2.10	0.48	0.18		0.38		0.48	0.74	30
50	8/01	2.63	0.45	0.22	0.06	0.50		0.61	0.79	48
52	8/01	2.25	0.37	0.15	0.08	0.47	_	0.40	0.79	38
53	8/05	2.15	0.46		0.17	0.56	_	0.31	0.65	33
54	8/14	2.38	0.32	0.19		0.34		0.69	0.86	72
54	8/05	2.12	0.40	0.25		0.57		_	0.89	104
55	8/06	2.95	0.52	0.35	0.11	0.66		0.56	0.75	297
56	8/09	2.25	0.39	0.22		0.37		0.52	0.75	0
56	8/04	3.15	0.41	0.43	_	0.75		0.65	0.92	33
57	8/10	2.60	0.43	0.20	—	0.53		0.54	0.81	91
57	8/05	2.50	0.53	0.53		0.45			0.99	66
58	8/13	1.70	0.25	0.17		0.30	0.30		0.66	105
59	8/08	1.70	0.49	0.40		0.22			0.60	80
60	8/10	1.70	0.26	0.32	—	0.36		0.35	0.42	84
60	8/14	1.80	0.26		0.52	_	0.38		0.64	35
61	8/12	2.75	0.51	0.44		0.74	_	0.39	0.66	206

Table 2. Guillemot  $\alpha$  chick plasma proteins and HP.

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#	Date	Total Protein	γ	β2	β1-2	β1	α2	α1	Alb/pre	HP
8	7/26	2.75	0.48	0.31		0.79		0.44	0.73	54
8	8/04	2.80	0.49	0.19		0.69	_	0.71	0.73	66
20	8/10	2.80	0.34	0.32	0.14	0.64		0.67	0.68	102
20	8/01	3.80	0.41	0.62	0.14	0.80	0.42	0.54	0.86	8
23	8/06	2.50	0.25	0.41	—	0.65	0.19	0.46	0.55	115
23	7/28	2.70	0.41	0.42		0.65	0.18	0.37	0.68	116
24	7/19	2.50	0.33	0.52		0.43		0.46	0.76	110
24	7/24	2.38	0.37	0.13	—	0.41	0.04	0.65	0.78	115
24	7/27	2.00	0.16	0.41	0.10	0.38	0.07	0.30	0.57	116
32	8/07	1.75	0.23	0.11	0.06	0.34	—	0.46	0.55	0
32	8/04	2.05	0.38	0.36		0.38		0.80	0.65	22
41	7/19	2.30	0.31	0.26	0.34	0.50		0.33	0.57	59
41	7/24	2.15	0.32	0.16		0.66		0.16	0.83	40
41	7/27	1.85	0.23	0.12		0.59	_	0.37	0.54	89
42	7/19	2.00	0.35	0.18		0.22	0.29	0.42	0.55	34
42	7/31	2.40	0.35	0.20	0.07	0.51	0.11	0.61	0.55	63
44	7/23	2.80	0.58	0.26	0.18	0.64	0.17	0.46	0.51	
44	7/29	3.00	0.47	0.72	_	0.82	_	0.36	0.93	202
44	8/10	2.15	0.45	—		0.68	0.08		0.25/0.19	79
46	7/18	2.45	0.71	0.22	0.41	0.50		0.26	0.54	70
46	7/23	2.15	0.27	0.25	—	0.29	—	0.55	0.79	0
46	7/29	2.53	0.47	0.20	—	0.19		0.32	1.34	48
47	7/20	2.73	0.46	0.44		0.37		0.42	1.05	54
47	7/31	2.50	0.52	0.33		0.44	0.02	0.42	0.77	157
53	8/05	1.80	0.28	0.54					0.74	75
56	8/04	1.58	0.20	0.09	0.29	0.21		_	0.64	35
58	8/13	2.63	0.55	0.44	—	0.25			1.39	53
61	8/12	2.45	0.27	0.19		0.61	0.13	0.43	0.82	243

Table 3. Guillemot  $\beta$  chick plasma proteins and HP.

Table 4. Adult guillemot plasma protein and HP.

#	Date	Total Protein	γ	β	β	β	α	α	Alb/pre	HP
24	7/21	2.85	0.50	0.24		0.68		0.03	1.40	197
37	7/29	3.15	0.50	0.13		0.44	0.39	0.93	0.91	138
41	7/27	3.70	0.33	0.59		0.73	0.65	0.42	0.99	177
62	8/12	3.23	0.73	0.27		0.43	0.57	<u> </u>	1.20	195

27 \_\_\_\_



Figure 9. Levels of serum haptoglobins as a function of growth performance index in pigeon guillemot nestling from Kachemak Bay, Alaska, 1994. The growth performance index was the residual about the regression of total body mass vs. wing length (a surrogate for body size). Nestlings with high positive residuals were heavy for their body size, while nestlings with low negative residuals were underweight for their size.







Fig 11. Nestling body mass as a function of wing length for pigeon guillemots from Kachemak Bay, compared with Jackpot Island in Prince William Sound, Alaska, during 1994. Body mass was square root transformed and wing length was transformed by taking the square root of the log. These transformations linearized the relationship between body mass and wing length, and produced homogeneous variance about the linear regression. These conditions are essential for meeting the assumptions of linear regression and allow slopes of regression lines to be compared. The slope of the regression line for guillemots raised at Jackpot Island was greater than that of guillemots raised in Kachemak Bay. Jackpot Island is in a part of Prince William Sound that was not oiled during the Exxon Valdez oil spill.

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29 \_\_\_\_

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31 \_

## A Study of the Adsorption and Biodegradation of Aromatic Hydrocarbons by Marine Sediments

#### Susan M. Henrichs Michelle Luoma

Institute of Marine Science University of Alaska Fairbanks Fairbanks, AK 99775-7220

## Abstract

Sources of acute and chronic discharges of petroleum to the marine environment include marine transportation, municipal waste and runoff, refineries, and offshore oil production. Aromatic constituents of petroleum are of special concern because their toxicities are greater than those of most other components. Because of their hydrophobic properties, aromatic hydrocarbons have a strong tendency to become associated with particles in sediments, and this association could affect their biodegradation. The adsorption of a specific aromatic hydrocarbon, phenanthrene, by intertidal sediments of Jakolof Bay, Lower Cook Inlet, was studied. Phenanthrene is strongly adsorbed by these sediments, and the adsorption is not completely, rapidly reversible by suspension of the sediment in clean seawater. Of the phenanthrene which is adsorbed irreversibly, 50–90% can be extracted by methanol. Adsorption is a linear function of dissolved phenanthrene concentration, and all sites for adsorption on the sediment surface appear to be equivalent. In collaboration with Joan Braddock, the effects of adsorption on the rate and extent of decomposition by bacteria are being studied.

## Introduction

#### **Overview**

The Alaskan Coastal Marine Institute (CMI) developed a group of "Framework Issues" which describe the concerns to be addressed by CMI-supported research. Among these, there was a need identified for "experimental studies for better understanding of environmental processes or the causes and effects of outer continental shelf (OCS) activities" (University of Alaska CMI Annual Report No. 1). Lower Cook Inlet is a site of offshore oil and gas production. Offshore production and marine transportation are sources of petroleum hydrocarbons to the ocean (National Research Council, 1985). Although aromatic hydrocarbons make up only a small part (approximately 10%) of the complex mixture of compounds in petroleum, they are of special concern because they are among the most toxic, mutagenic, and carcinogenic constituents (White, 1986; Pahlmann and Pelkonen, 1987). Sediments are important reservoirs for polycyclic aromatic
hydrocarbon (PAH) contamination in the marine environment (Wakeham and Farrington, 1980; Gschwend and Hites, 1981). Also, there is evidence that sediments are a major source of PAH and other hydrophobic pollutants to marine and lacustrine organisms (McElroy et al., 1989). Although sedimentary contamination decreases markedly with time after a spill, due to biodegradation and other processes, aromatic hydrocarbons can persist for at least 20 years in contaminated sediments (Teal et al., 1992). A factor in this persistence could be the strong association between sediment particles and aromatic hydrocarbons (Means et al., 1980) and a possible decrease in biodegradability associated with this adsorption (Guerin and Boyd, 1992).

Chronic discharges and small oil spills are a much larger source of petroleum contamination to the oceans, integrated globally, than are large spills (National Research Council, 1985). In any locality with human activity, chronic, low-level petroleum inputs are a virtual certainty, whereas a catastrophic spill is unlikely. Processes controlling the fate of a large oil spill in Lower Cook Inlet have already been investigated under the "Environmental Assessment of the Alaskan Continental Shelf" program, sponsored by the National Oceanographic and Atmospheric Association and the Minerals Management Service (Payne et al., 1984; Payne et al., 1989). Processes affecting smaller inputs of oil, and, in particular, the fate of lower-level contamination in sediments, have not been studied previously.

The present research is an experimental study of phenanthrene adsorption by Jackolof Bay, Lower Cook Inlet sediments to determine how adsorption may affect the persistence and fate of an aromatic hydrocarbon in these sediments. Two ways in which aromatic hydrocarbons can be lost from contaminated sediments are desorption, if the sediment is exposed to less-contaminated seawater, or biodegradation. Therefore this study included the investigation of the desorption of phenanthrene from sediments. In collaboration with Joan Braddock of the University of Alaska Fairbanks Institute of Arctic Biology, this project is also investigating how adsorption affects bacterial degradation rates. During the next year, the study will be extended to include two other aromatic hydrocarbons, benzene and naphthalene, and another sampling site, a salt marsh. When future results are combined with this year's findings, information will be available on how aromatic hydrocarbon structure affects adsorption, desorption, and biodegradation, and how sediment composition affects adsorption.

#### **Adsorption of Aromatic Hydrocarbons**

Aromatic hydrocarbons have low aqueous solubilities and a high affinity for surfaces in aquatic ecosystems. However, within the this compound class, both solubility and surface affinity vary. For example, the aqueous solubility (g/100 g solution at 298°K) is 0.18 for benzene, 0.053 for toluene, 0.0031 for naphthalene, and 1.1 x 10<sup>-4</sup> for phenanthrene (Shaw, 1989a,b). Adsorption coefficients (K<sub>p</sub>, or the adsorbed concentration/dissolved concentration ratio) of aromatic hydrocar-

33 –

bons on marine and lake sediments are negatively correlated with aqueous solubility (Means et al., 1982). Sediment or soil composition affects adsorption. K<sub>p</sub> increases with increasing organic content (Means et al., 1980; Schwarzenbach and Westall, 1981). Sorption of various aromatic hydrocarbons was correlated positively with the nonpolar-to-polar functional group ratio (Garbarini and Lion, 1986; Rutherford et al., 1992) and aromaticity (Gauthier et al., 1987).

In studies of the interactions of hydrophobic pollutants with soils and sediments, desorption is often found to be slower than adsorption, biphasic (rate decreasing with time), and incomplete (Isaacson and Frink, 1984; Brusseau et al., 1991; Pavlostathis and Mathavan, 1992). Slow desorption would be expected to decrease mobility and biodegradation of aromatic hydrocarbons in sediments and their biodegradation. Also, partition coefficients sometimes decrease with decreasing particle concentration, an effect which may be due to the binding of hydrocarbons by dissolved or colloidal components of experimental systems (Voice and Weber, 1985).

Studies of the adsorption of crude oil by several types of Kasitsna Bay (Cook Inlet, Alaska) sediments were reported by Payne et al. (1984) and Payne et al. (1989). Their goals and experimental methods were vastly different from those in this study. The Payne et al. studies were designed to determine the effects of suspended particulate matter on the removal of oil from seawater, in situations where the amount of oil in the system was fairly large compared to the amounts of water and sediment, and oil was mostly contained in a separate phase (surface slick). Payne et al. (1984) also investigated weathering and biodegradation of oil in severely oiled intertidal sites (1 liter oil/m<sup>2</sup>), where most of the petroleum was present in a separate phase or coating throughout the study. However, they did not address the effects of adsorption on the fate of petroleum in slightly- to moderately-contaminated sediments, which is defined in this study as those containing  $\leq 100 \,\mu$ g petroleum hydrocarbons per gram of sediment.

## **Adsorption Effects on Bioavailability**

Although there have been many previous studies of the microbial decomposition of hydrocarbons in the environment (reviewed by Leahy and Colwell, 1990), "the influence of sorption on the biodegradation of NOCs [nonpolar organic compounds] has been the subject of much speculation but little definitive experimentation. Sorption is generally thought to limit the availability of organic compounds, although the few published studies in this area ... arrived at alternate conclusions regarding whether or not sorbed compounds are available to microbial degraders" (Guerin and Boyd, 1992).

Several variables have been identified as affecting adsorption or its influence on decomposition rate or extent. Miheicic and Luthy (1991) found that naphthalene degradation rates were decreased by adsorption, and Manilal and Alexander (1991) found that adsorption slowed phenanthrene decomposition, but Fu and Alexander (1992) found that styrene adsorbed to soils decomposed rapidly. Guerin and Boyd (1992) found that two bacterial species had very different abilities to degrade naphthalene adsorbed to soils. Weisenfels et al. (1992) concluded that biodegradability and biotoxicity of PAH were decreased by adsorption and migration into an organic matter matrix. Adsorption effects on decomposition are difficult to quantify, since the chemical properties of the sorbate, sorbent, and aqueous solution, the mechanism of sorption, the time allowed for equilibration, and the characteristics of the organisms present all can influence biodegradation (Guerin and Boyd, 1992).

There is also no definitive answer to the question of whether adsorption to sediments affects hydrocarbon availability to higher benthic organisms. Sediments are apparently a major source of PAH and other hydrophobic pollutants to benthic invertebrates (McElroy et al., 1989). The evidence includes correlations between the concentrations and compositions of PAH in the organisms and their associated sediments. Also, it has been shown that feeding strategy affects hydrocarbon content and composition of invertebrates from a single site, indicating that a likely route of contamination is the digestive tract.

#### Summary

Aromatic hydrocarbons are strongly adsorbed by marine sediments, and adsorption is not rapidly, completely reversible, indicating that this process probably affects PAH retention by contaminated sediments. Organic matter appears to be the main adsorbent, but clay minerals could have a significant role in organicpoor sediments. The effects of adsorption on aromatic hydrocarbon biodegradation are not well understood and probably vary with the compound properties, bacterial community, and sediment composition. Results of most experiments indicate that adsorption decreases bioavailability and may contribute to the persistence of aromatic hydrocarbons in sediments.

# Methods

Sediments used in this study were collected in July 1994 from the northeastern shore, near the mouth of Jakolof Bay, which is located along the southwestern margin of Kachemak Bay in Lower Cook Inlet. These sediments were also used in studies of microbial degradation of phenanthrene reported by Joan Braddock. Sediments were collected from the oxic, upper 1 cm layer. The sediments primarily consisted of silt. The samples were stored under refrigeration until used in the experiments.

Adsorption was measured using  $9^{-14}$ C-phenanthrene (5–15 mCi/mmole) obtained from Sigma Chemical Company. The solutions were prepared with approximately 0.002 µCurie/mL (or 0.1 nmole/mL) of radiolabeled phenanthrene plus sufficient nonradioactive phenanthrene (also purchased from Sigma)

35 ——

to obtain the desired concentration. The radiolabeled compound purity was initially checked by thin-layer chromatography, which revealed no indications of impurities. However, recent analyses by a higher-resolution high pressure liquid chromatography (HPLC) method indicate that approximately 25% of the radiolabel, as received from the supplier, is not phenanthrene. Based on its chromatographic properties, the contaminant is probably another aromatic hydrocarbon. The data for this contaminant will be corrected once all HPLC analyses are complete. The values reported here are not corrected; however, because the contaminant and phenanthrene appear to have similar properties, a large change in the final results is not anticipated.

Formaldehyde (2%) was found to be effective in preventing respiration (as measured by evolution of  $^{14}CO_2$ ) of phenanthrene in non-sterile seawater solutions and sediments. It was used to prevent biological uptake of phenanthrene in all of the experiments described here.

Adsorption experiments involved suspending 0.05 g of wet sediment in 5 mL of phenanthrene solution in filtered seawater. The seawater was collected from 70 m depth in Resurrection Bay, Seward, Alaska. This water was used because it is more uniform in its properties than Kasitsna Bay water. Reaction times varied from 15 minutes to three days. Sediment was removed by centrifugation and filtration through a Whatman GF/F filter, then the activity remaining in solution was measured by liquid scintillation counting. For some experiments, adsorbed phenanthrene was then desorbed by suspending the sediment in phenanthrenefree seawater. Some sediments were also extracted with solvents in order to recover adsorbed phenanthrene.

# **Results and Discussion**

## Adsorption of Phenanthrene by Jakolof Bay Sediments

Adsorption coefficients have been measured at a total of eight concentrations, using three different reaction times at four of the concentrations and a single, one-day reaction time at the other four concentrations (Table 1). The adsorption coefficients (>400 mL/g dry sediment at the one-day reaction time) were large at all concentrations tested, showing a strong adsorption of phenanthrene by the sediment particles. As indicated in Table 1, experiments at four of the concentrations used homogenized sediment from a single subsample jar. When this was exhausted, another jar of sediment was homogenized and used for the subsequent four experiments. The adsorption coefficients for the two groups of experiments are significantly different, although they agree within a factor of two. We attribute this difference to variations in sediment properties, although the samples were collected within a few meters of each other.

\_ 36

	5	Sediment Subsample	Number 1	
Conc.ª (µg/L)	Reaction Time (days) (mL/g) <sup>b</sup>	Partition Coefficient Seawater <sup>b</sup>	% of Adsorbed Extracted by by Methanol <sup>b</sup>	% of Adsorbed Extracted
10	0.083	350 (10)	21 (0.9)	72 (3)
	1	460 (30)	22 (0.9)	71 (4)
	3	600 (60)	16 (0.7)	71 (4)
100	0.083	275 (20)	27 (0.2)	56 (2)
	1	510 (20)	20 (0.3)	57 (0.9)
	3	600 (40)	16 (2)	48 (4)
450	0.083	260 (9)	26 (2)	54 (0.7)
	1	500 (10)	21 (0.9)	56 (3)
	3	600 (20)	18 (1)	46 (1)
750	0.083	350 (20)	21 (6)	50 (2)
	1	440 (40)	20 (3)	55 (4)
	3	600 (70)	17 (2)	50 (5)
	S	Sediment Subsample	Number 2	
Conc.ª (µg/L)	Reaction Time (days) (mL/g) <sup>b</sup>	Partition Coefficient Seawater <sup>b</sup>	% of Adsorbed Extracted by by Methanol <sup>b</sup>	% of Adsorbed Extracted
44	1	850 (130)	11 (1)	44 (0.4)
200	1	1200 (440)	13 (1)	43 (2)
300	1	680 (140)	10 (2)	42 (5)
600	1	720 (170)	16 (1)	48 (3)

Table 1. Adsorption of phenanthrene by Jakolof Bay sediments.

\*Phenanthrene concentration in the seawater solution before adsorption.

<sup>b</sup>Values shown are the mean of three replicates, except that one replicate was lost (750  $\mu$ g/L, 0.083 day) and one replicate was disregarded (300  $\mu$ g/L, 1 day) because it differed by more than three standard deviations from the mean of the other two. Standard deviation is given in parentheses.

The strong adsorption of phenanthrene indicates that in these intertidal sediments, nearly all of any contaminant phenanthrene should be adsorbed, provided that petroleum contamination levels are not so high that substantial oil exists as a separate phase. For example, for a typical sediment consisting of about 70% water and 30% particles by weight, the adsorbed inventory would be 300 times the dissolved inventory. This, in turn, would mean that loss by desorption and diffusion to overlying water (even for reversibly-sorbed phenanthrene) would be slow. For example, if the upper 2 cm of a sediment were contaminated with 1  $\mu$ g/g of phenanthrene, it would require about three years for this contaminant to be desorbed and lost to overlying water, if no biodegradation occurred. The strong adsorption may also contribute to the processes leading to contamination of sediments. The research described here most directly addresses the situation where a thin (less than 1 mm thick) surface slick or other floating oil is grounded on the intertidal zone by a high tide, wind, and/or waves. If the oil is fairly "fresh," it will leach aromatic hydrocarbons into any overlying water and porewaters of underlying sediments. The strong adsorption of aromatic hydrocarbons by sediments will result in their contamination. Depending on how rapidly the adsorbed hydrocarbons are biodegraded (the subject of the research collaboration with Braddock), the adsorbed hydrocarbons may persist even after the floating oil is removed by physical processes, degraded photochemically, etc. In contaminated waters, particles would also acquire substantial concentrations of phenanthrene by adsorption, and this could affect concentrations in bottom or intertidal sediments. For example, if water phenanthrene concentrations were 1 to 6 µg/L, which was the concentration range for PAH observed after the Exxon Valdez spill (Wolfe et al., 1995), predicted sediment concentrations are 0.6-4 µg/g, well in excess of "baseline" concentrations in marine sediments (Wakeham and Farrington, 1980).

There is no consistent pattern of change in the phenanthrene adsorption coefficient with concentration, i.e., adsorption is a linear function of dissolved concentration. This pattern indicates that all adsorption sites occupied are equivalent; that is, none tended to bind phenanthrene more strongly than others. There is also no evidence of saturation of adsorption sites over the concentration range tested, which was limited by the solubility of phenanthrene in seawater.

Much of the adsorption occurred rapidly, within two hours (Table 1). In fact, an earlier set of experiments (data not shown) found the same adsorption at reaction times of 15 minutes and two hours. However, adsorption after one day was approximately 50% greater than that at two hours. Adsorption after three days was not significantly greater than that at one day in one experiment (data not shown), but was slightly greater in a second experiment (Table 1). Similar results for adsorption of hydrophobic organic chemicals by soils and sediments have been interpreted as due to rapid adsorption to sediment surfaces, followed by slower diffusion into sediment organic matter (Brusseau et al., 1991)

Three different sediment:solution ratios for the reaction mixtures were tested at each concentration (Figure 1). Although the mean partition coefficient was consistently lower for the largest sediment:solution ratio, there was no statistically significant difference in the partition coefficients. If partition coefficients are invariant with sediment:solution ratio, it is less likely that adsorption to colloids or complexation to dissolved organic matter are having a large effect on the behavior of phenanthrene.



Figure 1. Effect of sediment concentration on phenanthrene adsorption.

#### **Reversibility of Adsorption**

Experiments measured the amount of adsorbed phenanthrene that could be recovered by repeated 15-minute phenanthrene-free seawater extractions, essentially the reversibility of phenanthrene adsorption (Figure 2). The reversibility of phenanthrene adsorption was the same at the two concentrations tested, 10 and 750 µg/L, so the data are pooled in Figure 2. A maximum of about 45% of adsorbed phenanthrene was desorbed by three seawater rinses. Approximately 80% recovery would be expected if the adsorption were entirely reversible, i.e., the apparent partition coefficient for desorption was the same as that for adsorption. Recovery of adsorbed phenanthrene decreased for longer adsorption reaction times. Another experiment found that 10-20% of adsorbed phenanthrene could be desorbed by a single extraction with seawater for reaction times of two hours to three days, similar to the amount recovered in only 15 minutes (Table 1). The reversibility of phenanthrene adsorption decreased with increasing reaction time. The low amounts of phenanthrene recovered by seawater could be explained if both very strong and weaker adsorption sites were present on the particle surfaces. However, this would also be expected to result in increased adsorption with decreasing concentration of phenanthrene in solution, which was not observed (Table 1). The results are better explained by slow diffusion of phenanthrene into sediment organic matter (Brusseau et al., 1991); the reversal of that process would also be slow. The slow desorption means that contaminated sediments could be expected to retain substantial phenanthrene even if subsequently covered with or resuspended in clean seawater, at least on a time scale of days. In future work, longer times will be investigated.



Figure 2. Reversibility of phenanthrene adsorption by extraction of sediment with phenanthrene-free seawater. The reaction time given is for the adsorption of phenanthrene to the sediment. Each sediment sample was extracted three times. The first bar shows the recovery expected if the phenanthrene adsorption were completely reversible.

## **Extractability of Adsorbed Phenanthrene**

Most of the adsorbed phenanthrene remaining after a seawater rinse could be extracted with methanol (Table 1). Total recovery of adsorbed phenanthrene was nearly 100% at a low concentration, but approximately 70 to 80% at a higher concentration for the first set of experiments using one, homogenized sediment subsample. Total recoveries were much lower, 50 to 60% for the series of experiments done with the second sediment subsample. Acetone, acetonitrile, chloroform, 50:50 chloroform:methanol, methylene chloride, and toluene were also checked for their ability to extract adsorbed phenanthrene; none of these was more effective than methanol. All extractions were carried out over a relatively short time (0.5 hour). Incomplete solvent extraction could also be related to intraorganic matter diffusion; therefore longer extraction times will be investigated. One of the major objectives in investigating extractability of adsorbed hydrocarbons is to develop a method of recovering (and thus quantifying) adsorbed phenanthrene in biodegradation studies being conducted with Braddock.

## **Biodegradation of Adsorbed Phenanthrene**

As described in her report, Braddock has found that phenanthrene-degrading bacteria are present in the Jakolof Bay sediments studied. Because she began her work several months after this study began, investigation of adsorption effects on biodegradation are still in progress. However, the strong adsorption of phenanthrene by Jakolof Bay sediments indicates that, if adsorption affects microbial decomposition rates, this process will have a significant effect on the biodegradation of this aromatic hydrocarbon.

# **Preliminary Conclusions**

Phenanthrene is strongly adsorbed by Jakolof Bay intertidal sediments, and much of the adsorption is rapid, occurring within minutes. However, adsorption is not rapidly reversible, requiring more than several days in phenanthrene-free water. Of the adsorbed phenanthrene which is not desorbed in seawater, 50 to 90% can be extracted with methanol. Adsorption increases about 50% for reaction times of one to three days and becomes less reversible. Phenanthrene adsorption is approximately linear with concentration, indicating that all surface adsorption sites are approximately equivalent, but the adsorption coefficient varies by a factor of about two among sediment subsamples collected in a small area. Taken together, the characteristics of phenanthrene adsorption by the Jakolof Bay sediment are consistent with a process of rapid adsorption to the particle surfaces followed by slow diffusion into sediment organic matter. The strong adsorption indicates that this process could be an important factor in the acquisition and persistence of phenanthrene contamination in these sediments. The findings most directly address the situation where a thin surface slick or other floating oil is grounded on the intertidal zone and leaches aromatic hydrocarbons into the porewaters of underlying sediments. The strong adsorption of aromatic hydrocarbons by sediments will then result in their contamination.

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**4**I \_

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43 -

# Kachemak Bay Experimental and Monitoring Studies

# Raymond C. Highsmith Susan M. Saupe Tama L. Rucker

Institute of Marine Science University of Alaska Fairbanks Fairbanks, AK 99775-7220

# Abstract

Several differing intertidal research efforts occurred along the oil impacted shorelines following the Exxon Valdez oil spill. The lack of pre-spill baseline data made these sampling efforts both time consuming and cost intensive. The CMI-funded studies in lower Kachemak Bay were designed to obtain basic data on abundances and distributions of intertidal invertebrates and algae and to gather information throughout the year on community structuring mechanisms, such as recruitment and succession. The objectives were to gain an understanding of the relationships between and among intertidal invertebrates and plants, and some of the physical and environmental factors which drive these systems on exposed and sheltered rocky shorelines. In addition, it has been possible to compare several data collection methods commonly used during environmental impact studies of intertidal populations. Preliminary comparisons show significant differences between collection methods for many common invertebrates.

# Introduction

Marine intertidal communities were the largest habitat category affected by the *Exxon Valdez* oil spill, with damage from the oil and resultant cleanup activities occurring throughout the oil impacted area. The Coastal Habitat Injury Assessment (CHIA) study was initiated in 1989 to assess the injury to biological resources in intertidal habitats on oiled shorelines. That study encompassed the three major geographic areas impacted by the spill: Prince William Sound, Cook Inlet–Kenai Peninsula, and Kodiak–Alaska Peninsula. Due to the lack of any baseline data, oiled sites were randomly selected and matched with non-oiled reference sites for an After Control-Impact Pair study design. There are several drawbacks to this design. The number of site pairs must be high enough and randomly selected in order to extrapolate study results to the rest of the oiled shorelines. For instance, the currents which carried the oil to certain shorelines may carry recruits and food to these same shorelines in greater abundance than to non-oiled shorelines, or areas which are "shadowed" by other areas of the coast-

line. Shorelines near regions of active oil exploration and transport, such as Lower Cook Inlet, have the potential to be further impacted by oil and recovery activities in the event of another oil spill.

Several interesting results were revealed during the CHIA study, although no firm conclusions could be made as to their causes or long-term stability. The barnacle Chthamalus dalli was found in higher densities on oiled sites compared with control sites, especially in the mid- to lower-intertidal zones (Highsmith et al., 1994). In undisturbed systems, C. dalli tends to be restricted to the high intertidal, due to competitive exclusion from lower levels by the superior space competitors, Balanus glandula and Semibalanus balanoides (Connell, 1961, Wethy, 1985). C. dalli was the first species of barnacle to recruit into the free space created by the oil spill and cleanup activities. Settlement of C. dalli spat coincided with the availability of space resulting from the cleanup during the summer of 1989, giving this species a six-month advantage over the other prominent intertidal barnacles. Thus, the timing of the disturbance favored initial recruitment of one species over others. Mussel size and age data for sites sampled during the CHIA study (Highsmith et al., 1992) indicated that mussels of a given age tended to be larger on oiled sites relative to control sites. It was speculated that there may be inherent differences between oiled and control sites, and that the currents which delivered oil to certain shorelines may also concentrate recruits and fresh particulates in the intertidal zone in these areas.

The two main studies initiated for this CMI study in outer Kachemak Bay were designed to obtain "pre-spill" data on invertebrate and algal percent cover, abundance, recruitment, succession, and interactions. The major focus of the study has been to look specifically at recruitment and successional patterns in comparison with several physical factors, such as relative water motion and wave exposure. In addition, rates of recruitment and successional patterns are being studied on shorelines where disturbances (removal of all invertebrates and algae) occurred at different times throughout the year. These disturbances (cleared areas) were created at three different tidal elevations on each site, with sites chosen based on varying wave and current exposure. The clearings occurred in March, July, and October of 1994, and in March of 1995. They will be monitored over time to determine the recruitment of invertebrates and algae and to obtain data on the natural variabilities within undisturbed (non-cleared) communities. Information on intertidal communities for much of the Alaskan shoreline is sparse. The data collected in the CMI study will allow for a better understanding of the biological and physical processes that occur within intertidal communities and the potential effects of the nearby oil industry.

Data collections for the recruitment and succession study also allowed us to compare several methods commonly used for intertidal monitoring (Hawkins and Hartnoll, 1983; Foster et al., 1991; Hiscock, 1985), some of which were used for the *Exxon Valdez* oil spill studies (Highsmith et al., 1994; Gilfillan et al., 1993). These

45 ---

include point-contact methods for invertebrate and algal percent cover data collections, direct counts of invertebrates in the field, and collection of invertebrates with subsequent sorting and counting in the laboratory. In addition, for several organisms, two methods of counting were used in the field: total counts within a quadrat frame and random subsamples within the quadrat frame. The initial analyses of these data indicate that the accuracy of some methods is low, with many species being undercounted or completely missed in the field. The results of these method comparisons will be extremely useful for future development of damage assessment studies and intertidal community monitoring.

# Methods, Results, and Discussion

Two major studies have been conducted since March 1994. The first, a recruitment/succession study, was conducted on eight sites. The sites were chosen to represent a gradient of wave exposures in order to improve our understanding of the relationship between recruitment and post-recruitment events to wave and current exposure. Dissolution rates of calcium sulfate were measured on each site to determine relative water motion (Muus, 1968, Gerard, 1982, Petticrew and Kalff, 1991). This was accomplished by deploying molded cylinders of gypsum at the 2.5 meter of vertical drop (MVD) from the mean high high water (MHHW) line on each site for three days. The amount of dissolved calcium sulfate was determined by the weight difference before and after deployment. The sites used for these studies were subjectively selected to represent a range of wave and storm exposure on rocky shorelines. Dissolution measurements are also sensitive to water motion independent of wave action, such as tidal currents. When the sites are ordered in a graph by subjective rankings of wave exposure, based on fetch, aspect, slope and grain size, the dissolution rates of the molded calcium sulfate cylinders generally follow a trend of higher dissolution on more highly exposed sites (Figure 1). However, several sites have higher relative dissolution rates than expected based on wave exposure. At these sites, such as Jakolof Bay, there is usually a strong current due to tidal exchange through a constricted opening.

On the sites selected for the recruitment/succession study, a "disturbance" was created by removing all organisms from quadrats at each of three MVDs along four transects on each site. A set of clearings was made on each site in March, July, and October 1994 and in March 1995. During subsequent visits to each set of clearings, percent cover estimates and direct counts were made of invertebrates and algae that had recruited into each cleared area. In addition, similar data were collected from permanently established control quadrats in order to obtain data on inter- and intra-annual variability.

Data were obtained from the permanent control quadrats during the initial visit. The following invertebrates were counted from within each quadrat: the limpets *Lottia pelta, Tectura persona, T. scutum, L. borealis, L. digitalis,* and unidentifiable limpets (juveniles); the barnacles *Chthamalus dalli, Semibalanus balanoides* and

**AUGUST 1994** 



Figure 1. Mean dissolution (mg) of calcium sulfate cylinders deployed at the 2.5 MVD on recruitment/succession study sites over a three day period in August 1994. Sites are ordered from left to right in order of decreasing wave exposure. Four cylinders were deployed on each site. Vertical bars are one standard error.

Balanus glandula (combined), Semibalanus cariosus, and unidentifiable barnacles (juveniles); the littorines Littorina sitkana and L. scutulata; the mussel Mytilus trossulus; the dog whelks Nucella lima and N. lamellosa; and the chiton Katharina tunicata. A 40 point grid was used to obtain percent cover estimates. What occurred immediately under each grid intersection was recorded, including algae, bare rock, or any invertebrates. During July, invertebrate count and percent cover data were collected from both the control quadrats and the quadrats scraped in March. During a low-tide series in October, direct counts were made of all invertebrates except barnacles on the March scrapes and of all invertebrates including barnacles for the July scrapes. In addition, percent cover data were collected for all organisms from the controls, and the March and July scrapes.

The results of percent cover estimates of recruitment into scraped quadrats are summarized in Figures 2 and 3. One figure represents data from the same quadrats censused twice and the other figure presents data from two separate sets of quadrats each censused three months after initial clearing of the quadrats.

47 -----



Figure 2. The percent cover of recruited algae and invertebrates into quadrats that were scraped in March 1994. Recruitment was censused in July and October 1994. MVD = meter vertical drop.

Figure 2 shows data from quadrats scraped in March 1994 and censused in July (columns labeled "Jul") and censused again in October (columns labelled "Oct"). Initial analyses show high numbers of barnacle recruits (mainly *Semibalanus balanoides* and *Balanus glandula* at MVDs 1.5 and 2.5 and *Semibalanus cariosus* at MVD 3.5) on almost all sites following scraping in March 1994. Some die-off of barnacles had occurred between July and October, although total percent cover was still >50% on most sites. Barnacles accounted for almost 100% of initial recruitment in the high- and mid-intertidal zones, especially at MVD 1.5. At lower MVDs, "other" organisms accounted for a larger percent of total cover. The "other" species included littorine snails, small limpets, and species of filamentous algae, such as *Pilayella, Acrosiphonia*, and *Cladophora*. These algal species were also initial recruits at sites damaged during the *Exxon Valdez* oil spill (Highsmith et

. 48



Figure 3. The percent cover of recruited algae and invertebrates into quadrats that were scraped in March and July 1994. The histograms labelled "Mar" represent recruitment data obtained three months after scraping the quadrats in March. The quadrats labelled "Jul" represent recruitment data obtained three months after scraping the quadrats in July. MVD = meter vertical drop.

al, 1994). For quadrats scraped in July 1994 and censused three months later in October, recruitment was very low (columns labelled "Jul" in Figure 3). The column labelled "Mar" shows the data discussed earlier for quadrats scraped in March and censused in July, for direct comparison of recruitment after disturbances at different times of the year. The recruitment data in Figure 3 represents three months of recruitment time for each set of scrapes. Clearly, there is a large seasonal component in post-disturbance recruitment. The first field season data collections from this study show weak or no correlation of initial recruitment into

49 ---

cleared quadrats (Figures 2 and 3) with gypsum cylinder dissolution rates measured at each site (Figure 1). However, there are large differences in recruitment into cleared quadrats depending upon when the clearings were made. With continued monitoring in 1995 and 1996, the effects of wave exposure, current regime, and seasonal timing of a disturbance on long-term recruitment, survival, and succession can be more thoroughly evaluated.

Water currents are considered critical to the transport of recruited mussels from filamentous algae into adult mussel beds (Verwey, 1952). Eyster and Pechenik (1987) reported in a laboratory study that water agitation greatly enhanced larval attachment to filamentous substrata. Therefore, water motion appears to be an essential factor in both primary and secondary phases of recruitment for young mussels. A study was established in lower Kachemak Bay to compare the sizefrequency distributions of mussel populations during the spring and fall on eight sites with varying wave and current exposure. Along each of four randomly selected transects, a quadrat was placed at the 1.8 and 2.3 MVDs for mussel collections. The quadrats were 10x20 cm and usually enclosed several hundred to several thousand mussels. All the mussels within each quadrat were collected and bagged, and then frozen and returned to the laboratory in Fairbanks for analyses. In the laboratory, the mussels were thawed and washed over a 0.5 mm sieve. The mussels were measured from the tip of the umbo to the widest portion of the posterior shell. Using dial calipers, measurements were taken to the nearest 0.05 mm. Collections were made in March and October 1994. The laboratory analyses are completed for the March collection. The data from the October 1994 collections are currently being entered and analyzed.

Calcium sulfate dissolution rates for the eight mussel study sites generally agree with the wave exposure rankings (Figure 4). Mussel data collected from the sites were plotted on cumulative frequency curves to illustrate differences in population size-frequency distributions between sites (Figure 5). Straight cumulative frequency curves, with a y-intercept near zero, represent populations with evenly distributed mussel sizes. Curves with a steep slope at small sizes and a large yintercept, represent populations with a high percentage of small individuals. Also, curves that approach a frequency of 1.0 at small size categories represent populations with very few large mussels. For the cumulative frequency curves in Figure 5, the order of the symbols in the legend, from top to bottom, represent the sites ordered from left to right in Figure 4. The size-frequency curves for the three sites with the highest percentage of large mussels (Little Jakolof, Cronin I, and Little Tutka) are the sites with the lowest wave exposure rankings. Two of the three sites also have the lowest gypsum dissolution rates, or relative water motion. Perhaps at the sites with higher wave exposures, mussels are exposed to forces sufficient to rip byssus threads from the substrate. Large mussels have larger surface areas and protrude from the substrate more than small mussels, thus increasing drag forces. Therefore, sites with higher wave exposure may have a smaller percentage of large mussels. Mussel larvae generally recruit onto filamentous algae (Bayne, 1964; Seed, 1976). However, other investigators have observed small, <1.0 mm mussels within the byssus mat of adult mussel beds (Petersen, 1984; McGrath et al., 1988). The high numbers of very small mussels (<1.0 mm) on many sites suggest that larvae may have recruited directly into the adult beds. In addition, the presence of the <1.0 mm mussels in April indicate that a settlement occurred at some point in the winter of 1993/1994. Mussels may undergo more than one annual spawning event when environmental conditions permit (Seed, 1976; Lower et al., 1982), although in Alaskan waters the major settlement pulse occurs in late July to early August (Ralonde, pers. comm.). The samples collected in fall 1994 will allow us to determine if there are differences in mussel recruitment among the sites due to wave exposure or water current rates.

During the recruitment/succession study described previously, four sites were also "double-sampled." Before destructive sampling on the sites, we had an opportunity to take individual counts and percent cover data of specific invertebrates from those quadrats that were to be scraped and burned for monitoring of future recruitment and succession. During the scraping procedures, all invertebrates within each quadrat were collected and preserved. In the laboratory they were rinsed over a 1.0 mm sieve, sorted to the lowest possible taxonomic level, and counted. The personnel conducting the field and laboratory counts have been identifying Alaskan intertidal invertebrates since the oil spill in 1989 and are



Figure 4. Mean dissolution (mg) of calcium sulfate cylinders deployed at the 2.0 MVD on mussel study sites over a three-day period in July 1994. Sites are ordered from left to right in order of decreasing wave exposure. Four cylinders were deployed on each site. Vertical bars are one standard error.

51 -



Figure 5. Cumulative frequency curves for mussel populations on the eight mussel study sites. The curves illustrate the contribution of each successive size class to the population. The order of the symbols in the legend, from top to bottom, correlate to the sites ordered from left to right in Figure 4. MB1 = Hesketh I, MB2 = Hesketh II, MB3 = Yukon Isl., MB4 = Cronin I, MB5 = Little Jakolof, MB6 = Herring I, MB7 = Little Tutka, MB8 = Nubble Point.

highly experienced taxonomists. The samples are currently being sorted in the laboratory and the results are not complete. However, for those quadrats that have been analyzed, the field counts underestimate the number of organisms actually present. For example, the blue mussel Mytilus trossulus was undercounted by up to a factor of four. Mussels often grow in layers. Small mussels completely hidden by larger mussels above are missed during field counts. Other organisms, such as small limpets, hide inside large barnacle tests, Semibalanus cariosis, for example, and are undetectable during field counts. Barnacles often grow in "hummocks" where many barnacles are crowded together and it is difficult or impossible to identify individual barnacles. With the additional opportunistic field collections mentioned above, it will be easier to interpret data collected by either method and judge the benefits and drawbacks of each method. For instance, the nondestructive method of counting organisms directly on-site is much more cost effective as no additional laboratory procedures are necessary. However, abundances of many organisms may be underestimated in the field. Percent cover estimates miss many small or mobile species and the method is best for counting primary space competitors such as barnacles, mussels, and algae. Possibly, the ideal method for monitoring intertidal communities is a combination of all three methods.

# **Preliminary Conclusions**

The preliminary data shows that water motion may have an effect on recruitment and succession of intertidal invertebrates, especially for the blue mussel, *Mytilus trossulus*. Initial recruitment of barnacles was high on all sites after the quadrats were scraped in spring of 1994. However, after the quadrats were scraped in July, recruitment by fall was very low, often with zero percent cover in the quadrats. If early algal recolonization occurs in spring 1995, the bare space might not be available for barnacle recruitment to the extent seen for those quadrats scraped in spring 1994, possibly resulting in differing successional patterns. If the quadrats scraped in July remain bare, barnacles may be the main initial recolonizers in spring and summer. This would, in effect, have delayed the "recovery" of the cleared quadrats by nine months compared to the quadrats cleared in March. However, successional patterns may be similar in the long term.

The data collected during the "double-sampling" study strongly suggests that the method used for invertebrate monitoring may affect the results obtained. Field counts underestimate the abundance of many invertebrates, percent cover estimates using the point-contact method miss many small and mobile species, and laboratory counts, though more accurate, are time consuming and costly. The data collected during this CMI-funded study will be invaluable for judging the benefits and drawbacks of each method when designing future monitoring studies, both pre- and post-disturbance.

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53 —

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54

# **North Slope Amphidromy Assessment**

#### **Thomas C. Kline**

Prince William Sound Science Center Cordova, AK 99574

#### John J. Goering

Institute of Marine Science University of Alaska Fairbanks Fairbanks, AK 99775-7220

# Abstract

Natural stable isotope abundances measured in tissues of Alaska North Slope coregonine fishes reflect differences in feeding habitat relative to size and age. This methodology is being developed to use as a tool for proxy analysis of fish behavior in future North Slope environmental monitoring studies. Development of the methodology consists of sampling arctic and least ciscos, broad whitefish, Dolly Varden, and representative noncommercial fish and forage species through collaboration with existing sampling programs being conducted by the North Slope Borough Department of Wildlife Management and BP Exploration through contracts to LGL Alaska Research Associates, Inc. Intensive sampling at a limited number of sites is enabling an analysis of population structure using natural stable isotope abundance. Initial results of the first field season confirm that stable isotope data reflect amphidromous feeding modes.

# Introduction

## **The Problem**

Fishes of the genus Coregonus numerically dominate coastal and freshwaters of the Alaskan arctic coastline. In addition to being obligate freshwater spawners, they undergo feeding migrations into coastal waters during the summer. The migration passages used by these fishes have been subjected to physical alterations as a result of the industrial development necessary for oil exploration and extraction. Considerable effort has been made to ascertain whether structures such as causeways located in migration routes have had a detrimental effect on these species. Thus far, no detrimental effects have been detected using conventional research tools. However, subtle effects on migrational behavior with longterm consequences may not be detectable with the technologies currently being employed. An example of a potential effect of shifts in migrational behavior would be an alteration in life history feeding strategy. Evidence that different life history strategies exist on the North Slope is found in Teshekpuk Lake, where a portion of the least cisco and broad whitefish populations are thought to be nonmigratory because they grow at a slower rate compared to amphidromous populations (George et al., 1996). A potential consequence of industrial development

55 \_\_\_\_\_

could be increased dwarfism if migration is restricted. Fecundity loss leading to population reduction is another possible consequence if spawning ground access is made more difficult. New techniques that assess subtle changes in the migrational behaviors of these fishes could allay these fears.

#### The Approach

Large gradients in the natural abundance of  ${}^{13}C/{}^{12}C$  and  ${}^{15}N/{}^{14}N$  exist between freshwater and marine habitats on the Alaskan arctic coastline. This is due to the existence of massive terrestrial peat deposits that provide an important carbon source (directly via peat-consuming insects and indirectly via respiration of peat carbon) to food chains existing there and the distinctive isotopic signatures of the peat deposits. Because stable isotope ratios are conserved during feeding processes, they make effective tracers of food sources enabling researchers to distinguish between peat and marine carbon. An ability to detect food-source signatures in fishes also enables the researcher to ascertain whether migration has taken place by determining where the sample was collected in relation to potential feeding locations. Evidence that isotopic signatures differ depending on amphidromous behavior and consequently carbon source was found in a pilot isotope study in 1993 conducted by Kline in collaboration with the North Slope Borough Department of Wildlife Management (DWM). The purpose of this project, North Slope Amphidromy Assessment, is to investigate the use of isotope measurements as a proxy for amphidromous behavior. The goal is to determine the suitability of measuring the <sup>13</sup>C/<sup>12</sup>C and <sup>15</sup>N/<sup>14</sup>N ratios of a fish to quantify its migrational movements between freshwater and marine habitats. The project sampling design was made to achieve this goal.

## **Project Collaboration**

This project complements other research being conducted on the North Slope by the DWM and LGL Alaska Research Associates, Inc. for BP Exploration (Alaska) Inc. (BPX). Sampling was designed to complement these studies so that they can provide the basis for the Coastal Marine Institute (CMI) match requirements. Also by sampling alongside existing sampling, fish loss is minimized. That is, fish that are killed for another purpose are also being used for stable isotope measurement. This has helped to justify the required Alaska Department of Fish and Game collection permit.

## **Background and Relevance to Framework Issues**

The DWM has been undertaking research on harvested and potentially harvestable fish populations that occur concomitantly with oil on the Alaska North Slope. This research has involved investigators outside of DWM, as well as their own personnel. Kline is currently collaborating with the DWM in a multi-disciplinary research effort on coregonine fishes of Teshekpuk Lake (the largest lake on the Alaskan arctic coastal plain and third largest lake in Alaska) and the Chipp River. The latter, as well as other watersheds between Barrow and

the Colville River, have recently been discovered to be important habitats for the recruitment of Bering and arctic cisco (Benny Gallaway, LGL Ecological Research Associates, Inc., Bryan, TX, pers. comm.). Kline's part in this investigation is to determine whether differences in feeding habits among the different species and species forms can be evidenced with stable carbon (C) and nitrogen (N) isotopic analyses. Preliminary data suggested that the dwarf form of the least cisco deviates from the isotopic composition of normal and intermediate forms. Two carbon isotope ratio clusters suggest there are at least two separate food chains, whereas the shift in nitrogen isotope ratio suggests a one-step trophic level shift or a different nitrogen source. Although Teshekpuk Lake broad whitefish stable isotope data with respect to form were not as clear cut, there was a significant difference in the carbon isotope distribution. These results corroborate other stable isotope fish studies conducted in this region that suggest significant isotope ratio gradients. Through exploitation of these gradients, we will gain a better understanding of fish ecology.

This project will continue in collaboration with the DWM (Craig George et al.) and will expand the utilization of newly developed stable isotope abundance methodology at sites being monitored by BPX and ARCO Alaska (ARCO).

This project embraces item #1 of the CMI Framework Issues. The results from the Teshekpuk study were reported in a seminar sponsored by the Institute of Marine Science at the University of Alaska Fairbanks. This work which is currently in the from of a draft technical report submitted to the DWM is being reduced to a paper for submission to a peer-reviewed scientific journal.

## **Potential Benefits in Relation to Mineral Exploitations**

Monitoring the subsistence and commercially harvested fish species (i.e., arctic and least ciscos, and broad whitefish) has been an ongoing process because of potential impacts resulting from oil development on the Alaska North Slope. Much of the focus has been on the impact of causeways altering the migratory behavior of these species (Gallaway et al., 1991). These species are classified as amphidromous because they migrate between the freshwater and marine environments for nonreproductive activities, and anadromous because they must reproduce in freshwater. Maintenance of these populations depends on their continued ability to move back and forth between freshwater and marine environments. The migratory pathways are complex because of the low topography and thermokarst features of the Arctic Coastal Plain and the intricate and tortuous pathways among lakes, streams, lagoons, and open water as well as the varied tolerance to salinity by the various fish species. The fish monitoring program has consisted of extensive monitoring stations in the developed area (e.g. Prudhoe Bay) and existing fisheries (e.g. Colville River).

Exploration for oil to the west of Prudhoe Bay, including the Ikpikpuk watershed (including Teshekpuk Lake) has commenced (Craig George, DWM, pers.

comm.), suggesting that potential impacts in this coastal area are imminent. Previous exploration and exploitation of North Slope oil has required the construction of roads and causeways that have potential impacts on amphidromous and anadromous fishes such as the least cisco and broad whitefish. A tremendous effort has been made to assess oil development impacts on fishes in the developed area to date. However, many questions remain unresolved, in particular, the effects of constructions on the migratory behavior of fishes. It has been difficult to assess whether the causeways are having a detrimental effect on fish populations using conventional fisheries methods (Gallaway et al., 1991). The purpose of this study is to assess the use of stable isotope data to determine the impact of oil development on the ecology of important subsistence and commercial fish species.

Changes in the observed stable isotope gradients may be used to determine how migratory and consequently feeding behavior shifts in response to alterations of fish habitats. Habitat alteration from activities such as oil exploration and mining, e.g. causeway construction, may alter migratory pathways of anadromous and amphidromous fishes. Thus, population interactions may change. Potential evidence of change by stable isotope analyses may include:

- 1) Forced overlapping of populations that were formerly separate;
- 2) Alteration of the nature of nutritional sources;
- 3) Shifts in the relationship of population characteristics with respect to <sup>13</sup>C and <sup>15</sup>N and habitat use, enabling prediction of potentially sensitive stages to environmental perturbation; and
- 4) Shifts in other population characteristics such as parasitism, genetics and population structure (e.g. length at age and population age distribution) with concomitant isotopic change.

Stable isotope analysis elucidates aspects of North Slope fish populations not available by other methods through:

- 1) Furthering the isotopic characterization of fish populations in order to determine the spatial and temporal patterns of North Slope fish populations;
- 2) Determining the interaction among populations by trophic status and food web characterization; and
- 3) Assessing habitats and food chains important as nurseries for the recruitment of populations.

Analyses in this study follow established isotope chemistry laboratory and data analysis protocols. Methods for synthesis of data have been established (Kline et al., 1989; Kline, 1991). Laboratory work for this project provides student projects where students are directly involved in pre-mass-spectrometry laboratory aspects of the project. This project may also provide data for a possible Ph.D. thesis for C. George. Field studies are being conducted to address CMI Framework Issue #1 to coincide with DWM needs for fish stock assessment. Presently, efforts are

. 58

being concentrated in the western portion of the Ikpikpuk watershed (Chipp River). Similarly, field studies which address Framework Issue #2 are being conducted to coincide with BPX and ARCO needs for fish stock assessment and impacts. The fusion of these studies with collaborating agencies comes in part from their role in matching CMI funding as well as facilitating sample collections within the operational constraints that exist in working in the arctic (e.g., the very short season and high financial costs).

#### **Applications of Amphidromy Assessment**

Stable isotope ratios are being used to characterize where individual fishes have fed by comparing the ratios found in the fishes to ratios found in various natural habitats. By generalizing characteristic isotopic signatures of feeding in fresh- and saltwater habitats, a mixing model can be established (Kline, 1991, Kline et al., 1993) to quantify a "degree of amphidromy" on a per fish sample basis. By analyzing a reasonable sample size (number of fish), it will be possible to assess how a fish population is behaving. If behavior should shift over time, a concomitant isotope shift can be used to quantitatively assess the change. No other methodology currently exists that will provide such data or analysis.

It may be necessary to build roads with culverts and causeways during future oil development in the study area. Such construction may reduce the migratory ability of coregonines (for example) thus restricting a greater proportion of the population to freshwater. Possible consequences of development, such as an increased proportion of dwarfism and reduction of population size, could be the result of a deprivation of coastal feeding grounds if the fish lose the high <sup>13</sup>C marine signature that is now present. Similar shifts could occur within any of the watersheds of the Arctic Coastal Plain. Stable isotope chemistry shifts would thus provide direct evidence of oil development effects on fish populations. The use of the natural stable isotope abundance method enables a predictive capability in assessing oil development impact, thus embracing item #2 of the CMI framework.

# **Results and Discussion**

## **Objective (1)**

• Establish baseline isotope attributes of unaffected populations before any development occurs. During and following development, fish populations can be monitored by looking for alterations in their expected isotope chemistry to determine if they are being excluded from feeding habitats. Stable isotope chemistry is a very powerful tool for use in ecological studies because of naturally existing isotope gradients (Schell and Zieman, 1989; Hesslein et al., 1991) and the fidelity of consumer-stable isotope ratios with the diet (Fry and Sherr, 1984; Owens 1987; Peterson and Howarth, 1987; Wada et al., 1991).

— 59 —

- In 1994, this objective was met by the field study to the Chipp River. In 1995 and beyond, objective (1) will be met by similar field projects in collaboration with the DWM and LGL Alaska Research Associates, Inc.
- Species sampled include least cisco, broad whitefish, humpback whitefish, Dolly Varden and other salmonids, representative other fishes for comparison, including obligate freshwater species, and potential fish forage species.

### **Objective (2)**

- Expand existing isotope studies in the developed area: a) the Sagavanirktok River and causeways; and b) the Colville River. Although behavioral changes may have taken place in the developed area, evidence in the Sagavanirktok (Lisa Stuby, M.S. Candidate of Schell, University of Alaska Fairbanks, pers. comm.) and Colville Rivers (Schell and Zieman, 1989) suggests isotopic shifts similar to those observed in Teshekpuk Lake.
- In 1994, this objective was met through collaboration with BPX and their contractor LGL Alaska Research Associates, Inc. It is expected that this sampling will be continued.
- Species sampled include least cisco, arctic cisco, broad whitefish, Dolly Varden and other salmonids, representative other fishes for comparison, including obligate marine species, and potential fish forage species.

### 1994 Sampling

Sampling in the Chipp River system took place during the last ten days of July 1994. The personnel left by boat from Barrow at ice-out. The field camp was established at subsistence site PK13 where gillnet and fyke net sampling sites were set up. Excursions were made downstream to a fyke net site at a lake confluent with the Chipp River and a gillnet site at the river mouth (Figure 1). Fishes were collected daily and processed at the field camp. Processing included the determination of weight, length, sex, sexual maturity, and stomach content data for each fish. Otoliths were removed for age determination at a later date and tissues were sampled for stable isotope analysis. The stable isotope analyses of these samples are presented here.

## **Verification of Isotopic Gradients**

Large isotopic ranges have been observed in fishes of the North American Arctic (Schell and Zieman, 1989, Hesslein et al., 1991).  $\delta^{13}$ C from Schell and Zieman (1989) are plotted in Figure 2 for comparison with present data. Marine fishes (sculpins and cod) had more positive  $\delta^{13}$ C and freshwater fishes (grayling, longnose sucker, and burbot) had more negative  $\delta^{13}$ C, whereas the amphidromous coregonines were intermediate and had greater  $\delta^{13}$ C ranges consistent with the notion that  $\delta^{13}$ C reflects feeding habitat. To further explain the isotopic gradients, Schell and Zieman (1989) measured <sup>14</sup>C abundance. Broad whitefish were most <sup>14</sup>C-depleted consistent with obtaining peat fossil-carbon. Grayling were less <sup>14</sup>C-enriched, suggesting input of atmospherically-derived



Figure 1. 1994 project sampling sites at Chipp River and subsistence fishing sites.

carbon, most likely from feeding on insects utilizing terrestrial plants. Thus <sup>13</sup>Cdepleted signatures may reflect terrestrial plant or peat carbon in fishes. Although only limited marine fishes were sampled at the Chipp River in 1994 (Figure 3), the more positive  $\delta^{13}$ C of sculpin is consistent with an elevated marine  $\delta^{13}$ C signature. A number of marine fish were sampled at Endicott in 1994 which will be used to better establish the marine  $\delta^{13}$ C value. Other non-coregonine fishes sampled from the Chipp River in 1994 were  $\delta^{13}$ C-depleted relative to sculpins (Figure 3). The rainbow smelt (anadromous) had intermediate values. Other fishes were more  $\delta^{13}$ C-depleted. The most positive of these, the burbot, is slightly  $\delta^{13}$ C-enriched compared to the others because they are piscivorous and  $\delta^{13}$ C

61 ...







Box Plots for Columns X<sub>1</sub>.....X<sub>7</sub>

Figure 3. Box plots of 1994 Chipp River non-coregonine  $\delta^{13}$ C. Abbreviations are the same as Figure 2 plus 9SP = ninespine stickleback, PIKE = northern pike, RBSM = rainbow smelt.

increases by about one delta unit per trophic level feeding process (Fry and Sherr, 1984; Wada et al., 1991). The pike was small and, though a piscivore, was probably feeding on smaller, lower trophic level fishes than the burbot. These data and future data of this type will provide validation of the freshwater-marine  $\delta^{13}C$  gradient concept.

## **Chipp River Coregonine Fishes**

The  $\delta^{13}$ C data of the coregonine fishes sampled from the Chipp River in 1994 were lipid-corrected using the method of McConnaughey and McRoy (1979) and are expressed as  $\delta^{13}$ C in Figure 4 as a function of length. The size distributions reflect the size range attained by these species in this system. All fish > 300 mm show a consistent dichotomy whereby a portion of each species tends to deviate from a  $\delta^{13}$ C of -27, the apparent boundary between marine and freshwater carbon in the Chipp River system (Figure 4). Sub-100 mm least ciscos undergo a rapid change in  $\delta^{13}$ C with respect to size. Assuming that these fish are of the same year class, change in feeding regime as evidenced by  $\delta^{13}$ C may be reflected by size attained (100 mm fish being more negative in  $\delta^{13}$ C than 80 mm fish). There appears to be a shift towards greater marine dependence in least ciscos



Figure 4.  $\delta^{13}$ C of three 1994 Chipp River coregonines. Abbreviations are the same as Figure 2 plus HBWF = humpback whitefish. Each symbol represents averages of replicate analyses of individual fish.

63 \_

O BDWF

HBWF
 LSCS

between 100 and 300 mm. There is a similar pattern in humpback whitefish. Broad whitefish showed the inverse  $\delta^{13}$ C-length relationship at ~60 mm, then an increase in  $\delta^{13}$ C, as with the other species. The broad whitefish >400 mm (all mature adults) were strongly dichotomous, suggesting alternating marine and freshwater feeding with few intermediaries. This dichotomy also suggests that  $\delta^{13}$ C must change rapidly with migration between fresh- and saltwater and by extension, carbon turnover is rapid in large fish (< 1 year) contradicting the conjecture of Hesslein et al. (1993). The large broad whitefish are also considerably more negative than either the smaller broad whitefish or other coregonine species, probably reflecting feeding on carbon more directly related to peat (i.e. insect larvae, instead of zooplankton). From these data, a preliminary relationship between  $\delta^{13}$ C and feeding habitat can be drawn: a  $\delta^{13}$ C of coregonines > -26 is marine, and a  $\delta^{13}$ C < -28 is freshwater, with intermediate values corresponding to transitional or estuarine feeding.

## **Food Web Reconstruction**

Dual isotope plots enable the reconstruction of food webs as each isotope can independently confirm the flow of organic matter through feeding processes that have predictable effects on isotopic composition.  $\delta^{15}N$  also enables a good assessment of trophic position (Fry, 1988; Cabana and Rasmussen, 1994). Broad whitefish were divided into size groups and compared to non-coregonines sampled



Figure 5. Dual isotope plot of non-coregonines and broad whitefish sampled from the Chipp River in 1994. Each symbol represents averages of replicate analyses of individual fish.

\_ 64

from the Chipp River (Figure 5). The distribution of broad whitefish stable isotope data thus elucidates the diversity of feeding exhibited by this species with respect to size. There are significant clusters of broad whitefish at  $\delta^{15}N$  of ~ +12 at  $\delta^{13}C$  of ~ -26 and -33, suggesting a higher trophic position at times (predominant) and lower trophic positions at others, with the greatest marine carbon being at the lower trophic position (implied by lower  $\delta^{15}N$ ). The significant cluster at -33 departs strongly from other fishes, consistent with a specialist feeding mode.

Broad whitefish had a wide range in  $\delta^{13}$ C consistent with feeding migrations between habitats of low and high <sup>13</sup>C. Chipp River  $\delta^{13}$ C ranged from ~ -20 to -34, a much greater range than found in two broad whitefish populations near the Mackenzie River by Hesslein et al. (1991), i.e. ~ -21 to -25 in Kukjuktuk Creek and -24 to -28 in Travailent Lake. This difference in findings is partially explained by the more limited age classes and numbers sampled by Hesslein et al. (1991). The need for a large sample size of a given species is apparent with these data. This is particularly apparent in the dual isotope plot because of the broad range in values and the need for data clustering for interpretation.

#### Chipp River vs. Teshekpuk Least Cisco

There is good consistency in the <sup>13</sup>C of least ciscos in the comparison of Chipp River and Teshekpuk Lake data (Figure 6). Any site-specific differences can be



Figure 6. 1994 Chipp River least ciscos versus 1992 Teshekpuk Lake least cisco morphs. D = dwarf morph, I = intermediate form, N = normal form.

65 .

attributed to feeding migrations. The data suggests that Chipp River least ciscos reside in fresh water until ~250-300 mm when they become amphidromous. Normal-form least ciscos from Teshekpuk Lake commence marine feeding at ~150 mm. The dwarf form is more similar to Chipp River least ciscos in terms of the regression line (Figure 6). However, Teshekpuk Lake dwarf-form least ciscos are all <200 mm. Thus it was deduced that the dwarf Teshekpuk Lake least ciscos are exhibiting a "normal" behavior and that the normal growth form represents an adaptive change in life history for Teshekpuk Lake. The stable isotope data provided evidence of the significance of amphidromous migrations and the different behavior modes for various populations of a given species.

### **Prudhoe Bay Samples**

At the time of this writing, Prudhoe Bay samples collected in 1994 are being processed. These samples include three coregonine species, Dolly Varden, and other marine fishes (flounder and sculpin) as outlined in the project objectives.

The initial stable isotope analyses reported here suggest that this technique will provide useful data for amphidromy assessment, thus meeting the project's principal goal and providing a new tool useful for environmental assessment needed for oil exploration and development in the Alaskan Arctic Coastal Plain.

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67 -

# **Recruitment of Juvenile Flatfishes in Alaska: Habitat Preference at Kodiak Island**

#### **Brenda L. Norcross**

Institute of Marine Science University of Alaska Fairbanks Fairbanks, Alaska 99775-1080

# Abstract

The distribution and abundance of juvenile flatfishes within the inshore waters of the Kodiak Archipelago, Alaska, have been examined during this ongoing interannual study. Analysis of data collected in 1991 and 1992 suggest the following conceptual models of flatfish distribution based primarily on depth and substrate. Age-0 rock sole are found predominantly in water depths less than 50 m on sand or mixed sand substrate within 10 km of bay mouths. Age-0 flathead sole are found predominantly in water depths greater than 40 m on mud or mixed mud substrate throughout bays. Age-0 Pacific halibut are found predominantly in water depths less than 40 m on mixed sand substrate near or outside mouths of bays. Age-1 yellowfin sole are found predominantly in water depths less than 40 m on mixed substrate at upper reaches of bays. These habitat models were confirmed for Chiniak Bay during collections made in August 1993 and August 1994.

# Introduction

Flatfishes are of strong economic importance in Alaskan waters, due to directed commercial and sport fisheries as well as to commercial bycatch, yet knowledge of the early life history of flatfishes and their habitat in Alaska has been minimal prior to this study. Inferences about the life history of juvenile flatfishes have been made from studies of the same species along the coasts of Oregon and Washington, or from related species in other parts of the world. Twelve species of potentially competitive flatfishes are found near Kodiak Island, Alaska (Rogers et al., 1986; Norcross et al., 1993). The prime targets for commercial fisheries are Pacific halibut (Hippoglossus stenolepis), flathead sole (Hippoglossoides elassodon), rock sole (Pleuronectes bilineatus), rex sole (Errex zachirus), and Dover sole (Microstomus pacificus). There are also directed fisheries for English sole (Pleuronectes vetulus) and starry flounder (Platichthys stellatus). Bycatch includes yellowfin sole (Pleuronectes asper), Alaska plaice (Pleuronectes quadrituberculatus), butter sole (Pleuronectes isolepis), and sand sole (Psettichthys melanostictus), which are marketed secondarily. An attempt is currently underway to develop a fishery for arrowtooth flounder (Atheresthes stomias) for surimi. Commercial bycatch of Pacific halibut has been the cause of premature closure of groundfish fisheries many times during the past decade.
Intertidal zones, shallow coastal areas, protected bays, and estuaries are often considered vital as nursery areas for flatfishes (Tyler, 1971; Gibson, 1973; van der Veer and Bergman, 1986; Tanaka et al., 1989), and abundance and size distributions of juvenile flatfishes have often been related to depth, sediment size, and food availability. Flatfish nurseries are found in shallow waters, often in less than 10 m (Edwards and Steele, 1968; Allen, 1988; van der Veer et al., 1991). Substrates of flatfish nurseries are reported as silt, mud, and fine to coarse sand (Poxton et al., 1982; Wyanski, 1990). Shallow bays and estuaries are thought to serve as excellent nursery areas and ideal feeding habitat because of high insolation of the bottom, high water temperatures, the particular sediment types found in protected waters (Pearcy and Myers, 1974), and the good supply of nutrients from land drainage (Pihl and Rosenberg, 1982).

Initial selection of sampling locations and sites along the coast of Kodiak Island began with the acknowledgement that the coastline of the Gulf of Alaska is more rugged and has a greater tidal range than flatfish nursery areas in other locations around the world. Kodiak Island is representative of the coastline of the Gulf of Alaska because it provides a variety of habitat from shallow, fine-grained tidal flats to deep and rocky areas. Chiniak Bay is the only region of Kodiak Island that is easily accessible via road from the town of Kodiak. Chiniak Bay encompasses three inner bays which are protected to varying degrees (Figure 1: Womens, Middle, and Kalsin Bays); it also provides relatively accessible and unprotected zones outside the mouths of these bays. Prior to this funding, fishes and physical data were collected during 1991 and 1992 from all major bays and straits along the perimeter of Kodiak Island. Sample collection was continued during 1993 and 1994 as a result of this funding; however, collections were limited to Chiniak Bay during 1993, and to Chiniak Bay and Kazakof Bay (on Afognak Island) during 1994.

This project describes and analyzes the recruitment of juvenile flatfishes, especially as influenced by habitat, to inshore waters of Kodiak Island, Alaska. The overall objectives of this project have been to:

- 1) Identify nursery grounds for juvenile flatfishes;
- 2) Characterize those areas according to physical and biological parameters; and
- 3) Develop indices of relative abundance of recruitment for as many species as the data allow.

Analysis of data collected in Chiniak Bay during 1992, 1993, and 1994 was funded primarily by the Coastal Marine Institute (CMI). Dietary analysis of flatfishes captured during 1991 was partially funded by CMI.

During the first two years of sampling (1991–92), all major bays along the perimeter of Kodiak Island were sampled in order to form a broadly based characterization of flatfish nursery areas around Kodiak. The considered parameters



Figure 1. Permanent stations within Chiniak Bay, Kodiak Island.

included location in the bay, depth, bottom temperature, bottom salinity, and sediment type. There were overlapping distributions for the most abundant juvenile flatfishes ages 0 and age-1 rock sole, yellowfin sole, flathead sole, and Pacific halibut), yet patterns of peak abundance were unique to each species. The following conceptual models of the physical parameters most descriptive of flatfish nursery habitat have been developed based on the 1991–1992 distribution of flatfishes around Kodiak Island (Norcross et al., 1995a). Age-0 rock sole are found predominantly in water depths less than 50 m on sand or mixed sand substrate within 10 km of bay mouths. Age-0 flathead sole are found predominantly in water depths greater than 40 m on mud or mixed mud substrate throughout bays. Age-0 Pacific halibut are found predominantly in water depths less than 40 m on mixed sand substrate near or outside mouths of bays. Age-1 yellowfin sole are found predominantly in water depths less than 40 m on mixed substrate in upper reaches of bays.

CMI Framework Issues addressed by this study are as follows:

- 1) Studies to improve understanding of the affected marine, coastal, or human environment;
- 2) Modeling studies of environmental, social, and economic processes to improve predictive capabilities and to define information needs;
- 3) Experimental studies to improve understanding of environmental processes or the causes and effects of OCS activities;
- 4) Projects which design or establish mechanisms or protocols for the sharing of data or information regarding marine or coastal resources or human activities to support prudent management of oil and gas and marine mineral resources;

This study will result in a better understanding of the potentially affected marine environment and its resources (Framework Issue #1) by further examining past data collections (Issue #4) and using them to produce conceptual habitat models (Issue #2). In future years, these habitat models will be linked to experimental results of exposure of flatfishes to oiled sediments (Issue #3).

These conceptual models can be used to predict prime habitat for individual species of juvenile flatfish and, combined with possible trajectories of an oil spill or areas of chronic seepage, will suggest the most vulnerable geographic locations and species of fish. These models may be used to begin assessment of the possible impact of oil development on flatfish communities.

## **Results and Discussion**

Field work funded by this project included the continuance of annual collections within Chiniak Bay on Kodiak Island during August (1993: Cruise KI9301; 1994: Cruise KI9403), as well as auxiliary collections within Chiniak Bay (June 1994: Cruise KI9401) and within Kazakof Bay on Afognak Island (August 1994: Cruise KI9402). Additional data collected within Chiniak Bay during November 1993 were made available by the National Marine Fisheries Service (NMFS), Kodiak Laboratory. The primary objective for all collections was to evaluate the proposed conceptual models of juvenile flatfish distribution developed for Kodiak Island (Norcross et al., 1995a).

All collections in Chiniak Bay during 1993 and 1994 were land-based day trips which employed a 24-foot Boston Whaler owned and operated by NMFS Kodiak. Sampling at each station included a fish trawl, conductivity, temperature and depth (CTD) cast, and sediment grab. Fishes were collected during tenminute tows with a 3.0 m plumb staff beam trawl equipped with a double tickler chain, 7 mm square mesh bag, and 4 mm mesh codend liner. Fishes were identified and measured; ages of flatfishes were estimated based on length. Catch-perunit-effort (CPUE) was calculated based on a ten-minute tow time.

7<sup>1</sup> –

#### Tasks:

- 1) Complete analysis of 1992 samples, including graphical analysis, statistical analysis, reports, and publications *(completed)*.
- 2) Sample Kalsin, Middle, and Womens Bays in 1993 to keep time series of baseline data intact to quantify interannual variation (completed; additional collections in these bays and within Kazakof Bay, Afognak Island, were completed in 1994).
- 3) Analysis of 1993 samples, including graphical analysis (completed; an additional graphical analysis of 1994 collections was done) and statistical analysis (in progress).
- 4) Comparisons among 1991, 1992, and 1993 results to quantify interannual variation (comparison of 1991 and 1992 completed [Norcross et al., 1995b]; further comparisons in progress; additionally the 1994 collections will be compared with previous years).
- 5) Community analysis of all fishes, not just flatfishes, in relation to their physical environment *(in progress)*.

### **1993 Collections**

Twenty-eight stations were occupied within Chiniak Bay during cruise KI9301, August 1993, resulting in a quantitative (cumulative CPUE) catch of flatfishes of 2798. The quantitative catch from ten stations during KI9302, November 1993 was 968, for an average CPUE of 99 flatfishes per station during KI9301 and 97 flatfishes per station during KI9302. With minor deviations, data from both cruises confirmed the conceptual models of juvenile flatfish habitat (Norcross et al., 1995a).

#### **1994 Collections**

Twelve stations within Chiniak Bay were occupied during Cruise KI9401, June 1994. Objectives of this cruise included the collection of data for comparison with flatfish distribution models, a preliminary examination of flatfish presence in Chiniak Bay during early summer, and length at age estimations. The quantitative catch of flatfishes amounted to 989, for an average CPUE of 82 flatfishes per station.

Cruise KI9402 consisted of 32 stations in Kazakof Bay, Afognak Island, occupied during August 1994. The purpose of this cruise was to collect data for comparison with flatfish distribution models, and to produce a baseline survey of groundfishes in Kazakof Bay. The quantitative catch of flatfishes was 453, for an average CPUE of 15 flatfishes per station.

Cruise KI9403, August 1994, included collections at 30 stations in Chiniak Bay. Twenty-one permanent (previously occupied) stations in Middle and Kalsin Bays, within Chiniak Bay, were sampled for comparisons of annual species composition and abundance. Nine new stations were established to give breadth to the sampling design. Seven of these new stations were near or outside the

- 72

mouth of Kalsin and Middle Bays and deeper than had been previously sampled in this area. Sample depths ranged from 1 to 120 m. The quantitative catch of flatfishes captured during KI9403 was 13,993, and the average CPUE was 161 flatfishes per station. Due partially to increased sampling effort during KI9403, the number of captured flatfishes was considerably greater than had been caught in 1991, 1992, and 1993. The large number of captured flatfishes during KI9403 may also be due to increased recruitment.

### Multi-year Analyses of Chiniak Bay

The distribution of age-0 rock sole, flathead sole, Pacific halibut, and age-1 yellowfin sole captured within Chiniak Bay in 1993 and 1994 closely followed the conceptual distributions predicted based on 1991 and 1992 data from all of Kodiak Island (Norcross et al., 1995a). One addition to the modelled distribution, as observed in the 1994 field season, is that age-0 flathead sole are also found at the bay mouth and outside the bay. This results from the fact that greater distances outside Middle and Kalsin Bays were sampled in the 1994 season than had been previously sampled.

In addition to the trawls completed in Chiniak Bay during KI9301 and KI9403, stations were sampled with repetitive tows and scuba dives in order to statistically compare the efficiency of beam trawl versus scuba divers in identifying and quantifying flatfishes on different bottom types. Data collected at these trawl/dive stations sampled during 1993 and 1994 will be used to determine whether scuba divers may be used interchangeably with beam trawl tows in areas and on substrates where trawling is ineffective or impossible.

#### Manuscripts

Funding from CMI assisted with analysis of data collected in 1991 and 1992 for juvenile flatfish distribution and feeding. Thus far, three manuscripts based on these data have been accepted for publication and are available upon request. Copies have been forwarded to the project contracting officer's technical representative (COTR). The distribution and abundance of flatfishes captured during August 1991 was evaluated by discriminant analysis in the manuscript "Nursery area characteristics of Pleuronectids in coastal Alaska, USA" (Norcross et al., 1995a), which has been accepted for publication in the Netherlands Journal of Sea Research. Multivariate analysis of distribution and abundance of flatfishes captured during 1991 and 1992 was summarized in the manuscript "Habitat models for juvenile Pleuronectids around Kodiak Island, Alaska, USA" (Norcross et al., 1995b). Feeding diversity of juvenile flatfishes was described in "Diet diversity as a mechanism for partitioning nursery grounds of Pleuronectids" (Holladay and Norcross 1995). These last two manuscripts were presented orally at the International Symposium on North Pacific Flatfish, held October 26-28, 1994, in Anchorage, Alaska, and will be published in the proceedings of that symposium.

### **Related Projects Assisted Through This Funding**

Live juvenile flatfishes were captured during 1992–94 field work for a separate laboratory study (Moles et al., 1994; Moles and Norcross, 1995) to assess sediment preferences of various juvenile flatfish species. The lab study addressed the response of flatfishes to various concentrations of petroleum hydrocarbons in sediment, submitted fishes to physiological tests which assess the effect of nonavoidance of oiled sediment on the survivability of the fish, and provided a monitor for rates of growth, feeding, and respiration during long-term (chronic) exposure to low levels of oiled sediments.

## Preliminary Conclusions

The conceptual models of flatfish distribution (Norcross et al., 1995b) based on 1991-92 data were verified during collections within Chiniak Bay during 1993 and 1994. As in 1991-92, station locations were selected based on depth and sediment strata, and data were analyzed using the physical parameters of location in the bay, depth, bottom temperature, bottom salinity, and sediment type. The four years of data now available on physical parameters and flatfish abundances within Chiniak Bay has enabled us to narrow the scope of collections proposed for 1995 collections (Table 1, Figure 1). Comparisons of fish abundance and distribution from 1991 through 1994 show differences among species and among stations, as well as interannual differences. In particular, the distribution of rock sole and flathead sole appear to be strongly related to depth and substrate type (Figure 1, Table 1). There appears to be no relation to temperature and salinity, therefore these parameters are not shown in the attached table. In the future, depth and substrate patterns will be examined in more detail and other parameters which may affect the distribution of the abundant flatfishes will be investigated. Key habitat characteristics for each species will then be discerned so that their abundances can be monitored over time.

## Acknowledgements

Data analysis for 1993 and 1994 and report writing were performed by Brenda Holladay and Sherri Dressel. Data analysis for 1991–92 comparisons were done by Franz Müter. Field sampling was conducted by Brenda Holladay, Franz Müter, and Sherri Dressel of the University of Alaska Fairbanks, and Eric Munk, C. Braxton Dew, and Pete Cummiskey of NMFS Kodiak. The principal investigator was involved in all phases of the project.

Station	Depth S	Sediment			c sole 2 1993	3 1994			rfin so 1993	ole 1994			halil 2 1993	out 3 1994			ead s 2 1993	ole 3 1994
KA	0–5	S	80	66	35	64	5	0	2	0	0	0	1	16	0	0	0	0
KA	0–5	S	-	16	20	80	-	2	0	1	-	3	0	23	-	0	0	0
KB	5-10	S	84	136	40	124	30	3	1	3	3	7	2	25	0	0	0	0
KC	10-20	mS	102	251	200	429	28	24	16	16	3	23	8	40	0	1	0	0
KD	20-30	sM	24	-	32	248	32	-	2	0	30	-	0	8	2	-	4	15
KE	30-40	sM	-	-	0	6	-	-	0	2	-	-	0	0	-	-	166	42
KF	50-60	sM	-	-	0	0	-	-	0	0	-	-	0	0	-	-	139	209
KG	60-70	sM	-	-	2	1	-	-	0	0	-	-	0	0	-	-	104	22
KH	7080	sM	0	-	-	0	0	-	-	0	0	-	-	0	6	-	-	1
KI	8090	mS	-	-	-	0	-	~	-	0	-	-	-	0	-	-	-	33
КJ	90–100	sM	-	-	-	0	-	-	-	0	-	-	-	0	-	-	-	0
KK	110-120	) mS	-	-	-	0	-	-	-	0	-	-	-	0	-	-	-	0
ML	0–5	S	1	1	109	0	0	0	8	0	0	1	60	26	0	1	0	0
MM	5-10	mS	64	76	32	0	81	0	2	0	7	13	15	15	0	0	0	0
MN	5–10	mS	35	88	19	6	31	2	1	0	3	6	8	10	0	0	0	0
MO	10–20	mS	-	-	-	2	-	-	-	0	-	-	-	6	-	-	-	0
MP	10-20	mS	29	-	48	9	31	-	10	0	3	-	8	12	3	-	3	1
MQ	20-30	mS	71	188	53	220	12	4	1	2	1	16	0	68	1	18	0	0
MR	30–40	sG	22	-	3	25	0	-	0	0	0	-	0	0	0	-	3	5
MS	5060	mS	11	0	0	0	0	0	0	0	0	0	0	0	26	303	65	14
MT	60–70	mS	-	-	0	0	-	-	0	0	-	-	0	0	-	-	2	3
MU	70–80	mS	-	-	0	1	-	-	0	0	-	-	0	0	-	-	0	13
		Total	523	822	593	1215	250	35	43	24	50	69	102	249	38	323	486	358
		Mean #	44	91	35	55	21	4	3	1	4	8	6	11	3	36	29	16

Table 1. Summary of flatfish catch-per-unit-effort within Chiniak Bay, 1991-94. Stations beginning with "K" are in Kalsin Bay and with "M" are in Middle Bay. S=sand, s=sandy, M=mud, m=muddy, G=gravel.

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- 75 -----

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-- 76

# Juvenile Flatfish Habitat in Kachemak Bay: A Pilot Study

Addendum to

## Recruitment of Juvenile Flatfishes in Alaska: Habitat Preference at Kodiak Island

#### **Brenda L. Norcross**

Institute of Marine Science University of Alaska Fairbanks Fairbanks, Alaska 99775-1080

# Abstract

The objective of this study was to examine and identify nearshore habitat used as nursery areas by juvenile flatfishes in Kachemak Bay, Alaska, and to characterize the juvenile habitat based on physical oceanographic parameters (including depth, sediment grain size, temperature, salinity, and distance from shore) and biological factors (stomach contents and food availability). This pilot was intended as a feasibility study to survey an accessible portion of Lower Cook Inlet. Samples were collected September 24-30, 1994, at 37 stations (Figure 1). A total of 3435 fishes were captured with a 3.0 m beam trawl, of which 42.2% were flatfishes. The most abundant juvenile (age-0 and age-1) flatfishes were rock sole (Pleuronectes bilineatus), flathead sole (Hippoglossoides elassodon), Pacific halibut (Hippoglossus stenolepis), arrowtooth flounder (Atherestes stomias), and yellowfin sole (Pleuronectes asper). Distribution of rock sole, flathead sole, halibut, and yellowfin sole during September in Kachemak Bay was compared with conceptual models of flatfish distribution developed for August in Chiniak Bay, Kodiak (Norcross et al., 1995). Although flatfishes in Kachemak Bay were sometimes distributed over a wider range of depths than in Chiniak Bay, the highest abundances of each species were within the bounds of the conceptual models.

# Introduction

Areas in Cook Inlet, Alaska, are scheduled to be open for oil and gas leases soon. In 1996 the federal government will make portions of Kachemak Bay near Homer available for oil and gas lease. Cook Inlet is a productive estuary which supports many commercial and sport fisheries, including a thriving halibut charter industry. Flatfishes are vulnerable to oil exposure because of their close interaction with the sediment (Moles et al., 1994; Moles and Norcross, 1995). Flatfishes bury themselves in the sediment for protection (Tanaka, 1990), avoidance of currents, and conservation of energy (Gibson and Robb, 1992). Because feeding may involve taking sediment and prey in mouth and sieving the sediment (Hicks, 1984), flatfishes could be directly exposed to oil through ingestion of benthic prey



Figure 1. Consecutive station locations sampled during C19401, September 1994.

as well as contact with the substrate. To date, the limited research on flatfishes in Cook Inlet has focused on adult fishes. Prior to this pilot study there was no information about the role of Kachemak Bay as a nursery ground for juvenile flatfishes.

Most juvenile flatfishes are found in nearshore shallow areas which have the advantages of higher temperatures, lower predation risk, abundant food, and appropriate substrata (Gibson, 1994; Minami and Tanaka, 1992). Distribution within the nursery grounds varies with species, locality, season, and year (Minami and Tanaka, 1992). There are significant differences in the distribution of flatfishes between and within families; these differences allow coexistence of a multitude of flatfish species on the continental shelf and slope (Minami and Tanaka 1992). Depth strongly influences the distribution of some flatfishes, as observed in coastal areas of the Southern Bight of the North Sea, and is one of the most important determinants of sole (*Solea solea*) distribution, in conjunction with substrate type and prey availability (Rogers, 1992). Walsh (1992) found the distribution of yellowtail flounder (*Pleuronectes ferruginea*) to be dependent more on depth than temperature because the species can tolerate a wide range of temperature fluctuations.

Sediment size is an important factor influencing the distribution of flatfishes; individual species can distinguish between and select sediments on the basis of their grain size (Gibson and Robb, 1992; Walsh 1992; Gibson, 1994; Moles and

- 78

Norcross, 1995). Because juveniles actively choose grain size during settlement, grain size is a parameter which is useful for fine-scale description of flatfish distribution. Thus, measurements of grain size provide insight into mechanisms of predator avoidance, energy conservation, and feeding strategy. Although the underlying causes of the association between sediment grain size and flatfish habitat are unknown, physical conditions and benthic organisms are both correlated with sediment structure and probably play a role in the distribution of juvenile flatfishes (Gibson and Robb, 1992). Experimental evidence suggests that juvenile flatfishes actively choose substrate based on grain size because it relates to their ability to bury effectively and avoid predation. Burying behavior has been most frequently attributed to predation avoidance, but may also be a mechanism to conserve energy (Gibson and Robb, 1992).

In laboratory and field studies, flatfishes have a strong preference for specific grain sizes. In fact, they will remain on favored sediment even if that sediment is polluted with oil while an unoiled, less favored grain size is available (Moles et al., 1994). Rock sole prefer sand, yellowfin sole prefer mixed mud/sand, and Pacific halibut prefer mud/sand or sand (Moles et al., 1994; Moles and Norcross, 1995). Field research in Alaska provides examples of flatfish species correlated with sediment size. Norcross et al. (1995) found rock sole on sand or mixed sand substrate, flathead sole on mud or mixed mud substrates, Pacific halibut on mixed sand, and yellowfin sole on mixed substrate. In the North Sea, plaice (*Pleuronectes platessa*) are more densely populated on sandy flats than muddy areas (Berghahn, 1986; Pihl and Veer, 1992). Muddy sediments are preferred by flounder (*Platichthys flesus*) and sole (van der Veer et al., 1991).

Temperature affects feeding and growth rates, as each species has a critical thermal range. Because flatfish species may actively change their distribution to alternate depths for energy conservation and optimal temperature, the distribution of juvenile flatfishes in nursery areas varies spatially and seasonally by species. Frank et al. (1992) found horizontal distributions of age-0 American plaice (*Hippoglossoides platessoides*), yellowtail flounder (*Pleuronectes ferruginea*), and witch flounder (*Glyptocephalus cynoglossus*) to correspond significantly to the bottom temperature distributions around the southern Grand Bank.

The objectives of this pilot study were to:

- 1) Establish that nursery areas for juvenile flatfishes do exist in Kachemak Bay; and
- 2) Characterize the most important parameters which define flatfish nursery areas in Kachemak Bay.

This study addressed Framework Issue #2 in that the conceptual habitat models developed around Kodiak Island (Norcross et al., 1995) were used to design the sampling plan and were compared to the distribution of juveniles in Kachemak Bay. Age-0 rock sole are found predominantly in water depths less than 50 m on

- 79 —

sand or mixed sand substrate within 10 km of bay mouths. Age-0 flathead sole are found predominantly in water depths greater than 40 m on mud or mixed mud substrate throughout bays. Age-0 Pacific halibut are found predominantly in water depths less than 40 m on mixed sand substrate near or outside mouths of bays. Age-1 yellowfin sole are found predominantly in water depths less than 40 m on mixed substrate at upper reaches of bays. In the future, with the inclusion of Kachemak Bay seasonal distribution data, these habitat models will be refined, and should be useful in identifying flatfish nursery habitats which could potentially be impacted by oil and gas development.

Framework Issue #3 was addressed through this study and a related study conducted at the National Marine Fisheries Service (NMFS) Auke Bay Lab, where Adam Moles is conducting laboratory experiments on the physiological affects of juvenile flatfishes exposed to oiled sediments. Moles is currently a Ph.D. student working with Norcross and is funded by NMFS. The lab studies will include survival and growth rates on oiled and unoiled sediments (Moles, et.al., 1994; Moles and Norcross, 1995). The results of Moles' study will enhance and be incorporated in the critical habitat models developed resulting from the 1994–96 field work.

# **Results and Discussion**

Cruise CI9401 consisted of land-based collections conducted in Kasitsna, Jackolof, and Tutka Bays and Sadie Cove (Figure 1) from September 24-30, 1994. Based out of the University of Alaska School of Fisheries and Ocean Science / Institute of Marine Science Kasitsna Bay Laboratory near Seldovia, Alaska, 37 stations were sampled from a 9.3 m aluminum Munson skiff. Collections ranged in depth from 1 to 80 m. A total of 3435 fishes, including nine species of flatfish, were captured. Sampling at each station included a fish trawl, conductivity, temperature and depth (CTD) cast, and sediment grab. Fishes were collected during ten-minute tows with a 3.0 m plumb staff beam trawl equipped with a double tickler chain, 7 mm square mesh bag, and 4 mm mesh codend liner. All fishes were identified, and then counted and measured using a Limnoterra electronic fish measuring board (FMB IV). Ages of the juvenile flatfishes were estimated based on length. The catch-per-unit-effort was calculated based on a ten-minute tow time.

#### Tasks:

1) Sample nearshore areas in Kachemak Bay at sites associated with Highsmith's benthic sampling to locate juvenile flatfishes. Measure related physical parameters (completed). The physical parameters measured include depth, sediment grain size, temperature, salinity, tidal stage, light stage, and distance from shore. The biological factors include the measurement of volatile organics in the sediments and the presence of associated species. Since flatfishes in this area were distributed deeper than anticipated based

80

on Kodiak models, flatfish sampling did not overlap with the intertidal areas sampled by Highsmith as was intended. In addition, Highsmith's sites were very shallow and covered too small an area to be sampled adequately with the plumbstaff beam trawl. In order to sample juvenile flatfishes that were in deeper water, it was necessary to expand sampling to cover more of Kachemak Bay than was originally planned.

- 2) Conduct graphical analyses of the distribution of flatfishes with respect to physical variables (completed).
- 3) Analyze stomach contents of most abundant flatfishes (currently in progress).
- 4) Conduct graphical and statistical analyses of stomach contents with respect to physical variables and Highsmith's results on benthic composition (this task is dependent on Highsmith's analysis; however, the measurement of percent carbon for each station as a relative indicator of food availability has been completed, and these data may used in analyses related to stomach contents).
- 5) Relate the distribution and stomach content analyses of fishes to a relative measure of flushing used in Highsmith's project to indicate recruitment rate of benthic organisms (*the completion of this task is dependent on Highsmith's analysis*).
- 6) Relate laboratory experiments which quantify the effects of non-avoidance of hydrocarbon-laden sediments to field distribution of flatfishes (this is a long range goal based on comparisons with Moles' work and will be done at a later date; more information concerning juvenile flatfish distribution in Kachemak Bay will be obtained in a subsequent study being funded under a separate contract).

The habitats of juvenile (age-0 and age-1) flatfishes were analyzed to characterize nursery grounds as described below:

- One age-0 Pacific sanddab (*Citharichthys sordidus*) was captured at a mean depth of 31.5 m and a bottom temperature of 9.5°C. The bottom salinity was 30.3 PSU, and the sediment type was muddy sand.
- All age-0 and age-1 arrowtooth flounder (*Atheresthes stomias*) were captured at depths ≥ 30 m. They were in waters with a bottom temperature between 9.2-9.7°C and the bottom salinity was 30.3-31.1 PSU. The sediment was predominately muddy sand and gravelly muddy sand.
- Age-1 rex sole (*Errex zachirus*) were captured in depths of 50–65 m. They were only in waters with a temperature range of 9.5–9.7°C. The bottom salinity ranged from 30.7 to 31.0 PSU. Rex sole were found only on muddy sand and gravelly muddy sand.
- All flathead sole (*Hippoglossoides elassodon*) age-0 and age-1 were captured at depths of 30-70 m. The bottom temperatures ranged from 9.3 to 9.7°C. Bottom salinity ranged from 30.3 to 31.0 PSU, with a concentration at 30.7 PSU. The sediment was predominately gravelly mud for age-0. Age-1 flathead sole were usually found on mixed substrate of muddy sand and gravelly mud.

81 \_

- Age-0 Pacific halibut (*Hippoglossus stenolepis*) were captured at depths of 5 to 60 m, but were concentrated at depths ≤ 40 m. They were captured at bottom temperatures of 9.2–9.7°C. The bottom salinity ranged from 30.3 to 31.0 PSU, and they were mainly captured at 30.3–30.6 PSU. Age-0 halibut were found primarily on sand mixed with mud and/or gravel.
- No age-0 Dover sole (*Microstomus pacificus*) were captured, as they are plankton larvae until they reach 45 mm in length (Matarese et al., 1989). One age-1 Dover sole was captured at a station in Kachemak Bay (CS#19) with a bottom temperature of 9.6°C and bottom salinity 30.7 PSU Depth was between 50-60 m and the sediment type was muddy sand.
- One age-0 yellowfin sole (*Pleuronectes asper*) was captured. It was at CS#31 off MacDonald spit at a mean depth of 32 m, a bottom temperature 9.5°C, and bottom salinity 30.3 PSU, and sediment type muddy sand.
- Age-1 yellowfin were captured at depths of 10 to 60 m, but were concentrated around 30 m. They were mainly captured at a bottom temperature of 9.5℃ and a bottom salinity of 30.3 PSU, but were found in the 30.3–30.7 PSU range. It is noteworthy that age-1 yellowfin sole were captured at depths ≥40 m, because during the four years of collection near Kodiak Island (Norcross, 1991–94 unpublished data), they have never been captured at depths greater than 40 m. Age-1 were primarily captured on muddy sand.
- Age-0 and age-1 rock sole (*Pleuronectes bilineatus*) were captured to depths of 56 m, but were nearly all caught ≤ 40 m. Rock sole were captured throughout the ranges of bottom temperatures sampled (9.2–9.9°C) and were concentrated at 9.4–9.6°C. The bottom salinity ranged from 30.3 to 30.9. Most age-0 rock sole were captured over pure sand. Age-1 rock sole were distributed among several sediment types. Rock sole was the only flatfish captured on sediment with gravel; four juvenile rock soles were captured at a single location with gravel (CS#33).
- Age-0 English sole (*Pleuronectes vetulus*) were captured at depths ≤32 m. The bottom salinity ranged from 30.3 to 30.6 PSU. Age-0 English sole were captured mostly on gravelly sand.

Distribution of flatfish in relation to depth and substrate type was observed. Age-0 flatfishes were most frequently captured at depths ranging from 6–20 m and  $\geq$  31 m on sand, muddy sand, or gravelly mud substrates. Age-1 flatfishes were most frequently captured at depths  $\geq$  31 m on muddy sand substrate (Tables 1 and 2).

The five most abundant juvenile flatfishes collected in Kachemak Bay in 1994 (age-0 rock sole, flathead sole, Pacific halibut, arrowtooth flounder, and age-1 yellowfin sole) were identical to the most abundant species in Chiniak Bay, Kodiak; however, the percent species composition varied between locations. Kachemak Bay had approximately the same proportion of rock sole (43%) and flathead sole (36%), whereas Chiniak Bay had 73% rock sole and 11% flathead sole. Additionally, when tested against the depth distribution models from

...... 82

Depth (m)	# stations	# age-0 flatfish	# age-1 flatfish
0-5	6	19	18
6–10	4	210	22
11-20	5	212	18
21-30	2	5	3
31–40	4	129	79
41-50	5	317	138
51-60	6	83	61
61–70	4	79	26
	Tot	al 1053	365

Table 1. Distribution of flatfish in relation to depth in Kachemak bay, September 1994.

Table 2. Distribution of flatfish in relation to substrate type in Kachemak Bay, September 1994.

Main sediment	# stations	# age-0 flatfish	# age-1 flatfish
Boulder	1	0	0
Cobbly gravel	1	0	0
Gravel	1	0	4
Gravelly boulder	1	0	0
Gravelly mud	1	134	16
Gravelly muddy sand	5	66	41
Gravelly sand	2	24	14
Muddy gravel	1	2	3
Muddy sand	10	388	235
Muddy sandy gravel	3	3	4
Sand	8	434	42
Sandy mud	1	1	6

Kodiak (Norcross et al., 1995), some differences were observed for the Kachemak Bay collections from September 1994. In Kachemak Bay, rock sole were found at depths  $\geq 50$  m, flathead sole were found at all depths, Pacific halibut were found at depths  $\leq 40$  m, and yellowfin sole were concentrated at stations with depth 30–60 m. When compared to Kodiak models, all juvenile flatfishes in Kachemak Bay were distributed at greater depths. This difference may be due to spatial or temporal differences, as Kodiak sampling was conducted in August 1991–94 and Kachemak Bay was sampled in late September 1994. Therefore, based on these results, a two-year multi-seasonal study is planned to allow determination of juvenile flatfish distribution within Kachemak Bay intra-seasonally and intraannually. Future sampling will control for depth in an attempt to isolate the controlling factors in juvenile flatfish distribution. Factors that will be investi-

83 \_

gated in the future include substrate, temperature, and salinity as analyzed in the pilot study. An index of "open" versus "enclosed" bays and associated benthos will also be used.

# **Preliminary Conclusions**

The goals of this pilot study were to establish that nursery areas for juvenile flatfishes do exist in Kachemak Bay, and to characterize the most important parameters which define flatfish nursery areas in Kachemak Bay. Results show sufficient numbers of juvenile flatfish in the studied area, and suggest that Kachemak Bay is a nursery area for juvenile flatfishes. The most important parameters which define the nursery area are depth and substrate type. Findings from Kachemak Bay do not exactly fit the Kodiak models of distribution, as flatfish are found deeper in Kachemak Bay. However, these differences may be a result of season (sampling in August in Kodiak and September in Kachemak Bay) or the unique geomorphology of the two regions. Due to the sediment distribution and slope of the continental shelf in Lower Cook Inlet, the geomorphology of Kachemak Bay provides an excellent opportunity to isolate and rank the physical parameters that define juvenile distribution for each species of flatfish. The proposed two-year study will specifically address this issue.

From related work (Moles et al., 1994) it has been established that juvenile flatfishes are sensitive to oil in the substrate. Because of the potential for additional oil and gas lease development in Lower Cook Inlet, it is imperative to have knowledge of nursery areas of juvenile flatfish prior to industry expansion. Once the habitat and distribution of juvenile flatfish species is known, it will be easier to monitor them. Further investigation is required to understand what habitats are important to the distribution of juvenile flatfishes in Kachemak Bay. This information may be critical to the management of flatfish stocks relating to habitat preservation or restoration (Burke et al., 1991), especially in light of the impending oil leases.

## Acknowledgments

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85 \_\_\_\_

# **Testing Conceptual Models of Marine Mammal Trophic Dynamics Using Carbon and Nitrogen Stable Isotope Ratios**

#### Donald M. Schell

Water Research Center Institute of Northern Engineering University of Alaska Fairbanks Fairbanks, AK 99775

# Abstract

Steller sea lions and harbor seals are declining in numbers in the northern Gulf of Alaska and the Bering Sea, and the northern fur seal has apparently stabilized after a decline from previously higher numbers. Although no lease sales are currently planned in the Bering Sea in the near future, if either species is declared endangered, oil and gas exploration and production might be severely curtailed in the Bering Sea or the Gulf of Alaska continental shelf habitats. This project focuses on the food web dynamics supporting top trophic levels in the Gulf of Alaska and the Bering and Chukchi Seas with an emphasis on attempting to find linkages to explain these population declines. The natural abundance stable isotope ratios of carbon and nitrogen were used to trace trophic transfers of these elements to identify possible changes in trophic status or habitat usage over seasonal, annual, and decadal time scales. Preliminary data indicates large temporal changes in isotope ratios over the seasons which may reflect changes in diet and habitat usage. This initial data has also shown the presence of large isotopic gradients in zooplankton between the continental shelf biota of the Bering and Chukchi Seas versus those from the pelagic regions near the Aleutian Islands. The isotopic variations in marine mammal tissues showing temporal signals (e.g., whiskers and claws) reveal that fur seals change trophic positions in the food web on a seasonal basis. Little dietary information linked to isotope data is available to account for these changes. Over the next year, research will focus on expanding the prey isotope data base and identifying possible causes for the observed isotopic variations.

# Introduction

The impetus for this work arises from the rapid decline of pinniped populations in the Bering Sea and the Gulf of Alaska. In spite of apparent strong correlations between the rapid rise of fisheries in the Bering Sea and Gulf of Alaska and commercial pressure on herring and pollock stocks, no definitive links or causeand-effect relationships have been established. If the oil and gas industry is going to enter the northern Bering Sea or Gulf of Alaska, it is likely to become involved in marine mammal and bird concerns. Stable isotope ratio studies, which link both energetic and feeding studies as well as provide a means to validate conceptual food web models, are a powerful tool in deciphering both temporal and spatial variation in ecosystem energy and nutrient flow.

From 1984 to 1990, work supported by the Minerals Management Service on bowhead whales (*Balaena mysticetus*) and zooplankton in the Bering and Chukchi Seas led to the compilation of a comprehensive data base on the stable isotope abundances in the biota. This information led to drastic revision of the perceived natural history of bowhead whales and to the assessments of critical habitat for bowhead whale feeding. The results of the isotopic work are still being used to set model data for recruitment by the species and to test parameters on models being employed by the International Whaling Commission.

Recently the isotopic work has been extended to include sea lions, using a small amount of funding by the North Pacific Universities Consortium on Steller Sea Lions, to begin analyzing sea lion tissues. As part of the joint U.S./Russian ecological investigations of the Bering and Chukchi Seas, additional fish and zooplankton samples were collected over much of the study area in summer 1993. These samples were analyzed as part of this project.

#### Background

Over the past two decades, stable isotope ratio analysis has emerged as a powerful tool in ecosystem research both on the process scale and as a validation technique for large-scale ecosystem models (Michener and Schell, 1994). In relevant applications to this study, Schell et al. (1989a) described a geographic gradient in isotope ratios in biota across the Alaskan Beaufort Sea and the Bering and Chukchi Seas and showed that this gradient could be helpful in describing bowhead whale natural history. The isotopic gradient arises from the primary producers in the ecosystem and is passed up food chains to label consumers up to the top predators. Saupe et al. (1989) described the parallel shifts in  $\delta^{13}$ C in euphausiids and copepods across this region, and Schell et al. (1989b) and Schell and Saupe (1993) discussed the effects of the gradient in forming oscillations in isotope ratios in whale baleen. Springer et al. (1984, 1986) investigated food webs in the region and noted the interannual effects of climate on the success of seabird recruitment. Hobson and Welch (1992) used isotope ratios to describe the trophic relationships of birds and mammals to the available prey species in the Canadian Arctic. Further extension to benthos (Dunton et al, 1989) and to fishes (Vinette, 1992) has confirmed that the isotopic trends are evident across the entire food web. Polar bears residing north of Alaska and Canada in the Beaufort Sea are several ‰ depleted in <sup>13</sup>C relative to bears from the western Chukchi Sea (Schell and Amstrup, in prep.).

In contrast to the primarily geographic control on carbon isotope ratios, nitrogen isotope ratios are influenced by trophic level. Vinette (1992) has shown that the  $\delta^{15}$ N of euphausiids and copepods in the Bering, Chukchi, and Beaufort Seas are statistically indistinguishable, but when pelagic and benthic species of known feeding habits are compared, a predictable enrichment of about 3.3% per trophic level in <sup>15</sup>N occurs. By assembling the trophic spectrum of species within an ecosystem it is possible to ascribe trophic status within the ecosystem. Hobson and Welch (1992) used  $\delta^{15}$ N values in the Barrow Strait-Lancaster Sound region to identify the roles of arctic cod (Boreogadus saida) and other prey species to top consumers. Higher trophic levels showed little change in  $\delta^{13}$ C but varied by an average of 3.8‰ between levels. Recently, Sease et al. (1993) showed preliminary data which confirmed that sea lions occupy a high trophic status in North Pacific food webs and reflect a geographic gradient between Prince William Sound and the Washington coast. Schell (1996) has further shown that sea lions in the Bering Sea are not as high in trophic status as spotted seals and span a wide trophic range between individuals. Although a particular sea lion may show relatively small isotopic variation over time, among the few individuals sampled, very often large differences have been noted.

# **Results and Discussion**

## Isotope Ratios in Zooplankton

The analysis of zooplankton samples collected from a Russian ship, *R/V OKEAN*, were the initial focus of this study and this aspect of the project was completed





during the past year. Figure 1 represents the cruise track of the vessel. The averaged isotope data for copepods and euphausiids from each of the general test areas are listed in Table 1. No significant differences were noted between locations except for the southern pelagic polygon. Here the  $\delta^{13}$ C values were significantly more depleted than at other stations for all general taxa tested—euphausiids,

D		Euj δ <sup>15</sup> N	phausiids - ‰ <u></u>			
Region						
Α	n	4	4	5	5	
	avg	9.87	-21.46	9.31	-23.57	
	range	9.1 to 11.2	-22.7 to -19.5	8.6 to 10.3	-25.0 to -23.2	
	sd	0.72	1.14	0.54	0.85	
В	n	4	4	7	7	
	avg	6.20	-23.25	4.54	-23.88	
	range	5.5 to 6.6	-24.6 to -22.0	3.1 to 6.4	-25.3 to -2.98	
	sd	0.43	1.02	1.09	0.75	
С	n	2	2	4	4	
	avg	8.71	-20.16	7.99	-21.39	
	range	8.09, 9.32	-19.97, -20.35	7.7 to 8.4	-22.6 to -20.3	
	sd			0.26	1.00	
D	n	9	9	13	13	
	avg	8.59	-20.53	9.20	-22.85	
	range	7.4 to 9.9	-22.1 to -17.7	6.9 to 13.3	-25.6 to -19.9	
	sd	0.83	1.27	1.64	1.52	
E	n	1	1	4	4	
	avg	8.04	-20.05	8.56	-22.18	
	range			7.2 to 9.3	-22.9 to -20.8	
	sd			0.59	0.75	
F	n	3	3	5	5	
	avg	9.38	-20.7	10.83	-22.4	
	range	8.4, 9.2, 10.5	-20.0, -20.0, -22.0	9.2 to 12.0	-23.9 to -20.9	
	sd	0.88	1.07			
G	n	3	3	3	3	
	avg	10.63	-21.5	11.62	-23.4	
	range	9.4, 10.2, 12.3	-20.2, -21.4, -23.0	10.8, 11.8	-23.3, -23.	
	sd			12.2	-23.4	

Table 1. Isotope ratio data for euphausiids and copepods collected on the *R/V OKEAN* cruise, July-September 1993 for the regions shown in Figure 1. Individual data shown for n < 4.

89 .....

copepods, and chaetognaths. This remarkable difference will be looked for again in summer 1995 on a research cruise along the Aleutian Islands and into the southern Bering Sea. The  $\delta^{15}N$  and  $\delta^{15}C$  values were about 2‰ depleted in the heavier isotopes when compared to other areas of the Bering Sea. This depletion, if typical of areas off the continental shelf in the Bering Sea, may help explain some of the sharp oscillations in isotope ratios found along the lengths of whiskers, as described below.

The possibility of temporal changes in isotope ratios was tested by comparing the 1993 data against data from earlier collections. Figures 2 and 3 show the  $\delta^{13}$ C and  $\delta^{15}$ N values of copepods and euphausiids from the Bering and Chukchi Seas over the sample periods. The region spans the northern Bering Sea on both the Russian and U.S. sides. Specific station locations and detailed data for these cruises are reported in Schell's (1992) report. No significant differences were found except for the southern Bering Sea samples noted above for both  $\delta^{15}$ C values. Although it appears that a trend toward more depleted  $\delta^{13}$ C values is apparent over time, this trend is not significant at the p = 0.05 level, and no trends are evident in the  $\delta^{15}$ N data. This indicates that the interannual variations in isotope ratios are small within taxa collected from the Bering and southern Chukchi Seas.



Figure 2. Range, average and standard deviations of  $\delta^{13}$ C values of euphausiids and copepods collected from the Bering and Chukchi seas. Years of collection are shown beneath each data set. Collection dates, station locations and individual data for all cruises prior to 1993 are in Schell (1992).

--- 90



Figure 3. Range, average and standard deviations of  $\delta^{15}N$  values of euphausiids and copepods collected from the Bering and Chukchi seas. Years of collection are shown beneath each data set. Collection dates, station locations and individual data are in Schell (1993).

#### **Isotope Ratios in Marine Mammal Tissues**

The collection of samples of marine mammal tissues from native harvests and from archived samples has been more successful than anticipated. Tables 2 and 3 list current sample holdings and the status of the preparation and analysis for isotope ratios of specimens from the Bering Sea and western Gulf of Alaska, respectively. Only a limited portion of the representative data are discussed in this interim report.

The initial examination of the isotope ratios of carbon and nitrogen along the lengths of vibrissae from ringed seals, northern fur seals, and Steller sea lions revealed that movement across geographic regions with distinct isotope ratios resulted in the acquisition of these features by the marine mammals. This data is very preliminary at this time and the reasons for their occurrence cannot be unequivocally stated. Some of the signal is imparted by geographic effects—the primary producer regimes in offshore waters are more depleted in the heavier isotopes than those on the continental shelves as evidenced by the isotope ratios in herbivores. Figure 4 shows representative isotope ratio traces along vibrissae from the three marine mammal species tested. Ringed seals from the Chukchi Sea have the least amount of variation, indicating relatively small temporal changes in either overall isotope ratio regimes in the environment or in the trophic status of prey species taken.

Species	Year	N	Status
Northern fur seals (Callorhinus ursinus)	1993	5	P, A, R
Northern fur seals (Callorhinus ursinus)	1994	70 (7)	P, A, R
Steller sea lions (Eumetopias jubatus)	1993	5	P, A, R
Steller sea lions (Eumetopias jubatus)	1994	11	P, A, R
Steller sea lion (Eumetopias jubatus)	1994	1 (all)	Р
Steller sea lions (Eumetopias jubatus)	1995	3	
Ringed seals (Phoca hispida)	1994	5	P, A, R
Bearded seals (Erignathus barbatus)	archived		
Walleye pollock (Theragra chalcogramma)	1993		P, A, R
Pacific cod (Gadus macrocephalus)	1993		P, A, R
Capelin (Mallotus villosus)	1993		P, A, R
Sandlance (Ammodytes hexapterus)	1993		P, A, R
Squid (Beryteuthis spp.)	1993		P, A, R

Table 2. Pinniped and prey samples from the Bering Sea, 1993-95.

P = processed (dried and ground/cut)

A = analyzed (isotope ratios)

R = recorded (data files and plots)

Table 3. Pinniped and prey samples from the western Gulf of Alaska.

Species	Year	N	Status
Steller sea lions (Eumetopias jubatus)	1993	10	P, A, R
Steller sea lions (Eumetopias jubatus)	1994	7	
Harbor seals (Phoca vitulina)	1993	4	P, A, R
Harbor seals (Phoca vitulina)	1994	10	
Walleye pollock (Theragra chalcogramma)	1993		P, A, R
Walleye pollock (Theragra chalcogramma)	1994		Р
Pacific cod (Gadus macrocephalus)	1993		P, A, R
Pacific cod (Gadus macrocephalus)	1994		Р
Herring (Clupea pallasii)	1993		P, A, R
Herring (Clupea pallasii)	1994		Р
Capelin (Mallotus villosus)	1993		P, A, R
Capelin (Mallotus villosus)	1994		Р
Sandlance (Ammodytes hexapterus)	1993		P, A, R
Sandlance (Ammodytes hexapterus)	1994		Р
Squid (Beryteuthis spp.)	1993		P, A, R
Squid (Beryteuthis spp.)	1994		P

P = processed (dried and ground/cut)

A = analyzed (isotope ratios)

R = recorded (data files and plots)



Figure 4.  $\delta^{13}$ C and  $\delta^{15}$ N values from along the lengths of pinniped vibrissae taken from representative animals. Male Steller sea lions (top row) and northern fur seals (middle row) were collected from the Pribilof Islands, Alaska, May 1994. Ringed seal (bottom row) vibrissae from a female adult (left) and a juvenile (right) were collected from near Barrow, Alaska, July 1994.

In contrast, both fur seals and sea lions show pronounced oscillations in isotope ratios along vibrissae, indicating changes in either diet or location or both, over the temporal span represented by the length of the whiskers. The fur seals show isotope ratio variations characterized by regular oscillations at constant intervals along the lengths accompanied by much larger singular shifts in  $\delta^{15}N$ . These oscillations may be greater than 8‰ (Figure 4). Some fur seals from the Pribilof Islands showed very high  $\delta^{15}N$  values at the tips of the vibrissae followed by a decline in the more recently laid down tissue. The  $\delta^{13}C$  oscillations, although usually matching the  $\delta^{15}N$  changes in direction, are typically much smaller in magnitude. The implications of a relatively constant  $\delta^{13}C$  are that the  $\delta^{15}N$  changes arise from shifts in diet and not from migratory movements to areas of differing isotopic regimes.

Sea lions show widely varying values in isotope ratios along vibrissae (Figure 4). Some animals were found to have only one or two large variations (up to 6–8‰), whereas others had several smaller oscillations. Growth rates of these whiskers were not available to date the times that the changes occurred; however, experiments with captive sea lions currently ongoing at the University of British Columbia should provide us with these essential data. For this project, whiskers of captive animals were marked at varying lengths and the growth rates are periodically measured. Similar experiments are planned for captive fur seals at the Mystic Marine Life Aquarium in Connecticut.

Potential prey species also showed large isotope ratio variations depending on size and location of capture (data not shown). Pollock shift trophic levels as they increase in size and predation by marine mammals on these changing size classes will be reflected in the isotope ratios of tissues being laid down at that time. The specific correlations between predator and prey isotope ratios have not yet been made but are being tested as data from past cruises and current analyses become available.

## Conclusions

Although the data shown are very preliminary at this stage, several tentative conclusions have been postulated for further testing. These include:

- 1) Isotope ratios in zooplankton from the Bering Sea indicate that primary producers in pelagic waters have lower  $\delta^{13}$ C and  $\delta^{15}$ N values than samples collected from onshelf and continental slope waters. This difference, if persistent across seasons and years, may offer a useful marker for habitat usage by marine mammals.
- 2) Northern fur seals show marked regular oscillations in  $\delta^{15}N$  along the lengths of their vibrissae but not nearly to the same extent in  $\delta^{13}C$ , suggesting that they undergo major shifts in trophic position on a seasonal or interannual basis.

3) Steller sea lions also show oscillations in carbon and nitrogen isotope ratios but the  $\delta^{15}N$  values are usually higher than those of fur seals. This may indicate a trophic separation between the species, but data is insufficient at this time to draw any final conclusions.

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95 ---

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# **Circulation on the North Central Chukchi Sea Shelf**

#### **Thomas J. Weingartner**

Institute of Marine Science University of Alaska Fairbanks, AK 99775

# Abstract

The circulation on the north central Chukchi Sea shelf and its connection to other portions of this shelf are poorly known. Indeed, existing circulation scenarios differ significantly from one another in this regard. This report summarizes a mooring deployment program in the north central Chukchi Sea designed to resolve these issues. Specific goals of this program are to determine: 1) the mean and time-varying circulation and water mass property distribution of the region and 2) the extent to which local winds, forcing by the deep ocean along the shelfbreak, and/or buoyancy flux contribute to circulation variability. The results have direct bearing on understanding transport pathways in the Chukchi Sea and on verifying models which predict the dispersal of pollutants (e.g., hydrocarbons, trace metals, radionuclides) released and/or advected into the region. The north central shelf is biologically important insofar as it supports a large benthic biomass and large numbers of marine mammals which feed on the benthos. As several of the processes under study are not unique to the Chukchi Sea, it is anticipated that the results will have applications to other arctic shelves.

Three moorings were deployed on the north central shelf of the Chukchi Sea between Herald and Hanna Shoals. These moorings are complemented by simultaneous current meter measurements obtained from Bering Strait, Hope Sea Valley, and the northeast Chukchi Sea (offshore of Pt. Lay to Barrow Canyon), fall shipboard hydrographic surveys, and daily satellite imagery of the sea-ice field throughout the winter months. The existing mooring and hydrography components of the program are supported by the National Science Foundation (NSF), the Ofice of Naval Research (ONR), and the Japan Marine Science and Technology Center (JAMSTEC). All of the moorings were scheduled for recovery in September 1995. The remote sensing component of the program will be conducted by collaborating National Aeronautics and Space Administration (NASA) colleagues. A portion of the results are expected to serve as a basis for comparison with numerical model experiments for the understanding of outflows of dense bottom water formed on arctic shelves in winter.

# Introduction

The Chukchi Sea is unique among arctic shelf seas in that its circulation and physical properties are strongly influenced by waters of Pacific Ocean origin. Due to the mean pressure gradient between the Pacific and Arctic Oceans, these waters flow northward through Bering Strait and ultimately enter the Arctic Ocean. En route, they are modified by a variety of exchanges with the atmosphere, sea-ice, and bottom sediments. Both the inflow and these exchanges have a profound influence on the circulation and dispersal of pollutants potentially introduced onto this shelf. For reference, Figure 1 shows a regional bathymetric map which includes the location of the current meter moorings referred to later. The figure shows that while much of this shelf is relatively flat, there are a number of key geomorphic features which exert significant influence on the circulation and, implicitly, on the various water mass modification processes. These features include Bering Strait in the southern Chukchi Sea, Barrow Canyon in the northeast corner of the shelf, and Herald Valley in the northwest. Other important features include Hope Sea Valley, a broad depression extending from Bering Strait to Herald Valley, Herald Shoal in the central basin, and Hanna Shoal in the northeast.



Figure 1. Bathymetric Map of the Chukchi Sea. Solid circles indicate positions of current meter moorings deployed in September 1994. Moorings C1, C2, and C3 are supported by MMS-CMI. Station positions (marked by plus signs) used to construct the contour plots are shown in Figure 3.

Although considerable effort has been expended in the last decade to understand the oceanography of this shelf, several fundamental issues remain which have direct bearing on pollutant dispersal. The most important of these issues, and the objective of this program, concerns the nature of both the mean and time-varying circulation field on the outer shelf. Coachman et al. (1975) presented one of the earliest regional circulation scenarios (Figure 2) which suggested that north of Bering Strait the flow bifurcates. One branch, consisting primarily of highsalinity, nutrient- and carbon-rich water (Bering Shelf Water, or BSW, following the nomenclature of Coachman et al., 1975; Walsh et al., 1989), flows northwestward from Bering Strait through the Hope Sea Valley and Herald Valley (beyond which the path of this water is a subject of speculation). The other branch,



Figure 2. Near-bottom circulation of Chukchi Sea as inferred by Coachman et al. (1975). [Figure copied from Coachman et al., 1975]

consisting of more dilute and nutrient- and carbon-depauperate water, flows northeastward parallel to the Alaskan coast within the Alaska Coastal Current (ACC). The ACC enters the Arctic Ocean and the outer shelf of the Beaufort Sea through Barrow Canyon.

While the branched nature of the flow is well established, recent measurements (Weingartner, in prep.) indicate that there is, in fact, a third branch that flows northward parallel to the east flank of Herald Shoal—not southward (Figure 2) as inferred by Coachman et al. (1975). This third branch also consists of BSW as indicated in Figure 3. This figure shows plots of temperature, salinity, dissolved oxygen, and fluorescence (proportional to chlorophyll) contoured from data collected along an east to west hydrographic transect occupied in September 1992 between Barrow Canyon and Herald Valley (Figure 1). Relatively warm (> 3.0°C), low-salinity (<32.2) water with a high dissolved oxygen concentration (>7.0 mL L<sup>-1</sup>) and low fluorescence characterizes the ACC waters entering Barrow Canyon. In contrast, bottom water salinities are higher and temperatures colder in Herald



Figure 3. Contour plots of temperature (upper left), salinity (upper right), dissolved oxygen (lower left) and fluorescence (lower right) between Herald Valley and Barrow Canyon along the hydrographic transect shown in Figure 1. Data collected in September 1992.

\_\_\_\_ 100

Valley. Fluorescence levels are also substantially higher here and near-bottom concentrations of oxygen are low (<6.0 mL L<sup>-1</sup>). Near-bottom concentrations of nutrients are substantially greater in Herald Valley than in Barrow Canyon (not shown). All of these properties indicate that the water flowing through Herald Valley is the BSW water mass. The third branch is indicated by the prominent fluorescence signature over the gentle depression to the east of Herald Shoal where bottom water properties are also consistent with the BSW water mass. Data from stations to the south occupied on this same cruise shows that these water mass characteristics extend southward to the southern Chukchi Sea and Bering Strait. In fact, their properties indicate that they were formed from water masses advected from the deep Bering Sea basin. The existence of the branch along Herald Shoal is also verified by moored current measurements. Mean monthly velocity vectors from four current meter moorings deployed in the Chukchi Sea in 1991-92 are shown in Figure 4. Of particular interest are the vectors for mooring HS3 which was situated in the depression east of Herald Shoal. The remaining moorings all lie within the ACC (MBC12, UBC12 were



Figure 4. Mean monthly ocean current and wind velocity vectors between October 1991 and August-September 1992. Mooring CLE3 was located offshore of Cape Lisburne, moorings UBC12 and MBC12 were located in Barrow Canyon, and HS3 was located to the east of Herald Shoal at the position indicated by C1 in Figure 1. Corresponding mean monthly wind velocity vectors at 65°N, 170°W and 70°N, 165°W are indicated in the upper two time series plots.

located in Barrow Canyon and CLE3 was located offshore of Cape Lisburne). The vectors for HS3 show northward flow in all months, even when strong northeasterly winds produce a reversal or nearly stagnant flow in the coastal current. The annual average implies a northward transport of about  $2 \times 10^5$  m s<sup>-3</sup>, about 25% of the mean northward transport through Bering Strait (Roach et al., 1995). Because BSW carries a high load of organic carbon, this northward flow could be a significant source of carbon for the outer shelf. Indeed, it might be essential in sustaining the large benthic biomass observed here (Feder et al., 1994) and the marine mammals which feed on these organisms. (Fay, 1982; Moore and Clarke, 1986).

A fundamental goal of this program is to determine where this water flows once it is transported north of Herald Shoal. In the absence of significant wind forcing, dynamical considerations suggest that the flow would be eastward, and very limited current measurements (Johnson, 1989) indicate steady (subsurface) eastward flow northwest of Hanna Shoal even when subject to strong easterly winds. Circulation on the outer shelf might also be forced by the pressure gradient associated with the eastward flowing Beaufort Undercurrent (Aagaard, 1984). In contrast, winter sea-ice trajectories are westward under the prevailing northeasterly winds (Colony and Thorndike, 1984; Muench et al., 1991), although it is not clear if ice drift is a reliable proxy for subsurface circulation. As a consequence of these conflicting and limited observations it is not at all clear what the circulation field over the north central shelf is. This proposal hypothesized that the (subsurface) flow along the outer shelf is eastward and includes BSW advected along the east flank of Herald Shoal and a portion of the transport carried through Herald Valley. If a mean eastward circulation prevails, then this flow represents a pathway for pollutants discharged onto the Russian shelves to the west or northward from the southern Chukchi Sea. Regarding the former, East Siberian Sea waters are believed to flow eastward around the north coast of Wrangel Island and are known to flow southeastward in the Siberian Coastal Current (Coachman et al., 1975) which enters the Chukchi Sea through Long Strait (Figure 1). To date, nearly all of the oil exploration activity in the Chukchi Sea has occurred in the region north and east of Herald Shoal. However, additional lease sales are under consideration in both the Russian and U.S. Economic Exclusion Zones of Hope Sea Valley. Consequently, pollutants released in this region could be carried onto the north central shelf. If, on the other hand, the prevailing flow is westward, then pollutants from marine industrial activities in the Beaufort Sea could be advected into this region.

In addition to determining the regional mean flow and water properties, an important program goal is to characterize the temporal flow variability. Three mechanisms can be identified which would contribute to this variability: winds, low-frequency oscillations propagating along the shelfbreak, and thermohaline influences. Winds, which play a prominent role in the circulation of the Chukchi Sea including Bering Strait, explain a significant fraction of the current variance (Coachman and Aagaard, 1988; Weingartner, in prep.). However, given Johnson's (1989) observations it is not clear that their influence will be as prominent over the outer shelf. Here, we might find that a significant fraction of the flow variability is related to remotely forced shelf waves propagating along the continental slope as Aagaard and Roach (1990) maintain to be the case on the outer Beaufort shelf. An important issue pertinent to modeling considerations is to determine the extent to which current variability is coherent with the local wind field. For example, if local winds and current variations are incoherent, then regional wind-driven barotropic models are unlikely to provide accurate circulation forecasts.

Thermohaline (or buoyancy) effects might also be quite important in this shelf's dynamics. The primary focus of this program is on the effects of negative buoyancy forcing associated with brine rejected from growing sea-ice. Particularly strong thermohaline gradients, which result in strong baroclinic pressure gradients, are associated with coastal polynyas (Schumacher et al., 1983). In the northeast Chukchi Sea, the prevailing northeasterly winds of winter frequently effect an offshore drift of ice leading to the development of a large polynya system along the Alaskan coast and concomitant formation of large volumes of cold, saline (dense) water (Cavalieri and Martin, 1994). Gravitational spreading, bottom friction, alongshore convergence within the coastal current (Weingartner, in prep.) and/or instabilities associated with the density front formed along the perimeter of the polynya (Gawarkiewicz and Chapman, 1995) can force this dense, coastal water mass offshore and across the north central Chukchi Sea. All of these effects could lead to a midwinter flow regime which is markedly different from the other months of the year. An illustration of one possible manifestation of this thermohaline effect is seen in Figure 5, which shows time series of temperature and salinity between October 1993 and September 1994 from two current meter moorings located near mooring C1 and C2 (see Figure 1). The winter of 1994 is characterized by two month-long events during which very high salinity water (at the freezing point) is observed at this location. The rapid increase and decrease in salinity at the end and the beginning of each of these events suggest a front or eddy propagating through the region. The depth of the feature is probably about 10 m and it is believed that the dense water originated from the polynyas along the northwest coast of Alaska-some 200 km to the east of the mooring site. This data suggests that the circulation on the north central shelf might, to some extent, be influenced by nearshore processes occurring several hundred kilometers to the east.

From the pollution transport perspective, the significance of these plumes is that they represent a mechanism by which shelf pollutants can be trapped within a near-bottom layer and advected across the shelf into the deeper portions of the Arctic basin. As presently configured, existing pollutant transport models do not incorporate these thermohaline effects. This transport mechanism is suggested in

103 \_\_\_\_



Figure 5. Time series of temperature and salinity between October 1993 and September 1994 from two moorings located in the central Chukchi Sea. With respect to Figure 1, the mooring positions were approximately at C1 (bottom two time series) and C2 (top two time series).

Figure 6, which shows contours of temperature, salinity, and silica along a hydrographic section extending across the shelf and slope northeastward of Hanna Shoal. (The data were collected from the ONR-funded U.S. Coast Guard *Polar Star* cruise of August of 1993). A near-bottom plume of very cold (-1.7°C) water (highlighted by shading in Figure 6) is seen descending from the shelf to a depth of about 125 m. Over the shelf, the plume is separated from the surface by a strongly stratified pycnocline which inhibits vertical mixing. The high silica values associated with the plume imply that this is the BSW mass. This observation also provides evidence for an eastward subsurface drift along the outer shelf. The low temperatures and the salinities (~32.5) are characteristic of water formed on the Chukchi shelf in winter. The salinity data in Figure 5 suggests that, in winter, plumes or eddies of substantially greater salinity (density) can be formed and subsequently swept into the basin to penetrate to even greater depths.

\_ 104






Figure 6. Contour plots of temperature (top), salinity (middle), and dissolved silicate (bottom). Data were collected in August 1993 from the USCG *Polar Star* along a transect extending northeastward from Hanna Shoal.

105 \_

Finally, an ancillary goal of this program is to provide better documentation of interannual variability in the oceanography of the Chukchi Sea. Substantial progress has been made toward this goal with respect to understanding the variability in water transport through the Bering Strait (Roach et al., 1995). However, little is known about long-term variability over the remainder of the Chukchi shelf. For example, in comparing current meter data from 1991-92 (Weingartner et al., in prep.) with measurements made in 1986-87 (Aagaard and Roach, 1990) and 1981-82 (Aagaard, 1988), it appears that from spring through fall, flow within the ACC was similar in all these years. In contrast, the winter circulation differed significantly among these years. In the winter of 1991-92 the coastal flow reversed or remained stagnant for several months, while relatively steady flow was observed in 1981-82 and only a brief flow reversal occurred in January 1986. As another example, local alongshore winds were significantly coherent with current variations in Barrow Canyon in the winter of 1991-92, consistent with the response of a coastal current to alongshore winds. However, in 1986-87 current variations here were only weakly correlated with the local winds. This result plus other evidence led Aagaard and Roach (1990) to conclude that current variations were due to remotely-forced shelf waves with periods of about five days. Comparison of late fall-early winter spectra of the alongshore winds between these two years shows nearly twice as much energy in 1986-87 at periods between four and six days. Shelf wave amplitudes are expected to differ accordingly between the two years as a result. Finally, it was noted that in each of these winters the size and the persistence of the coastal polynyas (hence the volume of dense water formed and the strength of the buoyant forcing) were substantially different from one another. Given this variability and the few available data sets, it is impossible to determine which, if any, of these winters is typical.

The results of this project have a direct bearing on a number of CMI Framework Issues:

- They will address mechanisms pertaining to the transport and fate of pollutants stemming from offshore marine activity in the Chukchi Sea or advected from elsewhere, including Russian waters and/or the Beaufort Sea;
- 2) The hypothesized circulation might differ significantly from that predicted by existing barotropic, wind-driven circulation models (Observations are therefore required to verify these models and/or to guide modelers on the additional physics required to produce realistic simulations. Existing pollutant transport models do not include the effects of buoyancy or deepocean forcing along the shelf/slope margin.);
- 3) The central shelf of the Chukchi Sea appears to be important to bottomfeeding marine mammals which might therefore be impacted by pollutants via the food chain; and

4) Conclusions pertaining to the potential impact of marine industrial activity must be based on an appreciation of the inherent variability of the physical environment.

# Discussion

This CMI program complements moorings supported by NSF, ONR, and JAMSTEC. (The NSF and ONR programs are being conducted in collaboration with Knut Aagaard of the University of Washington). The moorings were deployed in the fall of 1994 and were scheduled for recovery in September 1995 from the University of Alaska's *R/V Alpha Helix*. Figure 1 shows the mooring positions, and Table 1 details the mooring particulars. The array is concentrated in the northeast Chukchi Sea but also includes moorings in the Bering Strait and Hope Sea Valley. It is designed to examine the circulation field between Herald

Table 1. Locations of Current Meter Moorings Deployed in September 1994. (The MMS-CMI moorings are indicated as C1, C2, and C3 in Figure 1 and in the table.)

Area	Latitude (N)	Longitude (W)	Bottom Depth (m)	Funding Agency
Central Chukchi Sea	70° 37′	167°5′	54	MMS <sup>1</sup> (C1)
	71° 20′	164° 27'	46	MMS <sup>1</sup> (C2)
	71° 41′	167° 11′	49	MMS <sup>1</sup> (C3)
	69° 59′	166° 14′	46	ONR <sup>1</sup>
	70° 45′	163° 17′	46	ONR <sup>1</sup>
Upper Barrow Canyon	71°4′	159° 29′	79	JAMSTEC <sup>2</sup>
	71°3′	159° 32′	76	NSF <sup>3</sup>
Middle Barrow Canyon	71°25′	157° 45′	120	NSF⁴
	71°21′	157° 37′	107	NSF-JAMSTEC⁴
	71° 15′	157° 31′	62	NSF-ONR <sup>1</sup>
Hope Sea Valley	68° 10′	168° 58'	56	NSF <sup>1</sup>
Eastern Bering Strait	65° 47′	168° 35′	54	NSF <sup>1</sup>

<sup>1</sup>Each mooring consists of an Aanderaa current meter (ACM) moored above a Seabird temperature/conductivity recorder (Seacat). The instruments are vertically separated by 1 meter with the Seacats located 6 meters above the seabed.

<sup>2</sup>One Acoustic Doppler Current Profile (ADCP) current meter with a Seacat 6 m above bottom. The ADCP collects current profiles averaged over 2 meter bins throughout the water column from a depth of 60 m to 12 m

<sup>3</sup>Two Seacat/ACM pairs: 7 and 25 meters above bottom.

<sup>4</sup>Four Seacat/ACM pairs: 7, 25, 40, and 60 meters above bottom.

\_\_\_ 107 \_\_\_\_

and Hanna Shoals and the connection to the flow over the north central shelf and the Alaska Coastal Current (including Barrow Canyon). During the cruises hydrographic surveys throughout the Chukchi Sea (supported by JAMSTEC and NSF) are also conducted that allowed determination of regional water mass characteristics using temperature, salinity, dissolved oxygen, nutrients and  $\delta O^{18}$ . Lee Cooper is performing the  $\delta O^{18}$  analyses with support from the Department of Energy.

# **Planned Analyses**

The data will be analyzed with the goal of addressing the hypotheses advanced in the proposal. These are:

- 1) The mean flow of subsurface waters on the outer shelf is eastward and consists of BSW, mixed with other Chukchi Sea water masses, flowing northward through Herald Valley and along the east flank of Herald Shoal.
- 2) In winter, some of the dense water formed within the coastal polynyas athwart the northwest Alaskan coast, spreads northwest into the central Chukchi Sea.

Included within the specifics of these hypotheses are:

- a) The determination of the mean and time-varying subsurface circulation on the north central shelf; and
- b) The determination of the spatial coherence of the current variations and their relation to wind and buoyancy forcing.

The data from the three CMI moorings will be analyzed in conjunction with those from moorings funded by the other agencies participating in this program. Where possible, comparisons will be made between these data and results from previous years. The approach to addressing hypothesis 2 relies on understanding the wind and thermohaline forcing on the Chukchi shelf. This will be addressed using SSM/I (Special Sensor Microwave/Imager) imagery (to be analyzed by D. Cavalieri at NASA's Goddard Space Flight Center), winds derived from surface pressure forecast fields, and meteorological data from Barrow. Determining the mean flow, its variability, and the relationship between flow and water property variations to the various forcing mechanisms will be addressed using standard statistical and time-series analysis techniques. The methods given by Cavalieri and Martin (1994) and Weingartner et al. (in prep.) will be used to analyze the satellite imagery and to compute heat and salt fluxes and the buoyant forcing. Evidence of eastward propagating shelf waves will be sought by examining the coherence and phase relationship between the CMI moorings and the Barrow Canyon moorings which are located about 250 km to the east. There has been a collaboration with Glen Gawarkiewicz and Dave Chapman of the Woods Hole Oceanographic Institution within the past year concerning the physics of dense

water outflows on arctic shelves in general and those of the Chukchi Sea in particular. They have received funding from NSF to continue dense water modeling studies with a focus on conditions representative of the Chukchi shelf. Consequently, this CMI program effectively represents one leg of a coordinated observational, modeling, and remote sensing program directed toward a better understanding of winter arctic shelf circulation processes.

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\_ 109 \_\_\_

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110

# Winter Circulation Processes in the Northeast Chukchi Sea

### **Thomas J. Weingartner**

Institute of Marine Science University of Alaska Fairbanks, AK 99775

# Abstract

Cross-shelf advection of dense water, formed as a result of salt rejection from growing sea-ice, might provide an effective mechanism by which pollutants are confined to the bottom and advected into the deep basin of the Arctic Ocean. Recent modeling efforts have contributed significantly towards understanding this process, but there are few direct observations available for comparison with the models. In the northeast Chukchi Sea dense water is formed in winter in the coastal polynyas adjacent to the Alaskan coast and flows through Barrow Canyon. A helicopter-based field program was attempted in March 1995 to obtain a suite of conductivitytemperature-depth (CTD) profiles from Barrow Canyon in order to determine the spatial structure and mixing history of a dense outflow. The data were to be compared with the model results. Unfortunately, poor weather limited the number of sampling days, and the project goals were not attained.

# Introduction

Winter oceanographic conditions on arctic shelf seas are profoundly influenced by the thermohaline effects associated with freezing and ice production. Salt is rejected from growing sea ice and increases the salinity (density) of the underlying seawater. Over shallow shelves, horizontal gradients in ice production and salt rejection result in the formation of a bottom layer of cold, saline water (dense plume). Such conditions are met on a number of arctic shelves, in particular on the Chukchi shelf (Cavalieri and Martin, 1994; Martin and Cavalieri, 1989). Here, the prevailing northeasterly winter winds form a system of polynyas along the northwest coast of Alaska. Large volumes of ice and salt are formed in these relatively narrow (5-30 km wide) features. Although the physics governing plume behavior are not well known, the plumes have important implications on shelf circulation and mixing and therefore on the dispersal of pollutants. For example, because they are strongly stratified, vertical mixing across the plume boundary is inhibited, hence pollutants incorporated into the plume are confined within a bottom layer. The horizontal density gradient provides a source of energy for flow instabilities and eddies (Gawarkiewicz and Chapman, 1995; Jiang and Garwood, 1995; Whitehead, 1993). Under the influence of gravity, the plumes and the eddies tend to flow downslope, i.e., along the bottom and across the shelf/slope margin.

III .

The dense water will sink along the continental slope, possibly mixing with ambient seawater en route, until it reaches a depth where the plume density equals that of the ambient seawater. At this depth, sinking ceases and horizontal spreading of the plume ensues.

Although this description glosses over a number of complicated issues, it illustrates the potential that this thermohaline process has for concentrating pollutants near the bottom and advecting them across the shelfbreak. As currently constructed, existing pollution transport models are primarily wind-driven and barotropic and therefore do not incorporate thermohaline physics. Moreover, the thermohaline models described above are based upon a number of assumptions and simplifications and consequently, the results need to be verified by winter observations. These have been difficult to obtain due to the logistical difficulties involved in conducting winter field campaigns in the arctic. This program sought to obtain a suite of measurements for model comparison by conducting a winter conductivity-temperature-depth (CTD) survey in Barrow Canyon on the northeast Chukchi shelf (Figure 1). The effort was geared toward determining the water mass properties, mixing potential, and spatial scales of dense shelf water



Figure 1. Bathymetric Map of Barrow Canyon and vicinity. Solid circles indicate positions of current meter moorings deployed in September 1994, and solid triangles indicate positions of CTD stations occupied between March 7 and March 10, 1995.

flowing through Barrow Canyon. The source of these dense plumes are the coastal polynyas of the northeast Chukchi Sea. Most of the flow on the northeast portion of the shelf drains through Barrow Canyon into the Arctic Ocean and over the outer shelf of the Beaufort Sea (Coachman et al., 1975). Dense plumes have previously been observed flowing down the canyon in middle and late winter (Aagaard et al., 1985; Weingartner et al., in prep.). These dense water outflows can last for one to two months. Furthermore, the density of the plumes was such that they were capable of descending to the bottom of the Canadian Basin if no mixing occurred between the plume and the ambient shelf/slope water. Theoretical arguments show that the mixing is a function of the density contrast between the plume and ambient shelf water and the magnitude of the bottom slope (Price and Barringer, 1994; Pratt, 1986). In particular, if the bottom slope magnitude exceeds  $\sim 3 \times 10^{-3}$  then the plume should begin to entrain ambient water. In agreement with this theory, Weingartner et al. showed that the plume undergoes virtually no mixing over the upper portion of Barrow Canyon where the bottom slope is  $\sim 7 \times 10^{-4}$ . They further hypothesized that if mixing occurs within the canyon, then it is initiated in the lower half of the canyon at about the position of the 200 m isobath. The objective of this program was to test this hypothesis by occupying a set of cross-canyon CTD transects along the axis of the canyon. Additional goals of the program included determining:

- 1) The spatial structure of the dense water flowing off the Chukchi shelf and through Barrow Canyon; and
- 2) The change in plume volume and density as it flows along the length of the canyon.

Measurements satisfying both objectives would be directly comparable with predictions made by the models and would therefore provide a means for verification. The CTD survey was complemented by an array of current meter moorings deployed in the canyon. The particulars of the moorings are indicated in Table 1 and their positions are shown in Figure 1. The moorings, supported by the National Science Foundation (NSF), the Office of Naval Research (ONR), and the Japan Marine Science and Technology Center (JAMSTEC) were all deployed in the fall of 1994 and were scheduled for recovery in fall 1995. The current meters collect hourly measurements of velocity, temperature, and salinity, and allow the CTD measurements to be interpreted within the context of the temporal and spatial evolution of the flow and density fields. Specifically, the moorings at the head of the canyon will provide information on the properties of the plume as it enters the canyon. The moorings deployed midway down the canyon will allow a determination of the extent to which mixing has altered the plume properties as it flows down the canyon. Both sets of moorings would provide information on the spatial structure of the dense water, although not at the spatial resolution of the CTD survey. Three cross-canyon CTD transects were planned: at the head of the canyon, midway down the canyon, and at the mouth of the

113 \_

Area	Latitude (N)	Longitude (W)	Bottom Depth (m)	Funding Agency
Upper Barrow Canyon	71° 4′	159° 29'	79	JAMSTEC <sup>1</sup>
	71°3′	159° 32′	76	NSF <sup>2</sup>
Middle Barrow Canyon	71° 25′	157° 45′	120	NSF3
-	71° 21′	157° 37'	107	NSF-JAMSTEC <sup>3</sup>
	71° 15′	157° 31′	62	NSF-ONR⁴

Table 1. Locations of Current Meter Moorings Deployed in Barrow Canyon in September 1994.

<sup>1</sup>One Acoustic Doppler Current Profile (ADCP) current meter with a Seacat 6 m above bottom. The ADCP collects current profiles averaged over 2 meter bins throughout the water column from a depth of 60 m to 12 m.

<sup>2</sup>Two Seacat/ACM pairs: 7 and 25 meters above bottom.

<sup>3</sup>Four Seacat/ACM pairs: 7, 25, 40, and 60 meters above bottom.

<sup>4</sup>Mooring consists of an Aanderaa current meter (ACM) moored above a Seabird temperature/conductivity recorder (Seacat). The instruments are vertically separated by 1 meter with the Seacats located 6 meters above the seabed.

canyon. The first two transects were established in regions where the bottom slope was less than the critical value, while the bottom slope at the mouth of the canyon exceeds the critical value.

## **Results and Discussion**

Field operations were conducted from an aluminum hut built by the Department of Search and Rescue of the North Slope Borough. The Borough's Search and Rescue helicopter airfreighted (via sling load) the hut with all the sampling and survival gear from the Barrow airport to the sampling sites. Unfortunately, inclement weather within the three-week time frame scheduled severely impeded our ability to conduct this survey. Ground blizzards, fog, and unseasonably low temperatures prevailed throughout the survey period, resulting in unsafe flying conditions. A further complication arose due to the heavily ridged nature of the ice which necessitated deviations from the planned station locations in order to find a broad and stable enough first-year ice floe upon which to work. As a consequence, only four of the approximately 30 stations planned were occupied. The position of these stations is indicated in Figure 1, and their position and sampling times are listed in Table 2.

While only four stations were occupied, the sampling was conducted safely, efficiently, and without any substantial problems. The sampling plan required all the gear and scientists to be airlifted to the site from Barrow. The helicopter would then depart after setting down the field team and return when requested to move the team to the next site. At each location a 10" diameter hole was drilled

Station Number	Date 1995	Time (GMT)	Latitude (N)	Longitude (W)	Bottom Depth (m)
1	March 7	0335	71 15.75	157 36.8	66
2	March 7	2136	71 18.7	157 38.7	93
3	March 10	2106	71 20.7	157 42.2	104
4	March 11	0135	71 24.7	157 42.1	120

Table 2. Locations and Times of CTD stations occupied in Barrow Canyon.

through the ice. A wall tent was erected over the hole with one end joined to the hut. The tent's ridgepole served as a mount for a block and the CTD was lowered through the ice hole using a line that ran through the block. The opposite end of this line was wound around the drum of an electric winch bolted to the floor of the hut. Heat to the hut and tent was generated from electric heaters and Coleman stoves. In addition, the hut contained all necessary winter survival gear and housed the electronics for a VHF radio, fathometers, and global positioning system. Electrical systems were powered by a 6.5 kilowatt portable generator. Critical to successful sampling was the ability to maintain above-freezing temperatures in the hut and wall tent so that neither the CTD nor the water samples collected in Niskin bottles froze during any phase of the operation. In particular, freezing of the CTD would lead to spurious data and possible damage to the conductivity cell.

The use of the hut made this task easy and allowed very efficient use of the time spent on the ice. The lowest ambient air temperature encountered during this project was -33°F, and hut and wall tent temperatures were easily maintained between 45° and 60°F throughout this time. In fact, it is believed that the sampling design would have easily permitted operations at ambient air temperatures to at least -50°F.

Because of the weather, sampling could not be performed in a synoptic fashion there were delays of several days between occupation of the first and last station. Sampling commenced on March 7, following the cessation of several days of strong northeasterly winds which opened a wide polynya offshore of Barrow. However, dense fog and strong northeasterly winds (30 knots) prevented safe operations on March 8 and 9. Wind conditions on March 10 were calm but temperatures had decreased to about -35°F, the approximate cutoff temperature for the Borough's helicopter. These wind changes most likely resulted in significant changes in the water mass property distribution over the transect. Thus it is difficult to interpret the results in the absence of the current meter data. Nevertheless the salinity data is plotted in Figure 2. Temperatures were at the freezing point throughout the water column and the salinities (which ranged between 31.7 and 32.5) were characteristic of the dominant winter water mass mode on



Figure 2. Contour plot of salinity across Barrow Canyon. The perspective is looking downcanyon with the westmost station (4) on the left side of the plot.

the Chukchi shelf. There is no evidence in this data of water upwelled into the canyon from the Atlantic layer of the Arctic Ocean, as occasionally happens. At approximately 95 m depth within the canyon there is a 20 m thick halocline, across which salinities increase by about 0.5. The data shows a doming of the isohalines centered on station 3, but it is suspected that this is an artifact due to the time delay in sampling at stations 2 and 3. The downward inclination in the halocline from east to west (between stations 3 and 4) is thought to be real. The tilt of the isohalines between these two stations implies that the geostrophic component of the along-canyon velocity component increases with height above the bottom. Although verification of this interpretation must await the current meter results, this interpretation does corroborate one result from Gawarkiewicz and Chapman (1995) who found a similar tilt in the isohalines associated with downcanyon flow of a dense plume.

\_ 116

### Acknowledgements

Although the goals of this program were not achieved, many individuals and entities contributed considerable effort to the project and deserve recognition. This program would not have been possible without the approval of Mayor George Ahmagoak who, on behalf of the residents of the North Slope Borough, approved the participation of the Borough and enthusiastically supported the science goals. This is a level of support by a municipal body which is believed to be unparalleled in oceanography. The Department of Search and Rescue should also be acknowledged for their support, which included helicopter logistics, the warm indoor facility necessary for the project work and gear storage, and the sampling hut. Chuck Caldwell (Director, Search and Rescue) designed the hut and assisted in numerous ways during our stay. Gordon Hutton, Jim Martin, Charles Stalker, and he piloted the helicopter during the field program. Richard Lord and Dave Knowles were the crew chiefs responsible for the sling load operation. We appreciate the expertise involved by the pilots and crews in the potentially hazardous slinging operation. The crew chiefs, along with Dan and Eli Bryant, assisted with various operations on the ground. In addition, the Department of Wildlife Management provided lodging in their Animal Research Facility and the use of a wall tent and miscellaneous equipment which defrayed costs and contributed to a safe and efficient operation. In particular, Craig George and Robert Suydam are thanked for their help in this regard. Craig George provided much advice and participated in the field operations on two occasions. Jens Leavitt kept an attentive lookout for polar bears and cheerfully assisted in all phases of the fieldwork. Richard Glenn of Barrow Technical Services provided advice on ice conditions and assistance in hiring a polar bear guard. Steve Sweet of the Institute of Marine Science assisted in Fairbanks throughout all phases of the program. Finally, John Smithhisler's tireless attention to all details contributed to a safe and reliable program.

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117 \_\_\_\_\_

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# **New Projects**

Two new CMI projects are being funded this fiscal year, along with the ongoing projects reported above. They are presented here so that the reader is aware of the latest work supported by the CMI.

# The Alaska Frozen Tissue Collection and Associated Electronic Database: A Resource for Marine Biotechnology

### Gordon H. Jarrell

University of Alaska Museum University of Alaska Fairbanks Fairbanks, AK 99775-6960

#### Abstract

The Alaska Frozen Tissue Collection (AFTC) is the primary regional archive for frozen zoological samples and a major contributor to biotechnology studies of Alaska and its waters. Advances in molecular biology are rapidly establishing the value of frozen specimens for detecting environmental change. Samples in the AFTC are essential for monitoring long-term trends in marine organisms. Though new material is acquired from several sources, important opportunities have been missed and, unfortunately, some investigators are still unaware of this resource. The objective of this project is to expand the AFTC's zoological scope of marine organisms and thereby establish the AFTC as a state-funded regional resource for monitoring long-term trends in Alaska's marine environment. The AFTC is also expanding collaborative efforts with the Alaska Marine Mammal Tissue Archival Project (AMMTAP) and other marine mammal projects throughout the state.

Support from the CMI will do three things:

- 1) Expand the scope of the collection by recruiting contributions of marine mammal, bird, fish, and invertebrate specimens from Cook Inlet, Shelikof Strait, and the Beaufort and Chukchi Seas. Specimens that are preserved now will be important baseline samples as analyses become more sophisticated and as environmental perturbation increases.
- 2) Develop an electronic database that is accessible through the internet, thus facilitating the transfer of information and sharing genetic resources among investigators.
- 3) Secure the AFTC Collection Coordinator as a permanent state-funded position thus insuring a long-term systematic record of marine populations in Alaska's marine ecosystems.

Hypotheses directly relevant to the Mineral Management Service's needs are now being tested with AFTC specimens. Two letters from researchers received since the submission of the original proposal demonstrate the potential of this resource for addressing important management questions. The genetic status of harbor seals is directly relevant to their uncertain protection status. That study has a far better chance of achieving significant results because of the availability of AFTC specimens. AFTC specimens are also being used to test several hypotheses concerning northern sea lions. Both of these species have evidenced steep population declines and their future management will figure importantly in the development of Alaska's offshore and coastal resources.

# Interaction Between Marine Humic Matter and Polycyclic Aromatic Hydrocarbons in Lower Cook Inlet and Port Valdez, Alaska

#### David G. Shaw

Institute of Marine Science University of Alaska Fairbanks Fairbanks, AK 99775-7220

### Abstract

Humic materials are a major component of naturally occurring organic matter which interact strongly with organic pollutants including polycyclic aromatic hydrocarbons (PAH). The resulting chemical associations influence the fate and effects of pollutants in the marine environment. While these factors are becoming increasingly appreciated by the environmental science and environmental management communities, there is insufficient understanding of how this interaction occurs or how changes in the chemical characteristics of humic matter influence its ability to interact with pollutants. This proposal suggests a project to provide such information directly applicable to Lower Cook Inlet and Port Valdez, Alaska. Specifically, this project will statistically examine the relationship between variations in the amount and chemical characteristics of humic acid from marine sediments with the concentrations of a suite of PAH from those same sediments. Measurements and interpretation will be coordinated with two other CMI-funded projects (S. Henrichs and J. Braddock, principal investigators) which are studying sediment sorption of PAH and microbial degradation of PAH, respectively.

# **Funding Summary**

# **Student Support**

The Cooperative Agreement specifically stressed the need for supporting education along with research. The following student support information is summarized for the ten projects presently supported by the Coastal Marine Institute (CMI):

	Funds from MMS	Match from other sources
Fiscal Year 94		
7 Master Students	\$ 67,000	\$ 37,400
2 Doctoral Students	23,000	9,200
Source Totals	\$ 90,000	\$ 46,600
Fiscal Year 95		
7 Master Students	\$ 115,400	\$ 57,200
4 Doctoral Students	59,600	12,800
Source Totals	\$ 175,000	\$ 70,000
Totals to Date	\$265,000	\$116,600

These data show a strong and growing commitment to the education of graduate students by the projects supported though the CMI.

# **Total CMI Funding**

The total MMS funding available for funding CMI projects through federal fiscal year 1995 is approximately \$2 million. Since all CMI-funded projects require a one-to-one match with non-federal monies, the total project commitment for this time period has been approximately \$4 million.

## Sources of Matching

Matching for the CMI-funded projects has come from a wide variety of sources. It was originally thought that it would be difficult to obtain \$1 million per year in matching to support CMI research. Identifying and verifying sources remains a major administrative problem in the development of the CMI proposals, but it is clear that match is available for those investigators who are willing to spend the necessary extra effort locating and securing the support. The following list of

12I .

"matching support" participants are presented to demonstrate the breadth of support that has been located:

Afognak Native Corporation Alaska Department of Fish and Game (ADF&G) Alyeska Pipeline Service Company British Petroleum Exploration (BPX) College of Natural Sciences, University of Alaska Fairbanks (UAF) Institute of Arctic Biology, UAF Institute of Marine Science, UAF Japanese Marine Science and Technology Center (JAMSTEC) Kodiak Island Borough North Slope Borough University of Alaska Fairbanks, Equipment Fund University of Alaska Museum, UAF University of Northern Iowa Water Research Center, UAF

122

Some of the CMI-funded projects are closely related to other federally funded projects which cannot be considered as match, but nevertheless augment and expand the value of a project. Related projects are funded by the National Science Foundation (NSF), the Office of Naval Research (ONR), the National Aeronautics and Space Administration (NASA), and the National Marine Fisheries Service (NMFS).

A positive relationship has continued to develop between MMS, the University of Alaska, and the State of Alaska since the formation of the CMI. All parties to the agreement as well as the residents of Alaska are expected to benefit from the cooperative research that has been and continues to be funded through the cooperative CMI effort.