

Juvenile Fish Habitat Identification and Assessment Project Submitted under the BAA

96-067BAA

Duration: 2 years

Cost FY 96: \$467,400

Cost FY 97: \$ 50,600

Geographic Area: Prince William Sound

ABSTRACT

Marine nearshore habitats are important nursery grounds for juvenile fish providing protection from predation as well other benefits. This study will sample nearshore habitats for juvenile fish. Embayments with eelgrass beds and shallow soft-bottomed coastal areas in Prince William Sound will be sampled in oiled and unoled areas. The study will help define important nursery grounds as well as demonstrate the amount to which these areas have been degraded by oiling.

INTRODUCTION

The life history of many species of marine fish includes planktonic egg and larval stages, and a juvenile stage spent in nursery grounds until the fish are big enough to move to the habitat they occupy as adults. This type of life history allows for high losses of young in the planktonic stage, but after settlement as juveniles, losses must stay low to sustain a healthy adult population. Nursery grounds are vital to the survival of juvenile fish. Any degradation in the availability or health of a nursery habitat could lead directly to heavy losses of juvenile fish and a decline in the adult population.

The nursery grounds for recently settled juvenile fish are often nearshore habitats. Nearshore marine habitats range from embayments with soft bottoms, eelgrass beds and mild currents to rocky open coasts with strong wave action. Previous studies have shown that juveniles of many species of fish tend to settle in shallow, protected and semi-protected soft bottomed habitats. These habitats, especially eelgrass beds, provide protection from predation in a relatively warm, nutrient rich and calm environment.

MBC Applied Environmental Sciences proposes a FY 96 research project to sample selected shallow nearshore habitats of Prince William Sound for juvenile fish in order to determine which habitats are important as nursery grounds and assess the amount to which some areas have been degraded as nursery grounds by exposure to oil. To sample these areas, MBC will use a specially constructed 1-m beam trawl equipped with 2.5-mm mesh netting. This trawl was designed specifically to sample for juvenile fish over soft

bottoms. The size allows for use in shallow water from a small boat, and minimizes tow path damage. All individuals of juvenile fish caught will be identified and counted.

Laboratory processing and report preparation will continue beyond the dates of field sampling. The final report of the study will be provided to the Exxon Valdez Oil Spill Trustee Council by 1 April 1997.

Follow-up research, monitoring and restoration projects may be proposed for future years if this study indicates a need. Potential future projects include: targeting specific species of juvenile (and other life stage) fish for population studies; sampling for nursery habitats over cobble and rocky bottoms; tracking recovery of degraded habitat; and habitat restoration, such as eelgrass transplants.

NEED FOR THE PROJECT

A. Statement of Problem

Larval fish often disappear out of the plankton and the individuals are not seen again until they join the adult population as small adults. It is assumed that the period during which they are absent is spent in a nursery habitat which differs from the adult habitat. The nature and availability of juvenile fish nursery grounds are, in many cases, more assumed than known. Evidence exists to support these assumptions, but local studies with the right type of sampling gear can better define the habitat.

In the case of Prince William Sound, it is critical to define local habitats. Without knowing the key fish life-stage habitats, it is impossible to determine the harm caused to fish populations by the Exxon Valdez oil spill. Once important habitats are identified, impacted and non-impacted areas can be compared, to assess the consequences of oiling and to monitor recovery.

B. Rationale

This juvenile fish habitat study will be relevant to continuing programs such as the Pacific Herring and Sound Ecosystem Assessment (SEA) projects. The broad spectrum will be helpful in identifying other species which may benefit from further, targeted investigations. Knowledge gathered may also lead to further suggestions for restoration projects, such as eelgrass transplants, to revitalize impacted nursery grounds in an attempt to improve fisheries.

C. Summary of Major Hypotheses and Objectives

General Hypothesis. Impacts nearshore habitats in Prince William Sound have reduced the nursery ground habitat available for juvenile fish.

Working Hypotheses:

Hypothesis 1. Embayments and soft-bottomed coastal areas act as nursery grounds for juvenile fish of various species.

Hypothesis 2. Oiling has degraded some nursery grounds, reducing or eliminating use by juvenile fish.

Objectives:

Objective 1. Determine which habitats are vital as nursery grounds to a variety of species of juvenile fish.

Objective 2. Assess any lingering damage to nursery habitats due to oiling.

Objective 3. Identify problems in need of further study, including recommendations for monitoring or restoration programs.

D. Completion Date

The field project is planned to occur during the Spring/Summer of FY 96. Laboratory processing and report preparation will continue beyond the dates of field sampling. The final report of the study will be provided to the Exxon Valdez Oil Spill Trustee Council by 1 April 1997.

COMMUNITY INVOLVEMENT

MBC's project plan includes rental of appropriate local boats, with a skipper and one deck hand for this project. The boats and crew will be needed for approximately two weeks each month for four months in the spring/summer of 1996. It is important that the same crew be available for all surveys, if at all possible. The crew will assist our two scientists in field collection and preparation of samples, including deck sorting of debris and sample preservation. Local knowledge of the seaways and oiling history of Prince William Sound will be essential; while the oiled and unoled embayment sites have already been identified, the coastal sites will be finalized by our scientists with the help of the local crew prior to the first survey.

FY 96 BUDGET

Personnel	193.5
Travel	18.3
Contractual	252.8
Commodities	2.8
Equipment	0
Subtotal	467.4
Gen. Admin.	0
Total	467.4

PROJECT DESIGN

A. Objectives

1. Determine which habitats are vital as nursery grounds to a variety of species of juvenile fish.
2. Assess any lingering damage to nursery habitats due to oiling.
3. Identify problems in need of further study, including recommendations for monitoring or restoration programs.

B. Methods

The hypotheses to be tested by the proposed research project are as follows:

1. Embayments and soft-bottomed coastal areas act as nursery grounds for juvenile fish of various species.
2. Oiling has degraded some nursery grounds, reducing or eliminating use by juvenile fish.

Data required to test these hypotheses will be generated by conducting field sampling at six embayments and six shallow soft-bottomed coastal areas with eelgrass beds in Prince William Sound. The project will help define important nursery grounds as well as demonstrate the amount to which these areas have been degraded by oiling.

In FY 96, MBC proposes to collect juvenile fish from twelve selected shallow nearshore stations. Three unoiled embayments (Bass Harbor, Crab Bay, and Eshamy Bay) and three oiled embayments (Herring Bay, Northwest Bay, and Sleepy Bay) are proposed as study areas which reportedly have established eelgrass beds which may serve as juvenile fish nursery areas. Three unoiled and three oiled open coast nearshore study areas will be established with the assistance of local residents knowledgeable of the seaways, oiling history, and eelgrass bed locations.

The study areas will be sampled utilizing a specially constructed 1-m beam trawl with 2.5-mm mesh netting. The beam trawl equipment is specifically designed to sample for juvenile fish over soft bottom substrates with minimal tow path damage to the study area. The beam trawl will be deployed from a small boat to enable shallow water sampling within eelgrass areas.

Three replicate, 10 minute trawls will be taken during daylight hours along two selected isobaths. The proposed replicate tows will be performed at 1-m and 3-m isobaths at each station for a proposed total of 72 samples per survey. All individuals of juvenile fish captured will be identified and counted. Since the proposed project targets only the juvenile fish catch, the larger fish caught will be sorted from the catch and returned overboard as quickly as possible. Small fish, invertebrates, and debris will be retained, fixed in 10%

buffered formalin-seawater, and returned to the laboratory.

In the laboratory the samples will be rinsed of formalin and transferred to 50% isopropyl alcohol. Juvenile fish will be sorted from the remaining catch, identified, and counted. A voucher collection will be established to provide species verification and for future reference.

MBC proposes to occupy each station for four consecutive months (June, July, August, and September). The data set generated for statistical analysis will consist of 12 stations with three replicates at each of two depths per study and four study periods. Therefore, a total of 288 samples will be analyzed. The sampling/analysis matrix will consist of 72 unoiled and 72 oiled embayment samples and 72 unoiled and 72 oiled coastal stations.

Juvenile fish data will be presented as frequency of occurrence and density (number of individuals per hectare). Catch parameters will be summarized by habitat (embayment or coastal), site, depth, and month. Catch data will be plotted against unoiled vs oiled study areas to attempt to identify juvenile fish nursery grounds and demonstrate / assess the consequences of oiling to the habitats.

The project will establish baseline data for future monitoring studies and may lead also to further suggestions for restoration projects, such as eelgrass transplants to revitalize impacted nursery grounds in an attempt to improve fisheries.

C. Contracts and Other Agency Assistance

MBC's project plan includes contracting a medium vessel, a smaller equipment deployment boat, and a two man crew from the local Seward area. Therefore obtaining the services of a local skipper and deck hand familiar with the local seaways and oiling history of the Prince William Sound survey area. The crew will assist our two scientists in finalizing the coastal study sites, field collection, deck sorting, and preparation of samples for shipment to the laboratory.

D. Location

The project will primarily encompass the western Prince William Sound area. The selected sites will typify all areas of unoiled and oiled nearshore shallow water habitats in Prince William Sound. Ultimately the communities directly benefited by increased fishery habitats will be positively affected by the increased knowledge of juvenile fish nursery grounds and which areas need to be revitalized.

SCHEDULE

A. Measurable Project Tasks for FY 96

Start-up to April 15:	Arrange logistics
April 15 - May 15:	Consult with local contractor to specify coastal sampling sites

May 15 - June 1:	Mobilize equipment and ship to holding area in Alaska
June - September:	Conduct field sampling
August - September 30:	Initiate data reduction and analysis

B. Project Milestones and Endpoints

FY 96

Start-up to April 15:	Arrange logistics
April 15 - May 15:	Consult with local contractor to specify coastal sampling sites
May 15 - June 1:	Mobilize equipment and ship to holding area in Alaska
June - September:	Conduct field sampling
August - September 30:	Initiate data reduction and analysis

FY 97

October 1 to December:	Final data reduction and analysis
December - January:	Report writing
January:	Annual workshop attended by the principal investigator
January - March:	Draft report out for review
March - April:	Report finalized and submitted
April 1997:	Annual report on FY 96 project

C. Project Reports

MBC will submit a "Final Annual Report" by April 15, 1997. The proposed study plan is not anticipated to be a multi-year project. However, knowledge gained by the proposed study may lead to further suggestions for restoration projects or additional study sites within the Prince William Sound.

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

The proposed project is a research project which may lead to restoration projects in the future. Therefore, future projects which may be pursued after completion of this initial study will address coordination and integration with other restoration projects in the future.

ENVIRONMENTAL COMPLIANCE

MBC will obtain the state and local permits required to collect and ship the samples for the proposed project. Other requirements will be addressed by contracting for local vessel and logistical support. No EA or EIS should be required for this project.

PERSONNEL

MBC proposes that Mr. Charles T. Mitchell serve as the Principal Investigator / Project Leader. Mr. Mitchell is the President of MBC Applied Environmental Sciences which he

founded in 1969. On a day-to-day basis he is responsible for directing and implementing environmental studies involving the monitoring and assessment of the effects of resource utilization on the coastal environment. He holds a BS from San Diego State University, and has been involved in graduate studies at San Diego State University, California State University at Long Beach, and the University of California at Irvine.


Prior to his position at MBC, Mr. Mitchell was on the staff of the California Institute of Technology, California Department of Fish and Game, National Marine Fisheries Service, and the Scripps Institution of Oceanography.

He has published over 20 scientific papers, and is the senior author or editor on more than 500 major reports for industry, government, and academia. Active in both the private and academic sector, he currently serves as an appointed member of the Biology Advisory Council at the California State University at Long Beach, and has served two terms as President of the American Academy of Underwater Sciences, a national organization dedicated to the advancement of scientific diving.

His major areas of expertise include habitat mitigation of coastal wetlands and kelp beds, coastal fisheries, and artificial reef ecology.

MBC proposes that Mr. Robert Moore serve as Project Leader. Mr. Moore is the Technical Coordinator responsible for all MBC field operations. He has over 15 years of experience in field sampling techniques. Mr. Moore was directly responsible for the field efforts related to a similar juvenile fish beam trawling investigation at two embayments and five coastal stations in southern California. He is responsible for supervising all field personnel, implementing specific field sampling regimes, QA/QC for all field efforts, and is experienced with various navigational location techniques.

MBC will be utilizing a wide variety of in-house scientists and technicians to assist in field, laboratory, data reduction and analysis, and in reporting. Complete resumes will be provided should the proposed project receive funding.



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April 27, 1995

Date prepared

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET
October 1, 1995 - September 30, 1996

96-047

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel		\$193.5						
Travel		\$18.3						
Contractual		\$252.8						
Commodities		\$2.8						
Equipment		\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$0.0	\$467.4	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
Indirect		\$0.0						
Project Total	\$0.0	\$467.4	\$50.6					
Full-time Equivalents (FTE)		42.0						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
Comments:								

1996

Prep Apr 1995

1 of 4

Project Number:
Project Title: Juvenile Fish Habitat Identification and Assessment Project
Submitted under the BAA
Name: MBC Applied Environmental Sciences

FORM 4A
Non-Trustee
DETAIL

1995

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET
October 1, 1995 - September 30, 1996

Personnel Costs:			Months	Monthly		Proposed
Name	Position Description		Budgeted	Costs	Overtime	FFY 1996
C. Mitchell	President / Principal Investigator		6.0	526	0	3.2
M. Mancuso	VP / QA/QC		6.0	336	0	2.0
M. Curtis	Senior Scientist		3.0	468	0	1.4
R. Moore	Technical Coordinator / Project Leader		6.0	9,889	0	59.3
D. Vilas	Laboratory Director		3.0	3,589	0	10.8
D. Johnston	Senior Technician		4.0	14,833	0	59.3
K. Morris	Technician II		4.0	4,687	0	18.7
R. Brannon	Technician II		4.0	9,657	0	38.6
D. Ram	Clerical II		6.0	38	0	0.2
						0.0
						0.0
						0.0
Subtotal			42.0	44,023	0	
Personnel Total						\$193.5
Travel Costs:			Ticket	Round	Total	Proposed
Description			Price	Trips	Days	FFY 1996
Orange Co. to Seattle / Field Crew / 2			398	4		1.6
Seattle to Anchorage / Field Crew / 2			1,067	4		4.3
Car Rental Anchorage to Seward			770	4		3.1
Travel Per Diem					112	9.3
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
Travel Total						\$18.3

1996

Project Number:
Project Title: Juvenile Fish Habitat Identification and Assessment Project
Submitted under the BAA
Name: MBC Applied Environmental Sciences

FORM 4B
Personnel
& Travel
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
Prince William Sound Charter / 2 Boats / 2 Crew to assist MBC Scientists		215.6
Postage / Communications		16.2
Telephone		0.8
Shipping / Equipment and Samples		7.2
Photocopies		0.1
Camera Rental and Film		1.7
Beam Trawls		4.6
Shipping Costs Equipment and Samples		6.6
Contractual Total		\$252.8
Commodities Costs:		Proposed
Description		FFY 1996
Field Sample Containers and Fixative		1.7
Laboratory Supplies		1.1
Commodities Total		\$2.8

1996

Project Number:

Project Title: Juvenile Fish Habitat Identification and Assessment Project

Submitted under the BAA

Name: MBC Applied Environmental Sciences

FORM 4B
Contractual &
Commodities
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET
October 1, 1995 - September 30, 1996

[illegible]

1996

Project Number:
Project Title: Juvenile Fish Habitat Identification and Assessment Project
Submitted under the BAA
Name: MBC Applied Environmental Sciences

FORM 4B
Equipment
DETAIL

Status and Potential Recovery of the Black Oystercatcher (Haematopus bachmani): An Apex Predator in the Nearshore Environment

Project Number:
 Restoration Category: Research and Monitoring
 Proposer: Richard B. Lanctot
 Lead Trustee Agency: DOI-NBS
 Cooperating Agencies: DOI-FWS
 Duration: FY96-FY99 (4 years)
 Cost FY 96: \$157.7
 Cost FY 97: \$156.8
 Cost FY 98: \$151.7
 Cost FY 99: \$87.1
 Geographic Areas: Western Prince William Sound, Glacier Bay, Kodiak Island, Southeast Alaska, Cook Inlet
 Injured Resource/Service: Black Oystercatcher (Haematopus bachmani)

ABSTRACT

This proposal questions the current status of the black oystercatcher as a recovering species, and presents a plan of action for improved monitoring of the species and evaluation of factors (e.g., demography, oil toxicity, food, genetic variability) that may be limiting recovery of the population. The species' unique role as an apex predator in the nearshore environment demands an ecosystem approach to our study that will reveal interactions among predator and prey.

INTRODUCTION

Direct mortality to black oystercatchers as a result of the *Exxon Valdez* Oil Spill (EVOS) was estimated to be 20% (280 individuals) of the population inhabiting the spill zone (Andres 1994a). Indirect effects of the EVOS have included aborted nesting, lower hatching and fledging success, ingestion of oil by adults and chicks, and slower growth rates of chicks (Andres 1994a). Population monitoring programs have revealed decreases in population abundance (Klosiewski and Laing 1994) and in breeding pair numbers on different islands throughout Prince William Sound (Andres 1994a). To enact appropriate restoration actions, we propose a study to first evaluate whether black oystercatchers are recovering, and second, to determine if oystercatchers are recovering as quickly as possible given the potential rates of population increase. We identify four factors that may be limiting recovery, including demography, oil toxicity, food availability, and genetic variation (after NVPP 1994).

Although this study focuses on one species, in many ways, it provides a window to the health of the nearshore ecosystem by studying one of the system's apex predators. Oystercatchers potentially compete with sea otters (*Enhydra lutris*), river otters (*Lutra canadensis*) and sea ducks for macrofaunal invertebrates (NVPP 1994) and are known to influence and be influenced by the structure of the nearshore vertebrate and invertebrate communities (e.g., Hahn and Denny 1989,

Falxa 1992). The oystercatcher's role as an apex predator means changes in its population directly or indirectly influence the invertebrates they prey upon, the invertebrates and vertebrates with which they compete, and the larger vertebrates that prey upon them. In our study, we will attempt to document these interactions, as we endeavor to establish workable restoration objectives.

NEED FOR THE PROJECT

A. Statement of the Problem

The status of the black oystercatcher population in PWS is debatable. An ecosystem based study, that goes beyond damage assessment, is needed to evaluate potential constraining factors limiting population recovery. Based on these findings, appropriate restoration actions need to be determined and implemented.

B. Rationale

Are black oystercatchers recovering?

Damage assessment studies (Projects Bird Study 12, R-17, 103C, 93035) conducted in 1989-1993 provide little evidence to suggest that black oystercatchers have recovered (Andres 1994a). Nevertheless, the EVOS Restoration Plan lists the black oystercatcher as a "recovering" species (EVOSTC 1994). We believe this classification was made incorrectly and needs reevaluation. According to the EVOS Restoration Plan (EVOSTC 1994), the "black oystercatcher will have recovered when Prince William Sound populations attain prespill levels and when reproductive success of nests and growth rates of chicks raised in oiled areas are comparable to those in unoiled areas."

Evidence that populations have attained prespill levels appears to be based upon an increase in the number of breeding pairs on the heavily oiled Green Island from 1989 to 1993. This island represents only a small portion of the breeding area of the black oystercatcher and does not appear to reflect population changes across Prince William Sound (PWS). Breeding pair counts on other islands in close proximity to Green Island have reflected either stable or decreasing population numbers (Andres 1994a). Also, alternative explanations, besides a population recovery, are available for the increase on Green Island. These include 1) reoccupation by oystercatchers displaced by oil spill disturbances, 2) occupation of vacated territories by previously non-breeding adults, 3) emigration of adults from other breeding locations, or 4) recruitment of immature adults into the breeding population (Andres 1994a). These explanations would reflect movement of individuals from within the **remaining** population rather than a recovering population.

The second recovery criteria compares reproductive success of nests found in oiled and unoiled areas. Nests have hatched and young have fledged at similar rates at oiled and unoiled areas (Andres 1994a). However, these comparisons are confounded by uneven predation levels. For example, nests located on unoiled Montague Island are exposed to substantially higher numbers of predators, such as the common raven (*Corvus corax*) and river otter, than nests located on oiled Green Island (Andres, unpubl. data). Thus the increased risk of predation to unoiled nests and young may bias comparisons. To date, the risk of predation has not been factored into any reproductive success comparisons (Sharp and Cody 1993; Andres 1994a, unpubl. data).

The third recovery criteria compares growth rates of chicks between oiled and unoiled areas.

In 1989 and 1990, chick mortality on Green Island was greatest on heavily oiled shorelines (Sharp and Cody 1993). Throughout 1991-93, chicks in oiled areas grew slower than chicks in unoiled areas (Andres 1994a, unpubl. data). In addition, preliminary analyses of fecal samples collected in 1992-93 indicated some chicks were being exposed to oil through contaminated food items brought to them by adults (Andres, unpubl. data). The indirect effects of lower fledgling weights and exposure to hydrocarbons may result in reduced juvenile survival and, could, ultimately lower recruitment of adults into the breeding population.

Are black oystercatchers recovering as quickly as possible?

Black oystercatcher recovery may be affected by demographics, continued exposure to oil, limited food availability, or genetic factors unrelated to food or oil. Future restoration actions will depend upon what factors are deemed to be limiting population recovery.

The rate at which black oystercatchers can recover may be constrained by a low intrinsic reproductive capacity. Oystercatchers have a low birth rate (1-3 young) and a delayed maturation period (adults begin breeding at 5 years of age; Andres and Falxa, in press). This reproductive lag means that the effects of low chick production during the initial spill years may not be seen until much later, when young would normally enter the breeding population. Sharp and Cody (1993) estimated a loss of 635 and 293 chicks in 1989 and 1990, respectively, from adults failing to breed. Fortunately, an evaluation of juvenile recruitment rate may be determined during proposed breeding pair surveys in 1996-98. Black oystercatcher juveniles banded in 1991-93 should begin entering the breeding population at that time. Also, an intensive monitoring and banding program, combined with high breeding and wintering site fidelity common to the species (Andres and Falxa, in press), will allow information on adult survival and reproductive success to be collected.

Black oystercatchers may be especially susceptible to continued oil exposure because of their complete dependence on the intertidal area for food and nesting (Andres 1994a). This area served as the deposition site for much of the *T/V Exxon Valdez* oil (ADEC 1992, Morris and Loughlin 1994). Prey items preferred by black oystercatchers, such as mussels (Andres and Falxa, in press), have been found to have chronically high levels of hydrocarbons in oiled areas (Babcock et al. 1994). Fecal samples from chicks also revealed exposure to oil through contaminated prey items (Andres, unpubl. data). Because black oystercatchers limit their activities to discrete foraging and nesting territories, evidence of oil exposure (e.g., hydrocarbon analyses) found in adults and juveniles can be easily compared to a site's historic level of oiling. Most other apex predators travel over large areas, making the level of oil exposure difficult to measure. Wintering birds may also be extremely susceptible to oil because of their dependence on protected bays and inlets. Mussel beds within these areas may retain high levels of hydrocarbons because of the lack of natural wave action (Andres, pers. obs.). The level of current and past oiling in these wintering areas can then be related to an individual's subsequent reproductive success and survival.

Many of the prey species of black oystercatchers, including limpets, chitons, and mussels were killed or contaminated by oil from the EVOS (Highsmith et al. 1993). Changes in prey type, density, and distribution of size classes as a result of oiling may explain lower growth rates of chicks seen in oiled areas (see above). Alternatively, competition among other apex predators in the nearshore region may indirectly affect food availability for foraging adults. The discreetness of oystercatcher foraging territories and the ease with which they can be monitored allows detailed measurements of prey availability and consumption (both by oystercatchers and other apex predators) to be made. Thus, food availability can be related to prey choice, survival of adults, and growth rates of chicks, while controlling for competition by other predators. Based on these relationships, a

PROJECT DESIGN

A. Objectives

1. Determine status of recovery of injured populations of black oystercatchers (Table 1).

HoA. There are no differences in population dynamics (number of breeding pairs, breeding site fidelity, juvenile recruitment) between oiled and unoiled areas.

HoB. There are no differences in reproductive capabilities (e.g., nesting effort, level of parental care, hatching success and fledging success) between pairs breeding in oiled and unoiled areas.

HoC. There are no differences in growth rates of chicks, while controlling for volume of food delivered by adults, occupying oiled and unoiled areas.

2. Determine if recovery of black oystercatcher is constrained by continued oil toxicity.

HoA. There are no differences in over-winter survival (measured by breeding site fidelity) between those adults breeding in oiled and unoiled areas.

HoB. There are no differences in over-winter survival (measured by breeding site fidelity) between those adults wintering in oiled and unoiled areas.

HoC. There are no differences between oiled and unoiled areas with respect to measures of health of adult and juvenile black oystercatchers.

HoD. There are no differences between wintering sites with respect to measures of health of adult black oystercatcher.

Corollary 1. Health indices for individual black oystercatchers sampled on wintering sites do not correlate with health indices for the same individuals on breeding sites (and vice-a-versa).

HoE. There are no relative differences in hydrocarbon concentrations in blood of chicks sampled at hatch, and when several weeks old, between oiled and unoiled areas.

3. Determine if recovery of the black oystercatcher is constrained by food availability.

HoA. There are no differences in food availability in black oystercatcher foraging territories and what is available in surrounding oiled and unoiled areas.

HoB. There are no differences in food provisioning to young, relative to what is available, between oiled and unoiled areas.

4. Determine if recovery of black oystercatcher is constrained by demographic and genetic factors unrelated to oil toxicity or food supply.

HoA. There are no differences in juvenile recruitment into the adult breeding population in oiled versus unoiled areas.

HoB. There are no differences in ages of breeding adults occupying territories in oiled versus unoiled areas.

Corollary 1. Adults may begin breeding earlier than normal (before 5 years of age) if territory vacancies occur (e.g., from direct mortality of adults due to the oil spill).

- HoC. There are no differences in the degree of genetic relatedness of individuals from different breeding locations within PWS.
- HoD. There are no differences in genetic diversity between breeding populations in PWS, Southeast Alaska and Cook Inlet.
- HoE. There are no differences in the location of winter ranges for adults that breed at different locations.

- 5. Determine appropriate restoration actions, based on answers to objectives 1-4. Possible actions may include cleaning of mussel beds, removal of competitors of food, artificially increasing hatching success, and transplanting young between islands to increase genetic diversity.

B. Methods

Below we briefly describe the general biology of the species, present general methods common to more than one objective, and finish with specific methodologies for particular objectives and hypotheses.

General Biology

Black oystercatchers are one of the few vertebrate animals completely dependent on the intertidal zone. Pairs nest just above the high tide line, and feed themselves and provision their chicks from invertebrates collected in the intertidal zone. Pairs are monogamous, long-lived, and establish well-defined, composite feeding and nesting territories that are occupied for years. Adults feed primarily on molluscs such as mussels and limpets. Chicks are dependent on their parents for food almost exclusively until fledging and may continue to receive food for >120 days after hatch (Williams 1927). Immature birds may take three or more years to become as proficient as their parents in acquiring food (Falxa 1992). Pairs in PWS abandon their breeding territories in winter; only about 25% remain in PWS (Andres 1994b). Post-breeding birds form flocks and aggregate in tidal flats of protected bays and inlets where mussel beds occur. Feeding and roosting areas such as these are often used winter after winter, possibly by the same individuals. Immature birds delay breeding for up to five years; flocks of nonbreeding and second-year birds are common in breeding areas throughout summer. Immature birds may form pre-breeding pairs and establish territories, but do not raise young. Adults feed and roost in the vicinity of their natal sites, but have not been reported breeding at their specific natal site (Andres and Falxa, in press).

Capture, banding, and blood sampling

Intensive efforts will be made to capture breeding adults and their chicks on oiled and unoiled study areas. Traps (i.e., walk-in mesh enclosures) will be placed on nests to capture adults returning to incubate, and mist nets will be used to capture adults that attack humans during brood-rearing. Nest snap-traps have been successfully used to capture black oystercatchers in British Columbia (R. Campbell, pers. comm.) and walk-in traps have been used successfully for American Oystercatchers (*Haematopus palliatus*) in Virginia (E. Nol, pers. comm.). Mist-netting has proven successful in capturing adults of other similarly aggressive shorebirds (Lancot et al. in press). Chicks will be captured at hatch in the nest site when possible, or opportunistically at a later age when found on beaches. Some chicks will be captured repeatedly to measure growth and collect blood samples at

predetermined time intervals. All adults and chicks will be weighed, measured (tarsus, culmen, wing lengths), and banded with a U.S. Fish and Wildlife Service metal band and a unique combination of color bands. Color bands will only be placed on chicks ≥ 7 days of age. Blood will also be collected from adults (5 ml) and chicks (@ 1% of body weight) by standard brachial or jugular venipuncture techniques to allow (Hoysak and Weatherhead 1991, Lanctot 1994) for hematology, petroleum hydrocarbon, and genetic assays.

Artificial Incubation

For a sample of nests found, we propose manipulating nests to increase hatching success. To avoid biasing hatching success results, we will randomly select an equal number of nests from the oiled and unoiled areas to be altered. The manipulation will require replacing real eggs with dummy eggs. Collected eggs will be artificially incubated until hatch, and young will be returned to the nest to allow adults to raise them. Artificial eggs will be made from plaster of paris and painted to mimic natural colors. This method has been used successfully on red phalaropes (*Phalaropus fulicaria*) and ruffs (*Philomachus pugnax*) (J. Dale and D. Lank, pers. comm.). Because of the potential invasive nature of this proposal, we will conduct a pilot project in 1996 in which eggs from ten nests (five from oiled and five from unoiled areas) will be treated as described above. If this method proves successful, an expanded incubation program will be instituted in 1997.

Artificial incubation of eggs has several advantages over allowing eggs to hatch naturally. First, hatching success may be increased dramatically. Studies conducted in 1991-93 indicated that 50-70% of nests hatch, and that local losses to predation can be $\geq 90\%$ (Andres, unpubl. data). Even nests with artificial eggs that suffer predation are likely to survive because predators may leave the nest after discovering fake, inedible eggs, and adults frequently continue to incubate nests after natural predators have removed or eaten a portion of the clutch (Andres, pers. obs.). Second, the increased hatching success of chicks will allow a larger sample of breeding pairs to be studied during chick maturation when the most detrimental effects from oil have been documented (Andres 1994a, Ho 1C). Third, increased hatching success of nests will allow more chicks to be sampled for blood from which health assays and genetic analyses can be conducted (Ho 2C, 4C,D,E). More importantly, these initial samples will provide a baseline level of hydrocarbon exposure with which later samples of blood from the same chicks can be compared. In this way, we can determine if chicks are ingesting hydrocarbons from food delivered by adults (Ho 2E).

Population Dynamics

Breeding

Two-person crews will search shorelines by boat or on foot to determine the presence of breeding oystercatchers. Searches will be conducted throughout the oiled and unoiled areas during four time periods between early May and mid-August to ensure all breeding attempts are recorded (e.g., failed breeders, renesters) and to document breeding success of less studied pairs. Similar counts were made in 1989 and 1991-1993 (Andres 1994a, unpubl. data). Standardized methods developed during these years will be used to conduct population comparisons among years. All locations of breeding territories found in previous years will be checked in 1996-98. The black oystercatcher's habit of reusing old nest sites (Andres and Falxa, in press) should maximize detection of breeding pairs between years. Intensive banding and periodic visits conducted in 1996-98 will provide information on breeding pair numbers, breeding site fidelity, and juvenile recruitment (Ho

1A), nesting effort, hatching and fledging success (Ho 1B), over-winter survival (Ho 2A+B), and most importantly, juvenile recruitment (Ho 4A+B). Biologists from the Alaska Maritime National Wildlife Refuge will also collect blood samples from breeding adults in Southeast Alaska (Forrester, Lowrie, and St. Lazaria Islands) and in Cook Inlet (Chisik Island) during the summers of 1996-98. These samples will provide outgroups with which health and genetic data can be compared (Ho 2C, 4D).

Wintering

Visits will be made to wintering sites within PWS during the fall (November) and winter (February) in 1995-96, and to wintering sites outside of PWS during fall and winter of 1996-97. These visits will be coordinated with foraging ecology work proposed in the Shorebird Invertebrate Predator Proposal. At each wintering site, we will record the total number of birds and the identity of banded birds (i.e. banded earlier at breeding or wintering sites). A sample of birds (15-30) will be captured with walk-in traps, cannon-nets or mist nets. Cannon nets have been successfully used to capture Eurasian oystercatchers (*Haematopus ostralegus*; Ens 1992) and mist nets have been used to capture non-breeding black oystercatchers in British Columbia (R. Campbell, pers. comm.). Captured adults will be processed as described above. Banded bird sightings and blood samples from these birds will be used in genetic (Ho 4C,D,E) and health assay (Ho 2D) comparisons.

Foraging Behavior

Within oiled and unoled areas, a sample of 10 breeding pairs each will be followed to determine feeding territory locations, and to monitor food availability, forage selection and delivery rate to young. At each feeding site, two to four variable-length transects will be placed parallel to the shoreline within the intertidal area during low tide (approximately in the location adults are seen to feed). Along each transect up to five 0.25-m² quadrats will be placed at random distances along the line. The contents of each quadrat will be collected, and the species composition and abundance of the dominant invertebrates eaten by black oystercatchers will be recorded. Further, two invertebrate species preferred by black oystercatchers (bay mussel, *Mytilus trossulus*, and horse mussel, *Modiolus modiolus*) will be quantified into age class distributions. A sample of the dominant invertebrates from each breeding territory will be collected and archived for potential hydrocarbon analyses. Analyses of archived invertebrates will only be conducted if adult or chick oystercatcher P450 blood assays demonstrate continued exposure to hydrocarbons. A comparison will be made between invertebrate presence and abundance sampled in oystercatcher foraging territories to that available in the surrounding area as measured by the larger-scale Nearshore Vertebrate Predator Project (Ho 3A).

Adult provisioning of food to young will be recorded using the protocol developed by Andres (1994a) during his studies in 1991-1993. This approach allows the quantification of delivery rate, and size, type, and biomass of prey fed young by adults. Comparisons of these parameters will then be made between pairs in oiled and unoled areas (Ho 1B+C). Information on prey type and size fed young will also be compared to food available on feeding territories (Ho 3B) and to chick growth rates (Ho 1C). Other studies have found that black oystercatchers tend to take reproductively mature mussels (i.e., medium to large animals between 25 and 45 mm in PWS; Andres and Falxa, in press). This preference may affect the recovery of the injured mussel population and indirectly affect the recovery of other vertebrate predators, such as juvenile sea otters, river otters, harlequin ducks (*Histrionicus histrionicus*), and other sea ducks that also rely on mussels for food (NVPP 1994).

Health and Exposure to Oil

Health of individuals will be evaluated through conventional hematology and morphometric measures (e.g., length, weight). Assays or biomarkers will include complete and white blood cell counts, serum chemistries, and haptoglobin assays. Exposure of oil (i.e., aromatic hydrocarbons) will be evaluated by measurements of a specific enzyme called cytochrome P450-1A. This enzyme has been used previously to document hydrocarbon exposure in birds (e.g., Rattner et al. 1993). An additional test of oil exposure will include an ELISA assay of plumage swabs. Indices of adult and juvenile health and exposure to oil will be compared among oiled and unoiled areas (Ho 2C,D,E).

The results of these studies will compliment and expand on research conducted in 1992 and 1993 involving hydrocarbon analyses of juvenile fecal samples from breeding pairs located in oiled and unoiled areas. Preliminary analyses of fecal samples indicate high variability in hydrocarbon concentrations among sites (Andres, unpubl. data). Specifics of hematology, P450, and ELISA techniques will follow the protocol described in the Nearshore Vertebrate Predator Proposal (1994) and will use the same contractual services.

Population Genetics

Estimates of the genetic diversity within, and of variation among, semi-isolated populations of breeding and wintering black oystercatchers will be determined with hypervariable minisatellite fingerprints on nuclear DNA (Jeffreys et al. 1985) and restriction fragment length polymorphism analyses on mitochondrial DNA (Awise 1986). Both methods have been used extensively for estimating genetic distance between populations (Kuhnlein et al. 1989), for understanding parentage and breeding structure (Burke and Bruford 1987, Westneat 1990, Haig et al. 1993), and tracing dispersal patterns (Rabenold et al. 1991). Genetic relatedness among and within populations are based on estimates of fragment similarities between individuals (S, Lynch 1990). Lack of independence among pair-wise *S* values will be addressed by calculating an unbiased estimate of the standard error of the mean following Lynch (1990). Kruskal-Wallis ANOVA, followed by Mann-whitney *U* tests, will be used to determine if band-sharing differed significantly within and between PWS breeding and wintering populations (Ho 4C,D,E). Wright's (1951) index of population subdivision, F_{ST} , will be used to measure the fraction of genetic diversity attributable to population differentiation using Lynch's (1991) formula. Laboratory methods are presented in detail in Bruford et al. (1992) and Sambrook et al. (1989). Both techniques have been used successfully on other avian species at the Alaska Science Center's Molecular Ecology Lab (K. Scribner, pers. comm.).

C. Contracts and Other Agency Assistance

Health assays

CBC, WBC, serum chemistries:

Cytochrome P450 immunohistochemistry:

Haptoglobin, ELISA assay:

Dr. Alan Rebar, Purdue University

Dr. Paul Snyder, Purdue University

Dr. Larry Duffy, University of Alaska, Fairbanks

Genetic analyses

Multilocus minisatellite fingerprints:

Restriction fragment length polymorphism:

Dr. Kim Scribner, NBS

Dr. Kim Scribner, NBS

D. Location

Most of the objectives proposed require comparisons between oiled and unoled areas. For this study, we chose two oiled sites located at Green Island and Northern Knight Island, and two unoled study sites at Montague and southwestern Knight Island (Fig. 1). Oiling status of these sites is based on information collected by Andres (1994a) during his intensive damage assessment studies. Each area is known to have a minimum of 10 breeding pairs. Two of these areas overlap significantly with oiled and unoled areas designated in the Nearshore Vertebrate Predator Proposal (1994), allowing maximum logistic efficiency and data sharing. Green Island was chosen especially because breeding pair increases in recent years (Andres 1994a) have been used as an indication that the species is recovering. Southwestern Knight Island is also important because declines in the number of breeding pairs have been recorded in recent years (Andres 1994a). Understanding the differences between the population dynamics of these two sites is central to several of our hypotheses. In addition, breeding adults will be captured in Southeastern Alaska (Forrester, Lowrie, and St. Lazaria Islands) and Cook Inlet (Chisik Island) by biologists at the Alaska Maritime National Wildlife Refuge (Fig. 2).

Several wintering sites in and outside of the PWS will also be visited to locate birds banded during the summer and to collect blood samples. Genetic analyses of these samples will provide information on winter site fidelity, and health analyses will provide information on winter condition. Wintering sites within PWS include Gibbon Anchorage on Green Island, Port Chalmers and Rocky Bay on northeastern Montague Island, Constantine Harbor on Hinchinbrook Island (Fig. 1). Hanning Bay, on southern Montague Island and Latouche Island will be also be surveyed for wintering flocks. Wintering sites outside the PWS include Chiniak Bay on Kodiak Island and Glacier Bay (Fig. 2).

SCHEDULE

A. Measurable Project Tasks for FY 96

Oct. 1 - Nov. 10, 1995:	Arrange logistics (boats, equipment, contracts, etc.).
Nov. 11-20, 1995:	Capture adults at wintering sites in PWS in coordination with foraging ecology work of Shorebird Invertebrate Predator Project.
Nov. 21, 1995 - March 1, 1996:	Arrange logistics; begin genetic, P450, and health analyses.
March 2- 10, 1996:	Capture adults at wintering sites in PWS in coordination with foraging ecology work of Shorebird Invertebrate Predator Project.
March 11 - May 10, 1996:	Arrange logistics; continue genetic, P450, and health analyses.
May 10-15, 1996:	Breeding surveys in PWS.
June 10-15, 1996:	Breeding surveys in PWS, capture of adults, early hatching chicks.
July 10-15, 1996:	Breeding surveys in PWS, capture reneesting adults and chicks.
Aug. 10-15, 1996:	Breeding surveys in PWS, capture late hatching chicks.
May 25 - Aug. 15, 1996:	Breeding pair work: capture of adults and chicks as needed; measurements of food availability, adult feeding, and food delivery to chicks by adults; food choice by adults; chick growth and survival; incubation of eggs to enhance hatching success.

Aug. 16 - Nov. 10, 1996:	Compilation and analyses of data; continue genetic, P450, and health analyses from breeding population, logistics for future winter work.
Nov. 11-31, 1996:	Capture adults at wintering sites at Kodiak Island and Glacier Bay.
Dec. 1, 1996 - April 15, 1997:	Analyze data; continue genetic work as needed, write annual report on FY 96 work.
January 1997:	<i>Reporting of project findings at Restoration Workshop.</i>
April 15, 1997:	<i>Submission of 1996 Progress Report.</i> Summary of health issues and genetic diversity within PWS completed (Objective 2, HoC; Objective 4, HoC).

B. Project Milestones and Endpoints

May - August, 1997:	Breeding surveys throughout PWS and specific breeding pair work as described above.
August 1997 - April 1998:	Summarize and analyze data to write annual report on FY96-97 work: genetic work completed (Objective 4, HoC+D).
January 1998:	<i>Reporting of project findings at Restoration Workshop.</i>
April 15, 1998:	<i>Submission of 1997 Progress Report.</i>
May - August, 1998:	Breeding surveys throughout PWS and specific breeding pair work as described above.
August 1998 - April 1999:	Summarize and analyze data to write final report on FY96-98.
April 15, 1999:	<i>Submission of Final Report.</i> All objectives accomplished.

Note: In December 1997, a full review of the 1996 and 1997 data will be used to assess the status of black oystercatcher recovery and to allow for changes in the study design (e.g., modified, and/or additional fieldwork). If the species meets all recovery aspects as designated in the EVOS Restoration Plan (EVOSTC 1994), the project would move into final data analysis and report mode.

C. Project Reports

January, 1997 and 1998	Report findings at annual Restoration Workshop.
April 15, 1997 and 1998	Annual reports submitted to Chief Scientist
April 15, 1999	Final report submitted to Chief Scientist

If possible, publication of project results in peer-reviewed journals will be substituted for portions of annual or final reports.

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

Every attempt was made to integrate this project with previously funded research projects. Intensive consultations with principal investigators of the Nearshore Vertebrate Predator Project led to the selection of similar study sites, sharing of data where possible, and common use of contractual services (e.g., oil exposure and health assays). Existing invertebrate data specific to black oystercatcher winter and breeding territories will be obtained through Project 95320, PWS

Investigation. Genetic analyses will be conducted at the Alaska Science Center's Molecular Ecology Lab, NBS, through a collaboration with Dr. Kim Scribner. Logistical expenses for the PWS winter ecology work will be shared with the Shorebird Invertebrate Predator Project. Coordination through subject-specific workshops (e.g. Intertidal) and the annual EVOS Restoration Workshop will facilitate further exchange of information and identification of further opportunities for project collaborations.

Research on black oystercatcher from 1989-93 was conducted through the U.S. Fish and Wildlife Service (USFWS) via EVOS funding (Projects Bird Study 12, R-17, 103C, 93035). Brad Andres, the principal investigator of this project, will continue to work part-time on this project providing historic information and continuity to the current study. The USFWS will also provide extensive logistical supplies. Information from the past and current project will be exchanged to promote the greatest understanding of the species.

The Alaska Science Center, NBS, has an extensive shorebird research program dating back to 1976. Studies have focused on both breeding and post-breeding shorebirds, mostly in littoral and coastal regions of Alaska. This extensive background will be invaluable to conducting the black oystercatcher study. NBS will also provide extensive logistical support in the field and office.

ENVIRONMENTAL COMPLIANCE

This project is subject to approval by the Alaska Science Center's Animal Care and Welfare Committee to ensure compliance with the provisions of the Animal Welfare Act. Special Use Permits will be requested from appropriate state or private organizations to work throughout PWS. Appropriate environmental impact statements will be written where necessary. The majority of field work is non-intrusive and relies primarily on observations. The proposed capture and sample collection techniques have been used routinely in shorebirds and are considered very safe (Hoysak and Weatherhead 1991, Lanctot 1994). Application of tarsus leg bands will follow the guidelines of the U.S. Fish and Wildlife Service, Migratory Bird Banding Lab, Laurel, Maryland.

PERSONNEL

Project Leader/Manager

Mr. Richard Lanctot -- Ph. D. candidate. Research Wildlife Biologist at the Alaska Science Center, NBS. His background includes 7 years experience studying shorebird breeding biology, including studies on Spotted Sandpipers, Piping Plovers, Bristle-thighed Curlews, and Buff-breasted Sandpipers. Three of those years were spent investigating the impacts of oil development on shorebirds at Prudhoe Bay. He also has 2 years practical experience in genetics research. Publications on reproductive biology of shorebirds, and census and blood sampling techniques.

Collaborators

Mr. Brad Andres -- Ph. D. candidate. Studied the effects of the EVOS on black oystercatchers breeding in PWS, Alaska from 1991-93, and has authored several damage assessment and restoration reports. He is currently coordinating migratory landbird and shorebird programs and conducting field projects within Region 7 of the USFWS. Prior to

working on black oystercatchers, he studied the potential impacts of oil development on migrating shorebirds on the North Slope of Alaska. Publications include papers on natural history, habitat requirements and management of shorebirds.

Dr. Kim T. Scribner -- Project Leader, Molecular Ecology Laboratory, Alaska Science Center, NBS. He has over 15 years of research experience and has published extensively in the fields of population genetics and ecological genetics.

Mr. Robert E. Gill, Jr. -- Supervisory shorebird biologist with the Alaska Science Center, NBS. He has studied shorebird breeding and migration ecology in most parts of Alaska for almost 30 years. His extensive publication record includes papers on conservation strategies, migration patterns, and breeding ecology of shorebirds.

Ms. Leslie Slater -- Wildlife Biologist, Alaska Maritime National Wildlife Refuge, USFWS. Her background includes over 15 years of research and management experience in Alaska primarily studying seabirds in the Aleutians, and in Southcentral and Southeastern Alaska.



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Date prepared

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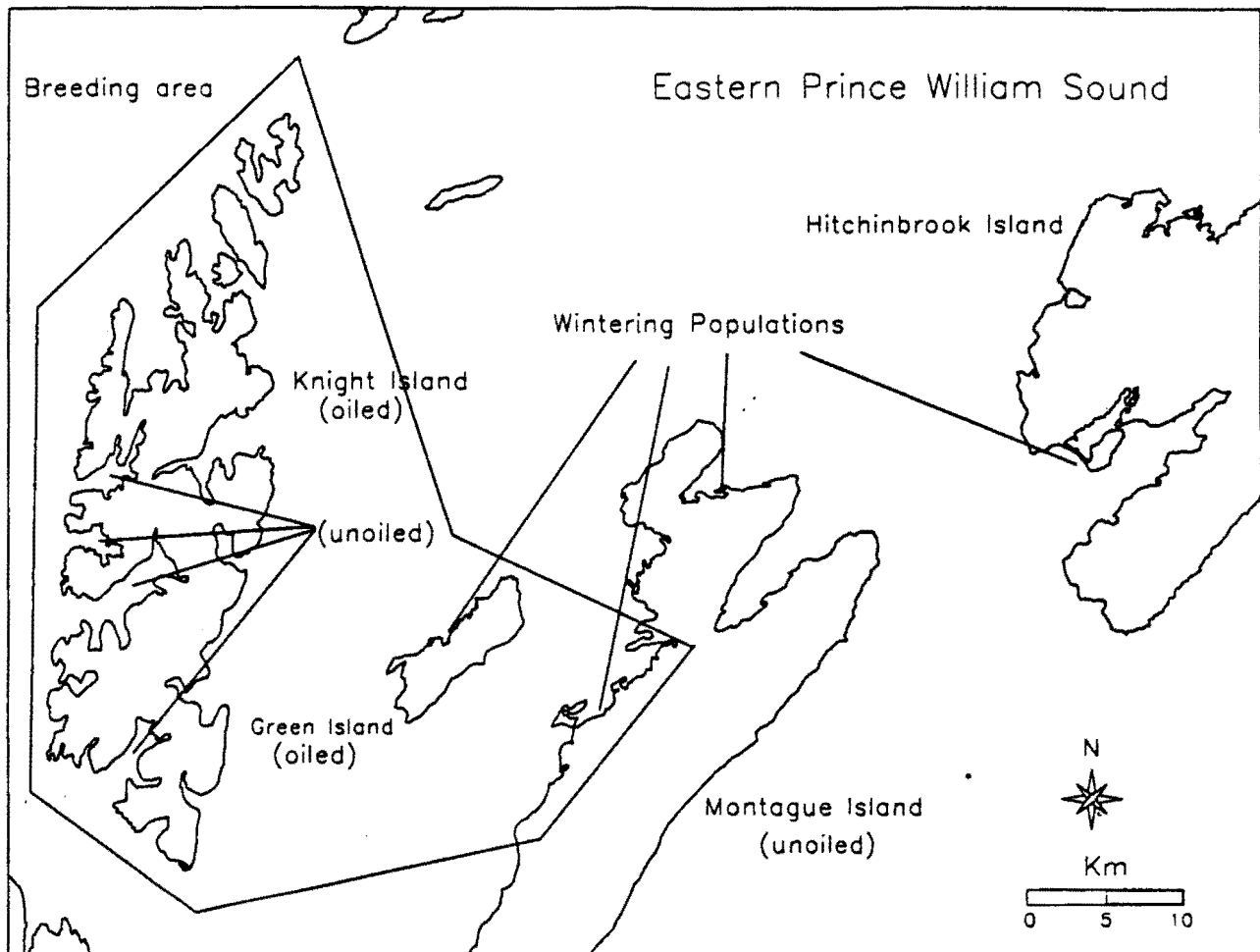


Figure 1. Location of breeding and wintering study areas for black oystercatchers in Prince William Sound, Alaska.

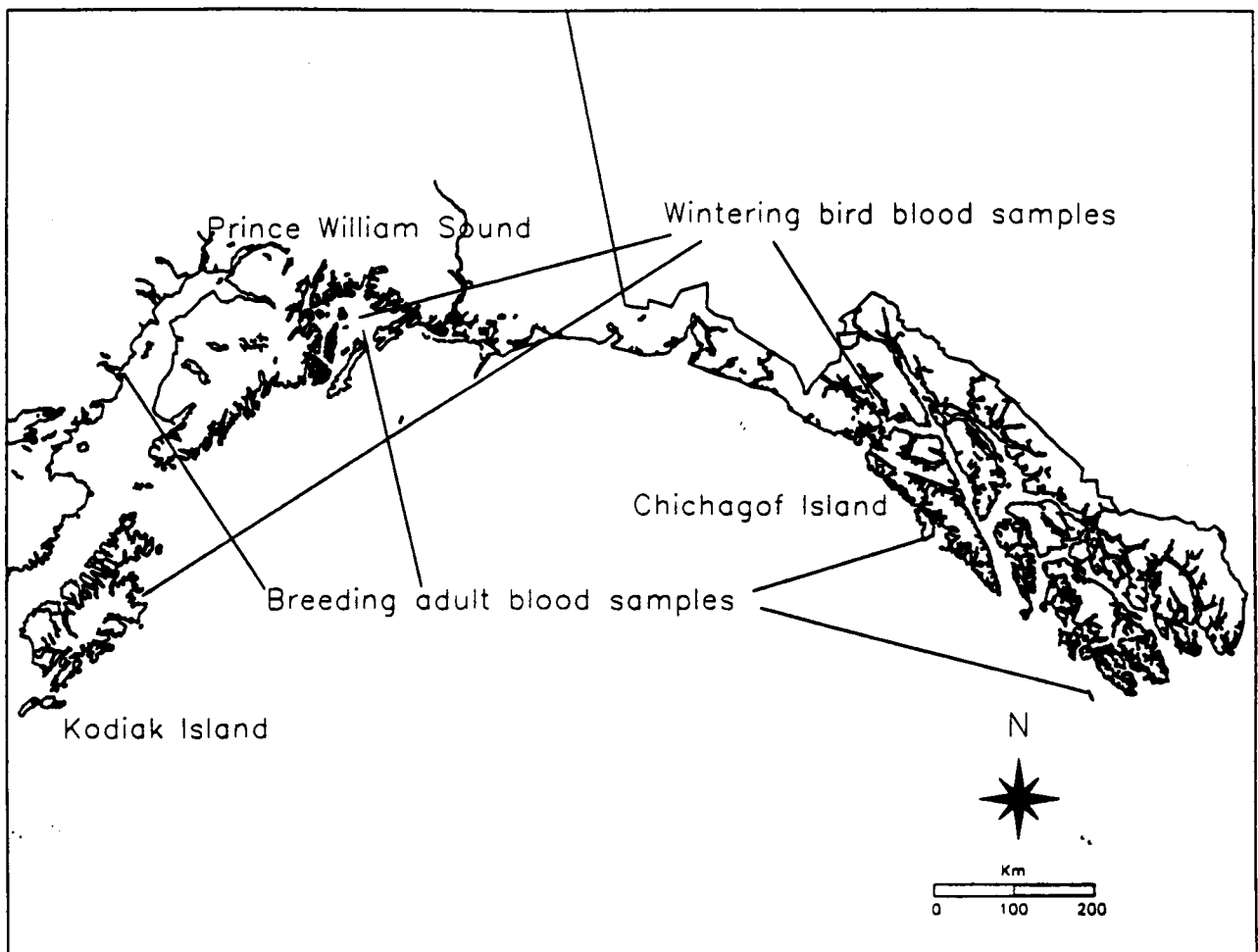


Figure 2. Locations of sites for collecting blood of breeding and wintering black oystercatchers, southeastern and southcentral Alaska.

Table 1. Injury and evidence for recovery of the black oystercatcher from the *Exxon Valdez* Oil Spill, 1989: demographic, oil exposure, trophic, and genetic factors.

Demographic

- Nine direct adult mortalities documented; oiled chicks found in 1989 (Sharp and Cody 1993).
- Andres (1994a) estimated 20% (280 individuals) of the Prince William Sound population died as a result of the EVOS.
- Winter and summer populations of oystercatchers were lower than expected in the oiled area of Prince William Sound when comparing pre- (1972-73) and post-spill (1989-91) data. A similar finding was found when comparing summer populations between 1984 to 1989-91 (Klosiewski and Laing 1994).
- Oystercatchers increased at a greater rate in the oiled zone than the unoiled zones of Prince William Sound during winter surveys from 1991 to 1993. However, the small number of birds sighted (1-2) makes this analyses biologically unmeaningful. Similar comparisons of population estimates during the summer showed no changes (Agler et al. 1994).
- Total number of breeding pairs increased on Green Island, remained the same on Montague Island, and decreased on Knight Island, from 1989 to 1993 (Andres 1994a).

Oil Exposure

- Hatching success increased on the heavily oiled Green Island between 1989 and 1992 (Andres 1994a).
- Brood loss was greater for pairs nesting in oiled areas in 1989, but did not differ between oiled and unoiled areas in 1991 (Andres 1994a).
- Weight gain of chicks raised at oiled sites was slower than for chicks raised at unoiled sites in 1991-93 (Andres 1994a, unpubl. data).
- Fledging success was lower on oiled Green Island relative to unoiled Montague Island in 1989 (Andres 1994a).

Trophic

- Mussels were smaller, died at a higher rate, and had higher levels of petroleum hydrocarbons on oiled Green Island than on unoiled Montague Island in 1989 (Sharp and Cody 1993, Andres 1994a). Persistent *Exxon Valdez* crude oil in dense mussel beds may pose a chronic oil exposure problem to breeding and wintering oystercatchers (Babcock et al. 1994).
-

Table 1. (continued)

- Black oystercatcher tend to take reproductively mature mussels (medium to large size; Andres and Falxa, in press). This preference may affect the recovery of mussel populations and indirectly affect the recovery of other nearshore vertebrate predators that also rely on mussels for food (NVPP 1994).

Genetic Structure

- The small population size of the black oystercatcher in Prince William Sound (ca. ≤ 1000 ; Agler et al. 1994) combined with high breeding and wintering site fidelity, and limited juvenile dispersal, may isolate breeding populations and result in inbreeding depression (Lacy 1987).
 - No genetic information is available on the species at present.
-

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996	PROPOSED FFY 1996 TRUSTEE AGENCIES TOTALS					
			ADEC	ADF&G	ADNR	USFS	DOI	NOAA
							\$157.7	
Personnel	\$0.0	\$89.9						
Travel	\$0.0	\$6.4						
Contractual	\$0.0	\$20.6						
Commodities	\$0.0	\$17.0						
Equipment	\$0.0	\$8.9						
Subtotal	\$0.0	\$142.8	LONG RANGE FUNDING REQUIREMENTS					
General Administration	\$0.0	\$14.9	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
Project Total	\$0.0	\$157.7	\$156.8	\$151.7	\$87.1	\$0.0	\$0.0	\$0.0
Full-time Equivalents (FTE)	0.0	2.6						
			Dollar amounts are shown in thousands of dollars.					
Other Resources	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0
Comments: The Alaska Science Center, National Biological Service, is responsible for coordinating all aspects of this project. Responsibilities include field work logistics, collection of field data and blood samples, shipment of blood samples to appropriate contractual agencies, data analyses, report writing, and publication of results. The US Fish and Wildlife Service will provide personnel and expertise to conduct the field work, provide logistical support for conducting the field work, and help in the preparation of reports.								

1996

Project Number:
Project Title: Status and potential recovery of the Black Oystercatcher (Haematopus bachmani): An apex predator in the nearshore environment.
Lead Agency: DOI - NBS

FORM 2A
PROJECT
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel		\$7.7						
Travel		\$0.6						
Contractual		\$2.5						
Commodities		\$0.3						
Equipment		\$0.0						
Subtotal	\$0.0	\$11.1	LONG RANGE FUNDING REQUIREMENTS					
General Administration		\$1.3	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
Project Total	\$0.0	\$12.4	\$14.7	\$11.9	\$10.0			
Full-time Equivalents (FTE)		0.2						
Dollar amounts are shown in thousands of dollars.								
Other Resources see below								
<p>Comments: Other Resources: Office supplies, computer equipment, secretarial assistance, and budgeting will be provided through the Migratory Bird Management Branch, DOI-FWS.</p> <p>Damage assessment work on black oystercatchers was funded in 1991-93 as Project numbers R-17, 103C, 93035.</p> <p>\$6,900 of total costs are for report writing, NEPA compliance, and community interaction activities.</p> <p>Program management will be conducted through the DOI - NBS lead agency.</p>								

1996

Project Number:
 Project Title: Status and potential recovery of the Black Oystercatcher
 (Haematopus bachmani): An apex predator in the nearshore environment.
 Agency: DOI-FWS

FORM 3A
 AGENCY
 PROJECT
 DETAIL

October 1, 1995 - September 30, 1996

<p>1996</p>	<p>Project Number: Project Title: Status and potential recovery of the Black Oystercatcher (<u>Haematopus bachmani</u>): An apex predator in the nearshore environment. Agency: DOI - FWS</p>	<p>FORM 3B Personnel & Travel DETAIL</p>
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1996 EXXON VALDEZ TRUST: COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
Equipment maintenance - outboards, camp supplies		1.5
Report writing, publication costs		1.0
When a non-trustee organization is used, the form 4A is required.		
Contractual Total		\$2.5
Commodities Costs:		Proposed
Description		FFY 1996
Food in camp (30 person days at \$10)		0.3
Commodities Total		\$0.3

1996

Project Number:
Project Title: Status and potential recovery of the Black Oystercatcher
(*Haematopus bachmani*): An apex predator in the nearshore environment.
Agency: DOI -FWS

FORM 3B
Contractual &
Commodities
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1996
Description				
				0.0
				0.0
				0.0
				0.0
				0.0
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				0.0
Those purchases associated with replacement equipment should be indicated by placement of an R.		New Equipment Total		\$0.0
Existing Equipment Usage:		Number of Units	Inventory Agency	
Description				
25' Boston Whaler fully outfitted		1	DOI-FWS	
Inflatable boats and outboards		1	DOI-FWS	
Camp gear (tents, sleeping bags, cook kits, etc.)		4	DOI-FWS	
Float coats/survival suits		4	DOI-FWS	
Weatherport		1	DOI-FWS	
Hand held VHF radios		2	DOI-FWS	
Propane freezer		1	DOI-FWS	
Generator		1	DOI-FWS	
Banding supplies (mist nets, bands, scales, rulers, etc.)		1	DOI-FWS	

1996

Project Number:
 Project Title: Status and potential recovery of the Black Oystercatcher
 (Haematopus bachmani): An apex predator in the nearshore environment.
 Agency: DOI-FWS

FORM 3B
 Equipment
 DETAIL

1996 EXXON VALDEZ TRUS COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel		\$82.2						
Travel		\$5.8						
Contractual		\$18.1						
Commodities		\$16.7						
Equipment		\$8.9	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$0.0	\$131.7	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
General Administration		\$13.6						
Project Total	\$0.0	\$145.3	\$142.1	\$139.8	\$77.1			
Full-time Equivalents (FTE)		2.4						
Dollar amounts are shown in thousands of dollars.								
Other Resources see below								
<p>Comments: Accounting, personnel hiring, purchasing, secretarial assistance, computer equipment, and other office supplies will be provided by the DOI-NBS.</p> <p>Damage assessment work on black oystercatchers was funded in 1991-93 as Project numbers R-17, 103C, 93035. \$27,600 of total costs are for report writing. NEPA compliance, and community interaction activities. Program management will be conducted by the project leader - R. Lancot of the DOI - NBS.</p>								

1996

Project Number:
 Project Title: Status and potential recovery of the Black Oystercatcher
 (Haematopus bachmani): An apex predator in the nearshore environment.
 Agency: DOI-NBS

FORM 3A
 AGENCY
 PROJECT
 DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Personnel Costs:			GS/Range/ Step	Months Budgeted	Monthly Costs	Overtime	Proposed FFY 1996
PM	Name	Position Description					
*	R. Lancot	Project leader/manager - lead field work during summer, responsible for all aspects of project	GS/11/1	12.0	4,300		51.6
							0.0
							0.0
	Biotechnician	assist in summer field work	GS/5/1	4.0	1,800		7.2
	Biotechnician	assist in summer field work	GS/5/1	4.0	1,800		7.2
	Biotechnician	assist in summer field work	GS/5/1	4.0	1,800		7.2
	Genetics technician	conducts genetics research	GS/5/1	5.0	1,800		9.0
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1996

Project Number:
 Project Title: Status and potential recovery of the Black Oystercatcher (Haematopus bachmani): An apex predator in the nearshore environment.
 Agency: DOI-NBS

**FORM 3B
 Personnel
 & Travel
 DETAIL**

1996 EXXON VALDEZ TRUST COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
Equipment maintenance - outboard motors, binocular and scope repair and cleaning, field gear repair and cleaning		1.5
Freight - shipment of field gear to Prince William Sound		0.5
Genetic analyses		5.6
Hematology and oil exposure assays		7.5
Publication costs		1.0
Statistical consulting		2.0
When a non-trustee organization is used, the form 4A is required.		
Contractual Total		\$18.1
Commodities Costs:		Proposed
Description		FFY 1996
Food		5.1
Computer software		1.0
fuel for boats		7.0
Blood, banding, boat safety supplies		1.1
rain gear, waders, hip boots		1.3
film supplies, color copying, rite-in-rain paper		0.6
Miscellaneous supplies		0.6
Commodities Total		\$16.7

1996

Project Number:
 Project Title: Status and potential recovery of the Black Oystercatcher
 (Haematopus bachmani): An apex predator in the nearshore environment.
 Agency: DOI-NBS

FORM 3B
 Contractual &
 Commodities
 DETAIL

October 1, 1995 - September 30, 1996

1996	Project Number: Project Title: Status and potential recovery of the Black Oystercatcher (<u>Haematopus bachmani</u>): An apex predator in the nearshore environment. Agency:	FORM 3B Equipment DETAIL
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HERRING REPRODUCTIVE IMPAIRMENT

Project Number: 96074
 Restoration category: Research
 Proposer: National Marine Fisheries Service
 Lead Trustee Agency: NOAA
 Cooperating Agency: State of Alaska, Department of Fish and Game, Division of Commercial Fisheries
 Duration: 2 years
 Cost FY 96: \$200,000
 Cost FY 97: \$69,500
 Geographic Area: Prince William Sound, and Southeast Alaska: includes laboratory research at Auke Bay, Juneau, Alaska
 Injured Resource: Herring

ABSTRACT

The purpose of this study is to examine the possibility of long term oil impacts on herring due to the *Exxon Valdez* oil spill using intense field and laboratory measurements. In FY 95, the study was partitioned into a field component (designed to search for possible reproductive impacts in PWS herring stocks caused by the *Exxon Valdez* oil spill) and a laboratory component (designed to determine if exposure of various life stages to oil can cause long term genetic damage), but in FY 96 only the field component will continue. This project was initiated in 1994 following the crash of herring populations in PWS, and represents the toxicological component of a suite of projects focused on causes of the crash and prospects for recovery.

INTRODUCTION

The 1993 crash in Prince William Sound herring resources stimulated a multi-disciplinary suite of studies to look at toxicological and ecological factors affecting long term recovery of the stocks. This proposal represents the toxicological part of an inter-agency herring package.

Herring stock in Prince William Sound (PWS) may have been reproductively impaired by the 1989 *Exxon Valdez* oil spill, and it is feared that continuing long-lasting effects could hamper restoration of the stocks that have crashed since the spill. Most or all of the life stages of herring may have been exposed to oil after the 1989 *Exxon Valdez* spill in PWS. Only a small proportion of fish from oiled areas showed significant histopathological damage in 1989 and 1990, but over 40% of the spawning areas were oiled (Brown et al. 1994). Many herring larvae collected from the water column exhibited morphological malformations, genetic damage, and small size that may have been caused by exposure to oil (Norcross et al. 1997), but when this damage was caused was not known (i.e., in pre-spawning adults, eggs, or larvae). Abnormal larvae were found as late as July, 1989 (Norcross et al. 1994), thus raising the possibility that abnormal larvae can survive for considerable periods of time (hatch in 1989 was in early May (Brown et al. 1994)), or that direct exposure of larvae to oil can lead to developmental

abnormalities. Exposure of herring eggs to petroleum hydrocarbon concentrations frequently results in abnormal larvae with poor survival potential (Kuhnold 1969; Linden 1976; Rosenthal and Alderdice 1976; Pearson et al. 1985; Kocan 1993). In the pectoral fins of herring embryos exposed to oil, anaphase aberrations were elevated (Hose et al. in prep.), giving some credence to the hypothesis that long term genetic damage was possible to the germ line. Some field research (Baker and Biggs, 1993; Kocan et al. 1994) suggests reproduction may have been impaired by previous exposure of adult or juvenile herring to oil in the water column, but with many uncontrolled (and unknown) factors, it is difficult to interpret these field data. Because year-class strength is heavily influenced by survival of herring larvae (Stevenson 1949; Taylor 1964; Outram and Humphreys 1974), contamination of pre-spawn adults, eggs, or larvae by petroleum hydrocarbons may have an adverse impact on herring populations.

Long term impacts remain a question, although immediate impacts were measured in the 1989-91 damage assessment studies. Recruitment failures appear to have continued and standing biomass has decreased; the toxicological influence of oil on either is unknown. The purpose of this study is to examine the possibility of long term impacts. Reproductively ripe adult herring will be collected from multiple sites in PWS, plus control sites in Southeast Alaska, and spawned by age class to determine if residual effects of the *Exxon Valdez* oil spill still persist in PWS stocks. Measurements include egg fertility, viability, and hatch timing, morphological abnormalities in the larvae and anaphase aberrations in the pectoral fins of larvae.

Time line. This four year project started in FY 94 with laboratory exposures to adult herring and impact measurements on larvae, and continued in FY 95 with direct exposure of incubating eggs to oil plus the first year of field observations. In FY 96, the oil toxicity laboratory studies will be closed out with a final report, and the field component will continue for a second (final) year. Table 1 gives the time line of the project.

Table 1

LABORATORY:

Year	Exposure	Measurement
FY94	pre-spawn adults	impacts on larvae
FY95	eggs	impacts on larvae
FY96	-	final report

FIELD:

Year	Exposure	Measurement
FY95	-	spawn viability by area and year class
FY96	-	spawn viability by area and year class
FY97	-	final report

Accomplishments in FY 94

In 1994 gravid adult herring were exposed to oil in water for a period of 8 or 16 days to determine the presence or absence of direct toxic effects, damage to gonads, reduced resistance to disease, and

heritable genetic damage in the progeny. Because it was not practical to measure germ line damage directly in the laboratory, efforts were focused on detection of chromosomal damage in the actively dividing somatic cells in the pectoral fins of larvae. Prediction of impacts on meiosis, therefore, was based on the premise that genetic damage in somatic cells would be correlated with germ line damage.

Exposure of gravid adult herring did not cause direct toxic effects to progeny, and gonads/gametes were not measurably damaged (as judged by egg fertility). Herring in this experiment were reproductively similar to those in PWS at the time of the *Exxon Valdez* oil spill. Arguably, gonads might be damaged if exposure had occurred earlier in maturation, such as at the time of gamete formation, but was beyond the context of the experiment. Resistance to disease in adults was decreased by exposure to oil, and mortality occurred in adults proportionate to oil dose. It is, therefore, plausible that disease resistance in herring populations in PWS was significantly reduced by exposure to oil, and that epizootics observed after exposure were an indirect manifestation of the spill. There was no indication that exposure of adult herring to oil caused chromosomal damage in somatic cells in progeny, and we infer that heritable genetic damage caused by exposure of adults was unlikely.

Accomplishments in FY 95

The FY 95 research and analysis is in progress. Adults were collected and spawned from seven sites (four within PWS and three in Southeast Alaska). Hatching was completed by mid June; tens of thousands of larvae were collected and will be examined in the next 4 months to determine size at hatch and abnormality rates. In the laboratory, two oil exposure experiments were completed: exposure caused premature hatch, reduced survival, and obvious morphological aberrations. When examination of preserved specimens is completed, we expect to confirm oil-induced increases in somatic chromosomal aberrations.

Proposal for FY 96

We propose to continue the field observational study for a second year to survey PWS herring stocks for evidence of reproductive damage caused by the *Exxon Valdez* oil spill. This will give two brood years of measurement for evaluation of long-term damage as expressed in reproductive impairment. We will close out the laboratory oil exposure part of the project with a final report.

NEED FOR THE PROJECT

A. Statement of Problem

Herring stock in Prince William Sound (PWS) may have been reproductively impaired by the 1989 *Exxon Valdez* oil spill, and it is feared that continuing long-lasting effects could hamper restoration of the stocks that have crashed since the spill. Most or all of the life stages of herring may have been exposed to oil after the 1989 *Exxon Valdez* spill in PWS. Major recruitment failures have occurred since the spill and standing biomass has decreased; the toxicological influence of oil on either is unknown. The standing stock in PWS crashed in 1993, thus herring are currently classified as not recovering. The stock crash in 1993 suggests the possibility that long term damage may have been a contributing factor in the crash or to lack of recovery. This study examines the toxicological basis for long term damage,

and seeks lingering evidence of such damage in Prince William Sound stock(s) by measuring reproductive impairment.

B. Rationale

The project will yield improved understanding of the impacts of the *Exxon Valdez* oil spill on the herring population in PWS, and add to the information base on the current status of reproductive fitness compared to other stocks. The monitoring portion of the study should continue in FY 96 before fish present at the time of the spill are all eliminated from the population through disease, predation, and senescence.

C. Summary of Major Hypotheses and Objectives

The goal of this study is to determine if herring reproduction in PWS stocks may have been impaired as a result of past oil exposures at one or more life stages. The primary test hypothesis is that fertility, percent hatch, larval viability, morphological abnormalities, and genetic aberrations were caused by the *Exxon Valdez* oil spill. These key reproductive parameters have been measured in 1995 and will be in 1996 from spawn taken from PWS and Southeastern Alaska control sites; multiple year classes will be sampled from each site. These key reproductive parameters have been measured in controlled laboratory oil exposures to adults and eggs in previous years.

D. Completion Date

Expected completion of the FY 96 field reproductive impairment research and report is April 15, 1997. The laboratory toxicology tests will be closed out and a final report submitted June 1, 1996.

COMMUNITY INVOLVEMENT

This project involves the collective local knowledge of the ADF&G employees living in Cordova and several towns in Southeast Alaska. Local hires have been and will be accomplished as needed to meet project goals.

FY 96 BUDGET

Personnel	73.3
Travel	23.6
Contractual	72.8
Commodities	14.2
Equipment	0.0
Hydrocarbon analyses	0.0
Subtotal	183.9
Gen. Admin.	16.1
Total	200.0

PROJECT DESIGN

A. Objectives

Survey herring in PWS for reproductive impairment by measuring larval viability by location and age class. Herring reproduction may have been impaired as a result of past oil exposures at one or more life stages. This is a continuation of the survey begun in 1995 to measure herring reproduction success from several age classes collected from several sites in PWS. Some of the age classes were exposed to oil, but post-1990 year classes were not. Spawn will be returned to the lab and reared until hatch to determine larval viability and abnormality rates. Additional eggs will be similarly collected and spawned from sites in Southeast Alaska as controls.

Primary test hypothesis: Fertility, percent hatch, larval viability, morphological abnormalities, and genetic aberrations were caused by the *Exxon Valdez* oil spill.

Assumptions: 1) The year classes prior to 1989 were potentially exposed to oil in 1989, 2) the 1989 year class was potentially exposed to oil in pre-spawning adults, eggs, and larvae, and 3) year classes after 1989 were not exposed to oil, except that the 1990 year class may have been exposed to residual oil in intertidal areas.

Controls will be post-spill year classes and two or three sites in Southeastern Alaska, including Sitka.

B. Methods

Observation requires extensive incubation of eggs, isolated by female, with monitoring for 30 to 40 days. We have the capacity to incubate eggs from approximately 950 individual females and will involve six technicians in addition to staff biologists at peak hatch. Some hatch staggering is possible, but spawning events are driven by reproductive condition of the fish and are site specific.

Procedures for collection, spawning, and transportation used in previous years will be followed. Reproductively ripe adult herring will be captured by cast net or gill net at four spawning sites in PWS and two with Southeast Alaska, separated by length into age classes, iced, transported to a laboratory facility, and spawned within 8 h of collection. Concurrent with our efforts at some sites, ADF&G will collect samples for age, weight, length, and disease. For age analysis, scales will be collected from each of 25 spawned females plus males providing milt. Ovarian membranes will be cut longitudinally and the eggs removed with a stainless steel spatula. From each female, eggs will be deposited on one 25 x 75 mm glass slides placed in seawater at the bottom of a shallow container. Approximately 100 eggs per slide will be deposited with gentle swirling. Eggs from each female will be placed in a staining rack and suspended in a beakers of seawater. Milt from 3 males will be prepared by cutting sections of testes into small segments. A few milliliters of milt will be added to beakers containing eggs. The eggs and milt will remain in contact 5 min. The eggs will be transported by air to Auke Bay in chilled seawater. Eggs will be suspended in seawater from monofilament line attached to arms driven by an offset cam to cause slow movement (1 rpm) through the water. Lighting will be natural, supplemented by overhead fluorescent light during daylight hours. Excess eggs will be removed from all slides by scraping, i.e., those along slide margins susceptible to mechanical damage, and clumps of eggs where not all eggs were exposed to water. This processing will be accomplished in water with a minimum of emersion. Eggs

will be counted to quantify fertilization success and stage development will be noted. Approximately one week before hatch, eggs from each female will be isolated in 1000 ml glass jars; slides will be held with plastic clamps and suspended with monofilament line from a mobile overhead rack. Jars will be placed in a flowing seawater bath. Hatch timing, hatching success, larval viability, and larval abnormalities will be observed daily for each fish. Hatched larvae will be assessed for swimming ability and gross morphological deformities. Larvae from each female will be preserved in buffered formalin for genetic analysis. Preserved collections will be arranged to group the first 10% of the hatch, the majority of larvae, and the last 10% of the hatch by female parent. Dead eggs and embryos will be quantified at the end of hatch.

Age classes from each location that could have been exposed to oil will be compared to those that were not: e.g., 1985-1989 vs 1990 (maybe trace oil or residual oil on beaches?) vs 1991-1993, and oiled locations will be compared with those that were not. Analysis of Variance techniques will be used to determine significance.

C. Contracts and other Agency Assistance

The primary plan to collect fish is to cooperate with ADF&G at the time each herring fishery occurs. However, as a contingency, it may be necessary to contract some commercial vessel time. For this purpose, \$6,000 has been requested for boat charters.

As in previous years, the genetic analysis will be contracted. The purpose is to search for cellular damage and chromosomal aberration in a manner consistent with previous *Exxon Valdez* oil-spill related herring research, including this project.

D. Location

Prince William Sound (PWS), Sitka Sound, and Auke Bay Laboratory (ABL). Herring impairment samples will be collected in PWS and in Southeast Alaska. All egg incubation will be at ABL.

SCHEDULE

A. Measurable Project Tasks for FY 96

1995 brood year: finish analyses and reports.

Mar 1996

1996 Brood year:

- | | |
|---|-----------------|
| 1. reproductive impairment survey in PWS: | Feb - Jun 1996 |
| 2. Chemical and contract analyses: | Jul - Nov 1996 |
| 3. Data analysis and final report: | Dec 96 - Apr 97 |

B. Project Milestones and Endpoints

Collection of data from live herring will be completed by July 1996: genetic and chemical analyses will

continue for approximately four months thereafter. Data analysis will require three months, and report writing an additional two. The FY96 project should be complete in April 1997.

C. Project Reports

A project report will be completed by April 1997. It is our intent to publish our data in peer-reviewed journals.

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

This project is an integral component of the suite of herring studies in the SEA plan. Reproductive impairment sample collection will be integrated with herring disease and spawn deposition research. This project is a continuation of project 94166 and 95074 and is part of an inter-agency cooperative study with ADF&G; it is the toxicological part of the 'Herring Package'. The other ADF&G/SEA plan projects are focused on the current status of the population and other factors that may limit recovery, including disease. Researchers from ABL will work closely with ADF&G to collect fish; age, length, weight, and VHN samples will be simultaneously collected from the same sites and year classes that reproductive impairment samples are collected, thus integrating state and federal research objectives.

ENVIRONMENTAL COMPLIANCE

EA or EIS are not required by this project.

PERSONNEL

GS-13 Physiologist - Stanley D. Rice

Received BA (1966) and MA (1968) in Biology from Chico State University, and PhD (1971) in Comparative Physiology from Kent State University. Employed at Auke Bay Fisheries Laboratory since 1971 as a research physiologist, task leader and Habitat Program Manager since 1986. Rice has researched oil effects problems since 1971, and has published over 70 papers, including over 50 on oil effects. Studies have ranged from field to lab tests, behavioral to physiological to biochemical studies, from salmonids to invertebrates to larvae to meiofauna. Rice has conducted and managed soft funded projects since 1974, including the Auke Bay Laboratory *Exxon Valdez* damage assessment studies since 1989. Activities since the oil spill have included leadership and management of up to 10 damage assessment projects, field work in PWS, direct research effort in some studies, establishment of state of the art chemistry labs and analyses in response to the spill, quality assurance procedures in biological-chemical-statistical analyses, establishment of hydrocarbon database management, servicing principal investigators and program managers in NOAA and other agencies with reviews and interpretations, provided direct input into agency decisions, interacted with other agencies in various ways (logistics coordination, critique experimental designs, interpret observations, etc.).

Dr. Stanley D. Rice
National Oceanic and Atmospheric Administration
National Marine Fisheries Service
Auke Bay Laboratory
11305 Glacier Highway
Juneau, AK 99801
phone: 907-789-6020
FAX: 907-789-6094
email: srice@abl.afsc.noaa.gov

GS-12 Fishery Biologist - Mark G. Carls

Received BA (1975) in Biology from Gustavus Adolphus College, St. Peter, MN, and MS (1978) in Biological Oceanography from Dalhousie University, Halifax, Nova Scotia. Mark has been employed at the Auke Bay Fisheries Laboratory since 1979. His principal involvement has been in research of petroleum hydrocarbon toxicology to marine fish and invertebrates, including egg, larval, and adult life stages. Mark has published 12 papers, and has 5 *Exxon Valdez* damage assessment papers pending publication. Since 1989, he has been involved as a principal investigator and co-investigator on several studies resulting from the *Exxon Valdez* oil spill.

Mark G. Carls
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National Marine Fisheries Service
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GS-11 Fishery Biologist - Scott W. Johnson

Received a BS (1974) in Zoology from San Diego State University, San Diego, CA, and a MS (1982) in Fisheries from Humboldt State University, Arcata, CA. Scott has been employed by NMFS for 15 years--3 years at the Southwest Fisheries Center in La Jolla, CA, and the last 12 years at the Auke Bay Laboratory. His principal studies have included research on 1) the habitat and ecology of juvenile salmonids, 2) riparian habitat issues, 3) effects of mining on marine resources, and 4) monitoring trends in marine debris abundance on the outer coast of Alaska. Scott has been a senior or contributing author on about 20 papers related to the above topics. Scott is presently Task Leader of two projects--Marine Debris and Mining Studies.

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1996 EXXON VALDEZ TRUS COUNCIL PROJECT BUDGET
 October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel	\$243.2	\$73.3						
Travel	\$20.5	\$23.6						
Contractual	\$64.0	\$72.8						
Commodities	\$33.5	\$14.2						
Equipment	\$5.0	\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$366.2	\$183.9	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
General Administration	\$40.9	\$16.1						
Project Total	\$407.1	\$200.0	\$69.5					
Full-time Equivalents (FTE)	4.6	1.3						
Dollar amounts are shown in thousands of dollars.								
Other Resources		\$112.2	\$98.1					
Comments: Other Resources: NOAA contribution is estimated: Habitat Investigations Prog Manager, J. Rice, 1 mo = \$10.9K Project Leader, M Carls, 12 mo = \$78.5K Senior Chemist, J. Short, 1 mo = \$7.8K Use of wet lab, biological processing lab, equipment (primarily ABL 6 microscopes) not otherwise covered estimated at \$15K								

1996

Project Number: 96074
 Project Title: Herring Reproductive Impairment
 Agency: National Oceanic & Atmospheric Administration

FORM 3A
 AGENCY
 PROJECT
 DETAIL

8/25/95 Version

October 1, 1995 - September 30, 1996

1996

FORM 3B
Personnel
& Travel
DETAIL

1996 EXXON VALDEZ TRUS COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
Vessel Charter for capturing adult herring		10.0
Contract to analyze herring larvae for genetic abnormalities		10.0
Exposure/Processing Crew (under NOAA Fishmarker Contracts)		
1 crew @ \$20/h	40h wk for 16 wks	12.8
2 crew @ \$12.50/h	40h wk for 20 wks	20.0
4 crew @ \$12.50/h	40h wk for 10 wks	20.0
NOAA considers Aircraft Charters as part of travel		
When a non-trustee organization is used, the form 4A is required.		
Contractual Total		\$72.8
Commodities Costs:		Proposed
Description		FFY 1996
Chemicals for biological tests (formalin, antibiotics, phosphate salts, etc.)		3.2
Biological processing (microscope slides, jars, film, disposables, dissecting tools, etc)		3.0
Incubation Equipment (component parts, plumbing parts, small nets, gravel, oil delivery supplies, etc.)		8.0
Commodities Total		\$14.2

1996

Project Number: 96074
 Project Title: Herring Reproductive Impairment
 Agency: National Oceanic & Atmospheric Administration

FORM 3B
 Contractual &
 Commodities
 DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1996
Description				
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
Those purchases associated with replacement equipment should be indicated by placement of an R.		New Equipment Total		\$0.0
Existing Equipment Usage:		Number of Units	Inventory Agency	
Description				
	Computers, Compaq	3	NOAA	
	Power Supply	1	NOAA	
	Camera	1	NOAA	
	Balance	2	NOAA	

1996

Project Number: 96074
 Project Title: Herring Reproductive Impairment
 Agency: National Oceanic & Atmospheric Administration

**FORM 3B
 Equipment
 DETAIL**

Effects of Oiled Incubation Substrate on Straying and Survival of Wild Pink Salmon

Project Number:	96076
Restoration Category:	Research
Proposer:	Alex Wertheimer and Stanley Rice NMFS, Auke Bay Laboratory
Lead Trustee Agency:	NMFS
Duration:	4 years
Cost FY96:	\$393.8
Cost FY97:	\$715.0
Cost FY98:	\$525.0
Cost FY99:	\$260.0
Geographic Area:	Little Port Walter, Baranof Island, Southeast Alaska
Injured Resource:	Pink salmon

ABSTRACT

This project examines the effects of oil exposure during embryonic development on straying, marine survival, and gamete viability of pink salmon. The primary objectives are to conduct a related series of controlled experiments on straying of pink salmon to determine the role of oil and other factors on straying so that field studies of straying in PWS after the spill can be interpreted, and to evaluate the significance of straying on management and restoration strategies.

INTRODUCTION

This project will examine the effects of oil exposure during embryonic development of pink salmon on the straying, marine survival, and gamete viability of returning adults. A series of controlled experiments will determine the impact of oil exposure on straying, as well as the effects of other factors (marking, stock, and transplant), so that measurements of straying in PWS after the spill can be interpreted, and the significance of straying on management and restoration strategies can be evaluated.

The project will require tagging several hundred thousand fry from wild and experimental treatment groups over two brood years, and examining returning adults in natal streams, other streams within 50

km of the natal streams, and an adjacent fishery. Pink salmon will be collected and spawned, and the fertilized eggs will be incubated in a controlled simulation of oiled intertidal habitat which occurred in Prince William Sound (PWS) after the *Exxon Valdez* oil spill. Fry from the oil-exposed and control groups will be marked to identify treatments when they emigrate from the incubators, and then released to migrate to the Gulf of Alaska. Corresponding groups of wild fry will also be captured and marked. Recoveries of tagged adults will be used to determine if oil exposure causes differences in straying and marine survival. Escapement and sampling rates in natal and non-natal streams will be estimated so that actual straying rates within the sampling region can be estimated, and the effects of oil, marking, population, and geographic factors on straying rate can be evaluated. Adults from the oil-exposure experiments that return to the release site will be identified as to treatment and then spawned. The fertilized eggs will be incubated in a clean environment to determine if oil exposure had affected gamete viability of fish from the original treatment groups.

This is a large multi-year study requiring significant logistic support for operations at remote sites. The study will be located in southeast Alaska because of the possible influence of prior or continuing oil contamination of pink salmon in PWS. The project was initiated in FY95 and will extend over five years. The incubation array for oil exposure is now under construction. Adults from the 1995 and 1996 returns will be collected and spawned in September of each year, and the fertilized eggs will then be placed in the experimental incubators (FY 95/FY96). In the spring of 1996 and 1997 (FY96/FY97), fry from the oil-exposed and control groups will be marked to identify treatments and released to migrate to the Gulf of Alaska; corresponding groups of wild fry will also be captured and marked. In the fall of 1997 and 1998 (FY97/98, FY98/99), adult returns will be sampled in natal and non-natal streams and in a large nearby fishery to recover marked fish from the experimental groups. Adults from the oil-exposure experiments returning to their release site in 1997 and 1998 will be spawned, and the survival of their progeny to the fry stage will be determined (FY98, FY99). Annual reports will be prepared each year. A final report will be prepared in 1999 summarizing the results of the study and the analysis of the restoration objectives. A synthesis of the results with previous field studies on pink salmon straying in PWS will also be prepared to evaluate the impacts of oil on straying of pink salmon, and to assess the implications of straying to management and restoration strategies for pink salmon in PWS.

NEED FOR THE PROJECT

A. Statement of Problem

Pink salmon were injured at several life-history stages during and shortly after the oil spill. Evidence of long-term damage from the toxic exposures of 1989 continues to build, and a thorough evaluation of the toxic contribution to pink salmon recovery problems became even more important when there was no explanation for the crash in pink salmon and herring in 1993. Three areas of continuing concern are the impacts of oil exposure on (1) homing and straying behavior; (2) survival of emergent fry in the marine environment; and (3) reproductive viability of exposed fish and their offspring.

Straying was a major concern during the spill; the Trustees supported a multi-million dollar effort to assess straying, and substantial straying of wild and hatchery stocks was observed. Unfortunately, the interpretation of that study is severely limited for several reasons. Consequently, the amount of straying

caused by oil is not known, natural straying rates are not known, and straying information cannot be used to develop or adjust restoration or management strategies.

B. Rationale

Pink salmon will be considered recovered when populations are healthy and productive and exist at pre-spill levels of abundance. Understanding the toxic effects of the 1989 oiling is a major component of the Trustee Council's program to restore pink salmon. This project contributes to the recovery of pink salmon by determining if oil exposure during incubation influences straying, marine survival, and gamete viability.

Results from Natural Resource Damage Assessment and Restoration Studies following the spill indicate that the toxic exposures of 1989 have caused persistent, long-term damage to pink salmon. Field studies in PWS after the *Exxon Valdez* oil spill have demonstrated differences in embryo survival between oiled and non-oiled streams. In addition, laboratory studies have shown that differences in survival between oiled and non-oiled streams are heritable (Restoration Study 94191A). Long-term (7-8 months) intra-gravel exposure of developing pink salmon eggs and alevins caused retarded development, altered emergence timing, decreased survival to eyeing and emergence, and an increased occurrence of gross lesions at emergence; it also had the surprising effect of delayed impacts on marine growth (Restoration Study 94191B). These developmental abnormalities from exposure to oil could persist and affect the behavior and fitness of the fish during subsequent life-history stages, including: (1) homing and straying, (2) survival of emergent fry in the marine environment; and (3) reproductive viability of exposed fish and their offspring.

Straying of pink salmon was a major issue following the spill. The Trustees supported a multi-million dollar effort to assess straying, and substantial straying of wild and hatchery stocks was observed. The ability of salmon to home (to return to their natal stream to spawn) is probably the most well-known and remarkable characteristic of these fish. Not all salmon return to their natal stream, however; some stray to non-natal streams to spawn. Some degree of straying is important to salmon populations; it is a mechanism for colonization of new habitat, as well as for the recolonization of habitat that has been damaged and subsequently restored. However, disruption in the normal amount of straying could have adverse impacts on the genetic structure of locally-adapted salmon populations. If high straying rates for pink salmon occur naturally in PWS, then the genetic structure of the populations in PWS should be relatively homogeneous, and large-scale mixing of wild stocks and the hatchery stocks derived from them should be of minor concern. Restoration of damaged pink salmon runs would thus be expected to occur naturally through recolonization from healthy stream systems. However, if the presence of oil increased straying from normally low levels, then the genetic diversity among and within wild stocks could be jeopardized from induced straying, and the genetic damage hypothesized to occur as a result of incubation in oiled substrate could be passed on to pink salmon in streams originally not oiled by the *Exxon Valdez*.

Straying rates for wild pink salmon observed in PWS in 1991 averaged 26% and ranged from 8-54% for fish from both oiled and non-oiled streams, based on coded-wire tag (CWT) recoveries in natal and non-natal streams. These straying rates seem high in relation to the concept that salmon normally home. Unfortunately, interpretations of that research are confused because even the wild stocks from non-oiled streams (controls) had to pass through oiled areas, and were thus not true controls. Also, marking the

fish with CWTs may have affected their straying behavior. Normal levels of straying are not known for pink salmon. Consequently, the amount of straying caused by oil is not known, and straying information cannot be used to adjust restoration or management strategies. This study will conduct controlled straying experiments to permit an evaluation of oil on straying, and to examine the effect of tagging, stock, and transplant on straying. To avoid the confounding effects of prior or continuing exposure to oil, the experiments need to be carried out in a geographic region remote from PWS. By identifying the effects of the various factors on straying, however, the results of these experiments can be directly applied to interpret the previous straying study in PWS.

C. Summary of Major Hypotheses and Objectives

The primary objectives of this study are to conduct a related series of controlled experiments on straying of pink salmon to determine the role of oil exposure of pink salmon embryos on their subsequent straying as adults; determine the role of other factors on straying so that the measurements of straying in PWS after the spill can be interpreted; and evaluate the significance of straying on management and restoration strategies in PWS. The study will also examine the effect of oil exposure during egg and alevin development on subsequent marine survival and gamete viability of pink salmon. Specific hypotheses to be tested include:

1. Oil exposure during embryonic development increases the straying of pink salmon.
2. Coded-wire tagging of pink salmon fry increases straying.
3. Stock origin (upstream vs. intertidal) affects the straying rate of pink salmon.
4. Transplant of gametes from a stream to a hatchery incubation and release site affects the straying rate of pink salmon.
5. Oil exposure during embryonic development decreases marine survival of pink salmon.
6. Oil exposure during embryonic development decreases the gamete viability of pink salmon.

D. Completion Date

This project will extend over the entire life-history of two brood years of pink salmon and will also include the egg/alevin life-history stage of their progeny. Oil exposures and marking of experimental groups will be completed by 1997. Recovery of returning adults will be completed by 1998. Evaluation of the viability of gametes of returning adults will be completed by 1999. The final report summarizing the results and detailing the accomplishment of the project's restoration objectives will be submitted in 1999.

COMMUNITY INVOLVEMENT

Scientists involved in this study will regularly present progress reports and results in scientific and public forums, including the annual workshop. They will be available to talk with interested public and will provide information for Trustee Council newsletters and annual reports as appropriate.

This project will be located in southeast Alaska out of the spill area because of the need to avoid the confounding effects of previous or continuing oil contamination in PWS. However, it will require substantial labor for fish marking and stream surveys, as well as contracts for vessel charters. Agency

hiring restrictions may limit us to contract hires for the intensive labor needs. We anticipate soliciting contract hires from communities in the area of the study (Juneau, Sitka, Petersburg, and Port Alexander). We encourage the Trustee Council to develop a labor pool from local communities in the spill area from which individuals could be either contracted or hired directly through the Council Administration for this and other Restoration projects. This would provide spill area residents better opportunity to participate directly in Restoration research.

FY 96 BUDGET

Personnel	\$184.4
Travel	39.4
Contractual	61.8
Commodities	76.2
Equipment	0.0
Subtotal	361.8
Gen. Admin.	32.0
Total	393.8

PROJECT DESIGN

Pink salmon were injured at several life-history stages during and shortly after the oil spill. Evidence of long-term damage from the toxic exposures of 1989 continues to build, and a thorough evaluation of the toxic contribution to pink salmon recovery problems became even more important when there was no explanation for the crash in pink salmon and herring in 1993. Straying was a major concern during the spill; the Trustees supported a multi-million dollar effort to assess straying, and substantial straying of wild and hatchery stocks was observed (Sharp et al. 1995). Unfortunately, the interpretation of that study is severely limited for several reasons. Consequently, the amount of straying caused by oil is not known, natural straying rates are not known, and straying information cannot be used to adjust restoration or management strategies. This project contributes to the understanding of the toxic effects of the oil spill and to the recovery process by examining the effects of oil exposure during incubation on the straying, marine survival, and gamete viability of pink salmon.

After the unexpected crash of pink salmon in 1993, two major research thrusts emerged: (1) evaluation of the ecosystem and its ability to support recovery of populations (SEA plan); and (2) evaluation of long-term damage from earlier oil exposure. Long-term damage was not originally suspected, even though there was ample evidence of short-term damage such as reduced embryo survival (Bue et al 1995), reduced marine growth (Wertheimer and Celewycz 1995; Willette 1995), and population effects (Geiger et al. 1995). Bue et al. (1995) found that elevated egg mortalities continued in oiled streams beyond the initial years of heavy oiling in intertidal spawning zones. They hypothesized that these persistent effects resulted from heritable damage passed on to subsequent generations. One model of how oil contamination could cause this damage is based on the biology of pink salmon egg-alevin development: Pink salmon spawn in contaminated intertidal zones of streams; the embryos incubate in contaminated streams for 7-8 months; and oil, which is extremely lipophilic, is readily absorbed into the large yolk reserves of the embryos. This exposure then causes both lethal and non-lethal damage to

developing embryos. The non-lethal damage can result in subtle developmental changes with potentially large implications in later life history stages, such as reduced marine survival and increased straying.

This model of exposure and damage is supported by controlled laboratory exposures to pink salmon eggs at Little Port Walter (LPW). This research, stimulated by the ADFG field studies, has shown that long-term (7-8 months) intra-gravel exposure of developing pink salmon eggs and alevins caused the predicted short-term effects (retarded development, altered emergence timing, decreased survival to eyeing and emergence, an increased occurrence of gross lesions at emergence) and also had the surprising effect of delayed impacts on marine growth (Restoration Study 94191B). These developmental abnormalities from exposure to oil could persist and affect the behavior and fitness of the fish during subsequent life-history stages, including: (1) homing and straying; (2) survival of emergent fry in the marine environment; and (3) reproductive viability of exposed fish and their offspring.

Substantial straying was observed in PWS after the oil spill in 1991 in a large tagging effort of both wild and hatchery pink salmon (Sharp et al. 1995). Interpretations of the study are confused because of concern that tagging caused some of the straying (pers. comm., J. Seeb, ADFG, Anchorage), and because even the wild stocks from non-oiled streams (controls) had to pass through oiled areas and were thus not true controls. Normal levels of straying are not known for pink salmon, and so it is difficult to evaluate the consequences of the observed straying. This study will conduct controlled experiments to permit an evaluation of the effects of oil incubation, tagging, stock, and transplant on straying. To avoid the confounding effects of prior or continuing exposure to oil, the experiments need to be carried out in a geographic region remote from PWS. By identifying the effects of the various factors on straying, however, the results of these experiments can be directly applied to interpret the previous straying study in PWS.

Straying rates for wild pink salmon observed in PWS in 1991 averaged 26% for fish from both oiled and non-oiled streams, based on coded-wire tag (CWT) recoveries in natal and non-natal streams (Sharp et al. 1995). Straying was highly variable, ranging from 8% to 54% for the six wild populations marked; straying rates were higher on average for wild fish than for hatchery fish. These high straying rates were surprising, but interpretation and use of the data were severely limited for several reasons. First, natural straying rates for pink salmon are not known for PWS or other areas. Second, the "controls" were wild stocks from non-oiled streams, but these fish had to migrate along contaminated shores, and were not true controls. Thus no measure of normal rates exists. Furthermore, if oil contamination continues, or heritable damage was indeed passed on, then "normal" rates cannot now be measured in PWS. Third, concern exists that placing CWTs in small pink salmon fry may cause damage responsible for some or most of the straying. Consequently, while substantial straying was measured in both oiled and non-oiled areas, clear interpretation of the results is not possible, and the significance of the measured straying remains unknown.

Straying rates of 26% seem high in relation to the concept that salmon normally home. However, virtually no other quantitative information exists on straying rates of wild pink salmon in their natural range for comparison. Reported straying rates in other species of salmon are highly variable. Examples are: Labelle (1992) observed an average straying rate of 2% for five stocks of wild and enhanced coho salmon, with a range of 0-11%; straying rates tended to be lowest for hatchery fish and highest for stocks subjected to certain supplementation practices. Pascual and Quinn (1994) reported highly precise homing of hatchery chinook salmon to the Columbia River, even if the fish were transplanted into the river

However, straying within the river was extremely variable among hatcheries, ranging from 1% to 95%, and was influenced by both environmental and genetic factors (Pascual and Quinn 1994). Tallman and Healey (1994) measured the straying rates for chum salmon in two streams located 2 km apart in the same bay; the straying rate from Walker Creek to Bush Creek was around 50%, while the straying rate from Bush Creek to Walker Creek was less than 2%.

The ability of salmon to home (to return to their natal stream to spawn) is probably the most well-known and remarkable characteristic of these fish. This tendency permits the establishment of discrete, locally adapted populations which are the basis of the stock concept in salmon management (McDonald 1981). Not all salmon return to their natal stream, however; some stray to non-natal streams to spawn. Straying is in itself a highly adaptive behavior. It is a mechanism for the colonization of new habitat (Milner and Bailey 1989), as well as for the recolonization of habitat that has been damaged and subsequently restored (Roys 1971; Leider 1989). Alexanderdottir (1987) and Quinn (1984) have speculated that pink salmon, which do not have overlapping generations because of their two year life cycle, may have relatively high rates of straying to provide a spatial population structure as a buffer against the risks inherent in a fluctuating environment.

The occurrence of strays in a spawning population does not necessarily mean that the strays are successful in transferring genetic information into the population. Tallman and Healey (1994) found that the gene flow was substantially lower than the straying rate among three populations of chum salmon, suggesting that strays have lower reproductive success than the native fish. However, higher gene flow was associated with higher straying rates. The rate and pattern of straying can still be considered indicative of the potential level of genetic interaction among populations and of the capacity of the species for recolonization of a site (Pascual and Quinn 1994).

Three possible explanations have been proposed for the high rates of straying observed for pink salmon in PWS. One is that oil exposure of the embryos induced high straying. No information exists on whether the developmental abnormalities associated with such exposure could also include deterioration of imprinting and homing. Previous research on the effects of oil on straying has focused on exposing returning adult salmon to oil for a short period of time (1-2 hours). Short-term exposure to oil had no deleterious effect on homing of either chinook salmon (Brannon et al. 1986) or coho salmon (Nakatani et al. 1985). Short-term oil exposure did cause temporary disorientation in migrating adult pink salmon, but did not prevent the eventual return to the home stream (Dames and Moore 1989). Straying rates observed in PWS by Sharp et al. (1995) were similar for fish from both oiled and non-oiled streams, however, the results were confounded because fry from non-oiled streams may have been exposed to oil as they migrated along oiled beaches.

The second explanation is that CWTs contributed to the observed straying rates. Morrison and Zajac (1987) reported that improperly injected CWTs can damage the olfactory nerves of small chum salmon. Pink salmon fry are smaller than chum salmon fry, and thus may be more easily damaged by tag injection. Seeb (pers. comm., ADF&G) found that many of the tags from pink salmon that had strayed in PWS were not in the ideal location in the head.

The third explanation is that the straying rates observed were indeed representative of wild stocks in PWS. Sharp et al. (1995) speculated that pink salmon originating from the intertidal reaches of streams may not imprint as strongly as do pink salmon spawned in upstream reaches of a stream, and may thus

return to a general region rather than a specific stream. Up to 75% of pink salmon spawning in PWS is in intertidal stream reaches. Pascual and Quinn (1994) also found that chinook salmon released into tributaries to the estuary of the Columbia River had higher straying rates than did the same group of fish released from locations higher upstream, suggesting that longer migration time or distance in freshwater may improve imprinting and homing.

The degree of straying of wild pink salmon is an important issue in the restoration and management of wild pink salmon populations in PWS. Information on the spatial patterns of straying, and the factors that affect them, can have direct bearing on such issues as the genetic interaction of wild and hatchery stocks (Pascual and Quinn 1994). If high straying rates occur naturally, then the genetic structure of the populations in PWS should be relatively homogeneous, and large-scale mixing of wild stocks and the hatchery stocks derived from them should be of minor concern. Restoration of damaged pink salmon runs would thus be expected to occur naturally through recolonization from healthy stream systems. However, if the presence of oil increases straying from normally low levels, then the genetic diversity among and within wild stocks could be jeopardized from induced straying, and the genetic damage hypothesized to occur as a result of incubation in oiled substrate could be passed on to pink salmon in streams originally not oiled by the *Exxon Valdez*.

The primary goals of this study are to conduct a related series of controlled experiments on straying of pink salmon to determine the role of oil and several other factors on straying so that the measurements of straying in PWS after the spill can be interpreted; and to evaluate the significance of straying on management and restoration strategies.

A. Objectives

This project has six major objectives related to straying of pink salmon. The design also permits evaluation of two additional objectives concerning the effects of oil exposure during incubation on marine survival and gamete viability.

1. Determine if oil exposure during incubation affects straying of pink salmon.
Hypothesis: Oil exposure during embryonic development increases the straying of pink salmon
2. Estimate natural straying rates of two stocks of pink salmon. Accomplishing this objective requires a sampling program that can estimate the total strays within a specific geographic area, and evaluation of the influence on straying of such factors as tagging, stock, and transplant (Objectives 3-6).
3. Determine if coded-wire tagging of pink salmon fry affects straying rate.
Hypothesis: Coded-wire tagging of pink salmon fry increases the straying of pink salmon
4. Determine if stock type affects the straying rate of pink salmon.
Hypothesis: Stock origin (upstream vs. intertidal) affects the straying rate of pink salmon
5. Determine if first-generation transplant affects the straying rate of pink salmon.
Hypothesis: Transplant of gametes from a stream to a hatchery incubation and release site affects the straying rate of pink salmon.

6. Develop a synthesis of pink salmon straying research, including the results of this study and use it to evaluate the implications for management and restoration strategies.
7. Determine if oil exposure during incubation affects the marine survival of pink salmon fry.
Hypothesis: Oil exposure during embryonic development decreases the marine survival of pink salmon.
8. Determine if oil exposure during incubation affects the gamete viability of pink salmon.
Hypothesis: Oil exposure during embryonic development decreases the gamete viability of pink salmon.

B. Methods

Overview

This project has been designed to examine the effects of oil exposure during embryonic development of pink salmon on: 1) straying rate, 2) marine survival, and 3) gamete viability of returning adults. Pink salmon gametes will be taken from fish returning to Lovers Cove Creek, an intertidally-spawning population on southeast Baranof Island (Figure 1). The embryos will be incubated at Little Port Walter (LPW) near the terminus of Sashin Creek in a controlled simulation of oiled intertidal habitat which occurred in PWS after the *Exxon Valdez* oil spill. Fresh water and salt water for incubation will be provided from Sashin Creek and the LPW estuary, respectively. Fry will be tagged with CWTs to identify treatments (Table 1, Objectives 1, 7, 8) and released to migrate to the Gulf of Alaska. Returning adults will be recovered at the release site, from the AKI hatchery brood stock return, and at other streams within 50 km of the release site. The cost-recovery fishery at AKI hatchery will also be sampled as a proxy for the commercial fishery. Recoveries of tagged adults will be used to determine treatment-specific straying rates and marine survival. Tagged adults returning to the release site will be held and spawned, and the fertilized eggs will be incubated in a clean environment to determine gamete viability of fish from the original treatment groups. The experiment will be conducted on two brood years (1995 and 1996) of pink salmon.

Because the effects of oil incubation on straying may be confounded by other factors that could affect straying, the influence of CWTs, stock, and transplant on straying will also be experimentally tested. These comparisons will utilize wild fry emigrating from both Sashin Creek and Lovers Cove Creek, as well as pink salmon fry from the control group of the oil-exposure experiment. The CWT effect will be examined by comparing straying rates of two groups of CWT fry with similar fish marked with fin clips only (Table 1, Objective 3). The stock effect will be tested by comparing straying rates of Sashin Creek wild emigrants and Lovers Cove Creek wild emigrants (Table 1, Objective 4). The transplant effect will be tested by comparing straying rates of Lovers Cove Creek wild emigrants with the control group of the oil-exposure experiment (Table 1, Objective 5). These comparisons will also be repeated for both brood years.

Sampling design: Assumptions and power

Assumptions. An empirical model was developed to determine the power to detect differences in straying between oil-exposure treatment groups at the release group sizes and sampling regimes proposed. A

number of assumptions were necessary to simulate the numbers of strays available for recovery, including marine survival, effects of oil exposure and marking and tagging on survival, straying rate, and sampling rate in non-weired streams.

Survival rates to return were based on the historical weir records for Sashin Creek (Olsen and McNeil 1967; Vallion et al. 1981). Survival to the weir ranged from 0.2 to 23.1%, averaged 3.7%, and had a median of 1.6% for 31 years for which data are available during the period 1939-1980. Because the distribution of survivals was highly skewed, the median was used as the assumption for "normal" survival. An estimate of 0.9% was used for "low" survival; over 70% of the observed survivals for Sashin Creek wild fry have been greater than or equal to this value.

Marking fish can be expected to reduce survival. No literature value is available for the effect of the CWT on small pink salmon. However, Bailey (1995) found that chum salmon fry marked with the adipose fin clip and CWT had 50% lower survival than unmarked fry. This rate includes the effects of tag loss subsequent to release. We used this as an adjustment to the survival assumptions, which gives a range of 0.5-0.8% survival to the weir. Wild pink fry marked with CWTs at Auke Creek, Alaska, for four brood years averaged 2.2% survival to return to the Auke Creek weir, with a range of 0.8-3.8% (Mortensen 1991). Our assumptions on survival are conservative relative to these observations.

Exposure to oil may also reduce marine survival (Hypothesis 2), which could affect our ability to detect differences in straying between treatments. We tested two levels of reduction in our survival assumption due to effects of oil: 20% and 50%.

The same survival rate was also assumed for adipose/pelvic fin-clipped fish. Although Blankenship (pers. comm., L. Blankenship, Washington Dept. Fish., Olympia, Wash.) observed lower survival for pelvic fin-clipped coho and chinook salmon smolts than for adipose fin-clipped, CWT smolts, Bailey (1995) observed higher survival for chum salmon fry with only pelvic fin clips and fish with both adipose and pelvic fin clips compared to adipose fin-clipped, CWT chum salmon fry. At Sashin Creek weir, returns of pink salmon marked with pelvic fin clips in 1976 ranged from 2.9 to 4.8% (Vallion et al. 1981).

We used two estimates of straying rates to project the numbers of strays that will be produced. The low rate (7%) was based on the observed rate of straying of hatchery pink salmon in the Juneau area (pers. comm., L. Macauley, Douglas Island Pink and Chum Salmon, Juneau). The high rate (26%) was the average observed in PWS by Sharp et al. (1995).

Based on the observations of stray pink salmon in PWS, we assume that the number of strays will decline with increasing distance from the natal stream. Sharp et al. (1995) recovered 79% of their total strays 30 km or less from the natal stream. We used this figure to estimate the number of strays that will be available in pink salmon streams within approximately 30 km from LPW, and developed a sampling design to intensively sample fish in streams within this distance. We assume that strays will be distributed proportionately to the escapement within this 30-km area. More distant sites will also be sampled, but at a lower effort.

Sampling effort will be allocated to four strata, with effort and methods varying between strata (Table 2). Stratum 0 includes streams with existing weirs where all returning fish can be sampled. Stratum 1 includes other pink salmon streams within 20 km of Sashin Creek, and Stratum 2 included streams 20-30

km from Sashin Creek. In strata 1 and 2, we assume we could sample at least 50% of the return as carcasses. Escapement to these streams will also be estimated to provide a measure of sampling fraction. This will allow expansion of tag recoveries to estimate actual number of strays to the surveyed streams, and will us to check our assumption of sampling rate. Stratum 3 includes streams 35-50 km from LPW. These streams will be sampled for tag occurrence rate only, to examine the assumption of declining occurrence with distance. Specific streams selected are listed in Table 3.

Streams were selected for sampling within Strata 1 and 2 based on the relative magnitude of the escapements within the sampling area. An index of escapement was generated using ADFG peak escapement counts from aerial surveys. The peak counts were expanded by a factor of 2.5 (from Sharr et al. 1993) to account for counting bias relative to the exact counts at the traps in Stratum 0. Based on this index, the streams selected provide >90% escapement coverage within the 30 km sampling region (Table 2). For stratum 3, one pink salmon stream was selected for sampling in each of the large bays occurring within 35-50 km of LPW. The specific stream selected was the one with the largest escapement within a particular bay, so that the number of carcasses checked will be maximized.

Power of sampling design. We ran simulations of the model predicting number of strays recovered, using the different combinations of survival, straying, and fishery exploitation estimates. Preliminary runs showed that the optimal number of CWTs per oil-exposure treatment was 70,000, given the limitations on the minimum number of treatment groups and the number of fry that could feasibly be marked (Table 1). Logistics of the wild fry marking limited treatment groups of wild fish to 60,000.

Simulations were then run to determine what level of difference between a dose and control could be detected with 95% confidence at the tagging and sampling levels proposed. Strays were assumed to be recovered only from strata 0-2 (30 km sampling region).

Results of the simulations showed that the magnitude of increase in straying detected is sensitive to the assumptions of survival and the straying rate of the controls (Table 4). At the median survival and low straying rate for the control, a 75-100% increase in straying (from 7% to 12-14%) can be detected, depending on the reduction in the marine survival of the treated group. At low survival and low straying for the control, a 100% increase in straying can be detected if the treated group has a 20% reduction in marine survival. At a 50% reduction in marine survival, the probability of detecting 100% increase declines to 94% (Table 4). For high straying rates, a 50% increase (from 26% to 39%) can be detected under all survival assumptions.

The ability to detect differences in marine survival under these same assumptions was also evaluated. We could consistently detect a 20% decrease in marine survival (from 0.8% to 0.6% or 0.5% to 0.4%) at the tagging and sampling levels proposed.

Gamete collection, egg incubation, and fry marking

Pink salmon gametes will be collected in the fall of 1995 and 1996 from Lovers Cove Creek, Baranof Island, southeastern Alaska. Ripe adult pink salmon will be seined from Lovers Cove Creek, and gametes from 245 females and 245 males will be collected and transported to the nearby NMFS research station at Little Port Walter (LPW) to be spawned. A randomized embryo pool will be created by: 1) spawning the

females into a common container, 2) randomizing the eggs within the container, 3) dividing the eggs into 245 aliquots, 4) fertilizing each aliquot with an individual male, and 5) recombining all fertilized eggs into a composite embryo pool. This composite embryo pool will then be divided into 92 aliquots of approximately 4000 eggs each. Each aliquot will then be randomly assigned to one of four treatments: high exposure dose, intermediate exposure dose, a control group to be marked with CWTs, and a control group to be marked with an adipose and left pelvic fin clip. A total of 92,000 eggs (23 aliquots) will be needed for each group. The individual aliquots will be incubated in individual pipe incubators filled with gravel.

Instream incubation will be simulated in the pipe incubators. These incubators will be constructed from 30-cm long sections of 20-cm diameter polyvinyl chloride pipe. The pipe will be stood on end, sealed, and fitted with a water intake at the bottom. The pipe will then be filled with appropriately-treated gravel. This design will allow water to upwell through the gravel and then out through an outlet fitting at the top of the incubator pipe. Fertilized eggs will be laid on top of the gravel to incubate. Upon hatching, the alevins will be permitted to burrow into the substrate.

Starting 24 hr after fertilization, embryos will be exposed to salt water for 4-hour intervals every 12 hours to simulate an intertidal environment. Water supply to the incubators will flow from a large head tank. During saltwater exposure periods, salt water with 26-30 parts per thousand salinity will be pumped from the bay into the head tank. At the end of each saltwater exposure cycle, saltwater flow into the head tank will be shut off, and freshwater flow into the head tank will be resumed. Salinity will thus rise and fall gradually at the beginning and end of each saltwater treatment period.

Dosing levels in the oil treatments will be based on the results from Restoration Study 94191B. Actual dosing will be established by analyzing hydrocarbon concentrations with gas chromatograph and mass spectroscopy (GC/MS) in incubator effluent and substrate at the beginning of the experiment and in incubator effluent, substrate, and fish tissue at each major developmental stage: eyeing, hatching, and emergence.

Various parameters will be recorded during incubation. Survival to eyeing and emergence, size at emergence and release, and emergence timing will be measured for each treatment group.

Upon emergence, fry will be moved to separate estuarine net pens for each treatment group to be held for tagging and fin-marking. Marking will begin as soon as sufficient fish (~ 10%) have emerged. In the spring of 1996 and 1997, a total of 210,000 pink salmon fry (70,000 per exposure treatment) from the gravel incubators will have their adipose fin removed and be coded-wire tagged (Table 1, Hypotheses 1-3). An additional 70,000 fry will be marked by removing the adipose fin and the left pelvic fin (Table 1, Hypothesis 1A). Approximately 10,000 fry can be marked daily.

Marking will be stratified into seven time periods to randomize the effects of handling and time of release. For each time stratum, a subgroup of 10,000 fry will be marked from each of the four experimental groups in a random sequence. Fry from all four subgroups within a stratum will be released at the same time, approximately 64 hr after the fourth subgroup is marked. Tag placement and clip quality will be checked regularly throughout each marking day. Subsamples for tag retention for each CWT subgroup will be taken after 24 and 48 hr. Mortalities following marking will be assessed daily.

until the fish are released. A composite group of 2000 fish will also be held for 14 days to monitor tag retention and mortality; these fish will not be released.

Capture and Tagging of Wild Fry.

Wild pink salmon fry emigrating from Sashin Creek and Lovers Cove Creek in 1996 and 1997 will be captured, marked, and released (Table 1, Hypotheses 1A, 1B, 1C). In Sashin Creek, fry will be captured using a floating screw trap. In Lovers Cove Creek, fry will be captured with screw traps or fyke nets. Capture methods for this component of the study will be tested in Spring 1995 to determine the most effective technique. From each stream, 60,000 fry will be tagged with CWTs in code lots of 10,000 tags (Table 1, Hypotheses 1B, 1C). At Sashin Creek, an additional 60,000 fry will be marked by removing the adipose fin and the right pelvic fin (Table 1, Hypothesis 1A). Equivalent numbers of fry will be CWT and pelvic fin-clipped at Sashin Creek on a given day. Fish will be held in pens adjacent to the traps for 64 hr. Tag placement and clip quality will be checked regularly throughout each marking day. Subsamples for tag retention for each CWT subgroup will be taken after 24 and 48 hr. Mortalities following marking will be assessed daily until the fish are released. Historical data from Sashin Creek show that emigration timing is highly variable, and can extend from early April until early June (Olsen and McNeil 1967), requiring an extensive trapping period to ensure sufficient coverage. From 2-4 people will be required at each site, depending on the number of fish to be handled and marked. Up to 3000 fry per day will be CWT at each stream.

Adult recoveries

Stream Recoveries. To assess the rate of homing vs. straying behavior, returning marked pink salmon will be recovered from natal and non-natal streams on Baranof Island and Kuiu Island (Figure 1). Sampling effort will be structured in four strata, with effort and methods varying between strata (Table 2). The sampling period will extend from mid-August through mid-October in 1997 and 1998.

Stratum 0 is comprised of the weir on Sashin Creek and the AKI hatchery brood stock raceway at Jetty Lake Creek in Port Armstrong. Close to 100% of the fish returning to these locations will be sampled. The expected returns to these streams represent 44% of the index escapement within 30 km of Sashin Creek (Table 2).

AKI Hatchery personnel will be contracted to examine all pink salmon that enter the facility and are spawned, in order to identify and recover strays from the various treatment groups. Any fish with a missing adipose fin will be retained for scanning for CWT and missing pelvic fins.

All pink salmon entering Sashin Creek will be checked for missing adipose fins. The weir will be operated so that fish cannot leave after entering, in order to provide a precise count of the number of fish in the creek. Fish with adipose fins will be passed into the creek. Fish without adipose fins will be checked for a missing pelvic fin. Pink salmon entering Sashin Creek that are missing a pelvic fin will be counted and killed. If both pelvic fins are present, the fish will be placed in a pen and held until mature for spawning for the gamete viability experiment. At that time, the fish will be killed, scanned for a tag, and the tag removed and decoded (if present). A CWT fish will be considered to have homed to Sashin Creek, unless the fish is from the Lovers Cove wild fry group.

Stratum 1 is comprised of pink salmon streams sampled within 20 km of Sashin Creek. All these watersheds will be on the east coast of Baranof Island (Figure 1). The streams sampled in this stratum represent 16% of the total escapement within 30 km of Sashin Creek (Table 2). Escapement of the sampled streams will be estimated to determine sampling proportion.

A three-person crew based at LPW will check carcasses on each of the streams once per week, with the exception of Lovers Cove Creek, which will be checked 2-3 times per week over a 6 week period. The streams will be accessed from LPW using a 5.1-m Boston Whaler skiff. The higher effort at Lovers Cove Creek is an attempt to sample as high a proportion of the escapement as feasible, because of the need to provide a precise estimate of both strays from the treatments released at LPW, and of homing fish from the Lovers Cove Creek wild fry. Each carcass will be counted, checked for a missing adipose fin, and marked with a jaw tag. The jaw tag will identify on subsequent surveys that the carcass has been already examined for marks. The jaw tag is also a critical component of the escapement estimation technique for each stream, described below. If a fish is missing the adipose fin, the head and the pelvic girdle (with fins attached) will be removed for later scanning for the presence of a CWT or a pelvic fin clip.

Stratum 2 is comprised of pink salmon streams approximately 20-30 km from Sashin Creek. These include watersheds on the east coast of Baranof Island and the west coast of Kuiu Island (Figure 1). The 30-km arc does not intersect all of Tebenkof Bay on Kuiu Island. Tebenkof Bay has four major embayments. We included in Stratum 2 streams in those embayments (Piledriver Cove and Thetis Bay) that are intersected by the 30-km arc, even if the streams were slightly (< 3 km) east of the arc. The streams sampled in this stratum represent 30% of the total index escapement within the 30-km sampling region (Table 2). Escapement of the sampled streams in Stratum 2 will be estimated to determine sampling proportion.

A four-person crew based on a charter vessel will survey each of the streams once per week over a six week period. The vessel-based operation will allow safe transit of Chatham Strait to sample streams in Tebenkof Bay, Port Malmesbury, and Patterson Bay (Figure 1). The crew will be able to sample during the day, then move safely to the next location after completing a survey.

Stratum 3 is comprised of pink salmon streams approximately 35-50 km from Sashin Creek. These include watersheds on the east and west coast of Baranof Island, and on the west coast of Kuiu Island. The stream with the largest ADFG escapement index count in each of five bays will be sampled: Red Bluff Bay, Rowan Bay, Bay of Pillars, Gut Bay, Branch Bay, and the inner portion of Tebenkof Bay. If time permits, streams in Table Bay on southwest Kuiu Island and Puffin Bay on southwest Baranof Island will also be surveyed (Table 3). The survey crews will not attempt to estimate escapement for these streams (with one exception); the emphasis will be on checking carcasses for tags and tag occurrence rate as a check of the assumption that stray recovery rate is proportionate to distance from natal stream. It may be possible, however, to get a rough estimate of sampling proportion using ADFG aerial survey counts for streams on which escapements were estimated, and generating an average expansion factor for the sampling year for the aerial surveys.

These streams (with the exception of Alecks Creek in Tebenkof Bay) will be sampled once per week over a three week period by a four person crew operating from a support vessel. The NOAA vessel R/V John N. Cobb will be used if available; otherwise it will be necessary to charter. The vessel-based operation will permit safe transit of Chatham Strait and the outer coast of Baranof Island to access sampling sites

The survey crew can sample during the day, then travel in the vessel afterwards to be in position for the next day's survey. During the survey, the crew will count and examine as many pink salmon carcasses as possible for a missing adipose fin. If a fish is missing its adipose fin, the head and the pelvic girdle (with fins attached) will be removed for later scanning for the presence of a CWT or a pelvic fin clip. The tail will be cut off of carcasses with adipose fins so that they can be identified on subsequent surveys as having been previously examined.

Alecks Creek in Tebenkof Bay will be sampled by the crew responsible for Stratum 2. Because this crew will already be sampling in the outer portion of Tebenkof Bay, this will be logistically efficient. Also, Alecks Creek is by far the largest stream in Tebenkof Bay, representing 30% of the peak escapement counts; no other stream has more than 10%. For this reason, Alecks Creek represents a good opportunity to determine if escapement magnitude or distance from natal stream influences occurrence of strays. Carcasses will be jaw-tagged for a population estimate for Alecks Creek in order to estimate sampling fraction on this system.

Tag Location. The location of CWTs within the heads of returning adult pink salmon will be examined to determine whether straying was influenced by where the tag was placed within the snout. Heads from adipose fin-clipped adults will be X-rayed so that tag location in fish that stray can be compared with tag location in fish that home. Samples of up to 100 heads will be X-rayed from each of three recovery categories: Sashin Creek, Lovers Cove Creek, and other area streams. The samples from Lovers Cove Creek and the other area streams will be from spawning or spawned-out fish. At Sashin Creek, however, because all adipose fin-clipped fish returning to the weir will be held alive after capture, and the tag will be removed and decoded at spawning in order to identify the treatment group, only fish that die in the holding net prior to spawning will be available to X-ray for tag location.

Estimation of Escapement. To estimate escapement into the stratum 1 and 2 index streams we propose to use the modified Jolly-Seber technique for carcass counts provided by Sykes and Botsford (1985). This population size estimator assumes an open population, and is relatively insensitive to violations of the assumptions of age-dependent catchability and survival (in our case, catchability and survival refer to the detectability and persistence of the carcasses over time). Standard errors are provided by simulation. Surveys of index streams will include recovering heads and ventral fins from adipose clipped fish, counting the number of carcasses in the stream, marking a representative fraction of the carcasses with snap ties, and noting the number of carcasses marked with snap ties on subsequent visits. The escapement to Sashin Creek in 1995 will be estimated using this technique and compared to weir counts. This initial analysis provides an opportunity to develop the simulation software prior to the first escapement surveys in 1997 and will produce an estimate of variance so designs for future escapement surveys can be refined.

Fisheries Recoveries

The number of fish harvested in the commercial fishery is not critical to our estimates of marine survival and straying if the assumed survival rates are representative of post-fishery survival, and if the treatment groups are equally distributed in the fisheries. However, if oil does affect homing behavior, then exposed groups might mill around more and thus be differentially vulnerable to the fishery. Pink salmon returning to Sashin Creek are thought to enter Chatham Strait from the south (Hoffman 1982). Adult tagging

studies indicate that some Sashin Creek fish move up Chatham Strait as far as Frederick Sound before returning to their natal stream. Fish harvested in lower Chatham Strait, however, are exclusively of lower Chatham origin (Hoffman 1981). Over the last four years, pink salmon harvest in area 109 from Frederick Sound to Cape Ommaney has averaged 17 million fish (pers. comm., H. Savikko, ADFG, Juneau). Fishery exploitation of Sashin Creek pink salmon is thought to be around 30% (pers. comm., Ben Van Alen, ADFG, Juneau). We estimate that the tag incidence rate for each treatment would be 1 in 30,000-50,000 fish in the general harvest area.

Because sampling this large and widely-dispersed fishery would be expensive and difficult, we propose instead to sample the AKI Hatchery cost-recovery fishery as a proxy for the common property fishery. Projected harvest for this fishery is 1,000,000 pink salmon (pers. comm., Dana Owens, Armstrong Keta Inc., Juneau). We can reasonably expect to cost-effectively sample at least 30% of this harvest to test whether treatment groups were exposed to differential harvest rates. No estimate of the power of the test is possible. Although fish harvested in lower Chatham Strait are predominately from the local area, we do not know if the cost-recovery harvest is representative of the general seine fishery in lower Chatham Strait. We propose to sample this fishery in 1997 (1995 brood returns) and use the results to (1) test for differential harvest of oil-exposed groups and (2) determine exploitation of Sashin Creek fish in the harvest. This information can then be used to determine the power of the sample to detect differences between treatments. Based on these results, and the geographic pattern of strays recovered by treatment, we can reconsider the option of broader fishery sampling in 1998 (1996 brood return).

At this time, AKI plans to deliver its fish to a floating processor located near the hatchery (pers. comm., Dana Owens, Armstrong-Keta Inc., Juneau). Sampling this harvest will require arranging with the processor to permit two samplers to examine pink salmon and remove those with a missing adipose fin as the fish are delivered to the processing lines. The samplers will be housed at Port Armstrong or on the processor for the duration of the harvest (3-4 weeks); heads and pelvic girdles from fish with missing adipose fins will be picked up and taken to LPW for examination for tags and fin clips and tag recovery and decoding at least twice weekly.

Analysis of Straying and Survival

The G-test of independence (Sokal and Rohlf 1981) will be used to test for statistical differences ($P = 0.05$) in straying between treatments for the oil-exposure and tagging-effects experiments (Objective 1.3). The number of strays observed in all escapement sampling strata and the number of homing fish recovered at Sashin Creek weir will be compared between treatments. For the oil-exposure test (Objective 1), if a significant difference is detected between the three groups, all three possible paired comparisons will be made, with the rejection criterion adjusted for multiple comparisons so that overall $P = 0.05$. For the effect of tagging experiment (Objective 3), two two-way contingency tables comparing the CWT and fin-marked releases will be analyzed.

Comparisons of straying rates between Lovers Cove Creek wild fish and Sashin Creek wild fish (Objective 4), and Lovers Cove Creek wild fish and transplanted Lovers Cove Creek fish (Objective 5) cannot be tested with the G-test because we will not have a complete count of the number of homing fish at Lovers Cove Creek. The total homing to Lovers Cove must be estimated by expanding observed tags by the sampling fraction. Comparisons for these objectives must thus be made using the estimated straying rates and associated variances, rather than observed recoveries.

Straying rates will be estimated for the various treatment groups by estimating the total number of strays, S , in non-natal streams within the 30-km sampling region, and the total number of homing fish, H , in the natal stream (Objective 2). S is calculated by

$$S_j = (\sum s_{ij}) / p,$$

where s_{ij} is the estimated number of strays for a particular treatment, j , in each non-natal stream surveyed, i , and p is the proportion of the escapement sampled within 30 km. Each s_{ij} is the observed number of strays expanded for the proportion of the escapement sampled for tags in stream i . H is the count of homing fish to Sashin Creek for all treatments, except Lovers Cove Creek wild fish; in that case, H is the observed number of homing fish in Lovers Cove Creek, expanded for the proportion of the escapement sampled for tags. The straying rate, f , is then

$$f_j = S_j / (S_j + H_j).$$

The variance of this proportion can be calculated from the variances of S and H . For S ,

$$\text{var}(S_j) = \sum \text{var}(s_{ij}).$$

The variance of each s_{ij} is derived from the variance of the escapement estimate used to calculate the proportion sampled for tags in stream i . For H , $\text{var}(H) = 0$ for Sashin Creek, because H is a total count. At Lovers Cove Creek, the variance of H is also derived from the variance of the escapement estimate used to calculate the proportion sampled. Variance of f is then

$$\text{var}(f_j) = [H^2(\text{var}(S)) + S^2(\text{var}(H))] / (S + H)^4.$$

A linear logistic model will be used to describe the relationship between straying rates and various factors, following the model used by Labelle (1992) for coho salmon. The objective is to predict the probability of straying for particular combinations of treatment, population, and geographic factors. The model used is

$$E[S/(S+H)] = \exp(b_0 + b_1 x_1 + b_2 x_2 + \dots) / [1 + \exp(b_0 + b_1 x_1 + b_2 x_2 + \dots)]$$

where f is the frequency of straying, b_n are parameters estimated by the model, and x_n are the predictor factors. We will use oil treatment, mark type, stock, transplant, distance from natal stream, direction from natal stream, and magnitude of non-natal stream as predictor factors.

Effects of oil exposure on marine survival (Objective 7) will be tested using the G-test. The contingency table for the comparison will be a 2×3 table, comprised of the three groups and the number of survivors and non-survivors for each group. The number of survivors for a treatment will be the sum of the observed number of tags at Sashin Creek weir, the observed number of tags recovered as strays, and the observed number of tags in the AKI fishery. The number of non-survivors for a treatment will be the number of "good" tags released (the number of fish tagged for a treatment adjusted for tag retention) minus the number of survivors. If a significant difference is detected between the three groups, the three possible paired comparisons will be made, with the rejection criterion adjusted for multiple comparisons so that overall $P = 0.05$.

h. Reproductive viability

Gamete viability will be determined for the oil treatment groups and the control (Objective 8). Tagged adults captured at Sashin Creek weir in 1997 and 1998 will be held for spawning. The CWT will be removed and decoded at spawning to identify the oil-treatment groups. Fish from each oil dose and from the control will be mated using a fully-crossed half-sib design (Falconer 1981). In this design, eggs from an oil-exposed female and a control female are each split into two aliquots. One aliquot from each female is fertilized with aliquots of sperm from the same oil-exposed male, and one aliquot from each female is fertilized with aliquots of sperm from the same control male. This 2 x 2 breeding matrix will be replicated 30 times for each treatment. Each half-sib family will be incubated in an individual container. Survival will be measured to fertilization, eyeing, and emergent fry stages. The numbers of defective or dead progeny will be compared between treatment groups. Because these gametes will not be incubated in an oiled environment, any observed increases in mortality or defective individuals can be attributed to oiling effects upon the first generation.

This approach will test if oil exposure lowers embryo survival, and will allow partitioning of the effect due to the male and female components. This will extend the results from Restoration Project 95191B, which will test for differences in gamete viability between within-treatment groups. Results obtained from the 1997 returns will be used to review and refine the breeding design in 1998.

C. Contracts and Other Agency Assistance

Personnel for the tagging and stream crews will be hired by contract. The AKI Hatchery will be contracted to screen their returning adult pink salmon for any tagged pink salmon from this study that have strayed to their facility. Contracts for vessel charters may be needed to transport crews to recover returning adult pink salmon from streams more than 30 km from LPW.

D. Location

The project will be implemented at Little Port Walter (LPW, Figure 1), a research facility of the NMFS Auke Bay Laboratory (ABL). This location is appropriate because of the logistic and infrastructure support the ABL and the LPW station provide for this complex array of experiments. It is also necessary to examine the response of pink salmon straying to oil exposure at a geographic locale remote from PW S, away from the confounding effect of prior or continuing oil exposure. Gametes will be collected from Lovers Cove Creek and Sashin Creek, Baranof Island, southeast Alaska. Eggs will be incubated, and pink salmon fry will be tagged at LPW, near the mouth of Sashin Creek, 10 km from Lovers Cove Creek. Returning adult pink salmon will be recovered from streams on the eastern coast of Baranof Island within 50 km of LPW.

Technical support provided at this location includes the use of the research station at LPW as a base for the fieldwork. This station will provide housing for project personnel, a wet lab for egg incubation, a weir across Sashin Creek for recovery of adult pink salmon, microscopes for the decoding of CWTs, and facilities for the spawning of adult pink salmon. The ABL will provide four tagging machines, vessel support, computer services, analysis of GC/MS samples, and communication and administrative support. Materials and personnel will be transported to and from LPW via the NOAA vessel R/V John N. Cobb, as well as contracted air taxi charters.

SCHEDULE

A. Measurable Project Tasks for FY 96

September to March: Incubate fertilized embryos (1995 brood); collect 36 GC/MS samples
Develop contractual labor agreements for tagging operations; evaluate fertility, early embryo survival in incubator.

March: Install traps for collection of wild fry (1995 brood)

March-April: Evaluate survival in incubators to fry emigration

April: Annual report

April-May: Tag and release hatchery and wild fry (1995 brood)

June: Clean incubators

July: Set up incubators for 1996 brood

August: Oil gravel

September: Collect and spawn pink salmon (1996 brood); Incubate fertilized embryos (1996 brood); Collect initial GC/MS samples (8) from incubator effluent and substrate

B. Project Milestones and Endpoints

<u>Milestone</u>	<u>Completion Date</u>
Spawning of 1995 brood adults	Sep 1995
Oil exposure of 1995 brood embryos	Apr 1996
Marking of 1995 brood fry	May 1996
Spawning of 1996 brood adults	Sep 1996
Oil exposure of 1996 brood embryos	Apr 1997
Marking of 1996 brood fry	May 1997
Spawning of 1997 brood adults	Sep 1997
Recovery of 1995 brood marked fish	Oct 1997
Estimation of 1997 natal, non-natal stream escapements	Oct 1997
Determination of 1997 brood gamete viability	Apr 1998
Spawning of 1998 brood adults	Sep 1998
Recovery of 1996 brood marked fish	Oct 1998
Estimation of 1998 natal, non-natal stream escapements	Oct 1998
Determination of 1998 brood gamete viability	Apr 1999

Endpoints

1. Objective 1: Determine if oil exposure during incubation affects straying of pink salmon
Completion Date: January 1999

2. Objective 2: Estimate natural straying rates of two stocks of pink salmon. Accomplishing this objective requires a sampling program that can estimate the total strays within a specific geographic area, and evaluation of the influence on straying of such factors as tagging, stock, and transplant (Objectives 3-6).
Completion Date: January 1999.
3. Objective 3: Determine if coded-wire tagging of pink salmon fry affects the straying rate of pink salmon.
Completion Date: January 1999
4. Objective 4: Determine if stock type affects the straying rate of pink salmon.
Completion Date: January 1999.
5. Objective 5: Determine if first-generation transplant affects the straying rate of pink salmon
Completion Date: January 1999.
6. Objective 6: Develop a synthesis of pink salmon straying research, including the results of this study, and use it to evaluate the implications for management and restoration strategies.
Completion Date: September 1999.
7. Objective 7: Determine if oil exposure during incubation decreases the marine survival of pink salmon fry.
Completion Date: January 1999.
8. Objective 8: Determine if oil exposure during incubation decreases the gamete viability of pink salmon.
Completion Date: July 1999.

C. Project Reports

Annual progress reports will be submitted in April of 1996, 1997, 1998, 1999.

- 1996 annual report: Details of the spawning of adult pink salmon in September, 1995, and the incubation of embryos (1995 brood).
- 1997 annual report: Details of the tagging and release of pink salmon fry (1995 brood); analysis of 44 GC/MS samples (1995 brood); the spawning of adult pink salmon in September, 1996; and the incubation of embryos (1996 brood).
- 1998 annual report: Details of the tagging and release of pink salmon fry (1996 brood); analysis of 44 GC/MS samples (1996 brood); the recovery and spawning of adult pink salmon (1995 brood) in September and October, 1997; and preliminary analysis of straying rates and marine survival of the 1995 brood.

1999 annual report: Details of the recovery and spawning of adult pink salmon (1996 brood) in September and October, 1998, and preliminary analysis of straying rates and marine survival of the 1996 brood and gamete viability of the 1995 brood.

The final report will be submitted in September, 1999.

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

Research by NMFS on effects of oil exposure to pink salmon has been closely coordinated with concurrent research efforts by ADF&G and UAF. This project directly complements Restoration Study No. 95191B and will be fully coordinated with its continuation.

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ENVIRONMENTAL COMPLIANCE

Compliance for NEPA will be met by a categorical exclusion. Collection and transportation of broodstock for the 1995 and 1996 broods will require an ADF&G Fish Resource Permit (FRP) and an ADF&G Fish Transport Permit (FTP). Fin clipping and tagging will be approved by the Pacific States Marine Fisheries Commission.

PERSONNEL

Project Leader

Alex C. Wertheimer - GM-13 Fishery Biologist. BS Fisheries Science, Oregon State University (1979). MS Fisheries Science, University of Alaska (1984). Currently employed by National Marine Fisheries Service, Auke Bay Laboratory as a Supervisory Fishery Biologist, Task Leader of Early Ocean Salmon Research. Author of over 20 peer-reviewed papers and 30 agency reports on various aspects of the biology and culture of Pacific salmon. Research on Pacific salmon has included determining early marine growth, distribution, and migration; in nearshore habitat utilization; predator/prey relationships; by-catch mortality; the effects of hydrocarbon contamination on juvenile salmon in the marine environment, the association of early marine conditions with year-class success of salmon; salmon aquaculture and

genetics; and status of stocks. Principle Investigator *Exxon Valdez* NRDA Fish/Shellfish 4, NMFS Component, 1989 through project completion in 1993.

Stanley D. Rice - GM-14 Physiologist. Received BA (1966) and MA (1968) in Biology from Chico State University, and PhD (1971) in Comparative Physiology from Kent State University. Employed at Auke Bay Fisheries Laboratory since 1971 as a research physiologist, task leader and Habitat Program Manager since 1986. Rice has researched oil effects problems since 1971, and has published over 70 papers, including over 50 on oil effects. Studies have ranged from field to lab tests, behavioral to physiological to biochemical studies, from salmonids to invertebrates to larvae to meiofauna. Rice has conducted and managed externally-funded projects since 1974, including the Auke Bay Laboratory *Exxon Valdez* damage assessment studies since 1989. Activities since the oil spill have included leadership and management of up to 10 damage assessment projects, fieldwork in PWS, direct research effort in some studies, establishment of state of the art chemistry labs and analyses in response to the spill, quality assurance procedures in biological-chemical-statistical analyses, establishment of hydrocarbon database management, servicing principal investigators and program managers in NOAA and other agencies with reviews and interpretations, provided direct input into agency decisions, interacted with other agencies in various ways (logistics coordination, critique experimental designs, interpret observations, etc.).

Ron A. Heintz - GS-11 Fisheries Biologist (Research). Ecology, University of Illinois (1979); MS Fisheries Science, University of Alaska (1986). He has worked for the National Marine Fisheries Service, Auke Bay Laboratory since 1985 concentrating his efforts on salmon enhancement research and salmon genetics. He is the principle investigator and co-investigator on several salmon genetics projects.

Adrian G. Celewycz - GS-11 Fisheries Biologist (Research). BS Biology, University of Illinois (1979); MS Fisheries Science, University of Alaska (1985). He has worked for the National Marine Fisheries Service, Auke Bay Laboratory since 1981, studying distribution, growth, habitat utilization, predator/prey relationships of juvenile salmon migrations. In addition to being recognized as "The Outstanding Student of Fisheries and Science" by the University of Alaska at Juneau in 1985, he was awarded Certificates of Recognition for superior performance by NOAA in 1989, 1990, and 1993. He served as co-investigator on *Exxon Valdez* NRDA Fish/Shellfish Study No. 4, and was awarded Certificates of Recognition by NOAA for outstanding contributions serving the public trust in response to the *Exxon Valdez* oil spill in 1989 and 1990.

Performance will be monitored by ongoing evaluation of time-specific milestones identified in the project schedule. Annual reports will document the accomplishment of project milestones. In FY-95, a GM-14 physiologist (Rice) will oversee and provide quality control for the whole project. A GM-13 biologist (Wertheimer) will be the project leader. A GS-13 chemist (Short) will establish a dosing protocol, determine hydrocarbon concentrations, and evaluate results of hydrocarbon analysis. A GS-11 biologist (Heintz) will help with the design of the project, and with data management and analysis. A GS-11 biologist (Celewycz) will be task leader for the artificial incubation and oil-exposure components, and a GS-11 biologist (John Thedinga) will be task leader for the wild fry capture and marking. Two GS-9 biologists (Bradshaw, Maselko) will assist in setting up the experiments, collecting data, analyzing data, and reporting results. Other ABL biologists (Orsi, Mortensen) will provide logistic coordination and serve as crew leaders on remote marking and stream survey operations, as needed. This project is undertaken as part of the research activities of the Auke Bay Laboratory (ABL) and will be supported by the laboratory infrastructure. The ABL will provide backups if any personnel changes occur. A GS-12

biologist (Wright) will be the agency project manager for coordination of this and other NOAA projects with the Trustee Council.

Alex Wertheimer

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April 30, 1995
Date Prepared

Table 1. Mark type and number marked for the experimental groups used to test specific hypotheses relating to the effects of oil exposure during embryonic development of pink salmon on straying behavior, marine survival, and gamere viability; and the effects of mark type, stock and transplant on straying behavior. See page 5 of the proposal for a description of the hypotheses. Highlighted treatment groups are used in more than one comparison. CWT = coded-wire tag.

Treatment	Mark Type	Number Marked	
		1995	1996
Objectives 1,6,7: Effects of oil exposure			
High Dose	CWT	70 K	70 K
Intermediate Dose	CWT	70 K	70 K
Exposure Control	CWT	70 K	70 K
Objective 2: Mark Effect			
Exposure Control	CWT	70 K	70 K
Wet Lab Control	Ad-Pelvic	70 K	70 K
Sashin Creek Wild	CWT	60 K	60 K
Sashin Creek Wild	Ad-Pelvic	60 K	60 K
Objective 3,4: Stock and Transplant Effect			
Lovers Cove Wild	CWT	60 K	60 K
Sashin Creek Wild	CWT	60 K	60 K
Exposure Control	CWT	70 K	70 K

Total Tags:

330 K

Total Fin Marks:

130 K

Table 2. Sampling strata for recovery in natal and non-natal streams of adult pink salmon returning marked as fry and released at Sashin Creek and Lovers Cove Creek.

Stratum Number	Description	Number of Cataloged Streams in Stratum	Proposed Number of Streams to Survey in this Stratum	ADF&G Estimate of Total Escapement to Stratum	Total Escapement to be Surveyed in Stratum	Fraction of Escapement in 30 km Region surveyed
0	Armstrong Keta Inc. and Sashin Cr. Weirs	2	2	177,037	177,037	44%
1	Streams within 20 km of Sashin Cr.	16	4	63,276	63,276	16%
2	Streams between 20 and 30 km from Sashin Cr.	27	7	157,168	118,988	30%
3	Streams more than 30 km from Sashin Cr.	*	*	*	*	*

* tag recoveries from this stratum will not be expanded, so sampling fraction has not been estimated.

Table 3. Stream number, name, and average peak aerial survey count for streams surveyed in each sampling stratum.

Stream Number	Name	Stratum Number	Average Peak Count
109-10-002	Armstrong Keta Inc. Weir	0	100,000
109-10-009	Sashin Cr. Weir	0	77,037
109-10-014	Port Herbert	1	500
109-10-007	Borodino Outlet	1	*
109-10-009	Lover's Cove Cr.	1	63,278
109-10-010	Newport Walter	1	*
109-10-021	Cliff Lk. Outlet, Deep Cove	2	12,250
109-10-023	Deep Cove NW Head	2	28,250
109-10-028	Parry Lk. Outlet, Patterson Bay	2	31,427
109-62-003	Piledriver Cove	2	15,472
109-62-028	William Cr., Thetis Bay	2	13,347
109-62-029	Wolf Cr., Thetis Bay	2	15,750
109-63-005	Joyce Cr., Malmsbury	2	14,740
109-20-006	Gut Bay	3	6,802
109-52-007	Rowan Cr., Rowan Bay	3	50,490
109-52-055	Kwatahein Cr., Bay of Pillars	3	17,410
109-62-012	Elena Bay - Tebenkof	3	15,965
109-62-020	Petrof Bay - Tebenkof	3	9,130
109-62-014	Sample Cr., Tebenkof	3	16,190
109-20-106	Red Bluff Bay	3	260,945
113-12-001	Branch Bay	3	*
113-11-009	Puffin Bay	3	*
109-61-011	Table Bay	3	812

* These streams are not regularly surveyed by ADF&G, no estimates of escapement exist.

Table 4. Estimated probabilities of detecting increases in straying at different assumptions for survival and straying rates.

# Controls Recovered in Natal Streams	# Control Strays Recovered	Survival as Proportion of Control	% Increase in Straying Rate	χ^2	P(x)	Treatment # Strays Recovered	Treatment # Recovered in Natal Stream
Control: Survival Rate = .008 Stray Rate = .07							
521	20	.80	50	2.082	0.851	24	401
			75	4.358	0.963 2	28	393
			100	7.256	0.992 9	32	385
		.50	50	1.621	0.797 0	15	251
			75	3.086	0.921 0	17	246
			100	5.84	0.984 3	20	241
Control: Survival Rate = .005 Stray Rate = .07							
326	12	.80	50	1.521	0.782 5	15	251
			75	2.734	0.901 8	17	246
			100	4.926	0.973 5	20	241
		.50	50	.976	0.676 8	9	157
			75	2.517	0.887 4	11	153
			100	3.565	0.941 0	12	150
Control: Survival Rate = .008 Stray Rate = .26							
414	74	.50	25	2.229	0.864 6	46	189
			50	9.411	0.997 8	56	171

		.80	25	3.052	0.919 4	74	302
			50	11.9	0.999 4	89	273
Control: Survival Rate =.005 Stray Rate = .26							
259	46	.50	25	1.547	0.786 4	29	118
			50	6.632	0.990 0	35	107
		.80	25	1.895	0.831 4	46	189
			50	7.720	0.994 5	56	171

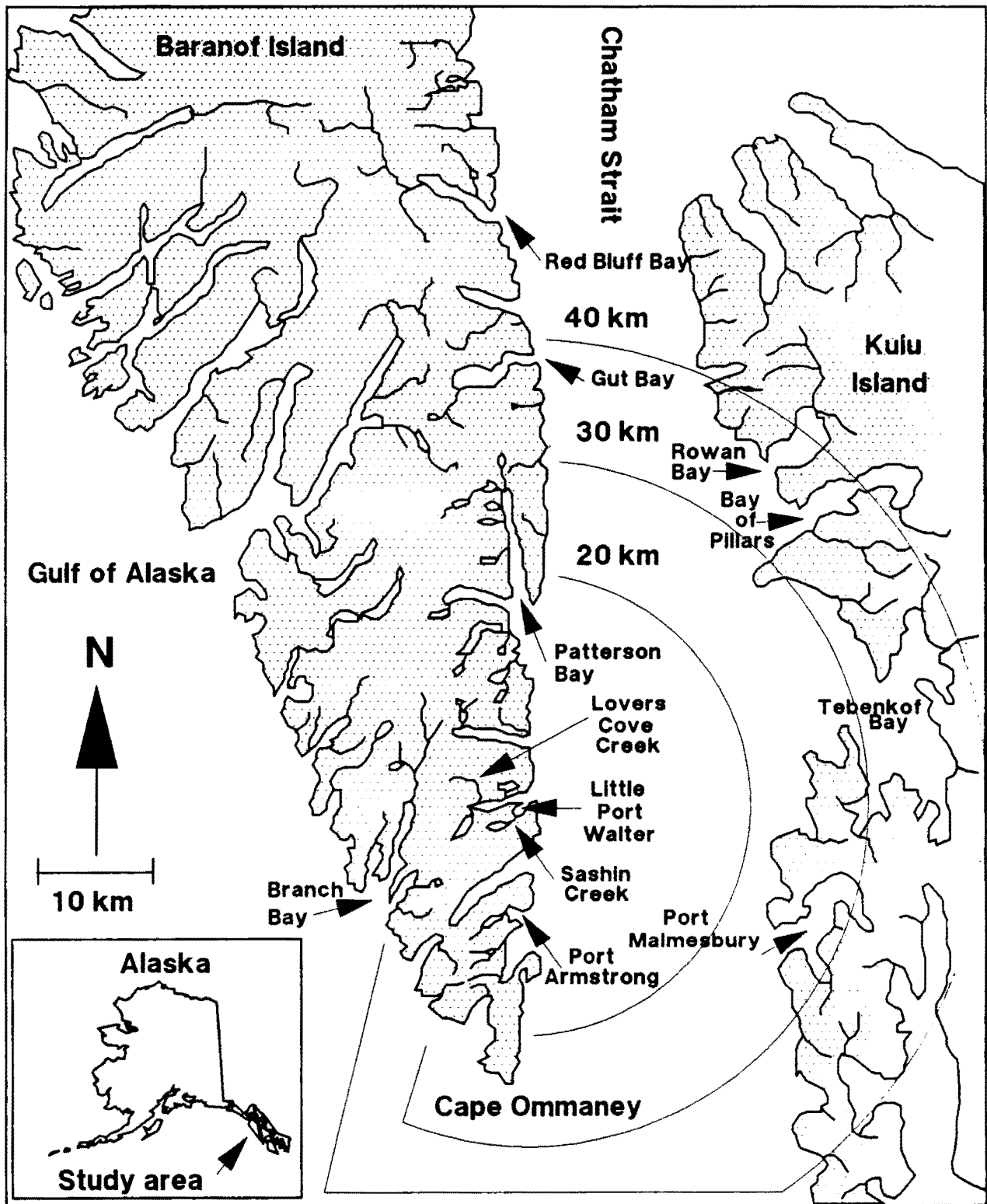


Figure 1. Southern Baranof Island and western Kuiu Island, southeastern Alaska.

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel	\$76.1	\$184.4						
Travel	\$5.8	\$39.4						
Contractual	\$5.8	\$61.8						
Commodities	\$56.4	\$76.2						
Equipment	\$24.0	\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$168.1	\$361.8	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
General Administration	\$11.8	\$32.0						
Project Total	\$179.9	\$393.8	\$715.0	\$525.0	\$260.0	\$0.0	\$0.0	\$0.0
Full-time Equivalents (FTE)	1.3	2.9						
Dollar amounts are shown in thousands of dollars.								
Other Resources	\$21.8	\$262.0	\$210.0	\$195.0	\$125.0			
<p>Comments:</p> <p>Other Resources:</p> <p>NOAA Contribution of Habitat Program Manager, J Rice. 2 mo = \$21.8K Project Leader, A Werthheimer, 12 mo = \$96.1 K Salmon Program Manager, B Heard, 2 mo = \$21.8K LPW Station Manager, R Martin, 4 mo = \$30.1K Fish Biologist, D. Mortensen, 3 mo = \$16.8K Fish Biologist, J Orsi, 3 mo = \$16.8K</p> <p>NOAA contribution of routine operating costs for the Little Port Walter Field Station estimated at \$25K</p> <p>This project's' activities and logistics are closely coordinated with #96161B</p>								

1996

Project Number: 96076
Project Title: Oil Effects on Pink Salmon Straying
Agency: National Oceanic & Atmospheric Administration

FORM 3A
AGENCY
PROJECT
DETAIL

Prepared: 1 of 4

5/1/95

COUNCIL PROJECT BUDGET

October 1, 1995 -

ember 30, 1996

[illegible]

1996

Project Number: 96076
Project Title: Oil Effects on Pink Salmon Straying
Agency: National Oceanic & Atmospheric Administration

FORM 3B
Personnel
& Travel
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
Tagging Crew (Under NOAA Fishmarker Contracts)		
1 crew @ \$ 15/hr for 60h/wk for 8 wks		7.2
6 crew @ \$12.50/h for 50h/wk for 6 wks		22.5
4 crew @\$12.50/h for 50h/wk for 8 wks		20.0
1 crew @\$12.50/h for 60h/wk for 6 wks		4.5
Tagger Rental: 2 for 2 mo @ \$1.9K/mo		7.6
NOTE: NOAA considers Air Charter costs under Travel		
When a non-trustee organization is used, the form 4A is required.		
Contractual Total		\$61.8
Commodities Costs:		Proposed
Description		FFY 1996
Groceries		14.5
Fuel		2.0
Rain Gear: 16 pr @ \$100		1.6
Chest Waders: 12 Pr @ \$200		2.4
Incubation gear/supplies		2.9
Tagging supplies (tags, cutters, head molds and scissors)		30.0
Computer software upgrades, repairs		2.0
Chem lab solvents, chemicals		6.0
Chem I Lab supplies (gloves, lab coats, Kimwipes, glassware, etc.)		4.0
Equipment maintenance & repair		10.8
Commodities Total		\$76.2

1996

Project Number: 96076
Project Title: Oil Effects on Pink Salmon Straying
Agency: National Oceanic & Atmospheric Administration

FORM 3B
Contractual &
Commodities
DETAIL

1996 EXXON VALDEZ TRUSTI JUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1996
Description				
	None			0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
Those purchases associated with replacement equipment should be indicated by placement of an R.			New Equipment Total	\$0.0
Existing Equipment Usage:		Number of Units	Inventory Agency	
Description				
All to be purchased in FY95				
Fyke Nets		4	NOAA	
Screw Traps		2	NOAA	
Saltwater intake pumps		2	NOAA	
Electronic Fish Measuring Board		1	NOAA	
Beach Seines		2	NOAA	

1996

Project Number: 96076
 Project Title: Oil Effects on Pink Salmon Straying
 Agency: National Oceanic & Atmospheric Administration

FORM 3B
 Equipment
 DETAIL

8/25/95 version
Interim Budgets for
96076
96191B
96163A-L

August 2, 1995

MEMORANDUM TO: Traci Cramer
Director of Administration

FROM: Bruce Wright
Program Manager

Re: Interim Budgets (Oct. 1, 1995 to Jan. 31, 1996)

Restoration Study 96076: Effects of Oil on Straying and Survival

Project 96076 is a continuing research program with on-going field and laboratory activities during the period October 1, 1995 through January 31, 1996. These activities focus on the maintenance, monitoring, and evaluation of the pink salmon embryos exposed to oiled gravel; the collection of water, gravel, and embryos for hydrocarbon analysis; and the development of methods for enumerating escapements from carcass mark/recapture techniques. The continuation of the project also requires planning and logistic arrangements for the large-scale tagging operations in the spring of 1996, data analysis and preparation of the annual report, and presentations at the January workshop. The interim costs include salary and contracts for personnel costs, travel to support field operations and to Anchorage for Trustee Council workshops and reviews, and essential supplies. A list of tasks to be accomplished over the interim period follows.

- 1) Maintenance and monitoring of incubation system. Daily monitoring of flow rates to 100 incubators, cleaning of incubators as necessary, and monitoring and maintenance of seawater and freshwater supply systems.
- 2) Evaluation of treatment effects. Assessment of survival of embryos to eyed-stage (late October-early November) and to hatching (January).
- 3) Hydrocarbon analysis. Collection of gravel, water, and embryo samples at eyed stage and hatch stage, and analysis of samples collected at earlier spawning in FY 95.
- 4) Sashin Creek weir operation. Continued operation of Sashin Creek weir until late October to enumerate total escapement of pink salmon to Sashin Creek. This requires daily fish counts and weir cleaning.
- 5) Stream Surveys. Weekly carcass surveys of Sashin and Lovers Cove Creeks in October

for estimation (mark/recapture) of pink salmon escapement. These surveys are needed to develop techniques and statistical models for enumeration of escapement in streams sampled for returning strays and must be done in conjunction with the weir escapement enumeration for the 1995 return.

6.) Planning, logistic support, data analyses, report preparation. Ongoing, October-January.

COSTS BY LINE ITEM, 96076

<u>Line Item</u>	<u>Costs (\$K)</u>
Personnel	\$ 6.8
Travel	10.9
Contracts	3.6
Commodities	13.8
Equipment	0.0
SUBTOTAL	97.1
General Admin	10.6
TOTAL	\$107.7

Restoration Study 96191B: Damage to Pink Salmon Fry and Pre-emergent Fry Incubated in Oiled Gravel (Laboratory Study).

Below are the interim budget requirements for this study. The primary objective of this project is to determine if pink salmon that incubate in oiled gravel ultimately experience impaired reproductive ability. Exposures for the 1993 brood were complete in the spring of 1994, and the adult fish will be mature at the end of FY95.

During the interim portion of FY96 we will begin evaluating the reproductive success of the 1993 brood. In addition, we will be writing our annual report, and preparing for the annual meeting in Anchorage. Labor costs include a half-time technician to culture the developing eggs, and PI to prepare the annual report. The travel budget reflects the need for two trips to Little Port Walter, to observe the survival to "eyeing" among the progeny of the 1993 brood, and later, to assist the hatchery technician when larvae begin hatching. In addition, we have included the cost of the trip to the annual meeting in Anchorage and the cost of another trip to Anchorage to meet with investigators from Restoration Study 96191A.

COSTS BY LINE ITEM, 96191B

<u>Line Item</u>	<u>Costs (\$K)</u>
Personnel	42.0
Travel	19.2
Contracts	0.0
Commodities	5.3
Equipment	0.0
SUBTOTAL	66.5
General Admin	6.3
TOTAL	\$72.8

Restoration Study 96163 A-L: Apex Predator Ecosystem Experiment (APEX)

Interim funding will be necessary for data analysis, report and workshop preparation, and development of the FY96 proposal based on the November 30-December 1, 1995 workshop review.

The attached APEX interim budget will allow for personnel and travel costs to accommodate data analysis, reporting, and travel (see tables below). The two contracts, 96163A and 96163G extend beyond January 31, 1996 already, so no interim funding will be requested. Program management costs will, however, be necessary. Project 96163H was not funded in FY95 so no interim funding will be requested.

COSTS BY AGENCY and BY LINE ITEM, 96163 A-L

<u>Line Item</u>	<u>DOI (\$K)</u>	<u>NOAA (\$K)</u>	<u>ADF&G (\$K)</u>	<u>TOTAL (\$K)</u>
Personnel	181.1	29.3	26.3	236.7
Travel	7.0	7.0	0.0	14.0
Contracts	0.0	0.0	0.0	0.0
Commodities	0.0	0.0	0.0	0.0
Equipment	0.0	0.0	0.0	0.0
SUBTOTAL	188.1	36.3	26.3	250.7
General Admin	27.2	4.4	3.9	35.5
TOTAL	\$ 215.3	\$ 40.7	\$ 30.2	\$ 286.2

cc: Dave Duffy
Ron Heintz
Dave Irons
Byron Morris
Jeep Rice
Sandra Schubert
Stan Senner
Joe Sullivan
Alex Wertheimer

96163 A-L Interim Budget for Oct. 1, 1995 to Jan. 31, 1996

PROJECT	TITLE	Personnel Costs (title/months)	Travel Costs	Total
95163 A	Fish Survey & Biology	\$ 5.8K (PM NOAA/1 months)	\$ 1.0K	\$ 6.8K
95163 B	Bird/Fish Interactions	19.2 (PI/4 months) 5.5 (PM DOI/1 month)	0.5 0.0	25.2
95163 C	Fish Diet Overlap	9.9 (PI/3 months) 20.3 (techs./3 months) 5.8 (PM NOAA/1 month) 1.2 (PM ADFG/.2 month)	4.5 0.0 0.0 0.0	41.7
95163 D	Puffins as Samplers	5.0 (PI/1 month) 6.0 (Assis. PI/3 months)	1.0 0.0	12.0
95163 E	Black-legged Kittiwakes	19.2 (PI/4 months) 4.9 (2 techs./2 months) 5.5 (PM DOI/1 month)	1.0 0.0 0.0	30.6
95163 F	Pigeon Guillemots	19.2 (PI/4 months) 4.9 (2 techs./2 months) 5.5 (PM DOI/1 month)	1.0 0.0 0.0	30.6
95163 G	Energetics	2.8 (PM NOAA/0.5 month)	1.0	3.8
95163 H	Proximate Composition	0.0	0.0	0.0
95163 I	Project Leader	35.0 (PI/2 months) 2.5 (PM DOI/0.5 month) 2.9 (PM NOAA/0.5 month) 15.0 (Fish Symposium)	1.0 0.0 0.5 0.0	56.9
95163 J	Barren I. Murres & BLKs	12.6 (PI/3 months) 6.9 (bio. tech./2 months)	1.0 0.0	20.5
95163 K	Fish as Samplers	4.2 (PI/1 month)	0.5	4.7
95163 L	Barrens & Historical	10.0 (PI/2 months) 2.1 (biologist NOAA/1 mo) 4.8 (biologists F&G/2 mo)	1.0 0.0 0.0	17.9
General Admin.				
TOTAL		236.7*	14.00	250.7*

PI = principal investigator PM = program manager

* General administration will be added to these costs. See summary tables below.

96086

Herring Bay Monitoring and Restoration Studies / Close out

Project Number: 96086C Revised
Restoration Category: Monitoring
Proposer: Drs. Raymond C. Highsmith and Michael Stekoll
Lead Trustee Agency: ADF&G
Cooperating Agencies: University of Alaska
Duration: 1 October 1995 - 30 September 1996
Cost FY96: \$156,100
Geographic Area: Herring Bay, Knight Island, PWS
Injured Resource/Service: Intertidal Invertebrates and Algae

ABSTRACT

In 1990, intertidal restoration studies were established in Herring Bay, Prince William Sound in response to the T/V *Exxon Valdez* oil spill. These studies have continued through the 1994 field season and show continued injury to *Fucus gardneri* and associated invertebrate populations, especially in the upper intertidal. The data collected during the 1995 field season will be incorporated into the existing Herring Bay data base and the rates and extent of recovery determined for injured resources.

INTRODUCTION

The Herring Bay Experimental and Monitoring studies conducted from 1990 through 1994, and continuing through the 1995 field season, showed damage to the intertidal invertebrate and algal population. Significant differences were detected between oiled and reference sites for grazing invertebrates such as *Tectura persona*, *Lottia pelta*, and *Littorina sitkana* and for the primary space competitor and main structural component of the upper intertidal, the alga *Fucus gardneri*. Recovery is under way in lower and middle intertidal zones and normal community interactions are returning. The upper intertidal, however, continues to exhibit damage. Data collected during the 1995 field season will allow estimates of the rates and extent of recovery in the upper intertidal. In addition, methods for actively enhancing *Fucus* recovery in severely damaged areas were tested and have been successful in reducing desiccation rates which may lead to increased germling survival. The evaluation of the erosion control fabric will continue through 1995.

NEED FOR THE PROJECT

A. Statement of the Problem

Five years after the Exxon Valdez oil spill, several intertidal species are still showing damage, including *Fucus*, the important structural component of the intertidal ecosystem. Monitoring of key intertidal species in Herring Bay will continue through the 1995 field season. However, additional funding is required to analyze these data and incorporate them into the existing Herring Bay data base, which will allow estimates of the degree and rates of recovery of injured species.

B. Rationale

Intensive research has shown that recovery subsequent to the oil spill is incomplete, especially for *Fucus* and associated invertebrate populations in the upper intertidal elevations. Populations of several key invertebrate and algal species will be monitored during the FY95 field season, requiring sample and data analysis. Funding is requested in FY96 for sample and data analysis and for the completion of the final report following the 1995 field season.

C. Summary of Major Hypotheses and Objectives

The objective of this study is to estimate the rates and extent of recovery of injured invertebrates and algae in the intertidal zone of Herring Bay. These results will be submitted in a complete report on the FY95 field season studies.

D. Completion Date

This work will begin in October 1995 and be completed in June 1996.

COMMUNITY INVOLVEMENT

This project will utilize University of Alaska personnel previously involved in Herring Bay Monitoring and Restoration Studies. They have extensive experience with these field and laboratory studies. The final report will be available to the public through the OSPIC office in Anchorage.

FY96 BUDGET

Personnel	106.3
Travel	6.4
Contractual	10.6
Commodities	1.6
Equipment	0.0
Subtotal	124.9
Gen. Admin. 25%	31.2
Total	156.1

PROJECT DESIGN

A. Objectives

The overall objective of the study is to determine the rates and extent of recovery of injured resources in Herring Bay, especially of *Fucus* in the upper intertidal. This will be accomplished through the analysis of data collected during the 1995 field season for invertebrate abundance and recruitment, and algal abundance, percent cover, and life history characteristics. These results will be presented in a final report in June 1996. In addition, new sets of erosion control fabrics will have been placed in the high intertidal as a means of reducing desiccation and of promoting *Fucus* germling survival. These data will be analyzed and reported in the final report.

B. Methods

Statistical analyses will remain the same as for previous years. The basic experimental design is an After Control-Impact Pair design in which an oiled site is statistically compared to a matched control or reference site that was unoiled. We will use fixed effects analysis between matched pairs (one-way ANOVA or t-tests) and Fisher's meta analysis tests for combining p-values within a habitat. By definition, "recovery" of a habitat or tidal height will be when the abundance or percent cover for oiled sites is the same as for control sites. Our operational definition for recovery for a particular taxon is when a taxon shows no significant difference between oiled and reference sites at $p < 0.05$ by Fisher's test.

C. Contracts and Other Agency Assistance

Principal investigators from the University of Alaska School of Fisheries and Ocean Sciences will cooperate to provide expertise on algal and invertebrate taxonomy and ecology.

The FY 95 field season will be implemented by the ADF&G. The University possesses the historical records of the Herring Bay sites and all of the raw and processed data from 1990 through 1994, and will be adding to the database through the 1995 field season. The University is in the best position to assure the consistency of the data to allow for comparisons to the previously collected data. The ADF&G is also currently involved in the monitoring and restoration of the nearshore through existing contracts with the University.

A contract will be issued to Coastal Resource Associates, Inc. (CRA) to assist in the analysis and report writing of *Fucus* monitoring and restoration data which will be collected with Dr. Stekoll in 1995.

D. Location

This study in FY 96 will take place in Fairbanks at the School of Fisheries and Ocean Sciences and at the Juneau Center School of Fisheries and Ocean Sciences in Juneau. The data which will be analyzed will have been collected in FY 95 from the intertidal zone in Herring Bay on Knight Island in Prince William Sound.

SCHEDULE

A. Measurable Project Tasks for FY96

Laboratory sample analysis, data analysis and graphics presentation will occur from October 95 to March 96. A draft final report will be submitted in June 1996. The final report will be submitted approximately four months after peer review.

B. Project Milestones and Endpoints

Objectives are to be met June 1996.

C. Project Reports

A draft final report will be submitted in June 1996. The final report will be submitted approximately four months after peer review.

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

This study will be conducted by a team of scientists with first hand experience in the monitoring of the effects of oil on intertidal communities. These investigators have worked together over the past five years to monitor the recovery of the intertidal community following the T/V *Exxon Valdez* oil spill. Principal investigators from the University of Alaska Fairbanks and University of Alaska Southeast will be coordinating efforts to study recovery of key algae and invertebrate species in the upper intertidal elevations.

ENVIRONMENTAL COMPLIANCE

This funding will support laboratory work and report writing and no permits or special requirements are anticipated.

PERSONNEL

Dr. Raymond Highsmith, Professor, Director West Coast National Undersea Research Center, UAF/SFOS, Fairbanks.

Dr. Highsmith has been the Coordinator and Principal Investigator of two *Exxon Valdez* Oil Spill projects; the Coastal Habitat Injury Assessment project and the Herring Bay Experimental and Monitoring Studies. His specialties include ongoing research of recruitment and population biology in the intertidal zone and is familiar with the effects of the oil spill on intertidal invertebrates throughout the spill impacted area.

1996 EXXON VALDEZ TRUST COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel	\$191.6	\$5.2						
Travel	\$17.5	\$0.0						
Contractual	\$108.4	\$156.1						
Commodities	\$6.3	\$0.0						
Equipment	\$0.0	\$0.0						
Subtotal	\$323.8	\$161.3	LONG RANGE FUNDING REQUIREMENTS					
General Administration	\$64.8	\$11.7	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
Project Total	\$388.6	\$173.0	N/A	N/A	N/A	N/A	N/A	N/A
Full-time Equivalents (FTE)	4.9	0.1						
Other Resources			Dollar amounts are shown in thousands of dollars.					
<p>Comments:</p> <p>This budget was revised on 7-24-1995 to incorporate revisions to the budget described in the letter from Molly McCammon to Raymond Highsmith and David Doudna. In his response, Mr. Doudna provided an explanation and justification for his budget and incorporating the newly negotiated 25% overhead rate. Please note that this budget has not officially gone through the University budget office for signature.</p> <p>In addition, ADF&G program manager cost now meet the one month per project program manager charge.</p>								

1996

Project Number: 96086-C (Close-Out)
 Project Title: Herring Bay Monitoring and Restoration Studies
 Agency: ADF&G

FORM 3A
 AGENCY
 PROJECT
 DETAIL

Prepared:

1 of 8

8/1/95

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Personnel Costs:			GS/Range/ Step	Months Budgeted	Monthly Costs	Overtime	Proposed FFY 1996
PM	Name	Position Description					
*	116110	LIB I	17J	0.0	5,530		0.0
	117064	FB III	18C	1.0	5,203		5.2
	Vacant	FB III	18B	0.0	5,047		0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
Subtotal				1.0	15,780	0	
Those costs associated with program management should be indicated by placement of an *.						Personnel Total	\$5.2
Travel Costs:			Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996
PM	Description						
	None						0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
Those costs associated with program management should be indicated by placement of an *.						Travel Total	\$0.0

1996

Project Number: 96086-C (Close-Out)
 Project Title: Herring Bay Monitoring and Restoration Studies
 Agency: AK Dept. of Fish & Game

FORM 3B
 Personnel
 & Travel
 DETAIL

1996 EXXON VALDEZ TRUS COUNCIL PROJECT BUDGET
 October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed FFY 1996
Description		
Contract with non-trustee agency		156.1
When a non-trustee organization is used, the form 4A is required.		Contractual Total
		\$156.1
Commodities Costs:		Proposed FFY 1996
Description		
None		
		Commodities Total
		\$0.0

1996

Project Number: 96086-C (Close-Out)
 Project Title: Herring Bay Monitoring and Restoration Studies
 Agency: ADF&G

**FORM 3B
 Contractual &
 Commodities
 DETAIL**

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET
October 1, 1995 - September 30, 1996

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1996
Description				
None				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
Those purchases associated with replacement equipment should be indicated by placement of an R.			New Equipment Total	\$0.0
Existing Equipment Usage:		Number of Units	Inventory Agency	
Description				
None				

1996

Project Number: 96089-C (Close-Out)
Project Title: Herring Bay Monitoring and Restoration Studies
Agency: ADF&G

**FORM 3B
Equipment
DETAIL**

1996 EXXON VALDEZ TRUST COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel	\$191.6	\$106.5						
Travel	\$17.5	\$6.3						
Contractual	\$108.4	\$10.5						
Commodities	\$6.3	\$1.6						
Equipment	\$0.0	\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$323.8	\$124.9	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
Indirect	\$64.8	\$31.2						
Project Total	\$388.6	\$156.1	N/A	N/A	N/A	N/A	N/A	N/A
Full-time Equivalents (FTE)	4.9	2.9						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
Comments:								
<p>NOTE:</p> <p>Report Writing Costs: The full amount requested for FY 96 is for the completion of a Final Herring Bay Monitoring and Restoration Studies report. No field activities are budgeted. The amount requested will cover approximately a nine month period.</p>								

1996

Project Number: 96086-C (Close-Out)
 Project Title: Herring Bay Monitoring and Restoration Studies
 Name: Contractor

FORM 4A
 Non-Trustee
 DETAIL

Prepared:

5 of 8

8/1/95

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Personnel Costs:				Months	Monthly		Proposed
	Name	Position Description		Budgeted	Costs	Overtime	FFY 1996
	Highsmith	Principal Investigator-Project Coordinator		1.0	10,366		10.4
	Stekoll	Principal Investigator		1.0	8,859		8.9
	Doudna	Project Manager		0.5	5,634		2.8
	Van Tamelen	Research Associate		6.0	4,686		28.1
	Lindeberg	Technician		4.0	4,177		16.7
	Saupe	Technician		3.0	4,497		13.5
	Rucker	Technician		1.5	4,947		7.4
	Egan	Lab Assistant		2.0	3,618		7.2
	Mickelson	Graduate Student		8.0	1,096		8.8
		Graduate Student tuition		8.0	338		2.7
							0.0
							0.0
Subtotal				35.0	48,218	0	
Personnel Total							\$106.5
Travel Costs:				Ticket	Round	Total	Proposed
	Description			Price	Trips	Days	FFY 1996
	RT FBKS-ANC (Oil Spill Workshop)			138	2	14	2.0
	RT JUN-ANC (Oil Spill Workshop)			311	2	14	2.3
	RT FBKS-JUN (Data Analysis)			400	2	10	2.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
Travel Total							\$6.3

1996

Project Number: 96086-C (Close-Out)
 Project Title: Herring Bay Monitoring and Restoration Studies
 Name: Contractor

FORM 4B
 Personnel
 & Travel
 DETAIL

1996 EXXON VALDEZ TRU: COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
Juneau	CRA-Fucus Monitoring/Restoration Subcontract for data analysis and report preparation	7.0
	Warehouse/Storage	0.7
	Publications/Printing	0.2
	Long-Distance Phone/FAX	0.6
	Computer Maintenance	0.2
Fairbanks	Warehouse/Storage	0.6
	Publications/Printing	0.6
	Long-Distance Phone/FAX	0.6
Contractual Total		\$10.5
Commodities Costs:		Proposed
Description		FFY 1996
Juneau	Office Supplies	0.4
	Computer Supplies (paper, disks, printer cartridges, software and upgrades)	0.4
Fairbanks	Office Supplies	0.4
	Computer Supplies (paper, disks, printer cartridges, software and upgrades)	0.4
Commodities Total		\$1.6

1996

Project Number: 96086-C (Close-Out)
 Project Title: Herring Bay Monitoring and Restoration Studies
 Name: Contractor

FORM 4B
Contractual &
Commodities
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1996
Description				
None				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
Those purchases associated with replacement equipment should be indicated by placement of an R.		New Equipment Total		\$0.0
Existing Equipment Usage:		Number of Units		
Description				
None				

1996

Project Number: 96086-C (Close-Out)
 Project Title: Herring Bay Monitoring and Restoration Studies
 Name: Contractor

FORM 4B
 Equipment
 DETAIL

FUCUS AS STRUCTURE FOR OTHER ORGANISMS

Project Number: 96088
Restoration Category: Research and Monitoring
Proposer: University of Alaska
Lead Trustee Agency: ADFG
Duration: 3 years
Cost FY 96: \$302.5
Cost FY 97: \$328.2
Cost FY 98: \$176.5
Geographic Area: Prince William Sound and Cook Inlet
Injured Resource: Intertidal Organisms

ABSTRACT

The structural and functional aspects of the upper intertidal community were severely disrupted by the EVOS and subsequent cleanup. The brown alga, Fucus gardneri, is the dominant organism in this community where it provides food, foraging areas, and shelter for a variety of other plants and animals. Experiments and observations made over the past four years in Herring Bay have shown that the Fucus populations in the upper intertidal are not recovering from the effects of the oilspill and cleanup. This is especially true of sheltered rocky habitats with a southern exposure. The upper intertidal in these regions has remained devoid of Fucus and other algal species. Invertebrate populations are also depauperate in this region because of the lack of algal cover.

The goals of this project are to 1) define the factors which have limited the recovery of Fucus populations, 2) test various techniques to accelerate the recovery of Fucus populations in the upper intertidal, and 3) determine the consequences for other organisms due to this slow recovery of Fucus, and 4) define the geographical extent of upper intertidal habitat throughout PWS that has not recovered.

INTRODUCTION

Fucus gardneri is the dominant intertidal organism in Prince William Sound and throughout much of the oil spill affected region. Fucus is a large (up to 50 cm in length), perennial brown alga that forms extensive beds in the intertidal. The dichotomous branching and bushy growth form of this species provides protection from heat, desiccation, and predation to a wide variety of intertidal organisms (Project 93039). Mobile subtidal and pelagic animals also use Fucus for foraging and spawning habitat. Studies of Fucus beds in other regions have shown that many invertebrate species, especially herbivores, are more abundant in Fucus beds than on adjacent barren substrate (Albrecht and Reise, 1994).

The importance of Fucus beds and their associated invertebrate species in providing food for subtidal

and terrestrial species has not been well defined. Food chain links between the Fucus beds and various fish and bird species have been identified, but it is unknown to what extent intertidal and subtidal species in Prince William Sound are dependent upon the Fucus beds for their survival. It may be possible that the declines observed for invertebrates and fish may have been due to the lost Fucus canopy and not the oil.

Despite large efforts directed at examining the effects of the oil spill on intertidal communities, there is little knowledge about the role of Fucus as structure for other biota. The major reason for this lack of understanding is that many of the studies were focused on the injuries caused by the oil spill. For example, if decreases in a limpet population dependent upon Fucus are observed along with decreases in Fucus populations, then it is unknown whether the limpet decline is due to effects of the oil spill or cleanup or due to the lost Fucus canopy.

Many intertidal organisms appear to be dependent upon Fucus as habitat (Project 93039, Herring Bay studies). Limpets and littorine snails are often found living on the fronds of Fucus, and amphipods and isopods also use Fucus fronds for habitat. In addition, other seaweeds may benefit from Fucus. We have observed that there is often a layer of small bladed and branched algae beneath the Fucus canopy.

Subtidal and pelagic animals may also make use of Fucus beds during high tides. Except for the eggs left by spawning fish, little evidence is left by subtidal and pelagic animals when they use Fucus beds at high tide. Yamada (Personal communication) documented that crabs are voracious predators on littorine snails in an intertidal habitat similar to Prince William Sound. We have also observed pink salmon swimming in the intertidal region during late summer.

NEED FOR THE PROJECT

A. Statement of Problem

Many intertidal organisms as well as subtidal animals use the structure provided by Fucus gardneri as habitat and foraging areas. These plants and animals, therefore, may have been injured by the reduction of Fucus canopy throughout the spill affected area. Recovery of organisms that use Fucus as structure is dependent upon recovery of Fucus and the presence of Fucus canopy. Fucus has recovered, or will recover soon, in low and mid tidal zones, but in the upper intertidal recovery has been slow due to short dispersal distances, lack of Fucus canopy, and high heat and desiccation stress (Project 93039).

B. Rationale

By examining the use of Fucus beds by other organisms at both low and high tides and comparing oiled to unoiled areas, we will be able to assess the extent to which Fucus serves as habitat for both animals and plants. By also monitoring the recovery status of Fucus we can assess the recovery of species dependent upon Fucus and determine why recovery has or has not taken place. If future oil spills or similar catastrophes occur, then we will be justified in assessing the recovery of intertidal

communities simply by monitoring the recovery of Fucus beds.

C. Summary of Major Hypotheses and Objectives

This project seeks to assess the recovery of Fucus canopy in the high intertidal and to ascertain what types of plants and animals use Fucus for habitat and foraging areas. We will continue to test various methods for accelerating the recovery of Fucus populations in regions where no Fucus recovery has been observed. We will also estimate the geographical extent of this recovery and identify the habitat types; based on geomorphology, wave exposure, and solar aspect; where Fucus beds have been slow to recover.

D. Completion Date

Monitoring of Fucus and users of Fucus canopy will be complete in FY 96 for Prince William Sound and in FY 97 for Cook Inlet and Kenai Peninsula. Reports will be completed in FY 98.

COMMUNITY INVOLVEMENT

No community involvement is anticipated at this time. We would, however, entertain the possibility of participating in informational meetings for some communities or submitting an article for the Trustee Council newsletter. We have not budgeted any funds for these community involvement projects.

BUDGET	FY96	FY 97	FY 98
Personnel	6.7	27.8	6.7
Travel	0.0	0.0	0.0
Contractual	276.8	277.0	147.8
Commodities	0.0	0.0	0.0
Equipment	0.0	0.0	0.0
Subtotal	283.5	283.7	154.5
Gen. Admin.	19.0	23.4	22.0
Total	302.5	382.2	176.5

PROJECT DESIGN

A. Objectives

This project is designed to determine the specific state of recovery of Fucus in the high intertidal and to ascertain the extent to which intertidal and subtidal biota utilize Fucus canopy for habitat and foraging areas. The specific questions to be answered by the experiments and observations are listed below.

1. What is the geographical extent of upper intertidal habitat in that is devoid of Fucus canopy?
2. What are the physical and biological factors regulating the recovery of upper intertidal communities in different geographic locations?
3. Can techniques be developed to accelerate the recovery of Fucus populations in regions where recovery has not yet occurred?
4. What is the temporal scale of recovery in different geographic locations?
5. What organisms are being or have been affected by the loss of food or habitat provided by Fucus?
 - a. Which intertidal invertebrates are dependent upon Fucus canopy for habitat and can we estimate recovery of these invertebrates by monitoring the recovery of Fucus beds? Is the composition of this community consistent in all geographic locations?
 - b. Do subtidal invertebrate predators forage in Fucus beds during high tides?

B. Methods

1. Distribution of Upper Intertidal Fucus Populations

The tidal level of the upper limit of the Fucus bed will be determined at each of the protected rocky sites originally monitored in the damage assessment studies. These sites will be revisited during 1996 and 1997 in project 96086 (Intertidal monitoring). Additional sites will be selected to obtain a sufficient number of replicates in at least four categories of solar aspect. Power analyses of existing data samples will be conducted to define the number of replicate sites that will be necessary. The upper limit of the Fucus bed will be determined by measuring the vertical distance from mean lower low water to the uppermost Fucus plant along a one meter wide transect. Each transect will begin at a randomly determined starting point, and the height to the uppermost Fucus plant will be measured with stadia rods and handheld sighting levels.

2. Physical and Biological Factors Regulating Recovery of the Upper Intertidal

Field experiments performed in Herring Bay over the past four years have identified various physical factors and biological interactions that structure the distribution and abundance of Fucus populations. Parts of these experiments will be repeated in other locations within Prince William Sound and the Kenai Peninsula regions to verify the generality of these results to the entire region covered by the EVOS. The focus of these experiments will be in the sheltered rocky habitat where we will look at egg dispersal processes and survival and growth of juvenile plants.

3. Acceleration of Fucus Recovery

For the past three years we have been testing various techniques to accelerate the recovery of Fucus

to an upper intertidal location in Herring Bay. One technique which involves to the use of coconut fiber erosion control fabric has shown some promise in promoting the recruitment of Fucus plants in this habitat. We will continue to monitor the experiments we have started in Herring Bay and test a few other techniques in new regions where Fucus has not recovered. The goal of this study will be develop low cost techniques that will provide small core populations of Fucus in the upper intertidal from which additional recruitment can occur. Our observations in Herring Bay have shown that new Fucus populations are limited by egg dispersal and new populations occur only near established groups of mature plants. The restoration techniques will involve the continued use of the coconut fiber fabric as well as the use of shredded fabric which will be applied to the intertidal substrate with adhesives.

4. Temporal Scale of Recovery in Different Localities

The recovery rates of the intertidal community in Herring Bay have varied with tidal height and beach aspect. Populations in the upper intertidal of beaches with a south facing aspect have shown the least amount of recovery. We will classify all the damage assessment sites that will be revisited in 1996 with respect to solar aspect. The state of Fucus recovery at these sites will be determined based on plant density, plant reproduction, and height of the upper Fucus boundary. Differences in these parameters within tidal height and solar aspect classes will be tested with an ANOVA. Additional sites will be chosen from the ADNRS GIS database to obtain sufficient number of replicates for each solar aspect class. From these data we will also develop a regression analysis where we will be able to extrapolate the rates to final recovery in all the different habitat types.

5. Effects of Fucus Loss on Associated Organisms

The invertebrates and algae living on Fucus will be determined and counted. Thirty randomly selected, large Fucus plants will be chosen at each of two tidal levels at each site. Transect tapes (100 m) parallel to the water line will laid in the high and mid tidal zones. The nearest plant, greater than about 40 g in wet weight, to 30 randomly selected points along each transect will be selected, carefully enclosed in a plastic bag, and removed from the rock. The tidal level of each plant will be recorded. These plants will then be transported to the support vessel and removed from the bags. All of the invertebrates and algae found on each plant will be sorted, counted, and weighed. The plant will then be weighed, measured for length, and assessed for reproductive status.

Some algae and invertebrates may not be living directly on Fucus plants. To assess the extent to which these epilithic organisms are dependent on Fucus structure, it will be necessary to perform experiments lasting more than one day. In areas in which both oiled and control sites are present in close proximity, ten pairs, oiled and unoiled, of sites will be selected. At each site a quadrat (25 x 25 cm) will be randomly located in a Fucus bed at the high tidal zone. The quadrat will be marked with stainless steel screws and all of the Fucus will be removed from the quadrat and within 0.5 m of the quadrat. The number and type of invertebrates in the quadrat will be immediately counted. Counts will then be repeated daily until numbers stabilize.

The use of Fucus beds by mobile subtidal predators will be determined by counting the number and type of these predators within 1 meter of a transect running from MLLW to MHW at high tide.

Transects will be observed by snorkeling from the low to the high zone, and the observations will be recorded by tidal level (low, mid, high zones). This procedure will be done at the ten pairs of sites used for the Fucus removal experiments described above. The transects will be located 2 m to the right of each clearing and marked with a rope anchored at the high and low zones and marked by tidal level.

If invertebrate predators are abundant, we will assess the impacts of their foraging by tethering littorine snails in the high and mid zones and inspecting the littorines after 1 and 2 days for evidence of predation. Invertebrate predators often leave distinctive foraging signs, so mortality can be attributed to specific predators. Snails will be tethered by gluing a small ring to the apex of the shell and tying fishing line to the ring and to a screw placed in the rock. Ten snails will be attached to each of ten screws and inspected daily for evidence of predation.

To determine the role of Fucus as structure alone, we will compare the use of artificial Fucus to that of live Fucus. In each of fifteen sites, two quadrats (25 x 25 cm) will be located in the high zone and cleared of all Fucus. Five braided nylon ropes with either artificial Fucus or live Fucus will then be attached to the sides of the quadrats such that Fucus plants are in the quadrat but the attached rope ends are outside of the quadrat. Two plants will be attached to each rope, giving a density of large plants similar to actual densities. Artificial and live Fucus will be randomly allocated to the quadrats. The number and type of invertebrates and algae found living on the plants and in the quadrat will be assessed after five days, one month, and two months.

C. Contracts and Other Agency Assistance

This project will be coordinated with the intertidal monitoring project 96086 to utilize the same vessel for most of the studies. In areas where experiments and sampling will be performed over multiple days, we will utilize air taxis or a separate vessel charter.

A contract will also be issued to Coastal Resources Associates (CRA) of Vista, California. CRA has been involved with both the Herring Bay studies and previous intertidal monitoring studies since their inception in 1989. They have developed some of the experimental procedures and sampling techniques described here.

We will need updates of the GIS model from the Alaska Department of Natural Resources to include various parameters for shoreline oiling classification, habitat type, and aspect.

D. Location

These proposed studies will be conducted in Prince William Sound in FY 96 and in both Prince William Sound and Cook Inlet in FY 97.

SCHEDULE

A. Measurable Project Tasks for FY 96

Start-up to May 15: Arrange logistics, final design of experiments and sampling procedures
May 15-July 30: Field work-Set-up and monitor experiments
July 30-November: Analyses of field data
November-April: Write annual report and prepare for next field season

B. Project Milestones and Endpoints

During the first summer of field work (1996) we will use the intertidal monitoring project platform to visit their sites in Prince William Sound. We will also charter our own vessel to visit additional sites and to set up and monitor experiments in Prince William Sound. Thus, by then end of the field season we will be able to complete Objectives 1 and 4 for Prince William Sound. We will also have set up and obtained preliminary data from experiments in Prince William Sound to attain objectives 2, 3, and 5. These will need to be monitored during the following field season. During the 1997 field season we will also use the intertidal sampling vessel to visit sites in the Cook Inlet and Kenai region allowing us to complete objectives 1 and 4 and to set up experiments evaluating objective 2 in that area. The sites with experiments set up on them will be revisited in fall by helicopter. We will also revisit sites using our own vessel charter in Prince William Sound to complete the experiments started there the previous season. Finally, in spring of 1998 we will visit the Cook Inlet and Kenai sites by helicopter to monitor the experiments set up the previous season.

October 1996 Objectives 1, 2 (part), 4, and 5 (part) will be completed for Prince William Sound

October 1997 Objectives 1, 3, 4, and 5 will be completed by this time.

April 1998 Objective 2 will be completed with a final sampling in Cook Inlet

C. Project Reports

We will submit an annual report to the Chief Scientist by April of 1997. A draft of the final report will be submitted by January 1998 with the final being submitted by April 1998. This final report will include data gathered from the final sampling in Cook Inlet.

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

This project will be closely coordinated with the intertidal sampling project, so that we can share the same vessel charter to visit previous sampled sites throughout Prince William Sound and Cook Inlet

ENVIRONMENTAL COMPLIANCE

We anticipate that this project will be categorically excluded from all NEPA regulations. State scientific collection permits will be obtained prior to the start of the field season from ADF&G.

PERSONNEL

1. Dr. Michael Stekoll, Professor, UAF/JCSFOS and UAS, Juneau - principal investigator

Dr. Stekoll has been a principal investigator on three Exxon Valdez oil spill projects: the Coastal Habitat Injury Assessment project, the Herring Bay Experimental and Monitoring Studies, and the Shallow Subtidal Assessment project. He has co-authored annual and final reports for these projects and has produced refereed publications on various aspects of these projects. His specialties include marine pollution biology, the biology and ecology of seaweeds in Alaska, especially Macrocystis and Fucus, and the mariculture of kelps and red seaweeds.

2. Dr. Lawrence Deysher, Sr. Scientist, Coastal Resource Associates, Inc. - Fucus restoration and ecology

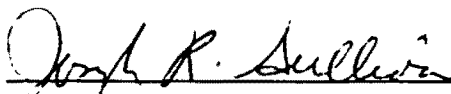
Dr. Deysher is a marine ecologist who has been with CRA since its inception. He directed field studies of intertidal algae as part of the EVOS damage assessment studies and participated in the original site selection for the CHIA monitoring sites. He has co-authored the annual reports on the intertidal algal monitoring studies and a paper on the use of DNR-GIS in selecting the intertidal monitoring sites for the CHIA studies. He is currently working on the experimental algal studies in Herring Bay as well as directing studies of kelp resources in California using a Geographic Information System.

3. Dr. Peter van Tamelen, Research Associate, UAF/JCSFOS, Juneau - Fucus ecology

Dr. van Tamelen has been working in Herring Bay on the intertidal algal studies since 1990. He has extensive experience in marine intertidal ecology, including studies on plant-herbivore interactions, succession, algal recruitment, and effects of physical factors on biological communities.



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Date Prepared

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996							
Personnel		\$6.7							
Travel		\$0.0							
Contractual		\$276.8							
Commodities		\$0.0							
Equipment		\$0.0	LONG RANGE FUNDING REQUIREMENTS						
Subtotal	\$0.0	\$283.5	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002	
General Administration		\$19.0							
Project Total	\$0.0	\$302.5	\$328.2	\$176.5					
Full-time Equivalents (FTE)		0.1							
Dollar amounts are shown in thousands of dollars.									
Other Resources									
Comments:									

1996

Project Number: 96088
Project Title: Fucus as Structure for other Organisms
Name: ADF&G

FORM 3A
AGENCY
PROJECT
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Personnel Costs:			GS/Range/ Step	Months Budgeted	Monthly Costs	Overtime	Proposed FFY 1996
PM	Name	Position Description					
*	11-5047	Fishery Biologist III	18B	1.0	5,047		0.0
*	11-6110	Librarian I	17J	0.3	5,530		5.0
							1.7
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
Subtotal				1.3	10,577	0	
Those costs associated with program management should be indicated by placement of an *.							Personnel Total \$6.7
Travel Costs:			Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996
PM	Description						
	None						0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
Those costs associated with program management should be indicated by placement of an *.							Travel Total \$0.0

1996

Project Number: 96088
 Project Title: Fucus as Structure for other Organisms
 Name: ADF&G

FORM 3B
 Personnel
 & Travel
 DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
Contract with non-trustee agency		276.8
When a non-trustee organization is used, the form 4A is required.		Contractual Total
		\$276.8
Commodities Costs:		Proposed
Description		FFY 1996
None		
Commodities Total		\$0.0

1996

Project Number: 96088
Project Title: Focus as Structure for other Organisms
Name: ADF&G

FORM 3B
Contractual &
Commodities
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1996
Description				
None				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
Those purchases associated with replacement equipment should be indicated by placement of an R.		New Equipment Total		\$0.0
Existing Equipment Usage:		Number of Units	Inventory Agency	
Description				
None				

1996

Project Number: 96088
Project Title: Fucus as Structure for other Organisms
Name: ADF&G

FORM 3B
Equipment
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996							
Personnel		\$110.9							
Travel		\$8.9							
Contractual		\$47.8							
Commodities		\$4.9							
Equipment		\$12.0							
Subtotal	\$0.0	\$184.5	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002	
Indirect		\$92.3							
Project Total	\$0.0	\$276.8	\$277.0	\$147.8					
Full-time Equivalents (FTE)		1.9							
Dollar amounts are shown in thousands of dollars.									
Other Resources									

Comments: Indirect Costs: Indirect costs are calculated at 50% TDC. Currently, the Contractor and the EVOS Trustee Council are negotiating a rate agreement for projects which are long-term, sole source in origin, and guarantee a minimum amount per year of funding to the Contractor. Therefore, this rate is subject to change pending the negotiation.

Cost associated with the report writing activities (in the FFY 97 budget) will be \$100.0 (includes personnel, supplies, and services for annual reports, final reports and refereed publications.

This project will utilize the vessel charter contracted by Project 96037: Coastal Habitat Intertidal Monitoring. If 96037 is not approved by the Trustee Council, the budget for this project MUST increase to include a vessel charter.

1996

Project Number: 96088
Project Title: Fucus as Structure for other Organisms
Name: Contractor

FORM 4A
Nor. Trustee
DETAIL

Prepared:

5 of 12

28/95

October 1, 1995 - September 30, 1996

1996

FORM 4B
Personnel
& Travel
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
Contract with sub-contractor to assist in the design of project, field work, data analysis and report preparation		25.0
Vessel Charter 60' vessel @\$1.6 X 10 days		16.0
RT air taxi from port to vessel 10 RT @ \$390/RT		3.9
Long distance telephone/FAX		0.5
Publication/printing for reports		0.5
Freight and Shipping		
Trucking supplies to the field (\$0.50lb)		1.0
Airline excess supplies to the field (\$50/item)		0.5
Warehouse for storing field supplies and equipment		0.4
Contractual Total		\$47.8
Commodities Costs:		Proposed
Description		FFY 1996
Office Supplies		0.4
Computer Supplies (disks, upgrades)		0.5
Field Supplies:		
Dry suit, mask, fins, snorkel, hood, weight belt (2)		1.6
Mikita cordless drill, screws and anchors		0.2
Raingear, boots		0.4
Surveyors tape, sight level, stadia		0.2
Write-in-the-rain paper and notebooks, metal tatums		0.2
5 gal plastic buckets, plexiglas, nylon rope, sampling quadrates, tools and spare parts		0.4
Float coats (3)		0.9
gas and oil		0.1
Commodities Total		\$4.9

1996

Project Number: 96088
Project Title: Fucus as Structure for other Organisms
Name: Contractor

FORM 4B
Contractual &
Commodities
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1996
Description				
N	Deskjet computer printer	1	500	0.5
N	Hand held VHF marine radio for use in the field	3	500	1.5
N	14' Zodiac and 25 hp outboard motor	1	10,000	10.0
Those purchases associated with replacement equipment should be indicated by placement of an R.		New Equipment Total		\$12.0
Existing Equipment Usage:			Number of Units	
Description				
	Computers and associated software	4		
	Survival suits	4		
	Laserjet printer	1		
	Temperature and salinity meter	1		

1996

Project Number: 96088
Project Title: Fucus as Structure for other Organisms
Name: Contractor

FORM 4B
Equipment
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996							
Personnel		\$14.7							
Travel		\$1.8							
Contractual		\$3.2							
Commodities		\$3.0							
Equipment		\$0.0							
Subtotal	\$0.0	\$22.7	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002	
Indirect		\$2.3							
Project Total	\$0.0	\$25.0	\$25.0	\$10.0					
Full-time Equivalents (FTE)		0.2							
Other Resources									

LONG RANGE FUNDING REQUIREMENTS

Dollar amounts are shown in thousands of dollars.

Comments: Indirect Costs: Indirect costs are calculated at 10% TDC.

The subcontractor will assist the contractor in the conduct of the study of the role of structure provided by fucus in providing habitat for other organisms in the intertidal zone. The sub-contractor will assist in the design of studies, take part in the field work, and assist with data analysis and report preparation.

1996

Project Number: 96088
Project Title: Fucus as Structure for other Organisms
Name: Sub-Contractor

FORM 5A
Non-Trustee
DETAIL

Prepared:

9 of 12

28/95

October 1, 1995 - September 30, 1996

1996

Project Number: 96088
Project Title: Fucus as Structure for other Organisms
Name: Sub-Contractor

FORM 5B
Personnel
& Travel
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
Postage and freight		0.2
Insurance		1.5
Office rent		1.5
Contractual Total		\$3.2
Commodities Costs:		Proposed
Description		FFY 1996
Office supplies		0.4
field supplies		0.5
computer software		1.5
computer supplies		0.5
copy costs		0.1
Commodities Total		\$3.0

1996

Project Number: 96088
 Project Title: Fucus as Structure for other Organisms
 Name: Sub-Contractor

FORM 5B
 Contractual &
 Commodities
 DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1996
Description				
	None			
Those purchases associated with replacement equipment should be indicated by placement of an R.				New Equipment Total
				\$0.0
Existing Equipment Usage:		Number of Units		
Description				
	None			

1996

Project Number: 96088
 Project Title: Fucus as Structure for other Organisms
 Name: Sub-Contractor

FORM 5B
 Equipment
 DETAIL

Mussel Bed Restoration and Monitoring

Project Number: 96090

Restoration Category: General restoration; monitoring

Proposers: Malin M. Babcock
NMFS, Auke Bay Laboratory

Gail Irvine
National Biological Service

Lead Trustee Agency: NOAA, DOI

Duration: 10 years

Cost FY96: \$209,700

Cost FY97: \$0

Cost FY98: \$280,000

Cost FY99: \$165,000

Cost FY00: \$0

Cost FY01: \$285,000

Cost FY02: \$170,000

Cost FY03: \$0

Cost FY04: \$290,000

Geographic Area: Prince William Sound, Kenai Peninsula

Injured Resources/Service: Mussels; indirectly Harlequin ducks, Black oystercatchers, Subsistence, Recreation

ABSTRACT

A comprehensive report will be produced synthesizing and summarizing 4 years of studies on the persistence of oiling in mussels beds in Prince William Sound and the Gulf of Alaska and restoration of 12

of these beds. Chemical analyses of mussel and sediment samples collected in 1995 will be completed early in 1996. No new sample collection or site visits are proposed for FY96.

INTRODUCTION

This project for FY 96 will produce a final report which will synthesize and summarize all research and activities conducted under Trustee Council authorization relating to *Exxon Valdez* oil-contaminated mussel beds, 1992-1995. Funds requested also include chemical analyses of samples collected in 1995. There is no proposed field work for 1996.

The persistence of *Exxon Valdez* crude oil underlying some dense mussel (*Mytilus trossulus*) beds in Prince William Sound (PWS) and along the Gulf of Alaska (GOA) began to cause concern in the spring of 1991 and was confirmed in surveys by NOAA's Auke Bay Laboratory (ABL) and the National Park Service (NPS). This project has been funded from 1992 through 1994 under Trustee Council Studies No. R103, 93036, 94090, and 95090.

Substantial amounts of petroleum hydrocarbons (HCs) from the *Exxon Valdez* oil (EVO) spill remain entrained in sediments underlying some dense mussel beds situated along the shorelines impacted by the spill. In 1992 and 1993 (only limited sampling survey sampling was conducted in 1994), ABL and NPS sampled mussels and sediments from 88 beds to determine the presence and level of oiling. Sediments collected from 31 of these beds in PWS had total petroleum hydrocarbons (TPH) concentrations greater than 10,000 $\mu\text{g/g}$ wet weight and 5 of the beds along the GOA showed greater than 5,000 $\mu\text{g/g}$. Decreases in HC concentrations between the years was only moderate and dependent on site location and exposure to storm activity.

In 1994, the Alaska Department of Environmental Conservation (ADEC) and ABL led an effort to manually clean 12 beds in PWS with the help of Chenega Corporation members. Preliminary evaluation of HC levels in sediments show promise that this activity, at least in the short term (<30 days), was successful in reducing HC concentrations in sediments underlying the mussels. Further evaluation is scheduled for 1995.

Other research conducted under this study, 1992-1993, included within-bed variability of oil distribution, stripping and patch removal of mussels to accelerate flushing of the oil, and examination of various indices of chronic exposure to EVO in mussels.

An annual sampling of sediments and mussel from previously documented oiled beds is scheduled in 1995 in order to determine rates of reductions in hydrocarbon concentrations in these beds. Rates will be compared between untreated and restored oiled mussel beds.

In addition to synthesizing and summarizing research conducted under this 4-year study, we will conduct a power analysis of the data to outline a future monitoring schedule to track HC concentrations at these sites to recovery levels.

NEED FOR PROJECT

- A. Statement of Problem and**
- B. Rationale**

The presence of substantial levels of petroleum hydrocarbons persisting under dense mussel beds in PWS and the Gulf of Alaska provides a continuing, potential source for HCs. Restoration (cleaning) of selected mussel beds (12 mussel beds were restored in PWS in 1994) should reduce potential exposure to HCs in subsistence users and higher predators such as harlequin ducks, oyster catchers and juvenile otters.

Data and results from previous years, and certainly 1995, need to be examined and analyzed for identifying trends in changes in oiling levels at the various documented oiled mussel beds. The manual cleaning effort in 1994 will require scrutiny of data produced by 1995 sampling to evaluate the efficacy of this activity in reducing HC levels associated with these mussel beds.

A final report will provide information needed for decisions relevant to future passive or aggressive actions with oiled mussel beds in both PWS and the GOA.

C. Summary of Major Hypotheses and Objectives

The projects objectives for FY 96 are to complete the chemical analyses of mussel and sediment samples collected in 1995 and prepare a comprehensive report covering the life of this study. Over the 4-year study, objectives have been 1) to establish the geographical distribution and intensity of oiling of mussel beds and to determine changes in selected beds over the years; 2) to examine within-bed distribution of oil; 3) to test minimally intrusive methods of decreasing petroleum hydrocarbon loads; 4) to manually restore 12 oiled mussel beds; and, 5) to examine various measures of stress in mussels under chronic exposure to EVO.

D. Completion Date

The comprehensive 4-year report will be completed during FY 96; further monitoring of HC levels in oiled mussel beds will likely be proposed on a 2- or 3-year schedule. *Exxon Valdez* oil may persist underneath these mussel beds greater than 20 years.

COMMUNITY INVOLVEMENT

For FY 96, we anticipate little community involvement during this data synthesis and summarization process. During previous years, we have participated in the Trustee Councils public processes where appropriate; and, in 1994 and 1995, actual work conducted under this study was assisted with the help of residents of Chenega, Alaska, the village most impact by the spill.

FY 96 BUDGET

	NOAA	DOI
Personnel	129.8	35.2
Travel	2.9	2.1
Contractual	0.0	0.0
Commodities	13.7	1.2
Equipment	0.0	0.0
Subtotal	146.4	38.2
Gen. Administration	19.5	5.3
Total	165.9	43.8

PROJECT DESIGN

A. Objectives

1. Conduct chemical analyses of samples collected in 1995.
2. Prepare a report synthesizing and summarizing all research and activities conducted under the 4-year history of this project.

Previous objectives have been:

3. Establish the geographic extent and intensity of oiling in contaminated mussel beds in PWS and GOA.
4. Determine within-bed distribution of crude oil in sediments underlying contaminated mussel beds.
5. Test minimally intrusive methods (stripping and patch removal) of decreasing the amount of EVO underlying oiled mussel beds.
6. Test for physiological and biological differences between chronically exposed mussels and clean mussels.
7. Manually restore selected oiled mussel beds with relatively high levels of contamination.

B. Methods

There will be no field work or sampling proposed for 1996. As part of the proposed report, we will conduct a power analyses to enable the establishing of a future monitoring schedule.

C. Contracts and Other Agency Assistance

None are anticipated.

D. Location

All work will be conducted at NOAA's Auke Bay Laboratory in Juneau, Alaska.

SCHEDULE

A. Measurable Project Tasks for FY 96

Start-up to 28 February 1996: Chemical analyses conducted

Start-up through Oct 1996: Data Analyses and Interpretation; report production

January, 1996: Summary of work presented at Trustee Workshop

B. Project Milestones and Endpoints

December 1995: 2 manuscripts published in EVOS Proceedings

This is included in Project Reports discussed below. The ultimate milestone/endpoint of this project will be when all documented oiled mussel beds have petroleum hydrocarbon concentrations in mussels and underlying sediments near background, or prespill, levels.

C. Project Reports

A comprehensive "final" report will be produced which will synthesize and summarize all research and activities conducted under this study (1922-1995). This report will also present a proposed monitoring schedule for future years to track decreases in levels of contamination in these mussel beds to a recovery status.

October, 1996: Comprehensive Report

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

This project is coordinated with several other Restoration studies. Data from study sites established under this project will be shared with the Nearshore Vertebrate Predator investigators and provided to the Shoreline Assessment group for comparison with their project. Other studies that have been associated with this project include the old Coastal Habitat 1B project, consumer species oriented projects (harlequin ducks, river otters, sea otters, black oyster catchers), and the subtidal sediment project.

NOAA's Auke Bay Laboratory has and will continue to contribute facilities, equipment including computer hardware, and dollars toward the success of this project. This project falls under National Marine Fisheries Service mandated responsibility for stewardship of living marine resources. It is not fully funded by our agency because of lack of base funding for successfully conducting the needed studies.

ENVIRONMENTAL COMPLIANCE

No Environmental assessment of impact statement is required.

PERSONNEL

MALIN M. BABCOCK

Education: Oregon State University, 1963. B. S., Zoology
University of Alaska Fairbanks, 1968. M. S., Zoology (Fisheries)

Experience: 1969-present. Researcher and Task Leader, Auke Bay Laboratory, National Marine Fisheries Service, Juneau, Alaska. Field, lab, and analytical expertise, and data analyses and interpretation particularly with effects of petroleum hydrocarbons on aquatic fish and shellfish. Studies have included Prince William Sound chemical baseline, short term and long term water-soluble fraction of crude oil and sediment toxicity tests assessing physiological and biochemical impacts - including growth and reproduction. I became Task Leader for the Coastal Habitat task within Habitat Investigations, ABL, in 1988 and directly supervise several staff scientists in varied research projects. I have strong participation in overall Habitat Investigations research planning, budget management and staffing.

After the *Exxon Valdez* oil spill, I was co-principal investigator for the EVOS Coastal Habitat Study "Pre-spill and post-spill hydrocarbon concentrations in mussels and sediments in Prince William Sound", becoming Principal Investigator of this project in 1991 and 1992; was also Principal Investigator for the NRDA study "Injury to Oysters" in 1989. In 1991, I participated in the interagency planning for investigating an evolving problem - that of the effects of contaminated mussel beds on higher consumer organisms, and led the preliminary field effort for identifying these beds and sampling parameters to establish the extent and intensity of petroleum hydrocarbons contamination.

I have been Project Leader for NOAA for the PWS portion of Mussel Bed Restoration and Monitoring - coordinating and leading a staff to investigate extent and intensity of oiling; distribution of HCs within a mussel bed; effects of minimally intrusive manipulative techniques to reduce HCs by increasing exposure of oiled sediments; effects of chronic oiling on mussels (byssal thread production, condition and reproductive indices, glycogen stores, feeding rates, growth, and histopathological abnormalities). In 1994, with staff from the Alaska Department of Environmental Conservation and Chenega residents, we manually restored 12 oiled mussel beds in Prince William Sound.

Additionally, staff under my direct supervision are involved in many aspects of EVOS Restoration program for several studies, training all NRDA study personnel in sampling for hydrocarbons, the NRDA/Restoration database, sample custody and tracking, etc.

Relevant Publications: Over 25 publication/reports - most of which involve effects of exposure to petroleum hydrocarbons on various Alaskan species of fish and shellfish. Over 20 public presentations of scientific studies.

GAIL V. IRVINE

Education: University of California at Santa Barbara, 1969. B.A.(honors), Zoology
University of Washington, Seattle, 1973. M.S., Zoology
University of California at Santa Barbara, 1983. Ph.D. Biological Sciences (Aquatic and Population Biology)

Experience: 1994 - Current. Position transferred to the new National Biological Survey, Department of Interior.

1990 - present. Coastal Resources Specialist, National Park Service. Research in marine community ecology; developing and directing a coastal monitoring and research program for the National Park Service. Thus far, the research has been concentrated in two national parks oiled by the *Exxon Valdez* spill, Kenai Fjords and Katmai National Parks. Supervised the oiled mussel bed, Gulf of Alaska project for the Trustee Council.

1984 - 1990. Marine Biologist, Minerals Management Service. Environmental analysis, including potential effects of oil and gas development on marine plants, invertebrates, and fishes (pelagic, nearshore and benthic communities). Research on coelenterate ecology in the Chukchi Sea.

My education and experience have been concentrated in the fields of community and population biology, with most research in marine systems. I have spent extensive amounts of time doing research at marine labs in Puget Sound (the Friday Harbor Marine Labs) and Panama (through the Smithsonian Tropical Research Institute). Since coming to Alaska, I have gained additional experience in the Gulf of Alaska (Kenai Fjords and Katmai National Parks), Cook Inlet (Lake Clark National Park), and the Beaufort and Chukchi Seas.

PATRICIA M. HARRIS

Education: University of Alaska Fairbanks; B.S. Biological Science 1966
Graduate work at U of A Fairbanks, U of A Southeast, University of British Columbia

Experience: 1986 - present. Researcher, Auke Bay Laboratory, National Marine Fisheries Service, Juneau, Alaska. As co-principal investigator of NRDA study Subtidal 3, I was responsible for field logistics and sample collection and assisted in data analysis and report preparation. I also assisted other NRDA projects in field collections. In 1992 and 1993, I participated in study design, field work, and proposal preparation for this project, formerly restoration Project R103 and 93036). Other areas of research have been habitat requirements of juvenile red king crab and sockeye salmon stock separation using parasites.

Relevant publications: Co-author of final reports for NRDA study Subtidal 3. Several public presentations of oil-related scientific research.

Malin M. Babcock

Project Leader
Malin M. Babcock
NOAA/NMFS Auke Bay Laboratory
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Project Leader
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Alaska Science Center
National Biological Service
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Anchorage, AK 99503-6199
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FAX: 907-789-3636

Bruce Wright for

Project Manager NOAA
Bruce Wright
NOAA/Office of Oil Spill Damage
Assessment and Restoration
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FAX: 907-789-6608
e-Mail: bwright@bl.afsc.noaa.gov

April 30, 1995
Date prepared

1996 EXXON VALDEZ TRUS COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

01-1

Budget Category:	Authorized FFY 1995	Proposed FFY 1996	PROPOSED FFY 1996 TRUSTEE AGENCIES TOTALS					
			ADEC	ADF&G	ADNR	USFS	DOI	NOAA
Personnel	\$265.0	\$165.0						
Travel	\$28.5	\$2.9						
Contractual	\$69.5	\$0.0						
Commodities	\$27.2	\$12.4						
Equipment	\$4.0	\$0.0						
Subtotal	\$394.2	\$180.3	LONG RANGE FUNDING REQUIREMENTS					
General Administration	\$44.6	\$24.8	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
Project Total	\$438.8	\$205.1	\$0.0	\$280.0	\$165.0	\$0.0	\$285.0	\$170.0
Full-time Equivalents (FTE)	4.3	2.8	Dollar amounts are shown in thousands of dollars.					
Other Resources	\$0.0	\$89.8						
<p>Comments:</p> <p>This project is scheduled for chemical analyses of all samples collected in FY95 and a comprehensive 4-year final report covering oiled mussel beds in Prince William Sound and the Gulf of Alaska. No field work is scheduled for FY96.</p> <p>Chemical analyses = \$ 111.1K Report (Both NOAA and NBS) = \$94.0K</p> <p>The FY95 approved amount for ADEC is shown in the FFY95 column; they will not participate in this project in FY96. Of the FFY Authorized amount approved for ADEC, they plan on not spending ~ \$50.0K, therefore, letting it lapse back to the Trustees.</p>								

1996

Project Number: 96090
Project Title: Mussel Bed Restoration & Monitoring
Lead Agency: National Oceanic & Atmospheric Administration

FORM 2A
PROJECT
DETAIL

Prepared: 1 of 13

7/11/95
Revised

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel	\$197.3	\$129.8						
Travel	\$16.3	\$2.9						
Contractual	\$39.6	\$0.0						
Commodities	\$22.5	\$12.2						
Equipment	\$0.0	\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$275.7	\$144.9	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
General Administration	\$32.4	\$19.5						
Project Total	\$308.1	\$164.4		\$200.0	\$120.0		\$200.0	\$120.0
Full-time Equivalents (FTE)	3.2	2.2						
Dollar amounts are shown in thousands of dollars.								
Other Resources		\$89.8		\$20.0	\$20.0		\$20.0	\$20.0
<p>Comments:</p> <p>Other Resources:</p> <p>NOAA's contribution includes Project Leader, Malin Babcock, 10 mo = \$75.0K Habitat Investigation Program Manager, Jeep Rice, 1 mo = \$10.9K Senior Chemist, Jeff Short, 0.5 mo = \$3.9K</p> <p>This project includes chemical analyses for ~450 samples to be collected from Prince William Sound (NOAA) and the Gulf of Alaska (NBS); cost will be \$111.1 and is distributed between Personnel and Commodities. The remainder is a for a comprehensive 4-year report on all work under this project for oiled mussel beds in Prince William Sound.</p>								

1996

Project Number: 96090
Project Title: Mussel Bed Restoration & Monitoring
Agency: National Oceanic & Atmospheric Administration

**FORM 3A
AGENCY
PROJECT
DETAIL**

1996 EXXON VALDEZ TRU COUNCIL PROJECT BUDGET
October 1, 1995 - September 30, 1996

Personnel Costs:			GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	P. Harris	Zoologist	9/5	6.0	4,600		27.6
	C Brodersen	Fish Biologist	11/7	6.0	6,000		36.0
	M. Larsen	Sr. chemist	11/6	1.0	5,800		5.8
	J. Lunasin	Chemist	9/5	12.0	4,600		55.2
*	B Wright	NOAA Program Manager	12/5	0.8	6,500		5.2
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
Subtotal				25.8	27,500	0	
Those costs associated with program management should be indicated by placement of an *.							Personnel Total \$129.8
Travel Costs:			Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
*	Anchorage, Workshop, 2		444	2	8	225	2.7
	Car Rental, Miscellaneous						0.2
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
Travel Total							\$2.9
Those costs associated with program management should be indicated by placement of an *.							

1996

Project Number: 96090
Project Title: Mussel Bed Restoration & Monitoring
Agency: National Oceanic & Atmospheric Administration

FORM 3B
Personnel
& Travel
DETAIL

October 1, 1995 - September 30, 1996

1996

FORM 3B
Contractual &
Commodities
DETAIL

1996 EXXON VALDEZ TRUST COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1996	
Description					
Those purchases associated with replacement equipment should be indicated by placement of an R.				New Equipment Total	\$0.0
Existing Equipment Usage:		Number of Units	Inventory Agency		
Description					
Computer, Compaq		2	NOAA		
Camera		1	NOAA		

1996

Project Number: 96090
 Project Title: Mussel Bed Restoration & Monitoring
 Agency: National Oceanic & Atmospheric Administration

**FORM 3B
 Equipment
 DETAIL**

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel	\$28.6	\$35.2						
Travel	\$4.1	\$0.0						
Contractual	\$26.4	\$0.0						
Commodities	\$4.0	\$0.2						
Equipment	\$4.0	\$0.0						
Subtotal	\$67.1	\$35.4	LONG RANGE FUNDING REQUIREMENTS					
General Administration	\$6.1	\$5.3	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
Project Total	\$73.2	\$40.7		\$80.0	\$45.0		\$85.0	\$50.0
Full-time Equivalents (FTE)	0.6	0.6						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
Comments: This will cover comprehensive 4-year data analyses and interpretation for the Gulf of Alaska oiled mussel beds which will be incorporated into the overall Final Report for this project.								

1996

Project Number: 96090
Project Title: Mussel Bed Restoration & Monitoring
Agency: DOI National Biological Service

FORM 3A
AGENCY
PROJECT
DETAIL

1996 EXXON VALDEZ TRUST COUNCIL PROJECT BUDGET
October 1, 1995 - September 30, 1996

[illegible]

1996

Project Number: 96090
Project Title: Mussel Bed Restoration & Monitoring
Agency: DOI National Biological Service

FORM 3B
Personnel
& Travel
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed FFY 1996
Description		
When a non-trustee organization is used, the form 4A is required.		Contractual Total
		\$0.0
Commodities Costs:		Proposed FFY 1996
Description		
<p>Film, graphic supplies</p>		0.2
		Commodities Total
		\$0.2

1996

Project Number: 96090
 Project Title: Mussel Bed Restoration & Monitoring
 Agency: DOI National Biological Service

**FORM 3B
 Contractual &
 Commodities
 DETAIL**

1996 EXXON VALDEZ TRUST COUNCIL PROJECT BUDGET
October 1, 1995 - September 30, 1996

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1996
Description				
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
Those purchases associated with replacement equipment should be indicated by placement of an R.		New Equipment Total		\$0.0
Existing Equipment Usage:			Number of Units	Inventory Agency
Description				

1996

Project Number: 96090
Project Title: Mussel Bed Restoration & Monitoring
Agency: DOI National Biological Service

FORM 3B
Equipment
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel	\$39.1	\$0.0						
Travel	\$8.1	\$0.0						
Contractual	\$3.5	\$0.0						
Commodities	\$0.7	\$0.0						
Equipment	\$0.0	\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$51.4	\$0.0	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
General Administration	\$6.1	\$0.0						
Project Total	\$57.5	\$0.0						
Full-time Equivalents (FTE)	0.5	0.0						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
Comments:								
<p>The Alaska Department of Environmental Conservation will not be participating in this project in FY96. This set of forms is included only for the purpose of showing approved amount for FY95. They will be returning ~50K of this amount to the Trustee Council.</p>								

1996

Project Number: 96090
 Project Title: Mussel Bed Restoration & Monitoring
 Agency: Alaska Department of Environmental Conservation

FORM 3A
 AGENCY
 PROJECT
 DETAIL

Prepared: 10 of 13

7/11/95

1996 EXXON VALDEZ TRUS COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Personnel Costs:			GS/Range/ Step	Months Budgeted	Monthly Costs	Overtime	Proposed FFY 1996	
PM	Name	Position Description						
Subtotal				0.0	0	0		
Those costs associated with program management should be indicated by placement of an *.							Personnel Total	\$0.0
Travel Costs:			Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996	
PM	Description							
Subtotal								
Those costs associated with program management should be indicated by placement of an *.							Travel Total	\$0.0

1996

Project Number: 96090
 Project Title: Mussel Bed Restoration & Monitoring
 Agency: Alaska Department of Environmental Conservation

**FORM 3B
 Personnel
 & Travel
 DETAIL**

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
When a non-trustee organization is used, the form 4A is required.		
Contractual Total		\$0.0
Commodities Costs:		Proposed
Description		FFY 1996
Commodities Total		\$0.0

1996

Project Number: 96090
 Project Title: Mussel Bed Restoration & Monitoring
 Agency: Alaska Department of Environmental Conservation

FORM 3B
Contractual &
Commodities
DETAIL

1996 EXXON VALDEZ TRUS COUNCIL PROJECT BUDGET
 October 1, 1995 - September 30, 1996

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1996
Description				
Those purchases associated with replacement equipment should be indicated by placement of an R.				New Equipment Total
				\$0.0
Existing Equipment Usage:		Number of Units	Inventory Agency	
Description				

1996

Project Number: 96090
 Project Title: Mussel Bed Restoration & Monitoring
 Agency: Alaska Department of Environmental Conservation

**FORM 3B
 Equipment
 DETAIL**

96091

Monitoring for Current and Potential Environmental Impacts of Oil Industry Activities in Cook Inlet, Alaska.

Project number: 96091

Restoration Category: Monitoring and Research, General Restoration

Proposer: Cook Inlet Regional Citizens' Advisory Council

Lead Trustee Agency:

Cooperating Agencies:

Duration: 2 years

Cost FY 96: \$135,000

Cost FY 97: \$135,000

Geographic Area: Cook Inlet: Trading Bay to Kamishak Bay

Injured Resource/Service: Sediment, shellfish, subtidal organisms, commercial fishing

ABSTRACT

This proposal requests assistance in funding the Cook Inlet Environmental Monitoring Study. For two years, Cook Inlet RCAC has devoted their entire environmental research budget as sole supporter of this critical program. Consequently, no funding has been available for smaller, more localized studies that promote community involvement. The goals of the monitoring program include: 1) establishing baseline hydrocarbon and biological data; 2) evaluating potential hydrocarbon accumulation in Cook Inlet sediments; and 3) evaluating potential environmental impacts of crude oil production and transportation in the Inlet.

INTRODUCTION

Cook Inlet RCAC was formed under Section 5002 of the Oil Pollution Act of 1990 (OPA 90). Our mission is to ensure the safe operation of the oil terminals, tankers, and facilities in Cook Inlet so that environmental impacts associated with the oil industry are minimized. The organization strives to provide a forum wherein citizens, government and industry may work together productively to fulfill this mission.

The Cook Inlet Environmental Monitoring Program was implemented to satisfy Council duties outlined by OPA 90. For the past two years a pilot study was conducted at several sites in the Inlet. The current project design reflects information gained in the pilot studies. For example, in the pilot study, mussels were used to evaluate bioaccumulation potential in upper Cook Inlet. Suspended sediments caused high mortality of the test animals so the project now includes use of artificial lipophilic devices instead of caged mussels. Data from the pilot studies has been used by industry and MMS.

Cook Inlet RCAC has developed a multifaceted study currently under bid for the 1995 field season. The study design combines biological and chemical analyses in evaluating environmental impacts in Cook Inlet. Specifically, the project includes analysis of sediment and water chemistry, focusing primarily on hydrocarbons. Sediments will be evaluated for toxicity which can be caused by many pollutants. Fish will be screened for indications of significant hydrocarbon exposure. Finally, lipophilic devices will be used to estimate chronic hydrocarbon exposure faced by organisms in the water column.

The Council would like to continue the study annually through 1997. At that time a solid three year foundation of consecutive annual samples will be available as excellent baseline data for use in comparative studies and in developing future research. One year of sampling only gives results relative to that particular season, day, temperature, etc. Multi-year sampling provides an estimate of annual variation. Annual variation must be considered in determining whether changes are occurring in the Inlet.

NEED FOR THE PROJECT

A. Statement of Problem

This project addresses restoration of sediments, shellfish, subtidal organisms, and commercial fishing in lower Cook Inlet as well as the reduction of marine pollution which may be impeding recovery of these resources. The Cook Inlet Environmental Monitoring Study also provides habitat protection by acting as an early warning system to environmental damage. Many citizens who depend on Cook Inlet believe that continued inputs from current oil activities are damaging Inlet resources. Cook Inlet RCAC believes it is important to document whether or not such pollution is actually occurring and to what degree it may be impacting organisms in the Inlet.

B. Rationale

The Cook Inlet Environmental Monitoring Study addresses two restoration objectives in the *Exxon Valdez Oil Spill Restoration Plan* as follows:

Reduction of Marine Pollution

The proposed project is designed to evaluate pollution inputs from current oil industry activities in Cook Inlet. Negative impacts may prevent resource recovery in lower Cook Inlet. Public perception,

particularly in the fishing community, is that the *Exxon Valdez* oil spill incurred great damages that are perpetuated by current industry activities.

Nearshore Ecosystem Projects

Our proposed project directly addresses one of the major objectives listed for the Nearshore Ecosystem Projects. Our study is designed to quantify hydrocarbon contamination and, if supported for consecutive years, it will provide persistence data for subtidal areas impacted by the *Exxon Valdez* oil spill.

The Cook Inlet Environmental Monitoring Study will also enhance intertidal studies already proposed in the *Exxon Valdez Oil Spill Restoration Plan* for FY 97. Our project will provide subtidal data for much of the area where the intertidal studies are planned.

C. Summary of Major Hypotheses and Objectives

There are two hypotheses tested by support of this proposal.

Ho1 There is no difference in samples taken from various sites.

Ho2 There is no difference in samples taken in different years.

The objective of the study is to characterize current impacts and pollution levels due to oil industry activities in the Inlet and to evaluate the potential for future problems.

D. Completion Date

Research supported by this proposal would take place June of 1996 with a final report submitted by February 1997. Similarly, funding for a second year would result in sampling in July 1997 with a final report submitted in February 1998.

COMMUNITY INVOLVEMENT

Cook Inlet RCAC also anticipates undertaking several smaller scale, localized research projects which are primarily dependent on the knowledge, experience and expertise of local citizens. Several examples of such projects include beach surveys, bioaccumulation studies, and discharge compliance. Our ability to engage in any of these additional projects is contingent upon our receiving funding for the final two years of the Cook Inlet RCAC Environmental Monitoring Study.

Community involvement specific to the proposed project would include educational outreach through meetings and news articles. Additionally we anticipate procuring vessel services locally.

FY 96 BUDGET

Cook Inlet RCAC is requesting funding of \$135,000 for FY 1996 and \$135,000 for FY 97. The funding will be spent completely on contracted services to continue research currently funded solely by the Cook Inlet RCAC. This appropriation will allow Cook Inlet RCAC to expend research dollars in a variety of smaller, community based projects that focus on oil industry issues at a local level. If funding is not provided, Cook Inlet RCAC will have to choose to abandon this critical database or to delay funding of community based projects. Cook Inlet RCAC welcomes partial funding as every dollar received allows budget allocations to smaller projects.

Personnel	0
Travel	0
Contractual	135
Commodities	0
Equipment	0
Total	135

PROJECT DESIGN

A. Objectives

1. Establishing baseline hydrocarbon and biological data.
2. Evaluating potential hydrocarbon accumulation in Cook Inlet sediments.
3. Evaluating potential environmental impacts of crude oil production and transportation in the Inlet.

B. Methods

Sediment PAH	GC/MS SIM mode
Water column PAH	GC/MS SIM mode
Sediment TOC	EPA 413.1M
Sediment PGS	dry sieve/pipetting
Sediment toxicity	Microtox or echinoderm larval assay
Pore water toxicity	Microtox or echinoderm larval assay
Biomarker analysis	Cytochrome P450 1A1 enzyme induction
(select fish sp.)	Bile PAH metabolites

Parametric and non parametric statistical analyses will be done to show whether sites: 1) were significantly different from each other; and 2) if sites changed over time.

Sites will be the same as those sampled the summer of 1995 and will most likely include Trading Bay, Kamishak Bay, Kachemak Bay, Tuxedni Bay and East Forelands. Sampling takes place annually.

C. Contracts and Other Agency Assistance

The entire project will be contracted to the private sector, universities or some combination thereof. It is important to maintain a consistent level of scientific expertise for the three year study. This is most economically attained by contracting out the entire project.

D. Location

The project will involve sampling several locations in Cook Inlet including Trading Bay, Kachemak Bay, and Kamishak Bay. Communities affected by the project include all communities who depend on the health Cook Inlet waters. These include shoreline communities from Anchorage to Kodiak as well as inland communities dependant on Cook Inlet fish stocks or recreational resources.

SCHEDULE

A. Measurable Project Tasks for FY 96

December 1, 1995: Develop request for proposal
January 1, 1996 - February 28: Receive proposals

March 1 - April 15:	Evaluate and select proposals
June - August:	Field sampling
June - September:	Laboratory and data analysis
September - November:	Monthly and Oral Reports Due
February 1, 1997:	Annual report for 1996 work

B. Project Milestones

The specific project objectives will be met on an annual basis and reported in an annual report for each year the study is funded. The project will end after FY 97.

C. Project Reports

An annual report will be submitted on February 1, 1997 for FY 96 and February 1, 1997 is funding continues into FY 97. The annual reports will be inclusive of all new results while addressing their relationship to data from previous years.

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

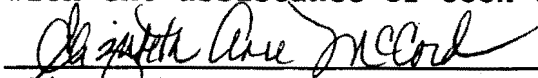
Cook Inlet RCAC has approached several state and federal agencies for funding or cooperative agreements in the past. These included the University of Alaska, ADEC and MMS. In 1994, MMS donated vessel services to the program but that vessel is no longer available. Cook Inlet RCAC remains the only organization supporting ongoing environmental studies in Cook Inlet. Currently there are no restoration projects that could coordinate with our efforts in Cook Inlet.

ENVIRONMENTAL COMPLIANCE

Several permits are required for the scientific sampling of fish and marine organisms. These are generally obtained by contractors.

PERSONNEL

This project will be conducted primarily by a contracted firm. Cook Inlet RCACs' Executive Director will oversee contractual obligations with the assistance of Cook Inlet RCAC scientific staff.


 Elizabeth Anne McCord
 Executive Director
 Cook Inlet Regional Citizens' Advisory Council
 910 Highland Ave.
 Kenai, AK 99611
 (907) 283-7222 phone
 (907) 283-6102 FAX

Date prepared 4/26/95

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel								
Travel								
Contractual		135						
Commodities								
Equipment								
Subtotal			LONG RANGE FUNDING REQUIREMENTS					
Indirect			Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
Project Total			135	0	0	0	0	0
Full-time Equivalents (FTE)								
Dollar amounts are shown in thousands of dollars.								
Other Resources (in kind services)		20	20					
Comments: <p>Monies requested in this proposal will be dedicated for contracted services required for successful completion of a monitoring project in Cook Inlet. Cook Inlet RCAC will donate contract administration services for the project.</p>								

1996

Prepared:

Project Number:

96091

Project Title: Monitoring for Current and Potential Environmental Impacts of Oil Industry Activity in Cook Inlet, AK

Name: Cook Inlet Regional Citizens' Advisory Council

FORM 4A
Non-Trustee
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs: 135		Proposed
Description Sampling and analyses services		FFY 1996
<p>The monies requested in this proposal will be 100% allocated to contracted services. Cook Inlet RCAC will donate in kind services for contract administration.</p>		135
Contractual Total		135
Commodities Costs:		Proposed
Description		FFY 1996
Commodities Total		

1996

Project Number: 96091

Project Title: Monitoring for Current and Potential Environmental Impacts of Oil Industry Activity in Cook Inlet, AK

Name:

Cook Inlet Regional Citizens' Advisory Council

FORM 4B
Contractual &
Commodities
DETAIL

P.03

FAX NO. 9072836102

WF CIRCUIT

APR-28-95 FRI 15:11

Restoration of PWS Pink Salmon by Diversion of Harvest Effort: Quantitative Genetic Assessment of Early-Returning Pink Salmon Broodstock

Project Number: 96093~~A~~-BAA
Restoration Category: Research
Proposer: University of Alaska Fairbanks
Lead Trustee Agency:
Cooperating Agencies:
Duration: 7 years
Cost FY 96: \$111,897
Cost FY 97: \$198,389
Cost FY 98: \$211,744
Cost FY 99: \$171,860
Cost FY 00: \$171,038
Cost FY 01: \$132,357
Cost FY 02:
Geographic Area: Prince William Sound
Injured Resource: Pink Salmon

ABSTRACT

Development of early-returning broodstock at hatcheries might beneficially reduce fishing on injured stocks. However, a risk is that early stock might interbreed with local salmon and hurt their fitness. Risk might be reduced by stock selection or broodstock management. This research uses quantitative genetics to assess 1) genetics of run timing in donors (predicts effectiveness of stock selection and broodstock management) and 2) fitness loss from interbreeding (exposes loss by laboratory breeding experiment).

INTRODUCTION

Restoration of injured pink salmon is being proposed by Prince William Sound Aquaculture Corporation (96093) through changing fish culture strategies. The aim is reduction of fishing effort during the time that injured stocks transit the fishing districts. Feasibility studies of the development of an early-returning stock are part of the proposed restoration project; refer to that proposal for a general introduction. This project will be a quantitative genetic evaluation of an early-returning stock cultured at a hatchery; that is, an evaluation that experimentally creates families of salmon and analyses their performance statistically.

Accomplishments to date: This project will use experimental designs based on successful research on pink salmon at Auke Creek, in Juneau, Alaska, in which quantitative genetic variability of run timing and other fitness-related traits have been analyzed, and in which outbreeding depression has been strongly suggested as the source of reduced fitness in hybridizations of genetically unrelated pink salmon.

Proposed in FY 96: This research will endure over six years, requiring repetition of experiments in odd-year and even-year lines of pink salmon. FY 96 (beginning in summer 1995) will be spent in preliminary experimental design and reconnaissance efforts.

Planned for the future: Beginning in summer 1996 the research will involve breeding experiments to be carried out in the laboratory each year from 1996 to 1999, involving releases of tagged pink salmon families in 1997 and 1998, and involving measurements of laboratory fish and returning tagged adults each year from 1997 to 2000. Analysis, modelling, and application of the results to program planning will be complete in 2001.

NEED FOR THE PROJECT

A. Statement of Problem

Mortality of pink salmon eggs has been attributed to effects of oiling of spawning beds and has persisted through several generations. Recovery may be inhibited by fishing mortality, a result of the economic exploitation of uninjured pink salmon resources that are only available for harvest at the same place and in the same time as the injured stocks.

B. Rationale

The parent project to this one, "Restoration of PWS Pink Salmon by Diversion of Harvest Effort", will investigate the development of potential early run pink salmon hatchery stocks (PERPS). Such stocks would have the benefit for restoration of diverting harvest of hatchery-produced salmon to a different time than when injured stocks are vulnerable. (Early run stocks move through fishing districts as much as 4 weeks before injured stocks). This research is necessary to assess the genetic interaction of PERPS with oil-injured and other wild pink salmon stocks in PWS, and to design broodstock management programs that would minimize such interactions.

C. Summary of Major Hypotheses and Objectives

The objective of the project is to assess, through quantitative genetic analysis of experiments, the probability and severity of interactions between PERPS at a Prince William Sound hatchery and oil-injured and other wild pink salmon stocks. Two major objectives will be sought: 1) genetic analysis of the variability of run timing in a PERPS (hypothesis: run timing is not inherited and may not be controlled by broodstock management practices) and 2) genetic analysis of outbreeding depressive effects of interbreeding between a PERPS and oil-injured and other wild resources (hypothesis: interbreeding entails no outbreeding-depressive loss of fitness in wild stocks).

FY 96 Objective: Preparation for first breeding experiments in summer 1996. Identification of early-run brood source (in summer 1995), experimental design, purchase of necessary equipment and supplies, hiring of temporary personnel, staging, will all be necessary activities.

D. Completion Date

The work will be complete in 2001 when results of two generations' experiments in each of the odd- and even-year brood lines of pink salmon have been analyzed and incorporated into resource management and broodstock management policies and plans. Important results will begin accruing in spring 1997 when the fish cultural performance of a PERP at a hatchery can first be evaluated and in summer 1998 when the first genetic analyses of run timing variation are obtained. Primary evaluation of a PERP will begin with those data.

COMMUNITY INVOLVEMENT

This project has been developed in collaboration and consultation with Prince William Sound Aquaculture Corporation (the regional private non-profit salmon aquaculture corporation for PWS) and the Native Village of Eyak Tribal Council. It will depend heavily on local knowledge and logistical support and assistance from the members of those organizations. The project has been developed with the consultation and general support of the Cordova District Fishermen United and the Prince William Sound Regional Planning Team.

FY 96 BUDGET

Personnel	64.2
Travel	1.3
Contractual	1.5
Commodities	1.5
Equipment	9.0
Subtotal	77.5
Gen. Admin.	34.2
Total	111.8

The project will increase in activity after the first year. In the first year the milestones will be experimental design work, reconnaissance (identification of candidate PERPs, etc.), purchasing and installation of incubation equipment (capable of incubating a large array of single families), and carrying out the first breeding experiment and beginning incubation of them.

PROJECT DESIGN

A. Objectives

The two major objectives of this research are

1. Estimate the genetic variability of run timing in a PERP and

- Describe outbreeding depression in hybrids of a PERP and a wild stock from the local region of the PERP.

A potential increased risk of an early-returning stock (compared to a local stock in the hatchery, one whose timing is synchronous with local stocks) is that, because of its ancestors' different ecological requirements, such a stock would be expected to be genetically different from local stocks and that any strays which interbreed with local stock would cause commensurately more severe outbreeding depression. The risk of outbreeding depression and the consequent loss of wild stock fitness and productivity is the basis of policy in Alaska that prohibits artificial introduction of salmon into local areas that originate in non-local stocks. Figures 1 and 2 illustrate the potential for hybridization between early- and late-returning hatchery stock strays and local stocks. A potential benefit of an early-returning broodstock, in addition to diverting fishing pressure from wild-stock, is that strays from an early-returning stock may have reduced or no opportunity to interbreed with local wild stocks.

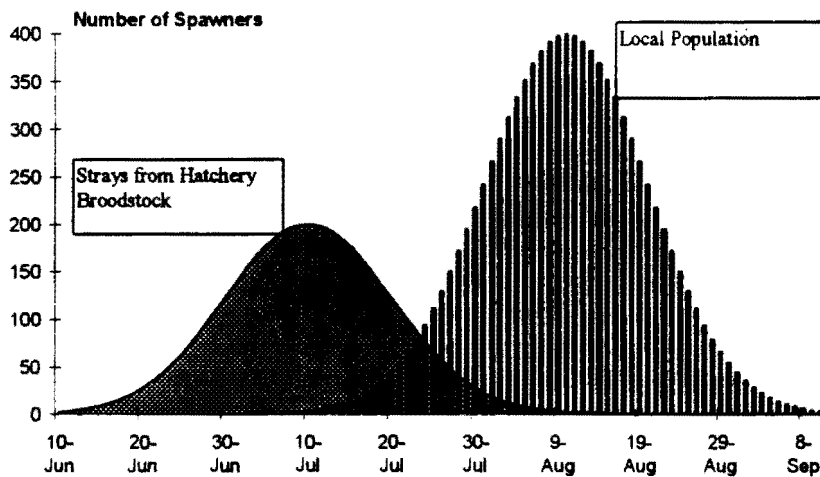


Figure 1 Hypothetical run timing distribution based on AFK (local population) and VFDA (introduced hatchery stock). The number of hatchery strays is one third of the total. The spawning time of the strays is on average four weeks ahead of the native fish in the stream and is assumed to be the same as that of the proposed early broodstock at the hatchery. The standard deviation of spawning times is ten days.

Opportunity for hybridization is low: note that overlap of tails in hybrid zone involves relatively few individuals; however hybridization (outbreeding) of strays and local wild stock may still occur and the potential for outbreeding depression in those hybridizations is high because of the disparate ancestries of the two stocks.

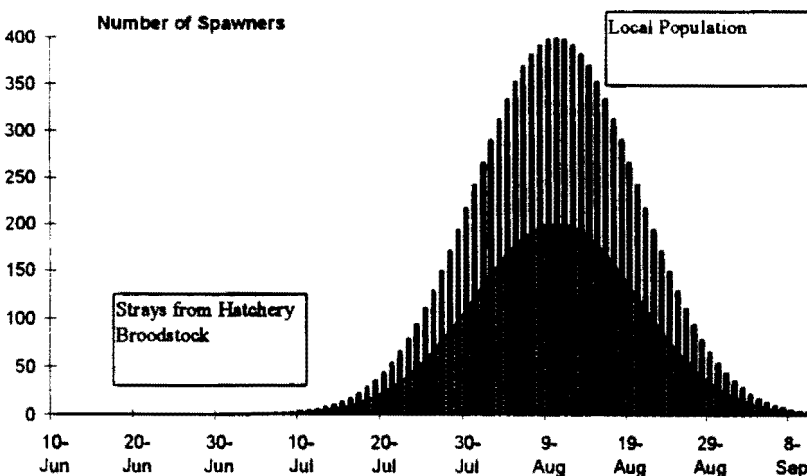


Figure 2 Hypothetical distribution when the spawning time of the strays is the same as the local stock.

Opportunities for hybridization—outbreeding—is high. Native fish have a high chance of mating with a stray; however risk of outbreeding depression in those

hybridizations is low because of the common ancestry of the two stocks.

Because early broodstock would be from an ancestrally different ecology, it would be likely that coadapted genomes in the broodstock and local stocks would be different. The risk of outbreeding

depression (Gharrett and Smoker 1991) in any hybrids that might occur, even though the frequency of hybridizations would be reduced, would be increased. Outbreeding depressive effects may not be fully expressed until the second generation of hybridization. Thus, if run timing is not very precisely inherited, first-generation hybrids may propagate later into the local stock in the second generation, with attendant risk to local stock fitness and productivity.

Evidence of increased outbreeding depression in the second generation has been observed in pink salmon. Hybrids of even and odd-year pink salmon showed little evidence of reduced fitness in first filial generation but exhibited increased fluctuating asymmetry and reduced marine survival in the second generation of hybridization (Gharrett and Smoker 1991).

The risk of outbreeding depression in local stocks may also increase if the run timing of an early-returning broodstock were to change to later dates over generations, in response to natural or artificial selective pressures. Run timing in salmonids has been observed to change over generations, for instance in response to artificial selection in a hatchery (Millenbach 1973) or to artificial selection imposed by commercial fishing (Alexandersdottir 1987.) In the first case steelhead run timing became earlier in response to breeding primarily early-returning fish; in the second pink salmon in Chatham Strait became later because the fishery systematically took early-returning fish. If late-returning fish in an "early" hatchery broodstock were favored a corresponding response toward later return timing would increase the risk of hybridization with local stocks over time. Broodstock management of an early-returning stock would have to guard against selection for late-return or might even explicitly select for early return in order to minimize hybridization with local stocks (Figure 3.)

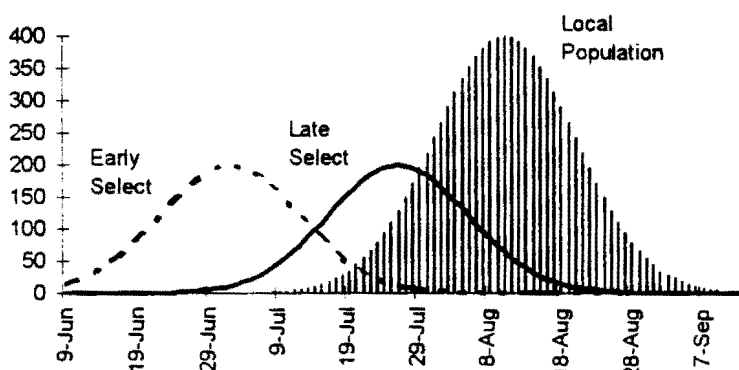


Figure 3. Consequence of 5 generations of breeding from late returns (selecting from second half of run, heritability moderate, 0.4) or of 5 generations from early returns (breeding only before the mid point, heritability low, 0.2) starting from the situation of Figure 1. Hybridization opportunity is increased markedly in the first case, illustrating the risk of inadvertent broodstock selection for later returning stock; these hybridizations would carry an increased risk of outbreeding depression compared to those illustrated in Figure 2 because of the disparate ancestry of the stocks. Hybridization is reduced moderately

in the second case, illustrating the potential benefit of intentional selection for early returns. Assumed heritability values are from Smoker et al (in press); they are chosen to illustrate high-risk situations for local stocks.

The desirable situation would be first that strays from an early-returning broodstock in local streams would have low fitness and not persist as a new population. This outcome might be expected in the absence of hybridization with local stock because embryo development rate would be too fast, resulting in early vernal emergence and fry death by starvation. Prediction of this desirable outcome assumes not only the inheritance of characteristic development rates in embryos, but also on the inheritance of seasonal run timing of pink salmon; experimental evidence from our studies at Auke Creek support these assumptions

(Joyce 1985, Hebert 1994, Gharrett and Smoker 1993a,b, Smoker et al In Press). Second it would be desirable that no viable hybridization occur.

B. Methods

Two kinds of genetic analysis will be applied to the feasibility assessment of an early-returning broodstock, quantitative genetic analysis and population genetic analysis. The quantitative genetic analysis will include experimental observation of particular families of known relationships, observations of phenotypic characters including run timing and embryo development rate. These analyses will rely on experimental designs developed at Auke Creek in Juneau. The population genetic analysis will rely on genetic tagging techniques also developed in pink salmon at Auke Creek in Juneau. These techniques will lead to direct observation of the rate of gene flow from early-returning brood to local stocks. Thus both the rate of hybridization (gene flow) and the fitness effect of hybridization will be observed.

With this information in hand it may be possible to design a broodstock management program, an artificial selection program, in which the broodstock can be maintained with early timing so that interbreeding with local stocks is prevented.

QUANTITATIVE GENETICS OF RUN TIMING

This experimentation will answer the questions

What are genetic mean and genetic variation of run timing in early broodstock, local stock, and in potential hybrids?

What are similar parameter estimates for spawning timing?

Is ancestral timing of early broodstock maintained in new environment?

Is there phenotypic or genetic correlation between run timing and spawn timing?

Could a selection program maintain or increase the differential of timing?

The design of this quantitative genetic evaluation research will be adapted from the experimental design of Smoker et al (In Press) and Smoker et al (1994) in which the heritability of run timing and genetic correlations with body size were estimated from 120 families of pink salmon at Auke Creek, Juneau, AK. In brief outline:

Hierarchical mating of 25 males and 50 females randomly chosen from peak of early broodstock, local broodstock, and from hybrid zone. Total of 150 families.

Incubate in separate incubators

Apply wire tags (1000 per family; 150 thousand total)

Release under optimized conditions (adjust fish culture conditions as necessary).

Recover tags from returning adults in broodstock. Observe timing of maturation (ovulation, spermiation.)

Replicate experiment in odd and even brood lines.

STATISTICAL MODELS AND PARAMETER ESTIMATION

Coded Wire tagging will enable the determination of genetic parameters based on family records. By having family identification, records can be corrected for known fixed effects such as stock of origin. Parameter estimates for all traits will be computed separately for stock groups by solving a sire-dam mixed model (Henderson, 1988) using derivative-free technique for estimating variance components employing restricted maximum likelihood (Graser et al., 1987). Meyer's (1988) derivative-free restricted maximum likelihood (DFREML) procedure will be used.

Genetic and phenotypic correlations will be estimated among all traits using the following formulas:

$$\begin{aligned}\text{Corr}_a(XY) &= \text{Cov}_a(XY) / \sqrt{[\text{Var}_a(X)\text{Var}_a(Y)]} \\ \text{Corr}_p(XY) &= \text{Cov}_p(XY) / \sqrt{[\text{Var}_p(X)\text{Var}_p(Y)]}\end{aligned}$$

where X and Y signify two traits, and Corr_a and Corr_p symbolize the additive genetic and phenotypic correlations, respectively. Additive genetic (a) and phenotypic (p) covariances for correlation calculations were estimated using the following formulas:

$$\begin{aligned}\text{Cov}_a(XY) &= [\text{Var}_a(X+Y) - (\text{Var}_a(X) + \text{Var}_a(Y))]/2 \\ \text{Cov}_p(XY) &= [\text{Var}_p(X+Y) - (\text{Var}_p(X) + \text{Var}_p(Y))]/2\end{aligned}$$

where Var and Cov symbolize the variances and covariances, respectively.

Heritability will be estimated as

$$h^2 = \text{Var}_a(X) / \text{Var}_p(X)$$

Where heritability predicts the response to selection of a given intensity.

OUTBREEDING EFFECTS ON FITNESS

This experiment will address the question of whether fitness effects of hybridization can be demonstrated, i.e. whether measurable outbreeding depression would occur as a consequence of hybridization between strays from an early broodstock and local wild stock fish. Failure of the experiment to demonstrate outbreeding depression will not indicate a lack of significant effects because the power of the experiment to detect effects may not be adequate. Our experience with pink salmon suggests that effects can be demonstrated in hybridizations of pink salmon as genetically different as odd-year and even-year populations in which second filial generation hybrids had lesser survival at sea and increased fluctuating asymmetry (Gharrett and Smoker 1991). Hybridizations of stocks from the same brood year and geographical region, but of different run timing, may not demonstrate equally detectable effects even though the real fitness effects of outbreeding may be significant.

FIRST GENERATION EFFECTS

This experiment will be adapted from the experiment design of Hebert (1994), in which larval fitness-related traits of pink salmon (embryo development rate) were analyzed in early- and late-returning pink salmon at Auke Creek, Juneau. In brief outline the design includes:

Factorial mating of 3 males and 2 females in a set. Interactive variation (dominance, epistasis) may be significant in fitness-associated traits.

Replicate the set 10 times sampled from peak of early brood, 10 times from peak of local brood, and 10 times from hybrid zone.

Observe embryonic development: timing of hatching; timing of yolk absorption.

Observe fluctuating asymmetry, if feasible (work in progress at Auke Creek; Gharrett, Smoker, Reisenbichler, et al.)

Analyze genetic (additive, interactive) components of variation.

Repeat in odd and even brood lines

SECOND GENERATIONS EFFECTS

The critical second generation will be examined for measurable effects of outbreeding by sampling parents from returns to Run-Timing experiment, above, and use of a similar breeding design. It will also be repeated in odd and even brood lines.

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- Smoker, W.W., A.J. Gharrett, and M.S. Stekoll. Genetic variation in timing of anadromous migration withing a spawning season in a population of pink salmon. *Canadian Journal of Fisheries and Aquatic Sciences Special Publication*. (Proceedings of the International Symposium on Biological Interactions of Enhanced and Wild Salmonids, Nanaimo, June 1991.) In Press..
- Smoker, W.W., A.J. Gharrett, and M.S. Stekoll. 1994. Heritability of size in an anadromous population of pink salmon. *Canadian Journal of Fisheries and Aquatic Sciences* 51 (Suppl. 1): In Press.
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Meyer, K., 1988. DFREML programs to estimate variance components for individual animal models by restricted maximum likelihood. User notes. Univ. of Edinburgh
Millenbach, C., 1973. Genetic selection of steelhead trout for management purposes. Intl. Atl. Sal. J. 4:253-257.

C. Contracts and Other Agency Assistance

Logistical and technical support will be provided by Prince William Sound Aquaculture Corporation and by the Native Village of Eyak as required. In FY 96 this will entail transportation and subsistence support for the PI, Quantitative Geneticist, and Fishery Technician from Cordova to the hatchery site of the experiment, assistance with the experiments (installation of incubation equipment, gamete collection, and fish cultural maintenance of experiments during incubation).

D. Location

The project will be undertaken at facilities of Prince William Sound Aquaculture Corporation, Cordova, Alaska.

SCHEDULE

A. Measurable Project Tasks for FY 96

Startup to June 30 1996:	Purchase equipment, Modify, install equipment Reconnaissance of early run time stocks, Experimental design,
July 1 to Sept. 15 1996:	Carry out breeding experiments for estimation of genetic variability of run timing. Carry out breeding experiments for measurement of outbreeding depression.
Sept. 15 1996 to end of FY 96:	Culture families in separate incubation.

B. Project Milestones and Endpoints

Please review the Appendix Figure for a timeline diagram of events and milestones for the entire project

C. Project Reports

Annual reports will be completed by December 1 of each project year. Activity toward each objective will be governed by the pink salmon life cycle in which returning adults are active in summer, spawning occurs in late summer, fry emigration occurs in spring. Fall is the available time for analysis of data and reporting.

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

This project is integrally coordinated with PWSAC's "Restoration of PWS Pink Salmon by Diversion of Harvest Effort" without which it cannot go forward.

The project has been reviewed and approved conceptually by the Regional Planning Team, Area E.

ENVIRONMENTAL COMPLIANCE

Compliance with all pertinent laws and regulations will be accomplished by PWSAC in carrying out the umbrella project "Restoration of PWS Pink Salmon by Diversion of Harvest Effort".

PERSONNEL

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Ph.D. Fisheries, Oregon State University

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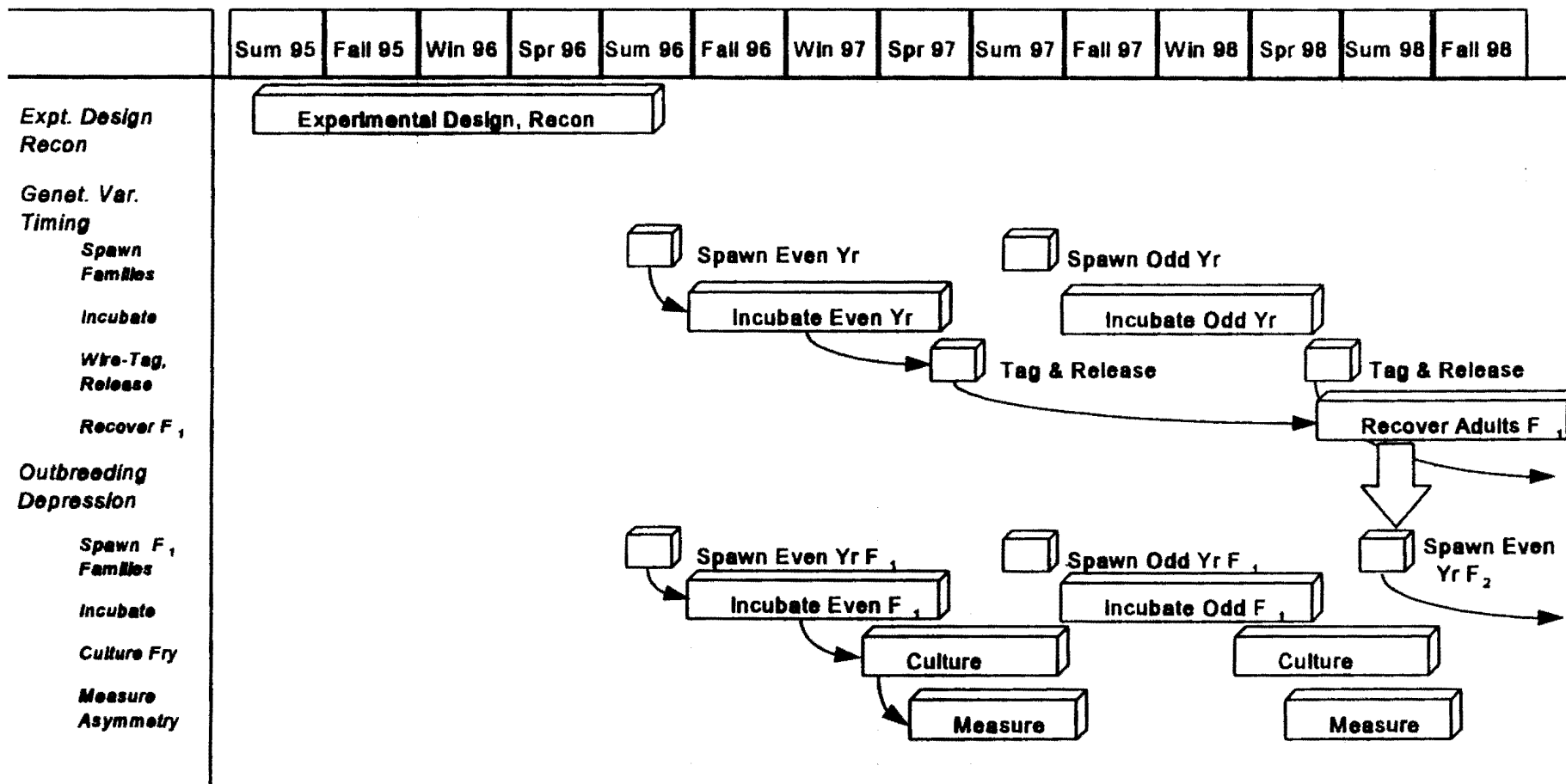
Ph.D Aquaculture Genetics, Biometrics Univ. of California Davis

Research on quantitative genetics of pink salmon, ploidy manipulation in Pacific salmon

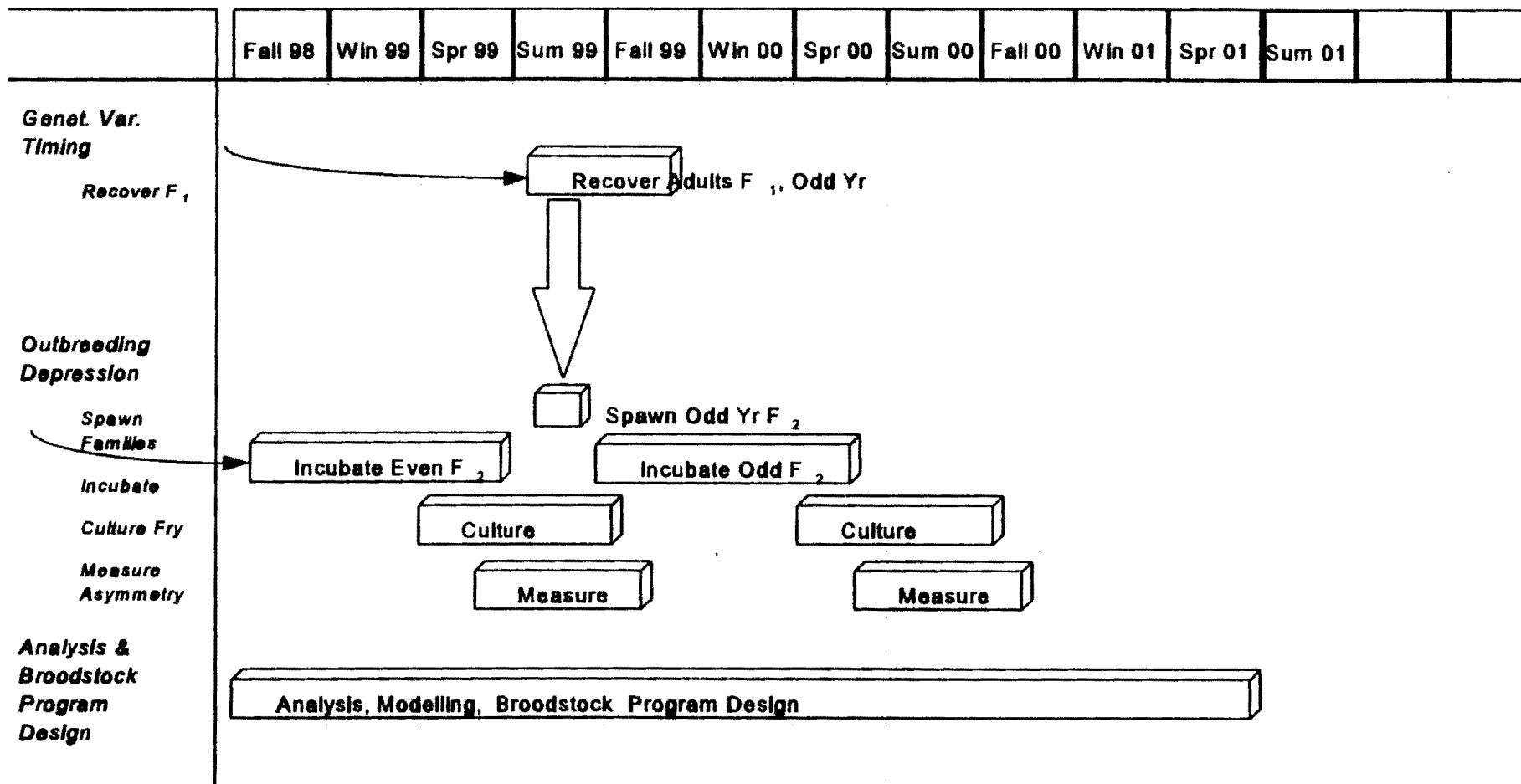
Expertise in experimental design and statistical analysis.

Date Prepared

Appendix Figure Quantitative Genetic Assessment of Early-Returning Broodstock. Timeline



Appendix Figure. Quantitative Genetic Assessment of Early-Returning Broodstock. Timeline. (Continued)



1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel		\$64.2						
Travel		\$1.4						
Contractual		\$1.5						
Commodities		\$1.5						
Equipment		\$9.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$0.0	\$77.6	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
Indirect		\$34.3						
Project Total	\$0.0	\$111.9	\$198.4	\$211.7	\$171.9	\$171.0	\$132.4	
Full-time Equivalents (FTE)		1.2						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
Comments:								
Indirect costs calculated at proposed (interim) negotiated rate of 50% of Modified Total Direct Costs. Audit agency is Office of Naval Research.								

1996

Project Number:
 Project Title: Restoration of PWS Pink Salmon...: Quantitative Genetic
 Assessment...
 Name: William W. Smoker

FORM 4A
 Non-Trustee
 DETAIL

Prepared:

1 of 4

4/29/95

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Personnel Costs:				Months	Monthly		Proposed	
	Name	Position Description		Budgeted	Costs	Overtime	FFY 1996	
	William W. Smoker	Principal Investigator		2.0	9,672		19.3	
				6.0	4,288		25.7	
				6.0	3,195		19.2	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
Subtotal				14.0	17,155	0		
Personnel Total						\$64.2		
Travel Costs:				Ticket	Round	Total	Daily	Proposed
	Description			Price	Trips	Days	Per Diem	FFY 1996
	R/T Juneau-Cordova			452	3			1.4
								0.0
								0.0
								0.0
								0.0
								0.0
								0.0
								0.0
								0.0
								0.0
								0.0
Travel Total						\$1.4		

1996

Project Number:
 Project Title: Restoration of PWS Pink Salmon...: Quantitative Genetic Assessment...
 Name: William W. Smoker

FORM 4B
 Personnel
 & Travel
 DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
Communications		0.5
Software Maintenance		0.5
Computer Maintenance		0.5
Contractual Total		\$1.5
Commodities Costs:		Proposed
Description		FFY 1996
Fishfood		1.0
Labware		0.5
Commodities Total		\$1.5

1996

Project Number:
 Project Title: Restoration of PWS Pink Salmon...: Quantitative Genetic
 Assessment...
 Name: William W. Smoker

FORM 4B
 Contractual &
 Commodities
 DETAIL

October 1, 1995 - September 30, 1996

1996	Project Number: Project Title: Restoration of PWS Pink Salmon...: Quantitative Genetic Assessment... Name: William W. Smoker	FORM 4B Equipment DETAIL
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4 of 4

4/28/95

Restoration of PWS Pink Salmon by Diversion of Harvest Effort: Population Genetic Assessment of Gene Flow from Early Return Stock

Project Number: 96093B-BAA
Restoration Category: Research
Proposer: University of Alaska Fairbanks
Lead Trustee Agency:
Cooperating Agencies:
Duration: 7 years
Cost FY 96: \$120,966
Cost FY 97: \$237,997
Cost FY 98: \$228,080
Cost FY 99: \$134,191
Cost FY 00: \$153,650
Cost FY 01: \$131,463
Cost FY 02: \$131,540
Geographic Area: Prince William Sound
Injured Resource: Pink Salmon

ABSTRACT

Development of early-returning broodstock at hatcheries might beneficially reduce fishing on injured stocks. However, a risk is that early stock fish might stray and interbreed with local salmon and reduce their fitness. The risk can be estimated by measuring gene flow experimentally. Potential early run pink salmon will be tagged with a natural gene marker and planted in a local stream, simulating straying. The effect will then be directly estimated over generations by measuring the genetic tag in the test stream and its gene flow to others.

INTRODUCTION

Restoration of injured pink salmon is being proposed by Prince William Sound Aquaculture Corporation through changing fish culture strategies. The aim is reduction of fishing effort during the time that injured stocks transit the fishing districts. Feasibility studies of the development of an early-returning stock are part of the proposed restoration project; refer to that proposal for a general introduction (Restoration of PWS Pink Salmon by Diversion of Harvest Effort, Project 96093). This project will be a population genetic evaluation of an early-returning stock cultured at a hatchery. It will use a biochemically detectable genetic tag and will directly estimate gene flow from an experimental introduction into a local stock.

Accomplishments to date: This project will use experimental designs based on successful research on pink salmon at Auke Creek, in Juneau, Alaska, in which biochemical genetic tags have been used to observe gene flow within a local population complex. This research will begin in FY 96 and be

complete in FY 2002 requiring repetition of experiments in odd-year and even-year lines of pink salmon. FY 96 (beginning in summer 1995) will be spent in inventory and assessment efforts.

Planned for the future: Beginning in summer 1996 the research will produce genetically marked fish. Analysis, modelling, and application of the results to program planning will be complete in 2001

NEED FOR THE PROJECT

A. Statement of Problem

Mortality of pink salmon eggs has been attributed to effects of oiling of spawning beds and has persisted through several generations. Recovery may be inhibited by fishing mortality, a result of the economic exploitation of uninjured pink salmon resources that are only available for harvest at the same place and in the same time as the injured stocks.

B. Rationale

The parent project to this one, "Restoration of PWS Pink Salmon by Diversion of Harvest Effort", will investigate the development of potential early run pink salmon hatchery stocks (PERPS). Such stocks would have the benefit for restoration of diverting harvest of hatchery-produced salmon to a different time than when injured stocks are vulnerable. (Early run stocks move through fishing districts as much as 4 weeks before injured stocks). This research is necessary to assess the genetic interaction of PERPS with oil-injured and other wild pink salmon stocks.

C. Summary of Major Hypotheses and Objectives

The null hypothesis to be tested here is that no gene flow will occur from a PERP to pink salmon populations (demes) in the local region. This research will use genetic tagging (Lane et al 1990) to estimate the rate of gene flow over several generations from A) a simulated straying event from an early returning broodstock to a local stock, and B) a simulated straying event from a late-returning broodstock to a local stock. Neutral genetic tags will be placed in early-returning brood and in late-returning brood at a hatchery, eyed eggs will be planted in local streams, and the fates of the inherited marker will be monitored for several generations as they "flow" into the local stocks.

A control experiment will be required. Changes in the frequencies of the marker gene may be due to either gene flow from the introduced fish or changes may be due to natural straying and gene flow between local stocks. Tests for natural selection on genetic markers are part of the simulation experiment. An estimate of the rate of natural gene flow will be required in order to estimate gene flow from the introduced stocks. This estimate will be made in an independent experiment and will involve genetic tagging at two wild-stock streams.

D. Completion Date

The work will be complete in 2002 when results of two generations' experiments in each of the odd- and

even-year brood lines of pink salmon have been analyzed and incorporated into resource management and broodstock management policies and plans. Important results will be available beginning in 1998 when the first genetically tagged adults are sampled. Primary evaluation of a PERP will begin with those data.

COMMUNITY INVOLVEMENT

This project has been developed in collaboration and consultation with Prince William Sound Aquaculture Corporation (the regional private non-profit salmon aquaculture corporation for PWS) and the Native Village of Eyak Tribal Council. It will depend heavily on local knowledge and logistical support and assistance from the members of those organizations. The project has been developed with the consultation and general support of the Cordova District Fishermen United and the Prince William Sound Regional Planning Team.

FY 95 BUDGET

Personnel	37.2
Travel	1.2
Contractual	.8
Commodities	20.8
Equipment	25.3
Subtotal	90.9
Gen. Admin.	30.0
Total	120.9

The project will increase in activity after the first year. In the first year the milestones will be experimental design work, reconnaissance, inventory and assessment (identification of candidate PERPs, etc.), purchasing and installation of equipment and baseline development.

PROJECT DESIGN

A. Objectives

The two major objectives of this research are

1. Plant genetically marked embryos in a test stream and monitor the geographic distribution of the marker gene over time.
2. Estimate natural gene flow between natural populations, also to be done with gene markers; a necessary control for the estimation of gene flow in objective 1.

B. Methods

Plant and follow progeny of genetically marked embryos in test stream.

1. Produce genetically marked and otolith marked embryos from introduced stock.
2. Plant eyed eggs in test stream.
3. At fry emigration estimate relative numbers of emigrant wild and introduced fry.
4. In generation 1 estimate relative contribution to natal stream from otolith mark.
5. In generation 1 examine neutrality of marker allele from genotypic frequencies in otolith marked fish.
6. At generation 1 estimate contribution to neighboring streams from otolith mark.
7. Assuming a nearly neutral marker, monitor long term contribution of supplementation on natal streams.
8. From demographics of nearby reference streams, infer demographic effects on test streams.

Estimate baseline gene flow:

1. Differentially gene-mark populations from two streams in close proximity to each other, but not strongly influenced by enhanced fish to estimate levels.
2. Estimate differences between emigrant fry and returning adults to confirm stability.
3. Estimate reciprocal exchange between these two systems.
4. Estimate gene flow into system over time.

Detailed Description:

Preliminary work

During the first field season, candidate streams will be examined and evaluated for suitability for the two portions of this work. Streams to be used for the introduction experiments must be accessible and have stream beds suitable for transferring eyed eggs. Streams to be genetically marked to study baseline gene flow levels must be suitable for weiring. Samples of the fish in these streams will be taken to supplement other genetic baseline information that is available and will be used in selection of appropriate marker alleles for the 1997 broodyear.

Simulation of straying experiment:

Candidate Streams

Two streams not heavily influenced by cultured fish will be chosen. These streams should have escapements of about 4,000 (2,000 to 6,000), and be accessible for collecting. This escapement, assuming a 3% marine survival (with 50% exploitation), indicates that ~ 300,000 fry would have emigrated. The streams will be supplemented with 300,000 eyed eggs which will require about 200 females. Both males and females will be screened: the effective population number, N_e , will be approximately 400. An appropriate marker will entail screening about 4000 fish (for a 1:10 screening effort).

A marker allele with a frequency of about 0.05 would be appropriate (Gharrett and Seeb 1990). Presumably, hatchery frequencies have diverged very little from wild frequencies. Also, no large differences have been observed in wild populations throughout the Sound. Data obtained in a genetic survey of PWS pink salmon populations and other historic data will be used to choose a marker allele expressed in muscle tissue:

	Genotype			Allele	
	AA	Aa	aa	A	a
Pre-marker frequencies	0.9025	0.095	0.0025	0.95	0.05
Generation 1 supplement (post marking)	0.237	0.500	0.263	0.487*	0.513*

* not in HW equilibrium

If there is a good return and mixture with the native population then:

Generation 1 stream(post marking)	0.570	0.297	0.133	0.719 *	0.281*
Subsequent generations	0.516	0.404	0.079	0.719	0.281

* not in HW equilibrium

Marking effort:

A crew of 6 people for 2 weeks for each marking effort will be required. Since they involve differently timed runs, the same crew can mark both lots. Tagging involves capturing fish, taking a muscle sample, tagging the fish with a uniquely numbered Floy cinch-up tag, holding the fish (holding pens) until the genotype has been determined electrophoretically. Targeted genotypes will be kept for breeding. Numbered tissue samples corresponding to the fish being held are analyzed electrophoretically at a preselected marker locus.

Embryos incubated in an isolated incubator will be otolith marked using thermal treatments. A random sample of 2000 eggs will be retained and reared to emergence to determine the frequency of the marker allele in the supplemented fish. About 4 man-weeks will be required for this analysis.

Simulated Straying:

Eyed eggs will be planted in gravel beds of the recipient streams. Fry sampling will be done on each recipient stream below the intertidal zone. Two samples of at least 100 fish (more at peak migration times) will be taken each week to determine the emigration timing and relative proportion of native and introduced fish in each stream. About 4 man-weeks will be required for this analysis. Neighboring streams will be censused to provide references for the abundance of pink salmon in the area.

Gene Flow Observations:

In the next generation fish will be sampled from the recipient stream and neighboring streams throughout the return. Approximately 500 fish will be taken from recent spawn-outs in each neighboring stream to estimate directly the number of fish straying from the supplemented stream. About 2000 fish will be sampled from the recipient stream to estimate the proportionate contribution of supplemental fish. Samples will include **paired** tissue samples and otolith samples. All samples will be examined electrophoretically to estimate genotypic and allelic frequencies of the marker allele. Samples carrying the otolith marker will also be used to look for changes in genotypic frequency which would indicate selection.

The collecting efforts will require logistical support and presence of samplers throughout the run, 2-3 samples per week from fresh spawn-outs. Tissues sampled for electrophoresis must be taken on ice and on the same day transferred to a freezer. Samples from spawnouts are compromised at the outset, so care must be taken with them to assure their quality.

Allozyme analysis of each 1000 samples for a single locus will require about 1 man-week. Otolith analysis will require about 100 man-hours per 1000 fish.

In subsequent generations, neighboring streams and the recipient stream will be sampled throughout the run. Samples will be electrophoretically analyzed to look for frequency changes that might indicate straying from the supplemented stream, and trace the subsequent success of supplemented fish in the recipient stream.

The collecting efforts will require logistical support and must include the entire run period, preferably in proportion to the abundance. Samplers should visit the stream at least 2 to 3 times per week and try to sample fresh spawn-outs.

Allozyme analysis of each 1000 samples for a single locus will require about 1 man-week.

Natural Baseline Gene Flow Control Experiment:

Requires 2 streams in close proximity (to increase the chances of detecting gene flow). Streams must not be heavily influenced by cultured fish or by the stream(s) used in the introduction experiment. Escapements should be between 5,000 and 10,000.

The streams must be completely weirable because it will be necessary to capture and electrophoretically

screen every returning male. Genetic marking will be accomplished by screening all returning males for an appropriate marker allele and using only the 1 in 5 that carry the allele for breeders. Each stream will be marked for a different locus.

Genetic Marker Alleles:

A marker allele with a frequency of about 0.1 would be appropriate. Data obtained in a genetic survey of PWS pink salmon populations and other historic data will be used to choose a marker allele expressed in muscle tissue:

	Genotype								
	AABB	AABb	AAbb	AaBB	AaBb	Aabb	aaBB	aaBb	aabb
Stream 1									
Pre-marker frequencies; <i>freq.</i> A=0.90; B=0.90	0.656	0.146	0.008	0.146	0.032	0.002	0.008	0.002	0.000
Gen. 1 post marking (not in HW equilibrium) <i>freq.</i> A= 0.95; B= 0.687	0.384	0.469	0.047	0.043	0.052	0.005	0	0	0
Subsequent generations	0.426	0.388	0.089	0.045	0.041	0.009	0.001	0.001	0.000
Stream 2									
Pre-marker frequencies <i>freq.</i> A=0.90; B=0.90	0.656	0.146	0.008	0.146	0.032	0.002	0.008	0.002	0.000
Gen. 1 post marking (not in HW equilibrium) <i>freq.</i> A=0.687 B=0.95	0.384	0.043	0	0.469	0.052	0	0.047	0.005	0
Subsequent generations	0.426	0.045	0.001	0.388	0.041	0.001	0.089	0.009	0.000

The Marking Effort:

Crews of technicians will be required at each stream throughout the return period. Two field crews will require living facilities and an enclosed laboratory structure for conducting protein electrophoresis. The laboratory will require a generator to run power supplies and analytical laboratory equipment (balances and pH meters); gas for preparing starch gels; low conductivity water, possibly stream water will suffice; a collection system for laboratory waste.

Each crew needs 5 to 6 people, two who can run and read gels, one to keep records and three to sample and tag fish. For each stream people will be needed for the entire return period.

Numbered tissue samples corresponding to the males being held will be analyzed electrophoretically at a preselected marker locus. Screened males possessing the marker will be released to spawn normally. Loss in productivity is small even when the ratio of males to females is less than 1:6.5. Males not released to spawn will be sacrificed.

References:

- Gharrett, A.J. and W.W. Smoker. 1993. Genetic components in life history traits contribute to population structure. Pp 197 - 202 In J.G. Cloud and G.H. Thorgaard, eds., Genetic Conservation of Salmonid Fishes. Plenum Press, NY
- Lane, S., A.J. McGregor, S.G. Taylor, A.J. Gharrett. 1990. Genetic marking of an Alaskan pink salmon population, with an evaluation of the mark and the marking process. Amer. Fish. Soc. Symp. 7:395-406..
- Gharrett, A.J., & J.E. Seeb. 1990. Practical and theoretical guidelines for genetically marking fish populations. Amer. Fish. Soc. Symp. 7:407-417.

C. Contracts and Other Agency Assistance

Logistical and technical support will be provided by Prince William Sound Aquaculture Corporation and by the Native Village of Eyak as required. In FY 96 this will entail transportation and subsistence support for the PI, Molecular Geneticist, and Fishery Technician from Cordova to the hatchery site of the experiment, assistance with the experiments (installation of incubation equipment, gamete collection, and fish cultural maintenance of experiments during incubation). Full field crew support for gamete collection and genetic marking will be needed.

D. Location

The project will be undertaken at facilities of Prince William Sound Aquaculture Corporation, Cordova, Alaska.

SCHEDULE

A. Measurable Project Tasks for FY 96

Startup to June 30 1996:

July 1 to Sept. 15 1996:

Sept. 15 1996 to end of FY 96:

B. Project Milestones and Endpoints

Plant and follow progeny of genetically marked embryos in test stream.

1. Produce genetically marked and otolith marked embryos from introduced stock.

Even year line:	Fall '96
Odd year:	Fall '97
2. Plant eyed eggs in test stream.

Even year line:	Winter '97
Odd year:	Winter '98

3. At fry emigration estimate relative numbers of emigrant wild and introduced fry.
 Even year line: Spring '97
 Odd year: Spring '98
4. In generation 1 estimate relative contribution to natal stream from otolith mark.
 Even year line: Fall '98
 Odd year: Fall '99
5. In generation 1 examine neutrality of marker allele from genotypic frequencies in otolith marked fish.
 Even year line: Winter '98
 Odd year: Winter '99
6. At generation 1 estimate contribution to neighboring streams from otolith mark.
 Even year line: Fall '98
 Odd year: Fall '99
7. Assuming a nearly neutral marker, monitor long term genetic contribution of PERP on local streams.
 Even year line: Fall '00, etc.
 Odd year: Fall '01, etc.
8. From demographics of nearby reference streams, infer demographic effects on test streams.

Estimate baseline gene flow :

1. Differentially gene-mark populations from two streams in close proximity to each other, but not strongly influenced by enhanced fish to estimate levels.
 Odd year line: Fall 1997
 Even Year: Fall 1998
2. Estimate differences between emigrant fry and returning adults to confirm stability.
 Odd year line: Spring 1998
 Even Year: Spring 1999
3. Estimate reciprocal exchange between these two systems.
 Odd year line: Fall 1999
 Even Year: Fall 2000
4. Estimate gene flow into system over time.
 Odd year line: Fall 2001, etc
 Even Year: Fall 2002, etc

C. Project Reports

Annual reports will be completed by December 1 of each project year. Activity toward each objective will be governed by the pink salmon life cycle in which returning adults are active in summer, spawning occurs in late summer, fry emigration occurs in spring. Fall is the available time for analysis of data and reporting.

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

This project is integrally coordinated with PWSAC's "Restoration of PWS Pink Salmon by Diversion of Harvest Effort" without which it cannot go forward. The project has been reviewed and approved conceptually by the Regional Planning Team, Area E.

ENVIRONMENTAL COMPLIANCE

Compliance with all pertinent laws and regulations will be accomplished by PWSAC in carrying out the umbrella project "Restoration of PWS Pink Salmon by Diversion of Harvest Effort".

PERSONNEL

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Molecular genetics techniques, Electrophoretic analysis of Allozymes, DNA analysis

Date Prepared

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996							
Personnel		\$42.7							
Travel		\$1.2							
Contractual		\$0.8							
Commodities		\$21.0							
Equipment		\$25.4							
Subtotal	\$0.0	\$91.1	LONG RANGE FUNDING REQUIREMENTS						
Indirect		\$30.1	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002	
Project Total	\$0.0	\$121.2	\$238.0	\$228.1	\$134.2	\$153.7	\$131.5	\$131.5	
Full-time Equivalents (FTE)		1.2							
Dollar amounts are shown in thousands of dollars.									
Other Resources									
Comments:									
Indirect costs calculated at proposed (interim) negotiated rate of 50% of Modified Total Direct Costs. Audit agency is Office of Naval Research.									

1996

Project Number:
Project Title: Restoration of PWS Pink Salmon...: Population Genetic
Assessment....
Name: Anthony J. Gharrett

FORM 4A
Non-Trustee
DETAIL

Prepared:

1 of 4

8/95

October 1, 1995 - September 30, 1996

Project Number:
Project Title: Restoration of PWS Pink Salmon...: Population Genetic
Assessment....
Name: Anthony J. Gharrett

4/28/95

October 1, 1995 - September 30, 1996

Project Number:
Project Title: Restoration of PWS Pink Salmon...: Population Genetic
Assessment....
Name: Anthony J. Gharrett

3 of 4

4/28/95

1996 EXXON VALDEZ TRUST COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1996
Description				
	500V/500mA power supply	3	700	2.1
	0.01g acc./500g gap. balance	1	1,100	1.1
	80C Freezer	1	5,500	5.5
	80C Freezer	1	9,000	9.0
	Centrifuge	1	2,475	2.5
	Incubator	1	4,500	4.5
		1	700	0.7
				0.0
				0.0
				0.0
				0.0
				0.0
Those purchases associated with replacement equipment should be indicated by placement of an R.			New Equipment Total	\$25.4
Existing Equipment Usage:		Number		
Description		of Units		

1996

Project Number:
Project Title:
Name:

**FORM 4B
Equipment
DETAIL**

Restoration of Prince William Sound Pink Salmon by Diversion of Harvest Effort

Project Number: 96093C

Lead Trustee Agency: Alaska Department of Fish and Game

Cooperating Agencies: Prince William Sound Aquaculture Corporation
Native Village of Eyak
University of Alaska
Department of Fish and Game
Prince William Sound Science Center

Start-up/Completion Dates: April 1, 1996 - September 30, 1998

Expected Project Duration: 7 years

Cost FY 96:

Cost FY 97:

Cost FY 98: More detailed proposal

Cost FY 99: to follow.

Cost FY 00:

Cost FY 01:

Cost FY 02:

Geographic Area of Project: Prince William Sound

Injured Resource/Service: Pink salmon, commercial fishing, subsistence

ABSTRACT

Pink salmon egg mortality attributed to oiling of anadromous streams from the *Exxon Valdez* oil spill has contributed to a reduction in adult pink salmon returns. Natural populations of pink salmon are harvested with large numbers of hatchery pink salmon in mixed stock fisheries, which may limit escapement to damaged streams and thereby delay recovery. This project will evaluate the feasibility of changes in hatchery production to reduce exploitation of injured wild stocks. Specific projects will focus on changing the location and timing of hatchery returns in western Prince William Sound. Funding for FY96-FY98 will be for Inventory and Assessment and logistical support for 96093A (Population Genetic Assessment of Gene Flow from Early Return Stock) and 96093B (Quantitative Genetic Assessment of Early Return Pink Salmon Broodstock). Funding for FY99-FY02 will be for logistical support for 96093b and 96093c.

IMPROVING RECOVERY RATES IN THE PRINCE WILLIAM SOUND USING ENHANCED BIOREMEDIATION

Project Number: 96094

Restoration Category:

Proposer: ADEC

Lead Trustee Agency: ADEC

Cooperating Agencies: NOAA

Duration: 3 FYs

Cost FY 96: \$965,600

Cost FY 97: \$600,000

Cost FY 98: \$600,000

Cost FY 99: \$0

Cost FY 00: \$0

Cost FY 01: \$0

Cost FY 02: \$0

Geographic Area: Prince William Sound

Injured Resources/
Services: Intertidal Organisms, Subtidal Organisms, Mussels, Clams, Sea Otters, Sediment, Subsistence Use, Passive Uses, and Use for Recreation and Tourism.

ABSTRACT

As part of its response to the *Exxon Valdez* oil spill EXXON, ADEC, EPA, and others applied nutrient enhancers at selected shorelines to accelerate the biodegradation of toxic portions of Alaska North Slope Crude (ANSC). This three year project will identify reasons why remaining portions of antagonistic subsurface oil have not biodegraded and assess the impact this having on shoreline recovery. Based on site characterization and risk the project will recommend use of appropriately selected non-intrusive, non-commercial bioremediation enhancement methods to accelerate stalled biodegradation. The study will demonstrate how

enhancement of biodegradable oil shortens the length of time shorelines need to return to pre-spill conditions. The bioremediation and recovery rates of selected sites will be studied and reported.

INTRODUCTION

In response to the *Exxon Valdez* oil spill EXXON, ADEC, EPA, and others added nutrient enhancers to oiled shorelines in order to accelerate biodegradation of the toxic portions of Alaska North Slope Crude (ANSC). This approach was used because toxic portions of ANSC directly interfere with recovery of impacted shorelines and because toxic portions of Alaska North Slope Crude (ANSC) can be made to biodegrade at faster rates.

More recent shoreline data and concerns expressed by local communities indicate that continued biodegradation of spilled oil may have stalled along portions of shorelines in the Prince William Sound. These sites are commonly referred to as some of the remaining "hot spots". This project will determine if bioremediation of residual oil at these sites stopped prematurely and determine if remaining biodegradable oil is contributing in any way to a delay in the recovery of the shoreline. It is important to emphasize that where ever possible this project will incorporate relevant data and findings of others.

In early FY 96 the team will review data from shoreline surveys collected by ADEC, NOAA, and others. Currently sites of most interests include approximately twelve low energy sites near the community of Chenega Bay that are referenced in the Draft Restoration Program dated March 24, 1995 and 12 to 14 other sites in western Prince William Sound said to be experiencing little reduction of subsurface oil.

Based on review of survey data and discussions with local residents, a total of 10 to 12 sites will be visited to learn the location, status, amount, and chemical make-up of remaining subsurface oil. This information is vital for determining the effect that residual oil is having on injured resources. Where appropriate selected shorelines will fully characterized according to geomorphology, hydrodynamic make-up, toxicity of remaining oil, status of recovery, and the sites' responsiveness to enhanced bioremediation. The team use this information to determine if, and why biodegradation stopped at these sites.

At this point effective bioremediation strategies will be developed for each site where evidence indicates that accelerating the biodegradation of remaining antagonistic oil will shorten shoreline recovery. To do this the team will conduct an ecological assessment that includes a biological and toxicological assessment. Each ecological assessment will be modeled as close as possible to similar work already being done by the University of Alaska. The team will use the assessment to help compare the expected benefits gained by accelerating biodegradation of residual oil against the ecological consequences of leaving the oil alone. Working with the Chief Scientist a joint recommendation on application of enhanced bioremediation methods at individual sites will be made. Saying it another way, the recommendation to proceed with using bioremediation enhancement will be based on

a comparison of the risks of not doing anything and the potential for gaining increased recovery rates.

In early FY 97 the team will discuss the benefits of using of non-intrusive, non-commercial techniques for accelerating biodegradation of antagonistic oil with the Chief Scientist and residents of near-by communities. The team will then present its recommendation on where enhanced bioremediation should be used to the EVOS Trustee Council. After receiving approval to proceed the team will assume responsibility for obtaining necessary permits and approvals.

During the summer of FY 97 the team will apply enhanced bioremediation methods at two or three of the best suited sites. Increased bioremediation and recovery rates will be studied in detail at each of these. Where justified the team is willing to consider application of appropriate bioremediation methods at other sites as well. But if so, the other sites would not be studied or monitored as closely as the 2 or 3 best suited sites.

During the summer of FY 98 the team will continue application of enhanced bioremediation methods and studying associated biodegradation and recovery rates. Approximately six months following the completion of the FY 98 field season a final scientific report will be submitted to the EVOS Trustee Council for their acceptance in accordance with PROCEDURES for the PREPARATION & DISTRIBUTION of FINAL PAPERS, October 1994, EXXON Valdez Oil Spill Trustee Council, Anchorage Restoration Office, 645 G Street, Suite 401, Anchorage AK, 99501.

The team is looking forward to hiring residents of Chenega Village and other Prince William Sound communities for this project. These local residents will be asked to contribute their knowledge of impacted shorelines, trained to conduct shoreline surveys and to collect samples, and, when possible, asked to provide boats for themselves and other members of this team.

Some preliminary work associated with this proposal began in early April, 1995 when contaminated sediment samples were collected at sites near the Chenega Village area. The samples were expressed mailed to the EPA Risk Reduction Engineering Laboratory in Cincinnati, Ohio for detailed chemical analysis. The results of this analysis will be made available to the EVOS Trustee Council as soon as it becomes available.

NEED FOR THE PROJECT

A. Statement of Problem

In response to the *Exxon Valdez* oil spill EXXON, ADEC, and others added nutrient enhancers to impacted shorelines to accelerate biodegradation of Alaska North Slope Crude (ANSC). This approach was used because toxic portions of ANSC directly interfere with recovery and because the toxic portions of Alaska North Slope Crude (ANSC) can be

encouraged to biodegrade at faster rates.

This project is designed to improve the recovery of resources which are either not recovering or are recovering at a slow rate because biodegradation of residual oil has stalled.

Resources directly effected include intertidal organisms, subtidal organisms, mussels, clams, sea otters, sediment, subsistence use, passive uses, and use for recreation and tourism.

B. Rational

As discussed in the Invitation to Submit Restoration Projects for Federal Fiscal Year 1996 knowledge of and elimination of residual oil is important for people's perception of recovery and for all of the injured resources that rely on near-shore ecosystems.

Surface oil on most Prince William Sound shorelines has disappeared over time through microbial degradation, photo-oxidation, mechanical abrasion, and other means. Subsurface oil is not amenable to most of these degradation processes. For example, subsurface oil in a low energy cove does not normally encounter mechanical abrasion caused by medium to high energy waves. Because of its location subsurface oil is not degraded by photo-oxidation. This leaves microbial degradation and mechanical removal as the two remaining means of eliminating antagonistic subsurface oil.

Given the volume of soil involved when applying mechanical abrasion to subsurface oil, logistical problems encountered with using heavy machinery, and the fact that removal of subsurface oil involves the destruction of shoreline recovery already achieved, use of mechanical techniques will not be considered by this study.

Since toxic and mutagenic portions of oil residue are susceptible to biodegradation, use of bioremediation technology offers the greatest promise of converting remaining toxic compounds into nontoxic products without disrupting recovery achieved.

As stated in the EVOS Trustee Council's Invitation to Submit Restoration Projects for Fiscal Year 1996, it has been learned that between 1991 and 1993 subsurface oil decreased at many sites through out Prince William Sound. Sites with little reduction in oil are primarily in "low energy" areas that have little wave action. The worst sites still sheen.

Many compounds in crude oil are environmentally benign, but a significant fraction are toxic or mutagenic. Studies conducted since the spill suggest biochemical effects are potentially related to oil toxicity. This finding along with other related findings establish that initial oil has a chemical effect on recovery. Further studies indicate that recovery rates can be impacted by residual oil left in the shoreline. Related studies have documented high concentrations of hydrocarbons in some mussel flesh, byssal thread mats, and underlaying sediments suggesting that leaching of non-degraded subsurface oil into shoreline waters may

be delaying recovery of resources.

This project will identify residual oil remains in shoreline hot spots and provide additional information on location, status, and amount of toxic and mutagenic oil that is there. A review will be conducted to determine what restricted the biodegradation of this antagonistic oil at each site. The effect that leaching residual oil is having on impacted shoreline resources will be measured and compared against recovery achieved in similar nearby shorelines. If it is found that remaining antagonistic oil is capable of contributing to a delay in recovery, non-intrusive bioremediation methods for destroying that oil will be identified.

The team will discuss the benefits of using of non-intrusive, non-commercial techniques to accelerate biodegradation of remaining antagonistic oil with the Chief Scientist and residents of near-by communities. A recommendation to use bioremediation enhancement techniques at appropriate sites will be based on a comparison of the risks of leaving that oil alone to the potential for increased recovery rates.

Monitoring changes to residual oil and the shoreline community before, during, and after application of selected bioremediation enhancement techniques is needed to quantify the relationship between enhanced bioremediation and improved recovery. Therefore recovery will be assessed prior, during, and after use of enhanced bioremediation at each study site.

C. Summary of Major Hypotheses and Objectives

In response to the *Exxon Valdez* oil spill EXXON, ADEC, EPA, and others added nutrient enhancers to oiled shorelines to speed up the elimination of toxic portions of Alaska North Slope Crude (ANSC). This approach was used because toxic portions of ANSC directly interfere with recovery of impacted shorelines and because toxic portions of Alaska North Slope Crude (ANSC) can be made to biodegrade at faster rates.

This project will inspect hot spot shorelines in the Western Prince William Sound for subsurface oil and determine if the remaining portions of subsurface oil are delaying shoreline recovery.

Since the poisonous portions of Alaskan crude oil are biodegradable we will attempt to understand why, after all this time, oil microbes have not degraded it. That is, try to understand what is stalling biodegradation of toxic oil. A method for restarting, or in some cases starting, its biodegradation will be found.

The go, no go, decision on applying enhanced bioremediation at appropriate hot spot locations will be made by comparing risks associated with leaving the oil like it is against gains expected from destroying harmful portions of subsurface oil that may be leaching into shoreline waters.

This study does not consider using commercial products like as Inipol or intrusive mechanical methods. Only generic bioremediation enhancers will be recommended. Improvements to damaged resources will be studied and reported on.

D. COMPLETION DATES

In early FY 96 the team will review data from shoreline surveys collected by ADEC, NOAA, and others. Members of the team will also meet with local communities to help identify shorelines with hot spots. Currently sites of most interests include approximately twelve low energy sites near the community of Chenega Bay that are referenced in the Draft Restoration Program dated March 24, 1995 and 12 to 14 other sites in western Prince William Sound said to be experiencing little reduction of subsurface oil.

From this information selected sites will be visited in mid-FY 96 and characterized by type, location, and amount of remaining subsurface oil. An assessment of its recovery rate and the impact that the subsurface oil is having on recovery will also be conducted.

In late FY 96 or early FY 97 the team will discuss the benefits of using of non-intrusive, non-commercial techniques for accelerating biodegradation of antagonistic oil with the Chief Scientist and residents of near-by communities. The team will then present its recommendation on where enhanced bioremediation should be used to the EVOS Trustee Council. After receiving approval to proceed the team will assume responsibility for obtaining necessary permits and approvals.

During the summer of FY 97 the team will apply enhanced bioremediation methods at two or three of the best suited sites. Increased bioremediation and recovery rates will be studied in detail at each of these. Where justified the team is willing to consider application of appropriate bioremediation methods at other sites as well. But if so, the other sites would not be studied or monitored as closely as the 2 or 3 best suited sites.

During the summer of FY 98 the team will continue application of enhanced bioremediation methods and studying associated biodegradation and recovery rates. Approximately six months following the completion of the FY 98 field season a final scientific report will be submitted to the EVOS Trustee Council for their acceptance in accordance with PROCEDURES for the PREPARATION & DISTRIBUTION of FINAL PAPERS, October 1994, *EXXON Valdez* Oil Spill Trustee Council, Anchorage Restoration Office, 645 G Street, Suite 401, Anchorage AK, 99501.

COMMUNITY INVOLVEMENT

As previously mentioned, this project involves close involvement with residents of local Prince William Sound communities. A number of local resident, particularly those living in or near Chenega Bay, have expressed concern about delays in recovery caused by remaining subsurface oil. Since the project is not yet accepted, ADEC has not discussed

this project with the leaders of the local communities because it did not wish to cause a false impression or mislead local residents.

Once the project is accepted by the EVOS Trustee Council ADEC will hold meeting in the local communities to get their help identifying shorelines having local hot spots. Data on these sites will be closely reviewed and appropriate sites investigated and appropriately incorporated into the study as warranted.

Residents of the Prince William Sound will be hired to fill needed field positions. Training and work experience gained from working on this project will provide local residents the skills and experience needed for employment with other marine related work that will be done by the Seward Marine Science Center and the Prince William Sound Science Center located in Cordova.

When possible it is the intension of ADEC to base this project out of Chenega Village.

FY 96 BUDGET

Personnel	88.9
Travel	12.5
Contractual	818.2
Commodities	3.8
Equipment	0.0
Subtotal	923.4
Gen. Admin.	44.2
Total	965.6

PROJECT DESIGN

Background

When oil spills occur, especially ones involving supertankers where huge amounts of oil are discharged into the environment, the environmental damage can be enormous. The affected area can be aesthetically ruined by the tarry, black residue, and the effects on wildlife and aquatic animals can be devastating. Local ecological diversity can be seriously threatened and food chain interactions significantly disrupted for years following the catastrophe. It is essential to accelerate cleanup to mitigate further damage to exposed plant and animal populations and to humans who consume animals that concentrate the contaminants through trophic level biomagnification. Many compounds in crude oil are environmentally benign, but a significant fraction are toxic or mutagenic. These are the compounds that we are most interested in destroying. Bioremediation is a technology that offers great promise in converting the toxic compounds to nontoxic products without further disruption to the local environment.

Crude oil is composed of tens of thousands of different compounds including normal and branched alkanes, cyclic alkanes (including the saturated fused ring biomarker compounds such as cyclic di-, tri-, and tetraterpanes), aromatic and polycyclic aromatic hydrocarbons (PAHs), heterocyclic sulfur-, nitrogen-, and oxygen-containing compounds, resins, and asphaltenes (Peters and Moldowan, 1993). The normal alkanes, which are the most readily biodegraded compounds, comprise 6 to 10% of the mass of the oil. They can be identified and quantified easily by gas chromatography (GC). The higher molecular weight alkanes are less biodegradable, more viscous, less soluble, and less volatile than the lower molecular weight alkanes. Aromatic hydrocarbons comprise about 1 to 3% of the oil. The 2-ring PAHs (naphthalenes) are the most easily degraded, while 3-ring, 4-ring, and 5-ring PAHs are progressively more recalcitrant. Also, alkylated PAHs are less biodegradable than are the unsubstituted parent compounds, and biodegradability decreases as the degree of alkyl substitution increases. Many of the PAHs can be resolved by GC and identified and quantified by mass spectrometry. Resins, asphaltenes, and biomarkers are the most resistant to biodegradation. The resins and asphaltenes cannot be analyzed by GC. These recalcitrant components remain in the residue after extreme biological and chemical weathering has taken place. Fortunately, however, these compounds have very little environmental impact. The highly branched and cyclic alkanes also cannot be easily analyzed by GC. These compounds comprise the unresolved complex mixture (UCM) or "hump" in the GC trace. The UCM rises significantly above the baseline and is especially pronounced in biodegraded petroleum [Killops and Al-Juboori (1990)].

When microorganisms metabolize petroleum hydrocarbons, the first step usually is reaction of molecular oxygen with the terminal methyl group of alkanes or an aromatic ring of PAHs to form alcohols. The fatty alcohols that are produced by oxidation of alkanes are further oxidized to fatty acids, which are subsequently metabolized via β -oxidation and the citric acid cycle to CO_2 , water, and biomass. The aromatic alcohols formed from PAH oxidation react with additional O_2 and ring fission occurs. Subsequent reactions can lead ultimately to mineralization. The intermediates that are formed during microbial oxidation of hydrocarbons are more polar and more water soluble than are the hydrocarbons themselves. Many of these intermediates are more biodegradable than the parent hydrocarbons. Also, they can be diluted to very low concentrations by tidal mixing. Thus, although the polar products of hydrocarbon metabolism can enter the water column more easily, they are unlikely to cause environmental damage or toxic effects to nearby biota.

Beach cleanup techniques, which include high and low pressure washing, dispersant application, burning, mechanical removal, and bioremediation, vary in effectiveness (Baker *et al.*, 1993). The effectiveness of specific cleanup methods is determined by the material composition of the beach and its exposure to wave action. Bioremediation is the only method that can be applied to the majority of beach types. In 1991, the congressional Office of Technology Assessment (OTA) published a background paper on the technology of bioremediation applied to marine oil spills [Office of Technology Assessment (1991)]. OTA discussed two approaches to bioremediation: biostimulation (nutrient enrichment) and bioaugmentation (addition of oil-degrading microorganisms). Biostimulation is viewed by the scientific community as more promising than bioaugmentation. Among its conclusions was

that the "...usefulness of bioremediation for marine oil spills is still being evaluated, and its ultimate importance relative to other oil spill response technologies remains uncertain."

Prior to the *Exxon Valdez* accident, oil bioremediation by nutrient enrichment in marine environments had received only limited field testing [Halmo (1985); Sendstad *et al.* (1982); Sendstad *et al.* (1984); Sveum (1987); and Sveum and Ladousse (1989)]. Experiments dating to at least 1973 have demonstrated the potential of this technology. Researchers, for example, have tested nutrient enrichment in nearshore areas off the coast of New Jersey, in Prudhoe Bay, and in several ponds near Barrow, Alaska. In each case, the addition of fertilizer was found to stimulate biodegradation by naturally occurring microbial populations [Atlas (1991)]. One smaller scale unsuccessful test using the same fertilizer as in Alaska was recently conducted on beaches in Madeira that were polluted by the Spanish tanker *Aragon*. Researchers speculated that the unsatisfactory results could have been due to differences in the type of oil, the concentrations of fertilizer used, the lower initial bacterial activity, and/or different climatic conditions [Biscoito and Moreira (1990)].

Taken together, the results of these studies suggest that bioremediation can be considered an effective response for the restoration of oil-contaminated beaches, but its effectiveness could not be unequivocally demonstrated by any of them, because the experimental designs all suffered from the same flaw: pseudoreplication (Hurlbert, 1984). *Pseudoreplication* refers to the use of inferential statistics to test for treatment effects (e.g., does bioremediation through nutrient enrichment enhance the rate of crude oil disappearance from an oil-contaminated beach compared to the inherent loss rate?) with data from experiments where treatments are not replicated or replicates are not statistically independent.

To make this point clearer, other definitions must be introduced. *Randomization* refers to the spatial assignment of treatments (experimental units) on the experimental plane in a way that mitigates or eliminates systematic natural bias or experimenter bias (e.g., effects of longshore currents, prevailing winds, underground water flow, light gradients, etc.). Its purpose is to eliminate biased placement of treatments that would impose one or more systematic effects on some treatments but not others. *Replication* refers to the repeated use of treatments and controls (two or more) to obtain an estimate of variability. Two types of variability must be considered: experimenter-induced variability (random error) or inherent variability among experimental units (e.g., initial hydrocarbon concentration, geomorphological characteristics, or exposure to waves). *Interspersion* is the act of mitigating the effects of regular spatial variation in the properties of experimental units through statistical randomization procedures (unrestricted or restricted) and segregation of treatments. For example, placement of four control plots in a row followed by four treated plots in a row fails the criterion of proper interspersion of treatments.

Two important examples of experimental designs that involved pseudoreplication are found in the Alaska oil spill project (Pritchard and Costa, 1991; Pritchard *et al.*, 1991; Pritchard *et al.*, 1992). In 1989, following the grounding of the supertanker *T/V Exxon Valdez* on Bligh Reef in Prince William Sound, EPA in conjunction with the Exxon Corporation and the State of Alaska embarked on the largest oil spill bioremediation project ever attempted. Two sites on Knight Island were selected for these experiments: Snug Harbor, located on the southeast

corner, and Passage Cove, located on the northwest edge of the island.

At Snug Harbor, six plots were established on two types of beach: one was a cobble beach and the other was composed of mixed sand and gravel. Three treatments were compared: oleophilic fertilizer (Inipol EAP 22), slow release solid fertilizer briquettes, and a no amendment control. Each plot was divided into 21 subplots for sampling purposes. At each sampling event, the samples collected from each subplot were analyzed individually for oil residue weight, normal and branched alkane concentrations, nutrients, and hydrocarbon-degrading bacteria by most probable number (MPN). The residue weight data were segregated according to beach type and analyzed by linear regression.

The statistical validity of the experimental design and subsequent analysis of data from the Snug Harbor experiment was deficient in several respects. First, the treatments were not randomized with respect to their relative positions: the oleophilic fertilizer plot was always between the control and the slow-release fertilizer plots, and both slow-release plots were adjacent to a running stream. This could have caused systematic errors that might have skewed the results in one direction or another. Furthermore, although a complete block design was used (albeit nonrandomized), the results were not analyzed by ANOVA. Instead, the investigators decided to treat each type of beach individually, using the 21 nonindependent data points that were obtained from each plot at each sampling event as pseudoreplicates in support of their regression analyses. The number of data points collected from within each plot are irrelevant, however, because the ANOVA only considers each plot datum when computing significant effects. The correct approach is to compute plot means from the 21 measurements from each plot and use an ANOVA to infer differences among the treatments. Finally, the plots were not identical in size, and the concentration of applied nitrogen was not the same for the oleophilic and slow-release fertilizer plots. All plots should be as nearly identical as possible in area and linear dimensions so that systematic fluctuations in environmental or climatic effects do not affect the plots differently. Furthermore, if a valid comparison of nitrogen source is desired, all treated plots should receive the same dose of nitrogen so that the only difference is the composition of the chemical being applied.

One of the most important lessons learned from the Snug Harbor experiment is that more independent replicates are required to discern statistically significant differences between treatments and controls, because the oil distribution was extremely heterogeneous. The result of having only two replicates of each treatment was that it was impossible to determine whether differences in biodegradation rate among the experimental plots were due to treatment or location (Pritchard *et al.*, 1991). The only way to separate these effects is to increase the number of independent replicate plots, randomly intersperse them on the experimental plane, and analyze the resultant data using appropriate statistical testing procedures.

A similar experiment was conducted at Passage Cove. The treatments included a control, a one-time application of Inipol combined with a slow-release granular fertilizer (Customblen™), and daily applications of water soluble fertilizer via a sprinkler system. Unfortunately, the treatments were not replicated, and the control and treatment plots were not treated the same way. It is possible that the enhanced biodegradation observed in the plots

that received the water-soluble fertilizer treatment could have been due to the daily watering instead of nutrient addition. Furthermore, since the plots were not replicated, there is no way to be certain that the effect was not location specific.

The data from the Passage Cove experiment were analyzed in the same manner as the Snug Harbor data. That is, the researchers used regression analysis of the log-transformed data to compare individual treatments to the control instead of using ANOVA to analyze the data from all three treatments simultaneously. Another statistical error in this analysis occurred when the researchers applied a zero-order decay model to the oil residue weight data, then excluded data that did not conform to the model before estimating degradation rate parameters. Outliers can be excluded from a data set only after performing an appropriate statistical analysis, and they cannot be excluded simply because they do not conform to an assumed model.

A third bioremediation experiment was conducted in 1990 by researchers from Exxon (Bragg *et al.*, 1994), who used hopane as a conservative biomarker (Peters and Moldowan, 1993) to eliminate much of the heterogeneity that confounded previous statistical analyses. Tests were carried out on three separate beaches on Knight Island. Each site was subdivided such that half received fertilizer (a combination of Inipol and Customblen) and the other half was untreated. Samples were collected periodically over a period of 109 days from the treated plots, but for only 72 days from the control plots. Replicate samples collected from a single plot were treated as statistically independent measurements for regression analysis with a complex multiparameter mathematical model. The researchers concluded that changes in hydrocarbon/hopane ratios were most influenced by the oil load, the concentration of the compounds comprising the polar fraction, and the cumulative nitrogen measured in the interstitial water. The cumulative nitrogen load was the most important variable.

Although the rate enhancement predicted by the model (approximately nine-fold enhancement over the control), and apparently observed, is impressive, the conclusions are based on a classical pseudoreplicate design. The three samples that were collected from each plot at each sample event were treated as statistically independent, but they were not. Thus, this artificially increased the degrees of freedom for the regression, but did not really affect the resolving power of the analysis. Although this experiment was closer to a true replicate design, because three separate independent sites were used, all of the treated plots were not treated alike. The fertilizer application frequencies were substantially different among the three sites.

It is clear from the foregoing discussion that most of the oil spill bioremediation studies reported in the literature, including the *Exxon Valdez* investigations, have been flawed to some degree. Thus, the conclusions from these studies are not supported by a rigorous analysis of the data. This has led to skepticism regarding the effectiveness of bioremediation among the scientific, engineering, and user communities. To prove that bioremediation can be used to restore Prince William Sound beaches to their prespill conditions, a well-controlled, statistically sound evaluation of the technology must be performed. This evaluation must involve adequate replication, proper interspersing of treatments, and the control plots must be treated in exactly the same way as the fertilized plots. The plots need

not be large. They should, however, be representative of the local terrain and provide a reasonable representation of the conditions at the test site (e.g., extent of tidal inundation, wave action, longshore currents, daylight exposure, stream flow gradients, and length of the impacted intertidal zone). In particular, the effect of tidal wash on performance must be considered, because of the large intertidal zones that are characteristic of Prince William Sound beaches. Plots that are 5 m wide by 10 m long (i.e., the longer dimension is perpendicular to the shoreline) are probably adequate.

The technology evaluation portion of this proposal mimics the experimental design that was used in a recent oil bioremediation study conducted on the shore of Delaware Bay (Venosa *et al.*, 1995). This study was the first to obtain convincing evidence that bioremediation occurs at substantial rates on a sandy marine beach. Oil removal rates due to physical processes, such as wave action and tidal wash, were quantified using hopane, a nonbiodegradable biomarker in oil, and the rates of biological transformation of oil constituents were determined by normalization to hopane. One of the conclusions of this study was that in a nutrient rich estuarine environment such as Delaware Bay, background nitrogen levels (which averaged greater than 0.8 mg/L) were sufficient to promote reasonable rates of bioremediation in the control plots. This is not the case in Prince William Sound, however, where the available nutrient concentration is relatively low. The project described in this proposal will be the first truly controlled demonstration of crude oil bioremediation in a nutrient-deficient environment. After an effective bioremediation strategy has been demonstrated, it will be implemented on a large scale to restore several oil-contaminated beaches to a state that is both ecologically sound and safe for human activities.

A. Objectives

The work described in this proposal has two major objectives. First, we will determine the most effective and economical method for applying bioremediation to restore oil-contaminated beaches in Prince William Sound to their pre-spill condition. Having identified the level of enhanced restoration that is achievable, we will apply the technology to beaches in Prince William Sound, including several near the Chenega Bay community. These two broad objectives will be achieved through the following series of specific objectives:

1. A detailed survey of candidate beaches will be conducted to identify those that are most likely to benefit from bioremediation. The beaches will be characterized with respect to geomorphology, hydrodynamics (e.g., exposure to wave activity and subsurface freshwater flow), the extent, location, and chemical composition of oil contamination, the toxicity of oiled sediments and beach pore water, and the ecological impact of the oil contamination present at these sites.
2. The rates and dominant mechanisms of nutrient transport will be investigated with a conservative tracer to design efficient and effective methods for applying fertilizer to contaminated beach material.
3. The effectiveness of bioremediation for restoring oil-contaminated beaches will be quantified at a minimum of two sites. The most promising oil bioremediation strategies will be tested during this phase of the project. We will use a study design that provides a statistically sound evaluation of treatment effects.

4. Several oil-contaminated beaches will be treated using the bioremediation strategy found to be most effective. The beaches selected for treatment will be chosen based on risk and potential for benefit. The Chief Scientist will be consulted during this decision-making process and a joint recommendation will be forwarded to the EVOS Trustee Council. To the extent possible, local residents will be employed and trained to apply selected bioremediation methods and collect monitoring samples.

B. Methods

The proposed project is expected to extend over three summers. The first summer involves a detailed site characterization and risk assessment of selected beaches in Prince William Sound. The sites will be characterized with respect to the extent of oil contamination, the abundance and diversity of their microbial, infaunal, and epibiotal populations, sediment toxicity, and the hydrodynamic conditions that control nutrient transport to and within the oil-contaminated zones. The chemical and ecological characteristics will establish a baseline that can be used to assess the effectiveness of the bioremediation treatments. The hydrodynamic characteristics will be used to determine strategies that are most likely to result in effective bioremediation.

Using this approach the most effective bioremediation strategy will be conducted in the second year. These experiments will enable us to make statistical comparisons of the biodegradation rates that result from the various treatments. The statistical analysis will allow us to determine whether bioremediation enhances the rate of oil degradation in the contaminated beaches and whether one treatment is more effective than the other. The null hypothesis that we will test can be stated as follows: treatment of oiled beaches with inorganic nutrients (nitrate and phosphate) does not accelerate the disappearance of contaminating hydrocarbons and consequently does not accelerate the restoration of the infauna and epibiota native to the shoreline.

The bioremediation strategy that is determined to be most effective and economical could be applied on a large scale during the third summer of this project. In this phase of the project, several oil-contaminated beaches will be restored to an ecologically healthy state that is suitable for appropriate human activities. This will involve fertilization of contaminated beaches at intervals that are sufficient to maintain maximal biodegradation rates. It will also involve chemical, toxicological, and ecological monitoring to assure that the bioremediation process is proceeding safely and effectively

1. Geomorphology and oil distribution

(a) Background

Since September 1989, the Hazardous Materials Response and Assessment Division of NOAA has sponsored surveys of the geomorphological changes and oil distribution at 18 stations throughout Prince William Sound. This work was designed to provide the scientific basis for decision making by the U.S. Coast Guard, as the Federal On-Scene-Coordinator of the Exxon Valdez oil spill, on shoreline treatment in 1990 and beyond. Other objectives evolved as the study progressed, including: 1) providing a physical and chemical framework for sites

included in NOAA's biological monitoring program; 2) monitoring the effectiveness of certain countermeasures, such as berm relocation; and 3) gaining a better understanding of the processes of oiling and natural cleansing of gravel beaches.

Although Prince William Sound is a relatively sheltered area, at least in comparison with the open north Pacific Ocean, the more exposed sites are subject to significant wave action during the passage of frequent extratropical cyclones. There are also many areas in Prince William Sound that are completely sheltered from significant wave action. The study sites of the original surveys were chosen to represent the entire spectrum of hydrodynamic energy levels that exist throughout the Sound. Shoreline environments included exposed cobble/boulder platforms with berms, bayhead beaches with mobile fine-gravel, pebble beach/tidal flat areas, sheltered rubble slopes, and sheltered bedrock. All of these shoreline types were heavily oiled during the spill.

(b) Methods of Study

A detailed description of the methods of study used during the NOAA field surveys is given in Michel and Hayes (1991). The field area has been visited 13 times to date during this project between September 1989 and July 1994. Before any new survey work is started in 1996, the NOAA surveys and other SCAT surveys will be reviewed to determine if any other potential sites should be examined for restoration work.

Based on our present knowledge, we are considering several sites for survey in 1996. First, we will investigate several sites near the Chenega Bay community. The Invitation to Submit Restoration Projects for FY96 (p. 72) states that: "There are approximately twelve 'low energy' sites near the community of Chenega Bay which have experienced little reduction in subsurface oil. The worst sites still sheen." Additional potential sites include EL056C(head of the east arm of Northwest Bay), EV036A (Northeast Evans Island), GR101B (North side of a detached island north of Green Island), KN136A (Bay of Death in Bay of Isles), KN213C (Rua Cove), LA015C and LA015E (Northeast coast LaTouche Island), LA 020C (West shoreline of Sleepy Bay), EV037A and EV039A (Northeast Evans Island, and ER020B (North end Elrington Island), NOAA station N-3 at the northwest end of Smith Island and NOAA station N-10 at the entrance to Herring Bay. Finally, the Bay of Isles may have a heavily oiled marsh in need of restoration.

Initial field surveys of the candidate sites will be carried out during the low tide portion of a maximum spring tide on 5-12 May, 1996. At each site, an intertidal zone segment 50-100 meters in length will be surveyed in detail and transects will be marked with permanent stakes. Approximately every 10 meters along the beach, topographic profiles will be run perpendicular to the beach extending from above the high tide line to the low water mark. Trenches will be dug to the bottom of the subsurface oil to determine the depth of oil penetration along each profile. Sediments (oiled and unoled) in the trench will be sampled and described in detail. The intertidal zone will also be sketched and photographed in detail. Each study site will have the following information available at the end of the initial survey:

1. Permanently marked transects;

2. Map of surface morphology and sediment distribution;
3. Map of surface oil distribution, if any; and
4. Three-dimensional map of subsurface oil, including distribution by oiling category (HOR, MOR, etc.).

The terminology and definitions of the oiling characteristics that will be used are those established during the 1991 interagency shoreline surveys of the oiled regions (MAYSAP). Surface oil was described using the following terms;

1. Asphalt pavement (AP): Heavily oiled sediments held cohesively together.
2. Coat (CT): Oil which ranges between 0.1 and 1.0 millimeter thick (can be easily scratched off with a fingernail).
3. Stain (ST): Oil less than 0.1 millimeter thick (cannot be scratched off by a fingernail).
4. Film (FL): Transparent or translucent film or sheen.

Subsurface oil was described according to the following terms:

1. Heavy oil residue (HOR): Pore spaces partially filled with oil; oil usually not flowing out of sediments. In July 1994 five sediments were HOR with actual TPH concentrations from 7700 to 7900 mg/kg.
2. Medium oil residue (MOR): Sediments heavily coated with oil; pore spaces are not filled with oil; pore spaces may be filled with water. Five of the July 1994 samples were described as MOR with actual TPH concentrations between 800- 4800 mg/kg.
3. Light oil residue (LOR): Sediments lightly coated with oil; water may bead on sediments. Five of the July 1994 samples described as LOR had actual TPH concentrations from 470 to 3300 mg/kg.
4. Oil film (OF): Continuous layer of sheen or film on sediments; water may bead on sediments. Three samples from the July 1994 series were described as OF with actual TPH concentrations of 80-1000 mg/kg.

Samples will be collected from both surface and subsurface points. For oiled surface samples, the top two centimeters will be collected for detailed characterization and analysis of weathering trends. Subsurface oiled sediment samples will be collected from discrete intervals, frequently from the bottom of the oiled sediments in the trenches. Other intervals will be collected as appropriate. No samples will be composited; all samples will be grab samples. All samples will be returned to the laboratory for analysis as quickly as possible.

2. Toxicology and Ecology

Objective

The effectiveness of bioremediation in large part rests on its ability to reduce injury to marine life caused by oil. If resources are injured prior to treatment they should be less so after treatment. Moreover, it should be demonstrated that treatment accelerates recovery of marine resources from injury relative to leaving the oil untreated. The application of nutrients or other materials to oil is not necessarily an innocuous activity and there remain many uncertainties about the effects of prolonged shoreline treatment on marine ecosystems. Certainly, it should be demonstrated that treatment does not increase injury or otherwise make things worse. In short, an effective bioremediation program will indeed accelerate the restoration of resources and resource values.

In some areas where oil remains, mussels and clams have remained contaminated to some degree raising concern about their consumption by subsistence families and wildlife such as shore birds and mammals (Houghton et al, 1993; other NRDA and Restoration references) This effort will attempt to determine if enhanced bioremediation can reduce this risk faster than some alternative action, including no action?

Another issue is the health, abundance and biodiversity of shoreline ecosystems. The goal of any technique is to return the area to be restored to a state equivalent to the pre-impact status. In many studies designed to assess the effectiveness of various shoreline oil removal techniques, the impact of the procedure on resident biota is often overlooked. Thus, a removal technique which rids the shoreline of oil but also destroys the intertidal biota would be considered effective even though the procedure caused more harm to the intertidal habitat than leaving the shoreline oiled. Bioremediation techniques offer the opportunity to overcome the destructive effects of mechanical cleaning of oiled shorelines (i.e., Stekoll et al., 1993; Houghton et al., 1993; Mearns, 1993) by temporally enhancing natural degradative processes. It is, however, unknown whether current bioremediation techniques are harmful to resident intertidal organisms or cause an alteration in the community composition. By monitoring the effects of various bioremediation techniques on intertidal communities this study will address the important question of the effects of this restoration technique on resident populations. Without this knowledge the effectiveness of bioremediation techniques at restoring intertidal communities to pre-spill conditions will remain unknown.

Related to this is the persistent toxicity of the oil to marine life and the extent to which that direct toxicity can be reduced.

Approach

A number of tools now exist to document the safety and ecological benefits of bioremediation and other treatments. Trends in oil bioaccumulation (and therefore seafood safety) can be monitored directly through a careful program of sampling mussels and clams before, during and after treatment in treated and untreated areas. This can be successfully done using resident organisms (i.e., Houghton et al., 1993; Shigenaka et al., 1993) or transplanted mussels and clams (Houghton et al., 1993.).

Trends in the recovery of the abundance and biodiversity of shoreline ecosystems can be monitored using non-destructive enumeration methods for the epibiota (plants and animals on the shoreline surface) and by enumerating infaunal (sediment dwelling) organisms screened from sediment cores (Stekoll et al, 1993; Houghton et al., 1993).

Trends in the actual toxicity of the remaining and treated oil can be documented using a small battery of well-tested bioassays on samples of sediment and pore water from treated and untreated shorelines (Doe et al., ; Mearns et al., in press). Accepted tests available today can document the effectiveness of treatment on reducing mutagenic and reproductive damage, and the mortality of various organisms.

Finally, this triad of biological assessment approaches will be coordinated so that a true assessment of the biological risks as well as benefits of treatment can be developed and used in the final treatment decision.

We propose a three-pronged strategy to make sure bioremediation accelerates the recovery of ecological and resource values and to make sure it does not increase injury. At treated and untreated sites we will monitor trends in mussel and clam bioaccumulation, epibiota and infaunal abundance and diversity and sediment toxicity.

We propose to use appropriate bioaccumulation, biodiversity and toxicology methods both to document the extent of existing injury and to monitor recovery trends. Existing ecological, bioaccumulation and toxicology conditions will be assessed as part of the 1996 shoreline characterization and correlated with oil weathering state. Because of the various oil weathering conditions, it is possible that two sites with equal amounts of oil may have very different ecological conditions. Sites that produce no toxicity, no excess bioaccumulation of oil and no signs of existing injury to infauna or epibiota should probably be left alone (not treated) because there will accrue no benefit, and possibly increased injury, by attempting to treat the remaining oil at these sites, either with bioremediation or any other treatment. However, sites exhibiting one or more signs of ongoing injury are candidates for effective treatment. We then propose to apply these biological assessment tools to monitoring the effectiveness and effects of treatment during the 1997 feasibility study and continue monitoring the effectiveness and effects of operational treatments in 1998 (year 3).

As mentioned earlier in addition to oiled sites, the biology risk assessment work will also be conducted at a few comparable unoiled sites for the purpose of determining the ultimate target for remediation.

2. Toxicity and Ecology Methods

A. Bioaccumulation

Mussels and/or clams will be surveyed, monitored and or transplanted for documenting petroleum hydrocarbon contamination. During Phase 1 (1996) populations of mussels and clams at target shorelines will be identified and up to 24 composites collected and analyzed

for over 45 polycyclic aromatic hydrocarbons (PAH's). During Phase II, composites of mussels and/or clams will be monitored for trends in PAH contamination at a minimum of three plots per treatment four times over the course of the treatment testing (36 samples). This may require transplanting mussels from an unoiled control site, a method used successfully in NOAA's Long-term Ecological Monitoring Program (Houghton et al., 1993). Finally, during full operations in Phase 3, mussels and/or clam composites will be collected before treatment, shortly after treatment and several months after treatment at three sites (27 samples) to document the extent to which bioremediation reduced contamination.

Mussels and clams used for bioaccumulation must be selected carefully for meaningful and comparable results. Protocols developed by NOAA (Salazar, 1994) will be used to collect and/or deploy transplanted mussels. Required transplanting will be conducted by NOAA HazMat personnel or others under NOAA guidance during cruises and trips scheduled by other investigators on this project. PAH's in mussel/clam soft tissue will be measured by Charles Henry, Louisiana State University, using standard extraction and GC/MS methods frequently employed by NOAA during spill responses and assessments. (described in Houghton et al., 1993). This will also ensure that data from this bioremediation program will be entirely consistent and comparable to data collected during NOAA's Long-term Ecological Monitoring Program (which will continue for 4 more years). Measure oil hydrocarbons in collected samples. Deploy mussels and or clams from clean reference sites.

Summary

July-August, 1996 - Recover deployed mussels and clams and measure petroleum hydrocarbons. Determine sites at risk (excess hydrocarbons)

Summer, 1997 - Deploy mussels and/or clams in treatment plots, retrieve and measure differences in bioaccumulation

Summer, 1998 - Deploy mussels and/or clams in treatment plots, retrieve and measure differences in bioaccumulation

B. Shoreline Ecosystem

To assess the response of epibenthic and infaunal communities to the application of various bioremediation techniques we will utilize the experimental design outlined in the main body of this proposal. All biological survey work will be carried out by a team from the University of Alaska under the direction of Dr. Michael Stekoll.

In Phase I, shoreline characterization will occur on candidate sites for restoration and on matched unoiled reference sites. Sites will be selected by other members of the bioremediation team. Sampling at the sites will consist of 15 quadrats (25 x 25 cm) placed

randomly over the entire shore to be sampled. These quadrats will be randomly placed with horizontal and vertical stratification to insure dispersion. This technique will allow us to assess the biological status of each shoreline being considered for treatment. By sampling unaltered reference shorelines nearby, the recovery status of these potential test beaches can be determined and a recovery "goal" can be defined. At each quadrat percent cover and density of the epibiota will be estimated. Percent cover will be assessed using a stratified random point contact method with 80 points. At each point the canopy, understory, and primary space occupant will be recorded. Farrell (unpublished data) has shown that at least 60 points are needed to obtain an accurate estimate of percent cover and that systematic points are not accurate if long, skinny organisms are encountered. In addition to percent cover, densities and sizes of key organisms will be estimated for each quadrat. Limpet, whelk, and *Fucus* numbers will be counted in each quadrat, and the size of the 10-25 individuals nearest the upper left quadrat corner will be recorded. Barnacles, littorine snails, and mussels will be counted in three randomly chosen 5 x 5 cm subsections of each quadrat. These more abundant invertebrates will not be measured. Finally, the abundance and size of large invertebrate predators, such as seastars, will be recorded in 6 larger plots (5 x 10 m).

If the site supports infauna, then cores (10 cm diameter) for fine sediment or collections of the upper 10 cm of sediment for coarser grained sediments will be taken. The same layout of quadrats will be used as described above. A single sample will be taken either 15 cm above or below (randomly determined) the left side of the quadrat. Subsequent samples, if any, will be taken from similar areas around the quadrat in a systematic manner to eliminate bias. For example, if the first sample is taken above the upper left quadrat corner, the next sample will be from below the lower right corner, then the next will be from above the upper right, and the final sample, if needed, would be preserved in formalin and transported to Juneau for sorting and counting. Infauna will be sorted into broad taxonomic categories and the number and biomass of individuals in each category will be recorded.

For Phase II bioremediation techniques will be experimentally tested on five (5 x 10 m) plots on each shoreline in a randomized block design. At least three shorelines will be used for the tests. Test plots will be separated by 10 m to reduce possible interactions, and all five bioremediation treatments plus an unmanipulated control plot will be randomly allocated on each shore. Sampling of each of the 5 x 10 m plots will be divided into three equal sections of 3.3 x 5 m, and each of these subplots will be sampled. In each subplot, three quadrats (25 x 25 cm) will be randomly located and permanently marked. To insure dispersion throughout the subplot, quadrat placement will be stratified both vertically and horizontally. Sampling will occur three times relative to bioremediation treatments: 1) all areas will be sampled prior to any treatment, 2) sampling will occur about one week after treatment, and 3) shores will be sampled again 6-8 weeks after treatment. The same procedure for quadrat and infaunal sampling will be used as that described above. The unaltered reference shorelines will continue to be monitored in the same manner to evaluate natural recovery and interannual variation. Comparisons will be made among the various bioremediation treatments as well as between the unaltered reference site and treated plots.

Phase III will involve bioremediation on a restoration scale where entire beach segments will

be treated using the most effective method from phase II. To assess the effectiveness of the technique employed in phase III, we will sample shorelines for epibiota and infauna using the same methods described for phase I. The entire treated shore will be sampled and compared to unoiled reference sites.

Summary:

Summer, 1996 - Quadrat and infaunal core surveys of epibiota and infauna at mid-intertidal zones of target shorelines. One survey each.

Summer, 1997 - Quadrat and infaunal core surveys of epibiota and infauna at mid-intertidal zones of treated and untreated plots, in replicate. Sample before, one week after and 6-8 weeks after treatment begins.

Summer, 1998 - Quadrat and infaunal core surveys of epibiota and infauna at mid-intertidal zones of treated and untreated plots, in replicate. Sample before, one week after and 6-8 weeks after

C. Toxicity

We propose the use of a series of toxicity tests to monitor and assess the ability of bioremediation to reduce harmful impacts of oil remaining in sediments at selected experimental and operational sites in Prince William Sound. Bioassays will be performed on sediment, sediment-elutriate and/or water samples by Dr. William Fisher (US EPA, Gulf Breeze, Florida), Kenneth Doe (Environmental Quality Laboratory, Bedford Institute of Oceanography, Dartmouth, Nova Scotia) and Dr. Kenneth Lee (Maurice Lamontagne Institute, Department of Fisheries and Oceans Canada, Mont-Joli, Quebec). Under coordination by Dr. Alan Mearns, NOAA, all worked together to successfully complete the first international assessment of the toxicology of shoreline bioremediation in Delaware.

A battery of tests using a variety of proven animal and microbial assays and a wide range of toxicity endpoints (see below), will be used initially to determine which tests will be most useful for experimental and operational monitoring. Most are accepted standardized marine toxicity tests but few have been applied to assessing the toxicity of either oiled or treated shorelines.

In Phase 1 we will use the entire battery of tests characterize/assess for risk to biota the various sites under consideration for possible bioremediation. The risks tested include survival, reproduction, hatching success, and (for Microtox) inhibition of photoluminescence. Toxicity characterization and screening tests will be carried out on sediments collected from oiled and unoiled sites in October 1995 (archived sediment samples from the NOAA HazMat 1995 Long Term Monitoring Program) and on sediments and porewater from oiled and unoiled sites during the summer of 1996 (approximately 10 sites from the geomorphology reconnaissance survey of target shoreline). From the results we will select the most sensitive and logistically-appropriate bioassay tests for use in Phase 2.

In Phase 2 we will assess change in toxicity (biological effects) in various treated and untreated plots throughout the summer of 1997 and so to determine optimum nutrient addition methods and nutrient addition rates for removal of toxicity. Select the most reliable bioassay tests for routine monitoring operations to identify site recovery.

In Phase 3 we will use selected bioassays to monitor the operational treatment of shorelines during the summer of 1998.

Tests

We will use several bioassay tests that have been used with success during the Delaware Bay Bioremediation Experiment: 1) Grass shrimp embryo hatch success test; 2) Microtox porewater test; 3) Microtox solid-phase (modified-elutriate) test; and 4) sea urchin fertilization porewater test

The Microtox pore water and sea urchin fertilization tests are standardized tests fully described in the toxicology literature and has been used in numerous environmental toxicology surveys by Environment Canada. The solid-phase Microtox test is also standardized but was modified in 1994 to assess the Delaware Bioremediation project.

The Grass shrimp embryo hatch survival tests is not standardized but proved to be one of the most sensitive tests used to document oil and treatment toxicity at the Delaware experiment. Subsequent development of this test system has greatly enhanced its applicability to field sites. Since the 1994 Delaware study, tests with oil water soluble fraction (WSF) have allowed us to reduce exposure time from two weeks to four days. Smaller samples can be used and a sediment elutriation method has been adapted but needs to be tested on sediments from an oiled shoreline.

Based on ecological relevance to the field site, additional tests that we propose to screen for use include:

5) An amphipod survival test will be conducted according to Environment Canada standard procedures, using the amphipod *Eohaustorius estuaries* (or another species, if more relevant). This test involves exposing amphipods to sediment samples and recording survival after a known exposure time. A sublethal response (reburrowing of surviving animals) will also be determined.

6) Mutatox test (measures mutagenicity). This is a new method to identify mutagenic effects that has just been released by Microbics Corporation. The test is based on a procedure similar to the Microtox Procedure.

7) Microtox solid-phase (modified-elutriate) test. A modified Microtox solid phase test using sediment elutriates will be developed/refined. The protocol will be defined for use with the Grass shrimp bioassay procedure.

8) Toxi-chromotest (bacterial enzyme activity/solid-phase) test. This bioassay uses a

mutant *E. coli* strain that produces a specific enzyme under healthy culture conditions. The bacterium is sensitive to a wide spectrum of toxicants that impair or inhibit the release of the enzyme. The exposure procedure includes growing the bacterium directly in contact with the sediment sample without solvent extraction. Inhibition of the bacterium to produce the enzyme due to the toxicant's action can be determined colorimetrically.

9) Bivalve mollusc survival test. Acute effects of the test sediment on embryos of blue mussel (*Mytilus edulis*) larvae during a 48-hour exposure period will be determined. The method involves exposure of <3-hour old mollusc larvae to pore water extracted from the sediment by centrifugation.

10) Juvenile polychaete (*Neanthes* sp.) survival test. Depending on the grain size of the sediment at the experimental sites under selection; a juvenile polychaete (e.g. *Neanthes* sp.) survival and growth test can be conducted. In this test the organism is in direct contact with the sediment and is allowed to ingest sediment in the process of feeding. Consequently, the growth response of the organism reflects the presence/absence of contaminants associated with the solid phase of the sediment that impair, inhibit or enhance food conversion to body mass.

Summary

Phase 1 (1995/1996)

- sediment toxicity to estuarine/marine amphipods
- Mutatox test
- Microtox porewater test
- Microtox solid-phase (modified-elutriate) test
- sea urchin porewater test
- Toxi-chromotest (bacterial enzyme activity/solid-phase) test
- Bivalve mollusc survival test
- Juvenile polychaete (*Neanthes* sp.) survival test

Phase 2 (1997):

- sediment toxicity to estuarine/marine amphipods
- Mutatox test
- Microtox porewater test

- Microtox solid-phase test
- sea urchin porewater test
- Toxi-chromotest test

Phase 3 (1998):

Bioassays will be selected based on their sensitivity, repeatability and applicability during the first two years of the program. For budget purposes it is assumed that amphipods and Mutatox will be selected.

References

Houghton, J.P., et al., 1993. Evaluation of the condition of Prince William Sound shorelines following the Exxon Valdez oil spill and subsequent shoreline treatment. Volume II. 1992 shoreline monitoring survey. NOAA technical Memorandum NOS ORCA 73. National Oceanic and Atmospheric Administration, Seattle, WA.

3. Hydrodynamic Characterization

(a) Objective

Bioremediation involves addition of rate-limiting nutrients, such as nitrogen and phosphorus, to oil-contaminated environments to stimulate the growth of indigenous hydrocarbon degraders. For bioremediation to be effective these nutrients must remain in contact with the oil-contaminated beach material at concentrations that are high enough to allow the hydrocarbon-degrading bacteria to grow at their maximum rates. When oil is stranded in the intertidal zone, several forces can remove water-soluble nutrients from the bioremediation zone. The periodic flooding that is associated with rising tides is one such force. Wave action can also affect nutrient transport in the intertidal region of beaches. In Prince William Sound, Alaska, the high annual rainfall results in high flow rates for fresh water through some beaches. All of these forces can interact to affect the pattern and rate of nutrient transport in oil contaminated beaches, and optimal nutrient delivery strategies can be devised only when the most important factors, or combination of factors, have been determined.

The objective of the first year of this project is to characterize the hydrodynamic forces that control nutrient transport in the beaches that will be targeted for restoration by bioremediation. This information will be used to design nutrient application strategies that can be tested during the second year and implemented on a large scale in the third year of

this project.

(b) Beach Hydrodynamics

Flow through the porous matrix of a beach is driven by a combination of three main factors, but the most important driving forces depend on the characteristics of individual beaches. The first factor is the rise and fall of the beach groundwater level due to tidal influences. Typically the water level in the beach tracks the level of the tide, but drainage lags behind the ebbing tide, because of resistance from the porous media. The degree to which beach drainage lags the falling tide depends on the hydraulic conductivity of the beach material. The second factor that influences the groundwater flow is input of water through wave action. This is usually considered to be the strongest effect (Brown and McLachlan, 1990), and it operates through two main mechanisms. First, water enters the beach from wave runup. This water percolates vertically into the sand above the water line and flows horizontally at the water table (Riedl and Machan, 1972). Waves can also affect groundwater movement in beaches by a pumping mechanism that moves water into and out of the beach in submerged areas. The pumping is driven by differences in head between wave crests and troughs (Reidl *et al.*, 1972). Finally, the flow of fresh groundwater seaward from inland might have a substantial effect on beach hydrodynamics by causing continuous advective horizontal flow from the beach face at or near the water line (Glover, 1959). This type of groundwater flow can interact with tidal fluctuations to produce complex variations in the groundwater level within the beach (Nielson, 1990).

The hydrodynamic factors that drive groundwater flow in beaches will influence the movement and persistence of nutrient that is applied to the beach. The relative importance of these factors is usually considered to be (in order of decreasing importance): waves, inland groundwater flow, and tidal inundation. The relative importance of these factors can shift dramatically with geographic location and local weather conditions, however. For example, at the proposed sites in Prince William Sound, the wave energies are typically low, but the tidal range is 15 to 18 feet. These macrotidal variations are expected to have more profound effects than normal tidal ranges, but the effect on groundwater flow and nutrient transport has not been studied. Also, the flow of fresh groundwater through Prince William Sound beaches is expected to be an important transport mechanism, because of the very high annual rainfall that is characteristic of the area. The nutrient application strategies that we will evaluate are designed to exploit different transport mechanisms. Some of the available nutrient application strategies have obvious appeal, because they place the nutrient exactly where it is needed, but they have extensive labor requirements and, consequently, high costs. Other strategies are less labor intensive, but the outcome of nutrient movement is less clear.

Waves and groundwater flow are known to produce horizontal advective flow within the matrix of the beach. The implication of this is that a tracer or nutrient applied to one area of the beach will be carried to another area by this flow. Wise *et al.* (1994) proposed a model for a Prince William Sound beach with inland groundwater flow that suggested that nutrient applied within a ditch above the high tide line, but deep enough to contact the groundwater table, would be carried into the bioremediation zone by horizontal advective flow, aided by the buoyancy of the less dense freshwater. An analogous model of a wave-driven system

suggests that a tracer applied to the subsurface within the upper swash zone would be carried into the bioremediation zone as the beach fills from tidal inundation (Strohmeier, unpublished, based on Riedl and Machan, 1972). Wise's method requires a substantial excavation to reach groundwater, which is not necessarily consistent with a beach restoration goal. Strohmeier's method requires only a shallow burial of solid nutrient at the upper level of tidal inundation, which provides a low impact, low labor option that is more consistent with the ultimate goal of this project.

A tidally dominated system is expected to produce less advective flow, and nutrient movement would be more a result of diffusion from the original source. This system requires the placement of the nutrient directly within the bioremediation zone as a solid or liquid. Under flat calm conditions and low magnitude tidal range without inland groundwater flow, the main mechanism for movement of the tracer is simple diffusion in all directions. However, in reality, this site is characterized by a high magnitude tidal range. If the beach drains slowly relative to the rate at which the tide ebbs, advective flow through the beach will result.

(c) Experimental Design

The nutrient transport study will investigate three nutrient application strategies: surface application of water-soluble nutrients, infusion of water-soluble nutrients in a trench located above the high tide line, and application of slow-release nutrients directly to the oiled sediments. The surface application method has been shown to be an effective mechanism for supplying nutrients to beaches contaminated with oil at the surface (Pritchard and Costa, 1991; Venosa *et al.*, 1995). The trench application method has been suggested for nutrient application on beaches with a high degree of fresh water input, such as those in Prince William Sound (Wise *et al.*, 1994), but it is also likely to be effective when wave activity is an important transport mechanism (Venosa *et al.*, 1994b). Slow-release fertilizers are probably the most economical nutrient application mechanism, because they don't require frequent reapplication. They have the potential for providing a constant supply of nutrient for relatively low labor costs. The effectiveness of slow-release fertilizers has been demonstrated in small-scale studies of crude oil bioremediation in the field (Lee *et al.*, 1993). The release rate is critical for the success of this approach (Merlin, *et al.*, 1995), however, and the fertilizer must be distributed such that the nutrient is transported throughout the contaminated region following its dissolution.

The most likely target beaches are contaminated mainly in the upper platform region. This region is completely covered by water only during spring high tides. At neap tide, the upper platform may not be covered at all during some of the high tide periods. This extreme variation in the extent of flooding that occurs during the lunar tidal cycle will affect nutrient transport. Water-soluble nutrients that are applied at the surface will be washed out quickly during spring tides, but they can persist in the bioremediation zone for several days following neap tides (Wrenn *et al.*, 1995a). Nutrients applied in a trench above the level of the spring high tide, on the other hand, will not be transported into the bioremediation zone unless the water table is high enough to entrain them. Therefore, nutrient transport from trench

application is likely to be inefficient during neap tides. Slow-release fertilizers that are applied directly above the contaminated sediments will suffer from similar transport limitations, but they are likely to be less severe, because they can also be transported by percolation of rainwater through the beach. We will quantify the effects of variations in the extent of tidal flooding on nutrient transport by applying water-soluble tracers during spring and neap tide periods. For each beach type, the water-soluble tracers will be applied at two spring and two neap tides to obtain better estimates of the variability in transport rates.

A randomized complete-block experimental design will be used to evaluate the three nutrient application methods, and a conservative tracer (lithium chloride) will be used to investigate the nutrient transport mechanisms. At least two blocks will be established on each beach type. Each block will contain five plots, which will be positioned near the middle of the upper platform at a position that is representative of the average exposure conditions for the upper platform. Two plots will be used for each of the water-soluble nutrient treatments (i.e., surface application and trench application), and one plot will be used to evaluate the slow-release fertilizer. For the water-soluble tracers, one of the two plots will be used for a spring-tide application and the other for application during neap tide. Thus, the five plots in each block will receive the following treatments: surface application of a water-soluble tracer at spring tide, surface application of a water-soluble tracer at neap tide, trench application of a water-soluble tracer at spring tide, trench application of a water-soluble tracer at neap tide, slow-release tracer applied at the first spring tide only.

The surface application and slow-release methods both involve addition of nutrient directly onto the contaminated area. The water-soluble and slow-release tracers will be broadcast over the surface of the plot, then watered in with 200 gallons of seawater. These plots will be wetted only by the rising tide and wave action for the duration of the experiments. In the trench application method, solid tracer will be applied in a trench located above the high water line. The trench will be deep enough to reach the level of the spring high tide water table. Tracer is transported through the contaminated zone by groundwater flow.

Because the application and nutrient transport mechanisms are fundamentally different, two types of plots will be used. The surface application and slow-release fertilizer plots will be 5 m by 10 m, located near the middle of the upper platform. For the trench application, tracer will be applied in trenches that are 5 m wide, and transects will be established through the upper platform. For positioning purposes, these transects will be considered to be 5 m wide. The plots and transects will be separated from adjacent plots by 10 m. Thus, a 55 m stretch of relatively uniform beach is required for each block.

The surface application and slow-release fertilizer plots will be divided into three subsections (α , β , γ) that measure 5 m by 3.3 m. Multi-port sampling wells will be installed in the center of each subsection. These wells are used to collect water samples from discrete depths within the beach. The screened sample ports will be placed at 12 inch intervals, and the wells will be installed such that samples can be collected from above, below, and within the oiled layer. If water samples cannot be obtained from within the bioremediation zone during neap tides, when large portions of the experimental plots will remain exposed at high tide, samples of the oiled sediment will be collected to monitor tracer concentration.

The trench application transects will use additional multi-port sampling wells for a more thorough characterization of the transport of nutrients added by this method. Monitoring wells for the trench plots will be placed at 5 m intervals along a transect through the upper platform. Five wells will be installed per transect. During neap tides, when water samples cannot be collected from the contaminated zone, samples of oiled sediment will be collected to monitor tracer concentration. The sediment samples will be collected from randomly selected positions in the vicinity of each of the monitoring wells.

Lithium chloride will be used as a conservative tracer for the hydrodynamic characterization. Water and sediment samples will be shipped to Cincinnati for analysis of the lithium concentration by atomic absorption spectrophotometry. We will use flame photometry for field monitoring of the lithium concentrations in these samples. The field measurements will be used to determine the progress of tracer movement so that sample collection intervals can be adjusted if necessary and the reapplication schedule can be optimized.

In addition to the samples that will be collected for chemical analysis of the tracer concentration, other measurements will be made to characterize the beach hydrodynamics. These will include continuous monitoring of water level in the beach along a transect through the upper platform and continuous measurements of salinity and dissolved oxygen (DO) in the oiled layer at two representative positions near the middle of the upper platform. The water-level measurements will be used to characterize fluctuations in the hydraulic gradients that drive nutrient transport in the beach subsurface. Water level will be recorded using piezometers that consist of one inch iron pipes containing pressure transducers. The pipes are screened at the bottom to allow the water level to respond quickly to changes in the water level in the beach. The pressure transducers will be connected to a data logger. The salinity measurements will be used to monitor the size and position of any fresh water lenses that exist at the sites, as well as the rate and extent of mixing between freshwater and seawater that occurs as a result of tidal fluctuations. The DO measurements will determine whether bioremediation can be limited by the oxygen transport rate into the contaminated zone. Salinity and DO will be measured using sensor packs installed at the bottom of screened wells. These sensor packs will have the capacity to store salinity and DO data for two weeks.

4. Bioremediation Technology Evaluation

Effective restoration of oil-contaminated beaches requires maintenance of nutrient concentrations that are high enough to support maximal growth rates of hydrocarbon-degrading bacteria. The optimal nitrate concentration for alkane degraders appears to be between 0.5 and 2.5 mg N/L (Venosa *et al.*, 1994a). A recent bioremediation field study conducted on a beach on Delaware Bay demonstrated that the background nitrate concentration, 0.8 mg N/L, was sufficient to maintain reasonably high biodegradation rates, but they could be increased further by daily application of water-soluble nutrients (Venosa *et al.*, 1995). These guidelines and the results of the nutrient transport studies that will be conducted in the first year will be used to determine appropriate nutrient application methods and schedules for reapplication.

A randomized complete block design with repeated measures will be used to evaluate

treatment effects. In this design, blocks consisting of several plots that receive different treatments are established on a relatively homogeneous stretch of beach. Each block contains all of the treatments that will be evaluated including a no-treatment control. The blocks are replicated and the relative positions of the treated plots are chosen randomly for each replicate block.

Geomorphological and hydrodynamic characteristics will be used to segregate the target beaches into several categories. At least three blocks will be established for each type of beach, and two or three types of beach will be studied. The blocks will be situated in a row on the beach parallel to the shoreline. Each block will contain three plots: two treatments and a no-treatment control. One of the treatments will probably involve application of a water-soluble fertilizer (e.g., sodium nitrate and sodium tripolyphosphate). The other treatment will probably involve use of a slow-release fertilizer.

The plots will be 5 m wide by 10 m long. The minimum distance between plots, and between blocks, will be 10 m. The plots will be positioned at the same elevation so that they all experience the same extents of submersion and exposure.

Nutrient will be applied by broadcasting solid fertilizer on the surface of the plots. Small gasoline-powered pumps will then be used to apply approximately 200 gallons of seawater to the plots treated with water-soluble fertilizer. Water-soluble fertilizer will be applied frequently, probably every day or two. Slow-release fertilizer will be applied approximately once every three weeks.

Samples of oiled sediment will be collected approximately every 14 days from all of the plots. Each plot will be divided into three equal sectors (labeled α , β , and γ , landward to seaward, respectively). Subsamples will be collected from each sector according to a predetermined randomized sequence that will disallow subsequent resampling from the same hole. The three subsamples will be composited into one, mixed and divided into a series of subsamples. The subsamples will be analyzed for residual oil by GC-MS, hydrocarbon degraders by MPN, microbial activity, and toxicity. An additional sample will be collected and stored frozen for archival purposes.

Samples collected for microbiological analysis will be cooled immediately on ice and transported to a field laboratory for processing. Alkane- and aromatic-hydrocarbon-degrading bacteria will be enumerated by MPN using a 96-well microtiter plate method (Wrenn *et al.*, 1995b). Heterotrophic microbial activity will be estimated using ^3H -thymidine incorporation and ^{14}C -glutamic acid mineralization (Lee *et al.*, 1995). Hydrocarbon biodegradation activity will be estimated using mineralization of ^{14}C -hexadecane and ^{14}C -phenanthrene (Roubal and Atlas, 1978; Brown *et al.*, 1991).

Residual oil will be measured by extraction of 100 to 500 g of sand with dichloromethane (DCM). The DCM extracts will be dried with anhydrous sodium sulfate, and the residual TPH will be determined gravimetrically. An aliquot of the DCM extract will be analyzed by GC-MS following solvent exchange into hexane. A Hewlett-Packard 5890 gas chromatograph equipped with an HP 5971 mass selective detector will be used to quantify the aliphatic and aromatic components of the residual oil. All analyte data will be normalized to the conservative, nonbiodegradable biomarker hopane [C_{30} 17 α (H),21 β (H)-hopane] (Peters and

Moldowan, 1993).

The variations in reported oil concentrations that are due to compositional differences in the beaches undergoing bioremediation will be minimized by reporting the oil concentration as mass of oil per unit surface area, rather than mass of oil per unit mass of beach material. This reporting convention requires analysis of the particle size distribution and surface area of the DCM-extracted beach material. The particle size distribution will be determined by separating the samples into twelve size fractions, ranging from greater than 75 mm (i.e., retained in a sieve with screen mesh of 75 mm) to less than 0.05 mm (i.e., passing through a screen with mesh size 0.05 mm). The average mesh diameter of two consecutive sieves is used to define the particle size for each size fraction. (For example, the average diameter of particles retained on the 2 mm sieve is 6 mm, because the next higher sieve has a mesh size of 10 mm.) The specific surface area, S (cm^2/g), for a given size fraction is (Patton, 1965):

$$S = \frac{6}{d\rho\phi}$$

where d is the average particle diameter (cm), ρ is the particle density (2.65 g/cm^3), and ϕ is the sphericity (dimensionless). The sphericity is a shape factor that corrects the surface area calculation for non-spherical particles. Sphericities will be estimated by direct microscopic examination using guidelines described in the Geomorphological Field Manual (Deckombe and Gardiner, 1983). The total surface area for a given sample is obtained by multiplying the specific surface area for each size class by the fraction of the total mass that is comprised by that size class, then summing over the entire particle size distribution.

The interstitial pore water nitrate concentrations will be measured in treated and control plots at regular intervals to assure that our nutrient concentration goal is maintained. Nitrate will be measured by the cadmium reduction autoanalyzer method (Technicon Industrial Systems, 1972).

Salinity, pH, and dissolved oxygen will be measured in interstitial water collected from each plot. These measurements will be made in the field immediately after the water samples have been collected. Portable field meters and probes will be used for each of these analyses. The accuracy of these measurements will be assured by performing appropriate calibrations in the field and by periodic recalibration of the portable meters in the laboratory.

Since response variables (MPN, oil analytes, and estimates of toxicity) will be measured repeatedly in treated and control plots at regular intervals during the course of the investigation, repeated measures analysis of variance (RMANOVA) (Freund *et al.*, 1976) will be used to analyze the performance data. When the repeated measures ANOVA indicates significant differences, univariate ANOVAs will be run on data at each time point. Where significant differences are indicated at a specific time point, protected least significant difference mean separations will be used to assess treatment differences. Regression analysis will be used to estimate the rate of oil degradation for each of the three treatments. These rates will be compared statistically. Statistically valid variability estimates will be obtained that will aid the design of future oil bioremediation studies.

The above statistical treatment of the data assumes that all samples are independent and have the same variability properties. However, samples closer to each other in space often are more autocorrelated (have less variability) than those separated by wider distances. If such spatial autocorrelation is confirmed, the nonindependence assumption becomes invalid, and thus the ANOVA results will be questionable. Powerful statistical techniques have recently been developed to address the effect of spatial variability (Cressie, 1994). This spatial data analysis will be used to model variability relative to the distance between plots. If necessary, new ANOVAs will be conducted on the data that has been corrected for spatial autocorrelation.

5. Bioremediation of Selected Sites in Prince William Sound, Alaska:

During the third year of this project, restoration of several oil-contaminated beaches in Prince William Sound by bioremediation will be initiated. This will involve large-scale application of fertilizer to selected beaches and a monitoring program to ensure that bioremediation is proceeding effectively and efficiently. The nutrient application strategy that will be used for this part of the project will be determined based on the results of the bioremediation technology evaluation that will be conducted during the second summer. The most appropriate technology will be selected after consideration of effectiveness and economics. Effectiveness will be characterized by chemical and toxicological endpoints. The chemical effectiveness will be determined by the extent to which known hazardous components of crude oil (e.g., PAHs) are reduced in the treated plots. The toxicological effectiveness will be based on reduction in the toxicity of sediments and beach pore water. The primary economic considerations will be the costs of the fertilizer and any equipment required to apply the fertilizer and the amount of labor involved. Labor costs are likely to be the most important economic consideration. Whenever possible, the workforce will be recruited from the Chenega Bay community so that local residents can learn to apply bioremediation.

The monitoring program, which will apply methods that were developed and tested in the first two years of this project, will have two major components. The first will support the engineering requirements of the bioremediation, and its primary focus will be to ensure that target concentrations of nutrients and oxygen are maintained in contact with contaminated beach material. The second component will document that beach restoration is proceeding at an accelerated rate without detrimental environmental impacts. The latter is a particularly important aspect of this phase of the project. Fortunately, any detrimental effects that occur are likely to be local and temporary, and the impact of these occurrences can be mitigated by imposing appropriate engineering or administrative controls.

Although the primary goal of the monitoring program is to ensure the safety and effectiveness of the beach restoration operations, this also presents an opportunity to evaluate the monitoring tools that were developed during the first two years of this project. If bioremediation becomes a widely accepted method for restoring oil-contaminated shorelines, monitoring tools that can accurately evaluate its performance must be available. Ideal tools

will be simple, rapid, and provide reliable estimates of the risk associated with remaining contaminants. Therefore, a secondary objective of the third year monitoring program will be to determine the most appropriate suite of measurements for routine assessment of the level of risk associated with residual oil and the restoration process. To accomplish this goal, simple, indirect measurements, such as visual inspection of the contaminated beach and chemical analysis of the remaining oil, will be correlated with more direct -- but more costly and difficult -- measurements of risk, such as toxicity of contaminated sediments and bioaccumulation of PAHs by shellfish.

The number and location of sites that will be restored during this phase of the project will be determined following the second year's technology demonstration. The most important factor determining the number of sites that can be restored during this phase of the project is the nutrient application technology that is selected. If a labor-intensive strategy is determined to be most appropriate, fewer beaches can be treated than if a strategy with minimal labor requirements can be used. The beaches that are selected for treatment during the third summer will be chosen based on an evaluation of where bioremediation is likely to have the most benefit. This decision will be made following consultation with the Chief Scientist with a joint recommendation presented to the EVOS Trustee Council. A wide range of potential sites will be considered, including many that will be characterized during the site survey that is to be conducted in the first summer. We will consider several factors in the site selection process, including the risk posed to humans and local ecosystems, the biodegradability of the oil, and any environmental characteristics that can affect the success of the bioremediation effort.

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C. Contracts and Other Agency Assistance

ADEC is the lead agency for this project. The project involves an international collection of government agencies, universities, contractors, and paid specialist all of which are recognized experts in one or more of the following the areas: enhanced bioremediation of spilled oils, risk reduction engineering technologies, environmental assessment, toxicology, shoreline geomorphology and hydromorphology, research laboratories, and oil spills.

Government Agencies: ADEC, NOAA, NMFS Auke Bay Lab, U.S. EPA Risk Reduction Engineering Cincinnati Ohio, Environment Canada Fisheries and Oceans Quebec Canada, EPA Gulf Breeze Florida Marine Research Laboratory.

Universities: Alaska University of Fairbanks School of Fisheries and Ocean Sciences, University of Cincinnati Department of Civil and Environmental Engineering, Institute for Environmental Sciences University of Louisiana

Contractors: Research Planing, Inc., Environmental Technologies & Solutions, Inc.

D. Location

Western portion of Prince William Sound. Based on present knowledge, we are considering several sites for survey in 1996. First, we will investigate sites near the Chenega Bay community. The Invitation to Submit Restoration Projects for FY 96 (p. 72) states that: "There are approximately twelve 'low energy' sites near the community of Chenega Bay which have experienced little reduction in subsurface oil. The worst sites still sheen." Additional potential sites include EL056C(head of the east arm of Northwest Bay), EV036A (Northeast Evans Island), GR101B (North side of a detached island north of Green Island), KN136A (Bay of Death in Bay of Isles), KN213C (Rua Cove), LA015C and LA015E (Northeast coast LaTouche Island), LA 020C (West shoreline of Sleepy Bay), EV037A and EV039A (Northeast Evans Island, and ER020B (North end Elrington Island), NOAA station N-3 at the northwest end of Smith Island and NOAA station N-10 at the entrance to Herring Bay. Finally, the Bay of Isles may have a heavily oiled marsh in need of restoration.

Project benefits will be realized at the above shorelines. Communities located in the Prince William Sound will all benefit to some degree. ADEC will ask for the approval of residents of Chenega Village to locate its operations in their community.

SCHEDULE

A. Measurable Project Tasks for FY 96

Start-up October 1, 1995	Confirm draft schedule for data reviews and field season, contract boats, et. al, Begin review of existing shoreline oil data located in EVOS Office, Anchorage Alaska
November, 1995	Meet with residents of Valdez and Chenega to present project, discuss location of shoreline hot spots, site characterization, assessment, and next season's work schedule
April, 1996	Confirm list of sites selected for field visits this coming summer
May thru Sept.15, 1996	Fully characterized selected shorelines to geomorphology, hydrodynamic make-up, toxicity of remaining oil, status of recovery, and the sites responsiveness to enhanced bioremediation.
Late FY 96 or early FY 97	Present benefits of using of non-intrusive, non-commercial techniques for accelerating biodegradation of antagonistic oil to Chief Scientist.
December 1996	Samples analyzed for chemical composition and toxicity

B. Project Milestones and Endpoints

1. Beaches that are candidates for restoration by bioremediation will be characterized with respect to geomorphology, hydrodynamics (e.g., exposure to wave activity and subsurface freshwater flow), the extent and location of oil contamination, the toxicity of oiled sediments and beach pore water, and the ecological impact of the oil contamination. Field work will be completed by September 15, 1996 and samples will be analyzed for chemical composition and toxicity by December 15, 1996. The assessment of ecological impact will be completed by December 15, 1996. A data base that can be used to rank oil-contaminated beaches with respect to relative risk and potential for restoration using bioremediation will be compiled in the final report for FY 96.
2. Nutrient transport rates will be measured using a conservative tracer, and the results will be used to design effective fertilizer application strategies to be conducted in FY 97. The field work will be completed by Sept. 15, 1996, sample analysis will be completed by Dec. 1, 1996, and the fertilizer application strategies will be selected by Jan. 15, 1997.
3. Two or three beaches best suited to demonstrate the effectiveness of bioremediation for restoration of oil-contaminated beaches in Prince William Sound will be recommended to the trustee council, following concurrence with the council's chief scientist, by April 30, 1997. The most effective and economical strategies will be tested by applying bioremediation on these beaches. The field work will be completed by Sept. 15, 1997. Analysis of oiled sediment and beach pore water for toxicity will be completed by Dec. 15, 1997, and chemical analysis of residual oil will be completed by Jan. 15, 1998. A

summary of the ecological monitoring program will be compiled by Dec. 15, 1997. Statistical analysis to demonstrate the existence of treatment effects will be completed by March 1, 1998.

4. A bioremediation strategy will be chosen for the restoration of several oil-contaminated beaches by April 30, 1998. Several beaches may be identified by the *Exxon Valdez* trustee council for restoration during the summer of 1997. Beaches targeted for bioremediation during FY 97 will be chosen by April 30, 1997. The beaches selected for restoration will be chosen from among those that are most likely to benefit from bioremediation. The beaches will be selected, following consultation with the trustee council's chief scientist, on the basis of their risk to humans and important natural resources. Bioremediation of selected beaches in Prince William Sound will be initiated by June 15, 1998. If sites are designated for bioremediation during FY 97, the operations will commence by June 15, 1997. To the extent possible, local inhabitants will be employed and trained to apply bioremediation so that the cleanup can continue after this project has been completed.

ENVIRONMENTAL COMPLIANCE

A number of applicable permits and regulations must be followed. These include applicable portions of Alaska Statute 46.03., Federal Clean Water Act, the Oil Spill Pollution Act of 1990, and perhaps NEPA. It is not known at this time which federal agency would conduct the NEPA review if its required.

Applicable state regulations include Alaska Water Quality Standards, Wastewater Disposal, and Oil Pollution Regulations.

PERSONNEL

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Juneau, Alaska 99801

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Consultant Environmental Engineer

for the

State of Alaska Department of Environmental Conservation

AREAS OF EXPERTISE

* Technical assistance to Local Communities and Industry on Development of Cost-Effective, Environmentally Sound Development Options

- * U.S. Environmental Protection Agency Policies and Procedures
- * Identification of Efficient, Cost Effective, and Environmentally Safe Oil Spill Clean-Up Methods
- * Directing Environmental Research and Related Field Surveys
- * Extensive Knowledge of Concepts, Practices, and Principals of Environmental Engineering Including Methods, Permitting, and Equipment
- * U.S. Oil Pollution Control Act, Clean Water Act, Clean Air Act, Safe Drinking Water Act, and Other Major Environmental Legislation
- * Design and Construction of Industrial and Domestic Wastewater, Oil Pollution, Drinking Water, and Air Pollution Control Systems
- * Regulatory Programs and Requirements Affecting:
 - Water Quality Standards
 - Wastewater Disposal
 - Cleanup of Oil and Hazardous Substance Spills
 - Air Quality Control
 - Land and Urban Development
 - Timber Harvesting and Processing
 - Seafood Processing
 - Petrochemical and Petroleum Industries
- * Industrial Development Policy for the State of Alaska
- * Broad Professional Network of State of Alaska Local Governments
 - More than 10 years of Working Directly with Local Governments, City Councils, and State Officials
- * Registered as a Professional Engineer in the State of Alaska and the State of Wisconsin

RELEVANT ACCOMPLISHMENTS

November 1992

At the invitation of the U.S. Environmental Protection Agency was a member of a team of United States scientists at the U.S./Israel Bioremediation Workshop held in Israel. The purpose of the workshop was to share technical information on use of bioremediation at oil spill and hazardous waste sites and to establish a working relationship with fellow research scientists in the field of bioremediation.

September 1992

Preparation of the "Field Bioremediation Study of Spilled Crude Oil on a Sheltered Cobble Beach in Prince William Sound"; Venosa, A.D., Viteri, A., Office of Research and Development, U.S.EPA Risk Reduction Engineering Laboratory, Cincinnati, Ohio 45268, September 1992. The intent of this study was to verify use of the normalized hopane ratio method as an accurate indicator of oil bioremediation; identify a cost effective method for remediation of subsurface shorelines, and complete TIER IV: Field Verification for Bioremediation at Cobble Beach Shorelines of the National Oil Spill Bioremediation Testing Protocol Methods Manual.

June 1992

At the invitation of the U.S. Environmental Protection Agency and the National Environmental Technology Application Corporation, proposed standardized methods for field verification of open ocean spill bioremediation products to a panel of national experts. The panel is currently evaluating this proposal for adoption into the National Oil Spill Bioremediation Testing Protocol Methods Manual and field study.

April 1991

At the invitation of the U.S. Environmental Protection Agency and the Treatability Protocol Development Subcommittee, drafted the National Oil Spill Bioremediation Testing Protocol Methods Manual used for identifying the worth of different types of bioremediation oil spill clean-up products.

November 1990

Received Letter of Commendation from the State of Alaska Department of Environmental Conservation for response work during the T/V EXXON Valdez oil spill.

October 1990

Completed the "STATE OF ALASKA PROTOCOL For Approving Chemical Products and Technical Clean Up Methods For Shoreline Clean Up of the T/V EXXON Valdez Oil Spill". This first of its kind oil spill protocol established risk assessment criteria for determining the net environmental gain of proposed oil spill clean up methods that have higher effectiveness and lower costs.

September 1990

At the request of the U.S. Environmental Protection Agency's Office of Research and Development selected to serve as a member of the national Advanced Oil Spill Bioremediation Products Protocol Development Panel. The purpose of the Panel is to evaluate and refine methods for selecting and field testing effective and environmentally safe oil spill bioremediation enhancement products.

June 1990

Completed a comprehensive summary report on the process used by the State of Alaska Oil Spill Response Center's Treatment Technology and Research and Development Sections to approve bioremediation enhancement products for clean up of the T/V EXXON Valdez Oil Spill.

September 1989 to April 1990

Manager of the State of Alaska T/V EXXON Valdez Oil Spill Response Center Treatment Technology Section. Developed the Treatment Technology Section's goals, related objectives, work tasks, and staffing and budget needs. Established and achieved the section's goal; to find efficient, cost effective, and environmentally safe oil spill clean up methods for the 1990 clean up season.

June 1989 to September 1989

Manager of the State of Alaska T/V EXXON Valdez Oil Spill Response Center Research and Development Section. Responsibilities included developing the State of Alaska approval criteria for use of proposed chemical and bioremediation enhancement oil spill clean up products and presented the scientific reasons for accepting or rejecting their use to the Federal On Scene Coordinator and Alaska Regional Response Team (RRT). Recommended modifications to mechanical clean up equipment to maintain cleanup efficiency of weathering oil.

- * Assisted the U.S. Environmental Protection Agency with field and toxicity testing of various bioremediation products and determining safe application rates. This included identifying and reviewing needed efficacy and toxicity data.
- * Developed efficacy and toxicity approval criteria for use of COREXIT 9580M2, a chemical shoreline cleaner, and
- * Supervised the state Oil Spill Response Center monitoring of a proposed chemical shoreline cleaner's efficacy and offered recommendations on its wide scale use to the Federal On Scene Coordinator's Research and Development Committee. This was the first time that a chemical cleaner was tested at an shoreline oil spill.

February 1979

In recognition of noteworthy contribution and outstanding service to the State of Alaska, received an **EXCEPTION SERVICE AWARD** for developing a comprehensive revision to the State of Alaska Water Quality Standards. Among other things, this revision developed water quality standards for marine waters and established criteria for petroleum hydrocarbons; oils and grease and other deleterious organic and inorganic substances; and, total residual chlorine.

February 1985

In recognition of noteworthy contribution and outstanding service to the State of Alaska, received an **EXCEPTIONAL SERVICE AWARD** for distinguished work in support of the state's effort to maintain environmental quality of the timber and pulp mill industries simultaneously with the economic health of the Communities of Ketchikan and Sitka.

OTHER ACCOMPLISHMENTS

- * Appointed lead State coordinator responsible for representing the interests of the Governor's Office and the Departments of Environmental Conservation, Fish and Game, Natural Resources, Commerce and Economic Development, and other state agencies. Responsibilities included directing and advising the State of Alaska on how to resolve conflicts between State of Alaska laws and the issuance of federal air and water quality permits to Southeast Alaska's two Pulp mills.
- * State of Alaska Project Manager of the first joint state, U.S. Environmental Protection Agency

multi-program inspection of a privately owned, complex industrial facility, in this case a magnesium bi-sulfite pulp mill. Developed the inspection's objective, to determine how the total amount of pollution being emitted by the pulp mill can be reduced without negatively impacting the operation of the pulp mill or individual pollution programs. Pollution programs involved in this 10 day inspection included: water pollution control, drinking water, air pollution control, oil spill prevention and spill control, leaking underground storage tanks, pesticides, Toxic Substance Control Act (TSCA), and Resource Conservation and Recovery Act (RCRA).

- * Called upon by the Governor to investigate serious public health hazards caused by failure of the City of Haines sewer system. Successfully lobbied 1.6 million dollars from the legislature and supervised repair of the city's sewer system.

- * Lead development of the department's Water Pollution Control Program.

- * By Appointment of the Governor of Alaska was the department's scientific and technical advisor on the North Pacific Rim's SEA USE COUNCIL.

- * Lead negotiator of a successful two year \$250 million dollar contract between the State of Alaska and 9,000 employees.

- * Project manager for numerous water quality surveys to determined the cause and amount of pollution in marine waters.

- * At the request of the Governor researched and wrote "Alaska Department of Environmental Conservation Comments on U.S. Environmental Protection Agency's Regulations on Canned and Preserved Seafood Processing Point Source Category", a full scale study on the economic impact of the proposed regulation on the state's seafood industry and comparative gains in environmental benefits. This paper was presented by the Governor to the Alaska Congressional delegation, U.S.EPA, and Alaska seafood industry representatives in Washington D.C.

- * At the request of the State of Alaska Legislature, reviewed and offered recommendations on the propriety of the department's On-Site Certification Program and offered follow up recommendations which have been adopted

- * Project Manager on the analysis of the state's administration of the Clean Water Act's National Pollution Discharge Elimination System program and offered recommendations.

PUBLICATIONS:

"Use of Bioremediation in Response to the T/V EXXON Valdez Oil Spill and Follow Up Research Efforts", Viteri, A., State of Alaska Department of Environmental Conservation, 410 Willoughby Avenue, Suite 105, Juneau, Alaska 99801-1794.

"Lessons and Future Needs: The T/V EXXON Valdez Oil Spill Response", Viteri, A., Piper, E., March 1992, State of Alaska Department of Environmental Conservation, EXXON Valdez Oil Spill Response Center, 410 Willoughby Avenue, Suite 105, Juneau, Alaska 99801-1794.

"Development Of Bioremediation Program In Response To The T/V EXXON Valdez Oil Spill, Prince William Sound, Alaska 1989-91" Viteri A., Piper E., June 1992, State of Alaska Department of Environmental Conservation, EXXON Valdez Oil Spill Response Center, 410 West Willoughby Avenue, Suite 105, Juneau, Alaska 99801-1795

"Color and Turbidity in Sawmill Cove and Adjoining Waters of Silver Bay, Near Sitka Alaska, on November 6-8 and December 8-9 1990", Viteri, A., September 1991, State of Alaska Department of Environmental Conservation, P.O. Box 0, Juneau Alaska, 99811

"State of Alaska Protocol For Approving Chemical Products and Technical Clean Up Methods

Proposed For Shoreline Clean Up Of The T/V EXXON Valdez Oil Spill" Viteri, A., Clark, J., pending publication.

"State of Alaska Approval Criteria For Use of COREXIT 9580M2 For Shoreline Clean Up of The T/V EXXON VALDEZ OIL SPILL", Viteri, A., July, 1989, State of Alaska T/V EXXON Valdez Oil Spill Response Center, Drawer 3149, Valdez, Alaska, 99686, pending publication.

"Ward Cove Water Quality Analysis", Kruze, A., Viteri, A., February 1988, State of Alaska Department of Environmental Conservation, Southeast Regional Office, 410 Willoughby Avenue, Suite 105, Juneau, Alaska 99801-1794.

"Petrochemical Development and Water And Air Quality in Alaska" September, 1981, State of Alaska, Department of Environmental Conservation, P.O. Box O, Juneau, Alaska, 99811.

"United State Environmental Protection Agency's Development of Effluent Guidelines for Alaska Seafood Processing Wastes", Viteri, A., June 1981, Department of Environmental Conservation, P.O. Box O, Juneau, Alaska, 99811.

"Investigation of the State of Alaska Department of Environmental Conservation On-Site Program and Suggested Recommendations", Viteri, A., September 1980, State of Alaska Department of Environmental Conservation, P.O. Box O, Juneau Alaska 99811.

"Tunnel and Reservoir Plan for Chicagoland Flood and Pollution Control", Viteri, A., 1973, Metropolitan Sanitary District Of Greater Chicagoland, presented at the third annual Japanese-USA Sewage Technology Conference, Tokyo Japan.

PROFESSIONAL HISTORY

April 1992 to Present

Manager, Domestic Wastewater Treatment Program. Statewide consultant in environmental engineering work concerning domestic wastewater treatment facilities and systems for controlling pollution and protecting the environment. Management and operation of the department's Domestic Wastewater Treatment Program and assist in preparing and tracking program budget reports. Conducts research before developing State regulations, guidelines, standards, policies and procedures pertaining to domestic wastewater facilities and systems. Identify minimum engineering design standards for wastewater disposal systems, subdivision plan review, and onsite sewage disposal systems, and establish them in State of Alaska Regulations.

In cooperation with the State of Alaska Board of Architects, Professional Engineers, and Land Surveys, Professional Engineering Community, and the public develop a State of Alaska engineering design manual for design, construction, and rehabilitation of onsite sewage disposal systems. Develop a program approach for privatizing the state's onsite sewage disposal bank loan certification program to Alaska Professional Engineers. Assist the Department to evaluation evidence and documents relating to alleged pollution incidents or environmental damage. Conduct or participate in hearings on legislation, and testify as an expert witness in court proceedings involving environmental matters.

June 1990 to April 1992

Senior level Environmental Engineer for the State of Alaska Department of Environmental Conservation. Duties include supervise State of Alaska involvement in a joint federal/state environmental evaluation of pulp mill pollutants in and near community of Sitka Alaska. Develop and lead reconnaissance surveys in and around the community of Sitka Alaska to identify and evaluate environmental health and public health risks. Participate as a member of the national Advanced Oil Spill Bioremediation Products Protocol Development Panel to evaluate and refine

methods for selecting and field testing effective, environmentally safe oil spill bioremediation enhancement products.

September 1989 to June 1990

Manager of the State of Alaska T/V EXXON Valdez Oil Spill Response Center Treatment Technology Section. Developed the Treatment Technology Section's goals, related objectives, work tasks, staffing, and budget. Responsibilities included:

- * Identified and achieved the section's goal: to find efficient, cost effective, and environmentally safe oil spill clean up methods for the 1990 clean up season.
- * Supervised development of procedures used for reviewing more than 1,500 products.
- * Supervised development and use of a procedure for identifying cost effective, efficient **mechanical clean up techniques**.
- * Developed a procedure for reviewing **chemical shoreline cleaners**. This procedure is nationally recognized as the most advanced method developed.
- * Supervised development of procedures used to select **bioremediation enhancement products**. This procedure has been adopted by the EPA

June 1989 to September 1989

Manager of the State of Alaska T/V EXXON Valdez Oil Spill Response Center Research and Development Section. Responsibilities included:

- * Developed State of Alaska approval criteria for use of proposed chemical and bioremediation enhancement oil spill clean up products and presented the scientific reasons for accepting or rejecting their use to the Federal On Scene Coordinator and Alaska Regional Response Team (RRT),
- * Recommended modifications to mechanical clean up equipment to maintain cleanup efficiency of weathering oil,
- * Assisted the U.S. Environmental Protection Agency with field and toxicity testing of various bioremediation products and determining safe application rates. This included identifying and reviewing needed efficacy and toxicity data.
- * Developed efficacy and toxicity approval criteria for use of COREXIT 9580M2, a chemical shoreline cleaner,
- * Supervised the state Oil Spill Response Center field monitoring of a proposed chemical shoreline cleaner's efficacy and offered recommendations on its wide scale use to the Federal On Scene Coordinator's Research and Development Committee. This was the first time an chemical cleaner has been tested at a shoreline oil spill.

May 1989 to June 1989

Work with the State of Alaska Oil Spill Response Center helping EXXON CORPORATION, USA to mobilize, and provide facilities for its 10,000 member clean up crew.

April 1989 to April 1977

ENVIRONMENTAL ENGINEER IV

State of Alaska Department of Environmental Conservation, Juneau Alaska. Responsibilities included:

- * Providing Southeast Alaska communities and industrial representatives with technical assistance

ad recommendations on cost-effective, environmentally sound development options

- * Preparing public hearing testimony for the Governor and the Commissioner of the Department of Environmental Conservation
- * Represent the interests of the State of Alaska during negotiations between industrial representatives and federal regulatory agencies
- * Author of the State of Alaska Water Quality Standards and Wastewater Disposal Regulations
- * Directing and advising the State of Alaska on how to resolve conflicts between the issuance of federal permit effluent limits for Southeast Alaska's Pulp mills and State Law
- * Consulting with Southeast Alaska city managers in developing, supervising, and obtaining Clean Water Act secondary sewage treatment waivers, and assisting in the rehabilitation of failed sewer systems
- * Directing and conducting legal, regulatory, and environmental research
- * Program manager for Southeast Alaska's Air Pollution and Water Pollution Control Programs
- * Providing technical environmental engineering assistance and consultation to the Department of Environmental Conservation regional and district office staff

March 1977 - September 1974

CIVIL ENGINEERING ASSISTANT III, State of Alaska, Department of Public Works. Responsibilities included:

- * Project engineer for major airport expansions and renovations in McGrath, Kodiak, Bethel, Deadhorse, and Fairbanks
- * Author of Operations Manual for the Deadhorse Airport Sewage Treatment Facility;
- * Design and survey engineer for several major airport construction and renovation projects
- * Developing engineering project specifications for design of airport terminals, runways, and lighting systems
- * Estimating construction costs and awarding contracts
- * On-site monitoring during construction phase of projects.

March 1971 - June 1974

ASSOCIATE CIVIL ENGINEER, Metropolitan Sanitary District of Greater Chicagoland.

Responsibilities included:

- * Develop comprehensive, technical program for the capture of combined sewer overflow for the 360 square mile Chicagoland area as part of the Tunnel and Reservoir Plan
- * Oversee major consulting firms contracted to conduct soil core sampling, research, develop rain overflow and capture data, phasing, and cost estimation of the combine sewer overflow project
- * Coordinating public relations and public information activities.

Environmental Technologies and Solutions, Inc.

Kevin L. Strohmeier President, Environmental Scientist

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Education

M.S. Environmental Science; University of Cincinnati, OH; 1994

B.S. Biology; University of Kentucky, Lexington, KY, 1982

Professional Experience

1993 to present	President, Environmental Scientist; Environmental Technologies & Solutions, Inc.
1991 to 1993	Graduate Research Assistant; University of Cincinnati, Department of Civil and Environmental Engineering
1989 to 1991	Assistant General Manager; TLC Garden Center, Cincinnati, OH
1986 to 1989	Business Manager; Still Waters Campground and Marina, Frankfort, KY
1982 to 1986	Graduate Research Assistant; University of Kentucky, Lexington, KY

Relevant Experience

- Provided field expertise, design consulting, and data interpretation for oil bioremediation study
- Developed and operated microcosms for modeling biodegradation of oil on beaches
- Design and implementation of nutrient transport studies in beaches
- Design, construction and operation of nutrient delivery systems for oil bioremediation study
- Knowledge and experience with field environmental sampling techniques
- Successful management experience for complex construction and research projects

Professional Societies

Water Environment Federation

American Chemical Society

Northern Kentucky Chamber of Commerce

Publications and Presentations

- Sih, A., P.H. Crowley, M.A. McPeck, J. Petranka and K.L. Strohmeier. 1985. Predation, competition, and prey communities: a review of field experiments. *Annual Review of Ecology and Systematics* 16:219-261.
- Strohmeier, K.L., P.H. Crowley and D.M. Johnson. 1989. Effects of red-spotted newts (*Notophthalmus viridescens*) on the densities of invertebrates in a permanent fish-free pond: a one-month enclosure experiment. *Journal of Freshwater Ecology* 5:53-65.
- Linton, M.C., P.H. Crowley, J.T. Williams, P.M. Dillon, H. Aral, K.L. Strohmeier and C.T. Wood. 1991. Pit relocation by antlion larvae: a simple model and laboratory test. *Evolutionary Ecology* 5:93-104.
- Venosa, A.D., M. Kadkhodayan, D.W. King, B.A. Wrenn, J.R. Haines, R.T. Herrington, K.L. Strohmeier and M.T. Suidan. 1993. Testing the efficacy of oil spill bioremediation products. *Proceedings of the 1993 International Oil Spill Conference*. American Petroleum Institute, Washington, DC. Pp 487-493.
- Wrenn, B.A., M.T. Suidan, K.L. Strohmeier, B.L. Eberhart and G.J. Wilson. 1995. Nutrient retention in the bioremediation zone of a sandy beach. *Proceedings of the 1995 International Oil Spill Conference*. American Petroleum Institute, Washington, DC.
- Haines, J.R., B.A. Wrenn, E.L. Holder, K.L. Strohmeier and A.D. Venosa. An automated most-probable-number method for estimating oil degrading bacterial populations. Manuscript in preparation.
- Strohmeier, K.L., A.D. Venosa and M.T. Suidan. A microcosm for the study of hydrocarbon biodegradation using respirometric techniques. Manuscript in preparation.
- Strohmeier, K.L. Effects of red-spotted newts on the benthic community structure of a fish-free pond. Presented at the 70th Annual Meeting of the Ecological Society of America, University of Minnesota, Minneapolis, MN, June 17-21, 1985.
- Strohmeier, K.L. Larval feeding rate and numerical response in the damselfly *Ischnura verticalis*. Presented to the University of Kentucky School of Biological Sciences, Lexington, KY, April 17, 1986.
- Strohmeier, K.L., A.D. Venosa, M.T. Suidan and J.R. Haines. A beach microcosm for the study of oil biodegradation. Presented at the Battelle In Situ and On-Site Bioreclamation Symposium, San Diego, CA, April 5-8, 1993.
- Strohmeier, K.L., A.D. Venosa, M.T. Suidan and J.R. Haines. A beach microcosm for the study of oil biodegradation. Presented at the 19th Annual Risk Reduction Engineering Laboratory Symposium, Cincinnati, OH, April 12-14, 1993.

Brian A. Wrenn

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Environmental Technologies & Solutions, Inc.
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Education

Ph.D. Environmental Science; University of Illinois at Urbana-Champaign; 1991

M.S. Biological Oceanography; University of Miami, Coral Gables, FL; 1984

B.S. Biochemistry/Chemistry; University of Illinois at Urbana-Champaign; 1980

Professional Experience

1995 to present	Environmental Scientist, Environmental Technologies & Solutions, Inc.
1992 to present	Post-Doctoral Research Associate; University of Cincinnati
1993	Instructor, University of Cincinnati
1986 to 1991	Graduate Research Assistant; University of Illinois at Urbana-Champaign
1987	Teaching Assistant; University of Illinois at Urbana-Champaign
1984 to 1986	Scientist; E. I. DuPont de Nemours and Co., Inc., Wilmington, DE
1980 to 1984	Laboratory Technician; National Oceanic and Atmospheric Administration, Miami, FL

Relevant Experience

- Design and implementation of petroleum bioremediation and nutrient transport field studies
- Conducted research on aerobic and anaerobic biodegradation
- Knowledge of microbial physiology and the biochemistry of biodegradation
- Design and operation of biological reactors and analysis of reactor kinetics
- Measurement of biodegradation kinetic parameters
- Modeling of substrate interactions during biodegradation in multi-substrate systems
- Measurement of microbial numbers and activity

Professional Societies

- American Society for Microbiology
- American Chemical Society
- International Association on Water Quality

Publications

- Wrenn, B.A., J.R. Haines, A.D. Venosa, M. Kadkhodayan, and M.T. Suidan. 1994. Effects of nitrogen source on crude oil biodegradation. *J. Industrial Microbiology* **13**: 279-286.
- Wrenn, B.A. and B.E. Rittmann. In press. A mathematical model for the effects of primary substrates on reductive dehalogenation kinetics. Biodegradation.
- Wrenn, B.A. and B.E. Rittmann. In press. Experimental evaluation of a mathematical model for the effects of primary substrates on reductive dehalogenation kinetics. Biodegradation.
- Wrenn, B.A., M.T. Suidan, K.L. Strohmeier, B.L. Eberhardt, and G.J. Wilson. Nutrient retention in the bioremediation zone of a sandy beach. *Proceedings, 1995 International Oil Spill Conference*, American Petroleum Institute, Washington D.C., pp. 896-897.

- Wrenn, B.A., K.L. Strohmeier, J.R. Haines, and A.D. Venosa. Selective enumeration of aromatic and aliphatic hydrocarbon degrading bacteria by MPN. To be presented at the 95th General Meeting of the American Society for Microbiology, May 21-25. Washington, D.C.
- Venosa, A.D., M. Kadkhodayan, D.W. King, B.A. Wrenn, J.R. Haines, T. Herrington, K. Strohmeier, and M.T. Suidan. 1993. Testing the efficacy of oil spill bioremediation products. Proceedings of the 1993 Oil Spill Conference, pp. 487-493.
- Haines, J.R., B.A. Wrenn, Edith L. Holder, K.L. Strohmeier, and A.D. Venosa. An automated most-probable-number method for estimating oil degrading bacterial populations. In preparation.
- Rittmann, B.E., E. Seagren, B.A. Wrenn, A.J. Valocchi, C. Ray, and L. Raskin. 1992. In Situ Bioremediation, Second Edition. Noyes Publications (Park Ridge, NJ).
- Rittmann, B.E. and B.A. Wrenn. 1992. Kinetics of Reductive Dechlorination of Trichloroethane (TCA) by Anaerobic Biofilms. Illinois Hazardous Waste Research and Information Center (Champaign, IL).

2. US Environmental Protection Agency, RREL, Cincinnati

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EDUCATION

Ph.D., Soil Science, Cornell University; minors in Microbiology and Environmental Quality, 1979
M.S., Soil Science, Cornell University, minor in microbiology, 1974
B.S., Soil Microbiology, Ohio State University, 1972

PROFESSIONAL EXPERIENCE

1980 to present Microbiologist, U.S. EPA, Cincinnati
Participated in a field test of bioremediation procedures in a controlled oil spill in Delaware, 1994. Project officer for study designed to characterize bacterial population of a trichloroethylene degrading biofilter, 1994. Developed and evaluated test procedures to measure utility of commercial bioremediation products. Participated in Alaskan Bioremediation Research Team following the Exxon Valdez Oil Spill,

1989-1990. Developed new methods for determining safety of public drinking water supplies; national certification officer for laboratory certification of drinking water laboratories; project officer for external grants; coordinated laboratory activities, ordered supplies and equipment

1978 to 1980 Postdoctoral research associate, University of Louisville, Louisville, Kentucky. Research on biodegradation of crude oil in Arctic marine ecosystems and denitrification activities of microorganisms in Alaskan coastal sediments. Participated in shipboard sampling cruises and landbased operations. Used and maintained capillary gas chromatograph and gas chromatograph-mass spectrometer. Coordinated technicians and ordered supplies and equipment.

PUBLICATIONS:

1. Wrenn, B.A., J.R. Haines, A.D. Venosa, M. Kadkhodayan, and M.T. Suidan. Effects of Nitrogen Source on Crude Oil Biodegradation. *J. Ind. Microbiol.* **1994**, 13, 279-286.
2. Haines, J.R., T.C. Covert, and C.C. Rankin. Evaluation of Indoxyl-b-D-Glucuronide as a Chromogen in Media Specific for *Escherichia coli*. *Appl. Environ. Microbiol.* **1993**, 59, 2758-2759.
3. Venosa, A.D., J.R. Haines, and D.M. Allen. Efficacy of Commercial Inocula in Enhancing Biodegradation of Weather Crude Oil Contaminating a Prince William Sound Beach. *J. Ind. Microbiol.* **1992**, 10, 1-11.
4. Venosa, A.D., J.R. Haines, W. Nisamaneepong, R. Govind, S. Pradhan, and B. Siddique. Screening of Commercial Inocula for Efficacy in Stimulating Oil Biodegradation in Closed Laboratory System. *J. Haz. Mater.* **1991**, 28, 131-144.
5. Covert, T.C., L.C. Shadix, E.W. Rice, J.R. Haines, and R.W. Freyberg. Evaluation of the Autoanalysis Colilert Test for Detection and Enumeration of Total Coliforms. *Appl. Environ. Microbio.* **1989**, 55, 2443-2447.
6. McDaniels, A.E., R.H. Bordner, P.S. Gartside, J.R. Haines, K.P. Brenner, and C.C. Rankin. Holding Effects on Coliform Enumeration in Drinking Water Samples. *Appl. Environ. Microbiol.* **1985**, 50, 755-762.
7. Haines, J.R. and R.M. Atlas. Biodegradation of Petroleum Hydrocarbons in Continental Shelf Regions of the Bering Sea. *Oil Petro. Pollut.* 1, 85-96.
8. Haines, J.R. and R.M. Atlas. *In-Situ* Microbial Degradation of Prudhoe Bay Crude Oil in Beaufort Sea Sediments. *Mar. Environ. Res.* **1982**, 7, 91-102.

9. Haines, J.R., R.M. Atlas, R.P. Griffiths, and R.Y. Morita. Denitrification and Nitrogen Fixation in Alaskan Continental Shelf Sediments. *Appl. Environ. Microbiol.* **1981**, 41, 412-421.
10. Haines, J.R. and M. Alexander. Microbial Degradation of Polyethylene Glycols. *Appl. Microbiol.* **1975**, 29, 621-625.
11. Haines, J.R. and M. Alexander. Microbial Degradation of High-molecular Weight *n*-Alkanes. *Appl. Microbiol.* **1974**, 28, 1084-1085.

Book Chapters

1. Tabak, H.H., J.R. Haines, A.D. Venosa, J.A. Glaser, S. Desai, and W. Nisamaneepong. Enhanced Degradation of the Alaskan Weathered Crude Oil Alkane and Aromatic Hydrocarbons by Indigenous Microbiota Through Application of Nutrients. *In: Gas, Oil, Coal and Environmental Biotechnology III*. pp. 3-38, C. Akin and J. Smith, eds., 1991.
2. Tabak, H.H., A.D. Venosa, J.A. Glaser, J.R. Haines, S. Desai and W. Nisapaneepong. Laboratory Studies to Evaluate the Enhanced Biodegradation of the Alaska Weathered Crude Oil Components Through the Application of Nutrients. *In: A Comprehensive Approach to Problems with Oil Spills in Marine Environments: The Alaska Story*. V. Molak, W. Davis-Hoover, S. Khan, and M. Mehlman, eds. pp. 139-146. Princeton Science Publishing Company, Princeton, NJ, 1990.
3. Venosa, A.D., J.R. Haines, W. Nisapaneepong, R. Govind, S. Pradhon and B. Siddique. Screening of Commercial Inocula for Efficacy in Stimulating Oil Biodegradation in Laboratory Microcosms. *In: A Comprehensive Approach to Problems with Oil Spills in Marine Environments: The Alaska Story*. V. Molak, W. Davis-Hoover, S. Khan, and M. Mehlman, eds. pp. 147-161. Princeton Science Publishing Co., Princeton, NJ, 1990.
4. Atlas, R.M., G.E. Roubal, A. Bronner, and J.R. Haines. Biodegradation of Hydrocarbons in Mousse from the IXTOC-I Well Blowout. pp 199-217. *In: Energy and Environmental Chemistry, Fossil Fuels*. L.H. Keith, ed. Ann Arbor Science, Ann Arbor, MI, 1982.

Meeting Papers

1. Breen, A., T. Ward, J.C. Loper, R. Govind, J. Haines, and D. Bishop. Trichloroethylene Degradation by Toluene Oxidizing Bacterial Colonizing a Vapor Phase Biofilter. *Environmental Biotechnology Keystone Conference*. March 16-22, 1995, Lake Tahoe, CA.
2. Venosa, A.D., J.R. Haines, B.A. Wrenn, K.L. Strohmeier, B.L. Eberhart, M. Kadkhodayan, E. Holder, M.T. Suidan, D. King, and B. Anderson. Bioremediation Study of Spilled Crude Oil on Fowler Beach, Delaware. *Int'l. Oil Spill Conf.* February 27-March 2, 1995, Long Beach, CA. Amer. Petrol. Inst., Washington, DC.
3. Breen, A., A. Rooney, T. Ward, J.C. Loper, and J.R. Haines. Ammonia Monooxygenase Gene Probe Tracking of Ammonia Oxidizing Populations in Environmental Samples. *Amer. Soc. Microbial. Nat'l. Mtg. 94th*. 1994, Las Vegas, NV.

4. Venosa, A.D., J.R. Haines, and D.M. Allen. Enhancement of Oil Biodegradation on a Prince William Sound Beach with Commercial Inocula. *Soc. Ind. Microbiol. Sec. Int'l. Marine Biotechnol. Conf.* 1991, Baltimore, MD.
5. Glaser, J.A., P.H. Pritchard, A.D. Venosa, J.R. Haines, and C. Costa. Bioremediation Treatability Trials Using Nutrient Application to Enhance Cleanup of Oil Contaminated Shoreline. *Air Waste Management Assoc. Mtg.* 1990, Pittsburgh, PA.
6. Glaser, J.A., A.D. Venosa, J.R. Haines, and S. McCutcheon. Nutrient Movement Through Beach Media: Problems and Field Results; Application to Enhance Cleanup of Oil Contaminated Shoreline. *Air Waste Management Assoc. Mtg.* 1990, Pittsburgh, PA.
7. Haines, J.R., S. McCutcheon, J.A. Glaser, A.D. Venosa, and D. Miller. Importance of Hydraulic Behavior of Subsurface Water in Affecting Biodegradation of Crude Oil on Oiled Beaches in Prince William Sound. *Amer. Soc. Microbiol. Ann. Mtgs. 90th.* May 13-17, 1990, Anaheim, CA.
8. Tabak, H.H., S. Desai, A.D. Venosa, J.A. Glaser, J.R. Haines, S.L. Hayes, and W. Nisamaneepong. Biodegradation of Alaskan Weathered Crude Oil Constituents. *Amer. Soc. Microbiol. Ann. Mtgs. 90th.* May 13-17, 1990, Anaheim, CA.
9. Brenner, K.P., P.S. Gartside, J.R. Haines, C.C. Rankin, and R.H. Bordner. The Precision and Relative Accuracy of the Membrane Filter and Most Probable Number Total Coliform Methods Using Several Water Types. *Amer. Soc. Microbiol. Ann. Mtgs.* March 23-28, 1986, Washington, DC.
10. Haines, J.R., E. Pesek, G. Roubal, A. Bronner, and R.M. Atlas. Microbially Mediated Chemical Evolution of Crude Oils Spilled into Differing Ecosystems. *Amer. Soc. Microbiol. Ann. Mtgs.* March 1-6, 1981, Dallas, TX.
11. Atlas, R.M., G.E. Roubal, and J.R. Haines. Biodegradation of Hydrocarbons in Mousse from the IXTOC-I Well Blowout. *Amer. Chem. Soc. Mtgs.* August 24-28, 1980, San Francisco, CA.
12. Haines, J.R. and R.M. Atlas. Denitrification Potential of Alaskan Continental Shelf Sediment Microorganisms. *Amer. Soc. Microbiol. Ann. Mtgs.* May 11-16, 1980, Miami, FL.
13. Haines, J.R. and M. Alexander. Reduction of Bacterial Activity by Glass Microbeads. *Amer. Soc. Microbiol. Ann. Mtgs.* May 8-13, 1977, New Orleans, LA.
14. Haines, J.R. and M. Alexander. Microbial Metabolism of High-Molecular Weight

Polyethylene Glycols and High-Molecular Weight *n*-Alkanes. *Amer. Soc. Microbiol. Ann. Mtgs.* April 28- May 2, 1975, New York, NY.

Refereed Papers

1. Breen, A.W., A. Rooney, T. Ward, J.C. Loper, R. Govind, and J.R. Haines. Characterization of Bacteria in a TCE-Degrading Biofilter. *Symp. Bioremediation Haz. Wastes Res. Dev. and Field Eval.* EPA/600/R-94/075 San Francisco, CA., 1994. USEPA, Washington, DC., pp 229-234.
2. Haines, J.R., M. Kadkhodayan, D.J. Mocsny, C.A. Jones, M. Islam, and A.D. Venosa. Effect of Salinity, Oil Type, and Incubation Temperature on Oil Degradation. *In-Situ and On-Site Bioreclamation, Second International Symposium*, San Diego, CA., Battelle, Columbus, OH., April 5-8, 1993.

Presentations

Strohmeier, K.L., A.D. Venosa, M.T. Suidan, and J.R. Haines. A Beach Microcosm for the Study of Oil Biodegradation. *In-Situ and On-Site Bioreclamation, Second International Symposium*. San Diego, CA, Battelle, Columbus, OH., April 5-8, 1993.

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EDUCATION

Ph.D., Environmental Science, University of Cincinnati, 1980
M.S., Environmental Engineering, University of Cincinnati, 1968
B.S., Microbiology, University of Cincinnati, 1967

PROFESSIONAL EXPERIENCE

1990 to present	Program Manager, Oil Spill Bioremediation Research Program, U.S. EPA, Cincinnati
1986-1990	Program Manager, Sludge Pathogen Program, U.S. EPA, Cincinnati.
1975-1986	Program Manager, Municipal Wastewater Disinfection Program, U.S. EPA, Cincinnati.
1968-1975	Research Microbiologist, U.S. EPA and predecessor agencies, Municipal Wastewater Disinfection Program, Cincinnati
1967-1968	Research Fellow, University of Cincinnati, Department of Civil and

1966-1968 Environmental Engineering, Cincinnati
Part-Time Clinical Bacteriologist, Cincinnati General Hospital, Central
Bacteriology Laboratory.

PROFESSIONAL SOCIETIES AND RECOGNITION

- Society of the Sigma Xi (full member), 1968-present
- American Society for Microbiology, 1970-present
- Society for Industrial Microbiology, 1990-present
- SIM Awards Committee (1993-present)
- Water Environment Federation, 1972-present
- Chairman, Disinfection Committee, 1987-1989.
- Chairman, Pathogen Equivalency Committee, U.S. EPA, 1990-1991
- Research Committee, Water Pollution Control Federation, 1983-1988.
- Member of National Environmental Technology Applications Corporation (NETAC) panel of experts on effectiveness of bioremediation agents (1990 to present)
- Gold Medal, U.S. EPA, 1990, work on the Alaska Oil Spill Bioremediation Project
- Outstanding rating for continued superior performance, 1990 to 1992
- Nominated for Federal Employee of the Year in May, 1991, U.S. EPA.

PUBLICATIONS: Partial Listing

Author of over 63 refereed publications and 75 conference proceedings.

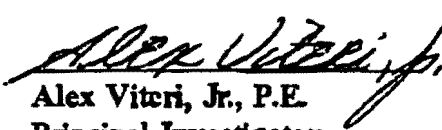
1. Venosa, A. D., J. R. Haines, W. Nisamaneepong, R. Govind, S. Pradhan, and B. Siddique. 1991. "Protocol for Testing Bioremediation Products Against Weathered Alaskan Crude Oil." Proc., International Oil Spill Conference. San Diego, CA, March 4-7.
2. Venosa, A. D., J. R. Haines, W. Nisamaneepong, R. Govind, S. Pradhan, and B. Siddique. 1991. "Screening of Commercial Inocula for Efficacy in Stimulating Oil Biodegradation in Closed Laboratory Systems." Proc., Bioremediation: Fundamentals and Effective Applications. Lamar University, Beaumont, TX, February 21-22.
3. Venosa, A. D., J. R. Haines, W. Nisamaneepong, R. Govind, S. Pradhan, and B. Siddique. 1991. "Screening of Commercial Bioproducts for Enhancement of Oil Biodegradation in Closed Microcosms." In: "Remedial Action, Treatment, and Disposal of Hazardous Waste. Proc., 17th Annual RREL Hazardous Waste Research Symp." EPA/600/9-91/002. U.S. Environmental Protection Agency, April 9-11, p. 388-403.
4. Venosa, A. D., J. R. Haines, and D. M. Allen. 1991. "Effectiveness of Commercial Microbial Products in Enhancing Oil Degradation in Prince William Sound Field

- Plots." In: "Remedial Action, Treatment, and Disposal of Hazardous Waste. Proc., 17th Annual RREL Hazardous Waste Research Symp." EPA/600/9-91/002. U.S. Environmental Protection Agency, April 9-11, p. 353-369.
5. Venosa, A. D. and John R. Haines. 1991. "Screening of commercial inocula for efficacy in stimulating oil biodegradation in closed laboratory reactors." *J. Haz. Materials*, 28:131-144.
 6. Venosa, A. D., J. R. Haines, W. Nisamaneepong, R. Govind, S. Pradhan, and B. Siddique. 1992. "Efficacy of Commercial Inocula in Enhancing Oil Biodegradation in Closed Laboratory Reactors." *J. Ind. Microbiol.*, 10:13-23.
 7. Venosa, A. D., J. R. Haines, and D. M. Allen. 1992. "Efficacy of Commercial Products in Enhancing Biodegradation of Weathered Crude Oil Contaminating a Prince William Sound Beach." *J. Ind. Microbiol.*, 10:1-11.
 Venosa, A. D., J. R. Haines, W. Nisamaneepong, R. Govind, S. Pradhan, and B. Siddique. 1992. "Screening of Commercial Inocula for Efficacy in Stimulating Oil Biodegradation on Laboratory Microcosms." In: V. Molak, W. Davis-Hoover, S. Khan, and M. Mehlman, eds., "A Comprehensive Approach to Problems with Oil Spills in Marine Environments: the Alaska Story." Princeton Scientific Publishing Co., Inc., Princeton, NJ.
 9. Wrenn, B., J. R. Haines, A. D. Venosa, M. Kadkhodayan, and M. K. Suidan. 1994. "Effects of Nitrogen Source on Crude Oil Biodegradation." *J. Ind. Microbiol.* 13:279-286.
 10. Venosa, A. D., M. Kadkhodayan, D. W. King, B. A. Wrenn, J. R. Haines, and M. T. Suidan. 1993. "Testing the Efficacy of Oil Spill Bioremediation Products." 1993 International Oil Spill Conference, Tampa, FL.
 11. Herrington, R. T., G. D. Sayles, C. E. Furlong, R. J. Richter, and A. D. Venosa. "Evaluation of Liposome Encapsulated Casein as a Nutrient Source for Oil Spill Bioremediation." In: In-Situ and On-Site Bioreclamation, the Second Battelle International Symposium, San Diego, CA, April, 1993.
 12. Haines, J. R., M. Kadkhodayan, D. J. Mocsny, C. A. Jones, M. Islam, and A. D. Venosa. 1994. "Effect of Salinity, Oil Type, and Incubation Temperature on Oil Degradation." In: "Applied Biotechnology for Site Remediation," R. E. Hincee, D. B. Anderson, F. B. Metting, Jr., and G. D. Sayles (eds.), Lewis Publishers, Boca Raton, FL.
 13. Venosa, A. D., M. T. Suidan, B. A. Wrenn, J. R. Haines, K. Strohmeier, E. Holder, and B. L. Eberhart. 1994. "Nutrient Application Strategies for Oil Spill Bioremediation in the Field." In: "Twentieth Annual RREL Research Symposium."

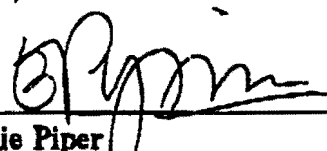
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14. Venosa, A. D., J. R. Haines, M. T. Suidan, B. A. Wrenn, K. L. Strohmeier, B. L. Eberhart, E. L. Holder, and X. Wang. 1994. "Research Leading to the Bioremediation of Oil-Contaminated Beaches." In: "Symposium on Bioremediation of Hazardous Wastes: Research, Development, and Field Evaluations." EPA/600/R-94/075, June 1994.
 15. Venosa, A. D. 1995. "Oil Spill Bioremediation on Coastal Shorelines: A Critique." In: Bioremediation: Principles and Practices. S. K. Sikdar and R. Irvine (ed.), Technomic Publishing, Lancaster, PA (submitted).
 16. Venosa, A. D., J. R. Haines, B. A. Wrenn, K. L. Strohmeier, B. Loye Eberhart, M. K. Suidan, and B. Anderson. 1995. "Bioremediation of Crude Oil Released on a Sandy Beach in Delaware." 2nd International Oil Spill Research and Development Forum. May, 1995 (accepted).
 17. Venosa, A. D., B. A. Wrenn, K. L. Strohmeier, J. R. Haines, B. Loye Eberhart, M. Kadkhodayan, E. Holder, M. T. Suidan, D. King, and B. Anderson. 1995. "Bioremediation Study of Spilled Crude Oil on Folwer Beach, DE." Poster presentation at the International Oil Spill Conference, Long Beach, CA. February, 1995.
 18. Venosa, A. D., M. T. Suidan, J. R. Haines, B. A. Wrenn, K. L. Strohmeier, M. Kadkhodayan, B. Loye Eberhart, E. Holder. 1995. "Bioremediation of an Experimental Crude Oil Spill on the Shoreline of Fowler Beach, Delaware." In: "In Situ and On-Site Bioreclamation Symposium, San Diego, CA."
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SUBMITTED ON BEHALF OF THE STATE OF ALASKA DEPARTMENT OF ENVIRONMENTAL CONSERVATION THIS MAY 1, 1995


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PROFESSIONAL BIOGRAPHY

Mr. Henry is a Research Associate V at Louisiana State University's Institute for Environmental Studies (IES) and is currently Senior Scientist for response and chemical assessment projects. Actively involved in environmental and analytical chemistry since he joined IES in 1985 as a staff member, Mr. Henry has developed an in depth understanding and working knowledge of environmental issues from a multidisciplinary perspective. This is a result of his academic interest and collaboration in research studies with Microbiologists, Toxicologists, Marine Scientists, and Marine Geologists. He has nine years experience applying analytical chemistry to environmental problems relative to hazardous waste, crude oil and refined oil product spills, chronic discharges from oil production activities, incineration, uptake of pollutants by aquatic species, responses of aquatic species to pollution, and recovery of environmentally stressed ecosystems. Prior to the T/V *Exxon Valdez* oil spill (24 March, 1989), Mr. Henry's position within IES was to provide nonroutine analytical support, primarily using mass spectrometry, for research projects conducted by LSU. During this time, he managed IES's laboratory resources, was involved in instrument development research, and provided laboratory support to NOAA-HMRAD as required. During the T/V *Exxon Valdez* oil spill, Mr. Henry provided on-scene technical support including chemical characterization of the spilled oil, source-fingerprint analyses, management of field laboratory support, risk assessments relative to clean up techniques, monitoring experiments using beach cleaners and bioremediation agents, and responded to a variety of fisheries issues. In addition to on-site support, Mr. Henry has managed and conducted the analytical support provided by LSU during the subsequent Winter (1989-1990) and Summer Shoreline Monitoring Programs (1990-1993) in Prince William Sound.

Post T/V *Exxon Valdez* spill, Mr. Henry has been manager of the NOAA response support activities provided by LSU. In this position, he routinely provides chemical and oil spill support to NOAA-HMRAD SSCs through telephone communication, electronic mail, and on-scene response. In addition to providing 24 hour on-call support to NOAA, Mr. Henry routinely teaches training courses in hazardous chemical assessment and oil chemistry. He is actively involved in the training that LSU provides to NOAA-HMRAD and provides additional nonincident support such as advising on chemistry issues during the CAMEO development (e.g. classification of greater than 4000 chemicals into the chemical protective clothing (CPC) recommendations).

Mr. Henry designs and implements his own research including the use of a source-fingerprinting method to assess chronic impacts of oil and tar along the coast of Louisiana. The goal is to develop an analytical tool to aid resource management agencies in identifying pollution sources and target future regulations and policies. Mr. Henry is currently writing the standard method for evaluating the efficacy of commercial bioremediation products using a detailed, compound specific GC-MS approach; this work is being conducted in association with the EPA and NETEC. Mr. Henry has responded on scene to more than 10 major (>100,000 gallon) oil spills in the last 4 years and is currently involved with several marine pollution monitoring projects including an assessment of chemical weathering patterns observed in stranded oil within Prince William Sound.

Resume/Vita: Henry

QUALIFICATIONS

- ◊ Nine years experience in Environmental/Analytical Chemistry investigating anthropogenic environmental stress in relation to hazardous waste, crude oil and refined oil product spills, chronic discharges from oil production activities, incineration, uptake of pollutants by aquatic species, and recovery of stressed environments. Experience in standard analytical procedures written by EPA, NOAA, ASTM, NIOSH. Experience in developing nonroutine methods to solve analytical problems such as microextraction techniques for whale blubber, methods for source-fingerprinting spilled crude oils and petroleum products, and rapid thermal extraction techniques to replace laborious solvent extraction procedures.
- ◊ Actively involved in multidisciplinary research at Louisiana State University in Chemistry, Toxicology, Microbiology, Environmental and Marine Sciences. Actively involved in developing analytical methods for monitoring the fate of anthropogenic-sourced pollutants in the terrestrial and marine environment, assessing oil bioremediation efficacy, and source-fingerprinting of spilled oils.
- ◊ Experience in managing large projects and resources including analytical laboratories. Good communication, teaching, and technical writing skills. Experience in writing project reports, scientific papers, and research proposals. Routinely teach courses in oil chemistry and chemical hazard assessment.
- ◊ In depth knowledge of field sampling techniques and procedures for water, sediments/soils, biota, and hazardous waste. Experience in designing and implementing sampling programs. Routinely teach training courses in sampling strategy and techniques. Experience in providing sampling plans during chemical and oil spill responses.
- ◊ Experience in setting up, operating, and supervising analytical field laboratories and mobile laboratories often during emergency responses with little advance preparation or resources in remote locations (e.g., Cold Bay, Alaska). Experience in developing analytical methods on-site for nonroutine analyses including methods for monitoring the fate of chemical beach cleaners applied during oil spill mitigation.
- ◊ Nine years mass spectrometry experience (GC/MS, LC/MS, ITDS, MS/MS). Experience with most conventional instrumentation for the analysis of organic pollutants (GC-FID, ECD, PID, HALL™, TCD; UV-Fluorescence; IR). Experience with most conventional field instruments (HNU™, OVA™, Photovac™, Microsensor P-200™, colorimetric tubes such as Dräger™, air sampling equipment, etc.)
- ◊ Strong background in sophisticated electronics which compliments instrumental analysis skills as well as instrumental development research and computer skills. Routinely use Macintosh™, IBM™, Hewlett Packard (pascal), and the NOAA Prime computer systems.
- ◊ Strongly committed to developing a broader understanding of the effect of anthropogenic-sourced pollution in the marine environment, developing an understanding of the "big picture," and making a difference.

Resume/Vita: Henry

EXPERIENCE

July, 1985 to present Research Associate V, Inst. for Environmental Studies, Louisiana State University, Baton Rouge, LA 70803. Research in applied analytical/environmental chemistry. At present, manager of response and environmental assessment and impact studies conducted at IES at LSU. Experienced in hazardous material spill response, oil spill response and damage assessment, non-routine methods development and analysis of complex sample matrixes by gas chromatography/mass spectrometry (GC/MS), evaluation and development of new instruments and techniques in environmental analysis. Senior Scientist in charge of the NOAA Hazmat contract to LSU for chemistry support.

Major projects while at LSU

- 1993 Characterization of the relative biodegradation potential of a variety of crude oils and refined oil products (PI, NOAA Funded).
- 1993 Provide on-scene support to the Greenhill well blowout, fire, and oil spill near East Timbalier Island, Louisiana. Support included establishment and management of an on-site GC/MS laboratory, source-fingerprinting and tracking the weathering of the spilled oil, ad shoreline clean-up assessment.
- 1992 Provided on-scene support during the Burlington Northern rail car aromatic concentrate spill on the Nemadji River near Superior, Wisconsin. Support included hazardous chemical assessment, development of monitoring strategies, and field surveys.
- 1992 Provided on-scene response to the 87,000 gallons styrene monomer spill in Morgan City, LA. Support included hazard assessment of spilled material relative to occupational worker safety (i.e., tug personnel, local residents, and clean up contractors), resource risk assessment, tracking the fate of the spilled chemical, and on-scene compound specific chemical analysis of air and water samples to assess human health and aquatic hazards. (NOAA)
- 1992 Provided on-scene response and support during the C/V Santa Clara I hazmat incident. Support included detailed risk hazard assessments of all cargo, entry monitoring recommendations, health and safety recommendations for on-site personnel, decontamination procedures, deactivation and/or disposal recommendations, and trajectory modeling using ALOHA and real-time weather data from a MET station to determine safe perimeter zones during mitigation activities. The C/V Santa Clara I had spilled two dangerous chemicals: arsenic trioxide, a highly toxic poison, and magnesium phosphide, an extremely explosive chemical when in contact with water in confined spaces. Greater than 500 pounds of magnesium phosphide was spilled in hold 1 of the C/V Santa Clara I. (NOAA)
- 1992 Characterization of chronic sources and impacts of oil and tar along the Louisiana coast (co-PI, MMS funded).

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- 1992-1993 Methods evaluation, methods development, and detailed chemical analysis of oils treated with commercial bioremediation products (EPA tier 2 testing) in conjunction with EPA Cincinnati, OH. (co-PI, EPA funded)
- 1991 T/V Exxon Valdez: Continued Summer Shoreline Monitoring Study of PWS. The purpose of this study was to evaluate the recovery of intertidal areas impacted by the T/V Exxon Valdez oil spill. (NOAA)
- 1991 Conducted experiments using field portable gas chromatographs to study the weathering of spilled crude oil; specifically, the volatilization of the low molecular weight hydrocarbons, nC-5 thru nC-9, including benzene, toluene, and the C-2 benzenes. These experiments were conducted in association with the NIST in-situ burn experiments. (NOAA/LSU)
- 1991 Designed and conducted field studies to assess the potential human health threat from ground-level exposure to integrated smoke plumes from the Kuwait oil fires. Characterized depositional oil/soot from the Kuwait fires. (NOAA)
- 1990 Response to the Apex spill in Galveston Bay, July, 1990 (700,000 gal. oil spill). Support included source fingerprinting, field assessments, developing an oil budget, and monitoring bioremediation studies. (NOAA)
- 1990 Provided laboratory support for source-fingerprinting analysis and damage assessment of the T/V Mega Borg oil spill for the State of Louisiana (LDF&G, LDEQ).
- 1990-1993 T/V Exxon Valdez: Shoreline Monitoring Study of PWS. The purpose of this study was to evaluate the recovery of intertidal areas impacted by the T/V Exxon Valdez oil spill. (NOAA)
- 1990 Net Environmental Benefit Analysis (NEBA) project which analyzed the use of intrusive technologies to remove subsurface oil from beaches impacted by the T/V Exxon Valdez oil spill in PWS, AK. The study was designed to determine if there were net environmental benefits from the proposed excavation and washing of oiled sediments, and return of treated sediments to the excavated site over natural cleansing processes and the use of approved treatments for the 1990 Summer cleanup. Chairman of the Chemistry Subcommittee and coauthor of final report. (NOAA)
- 1990 Source-fingerprinting of impacted mussels in conjunction with the Apex Houston, damage assessment. (NOAA)
- 1989-1990 T/V Exxon Valdez: Shoreline Winter monitoring study in PWS. The purpose of this study was to monitor the effects of winter storms on the natural removal (cleansing) of stranded oil from heavily impacted beaches. (NOAA)
- 1989-1990 Part of the NOAA/HAZMAT response team providing scientific support for the T/V Exxon Valdez Oil spill response and clean up effort in Prince William Sound (PWS), AK. These efforts include the development of analytical procedures for fingerprinting oil, management of field laboratories, assisting in bioremediation studies, and fishery issues.

- 1989 Fingerprinting of oiled birds and animals associated with the T/V *Exxon Valdez* oil spill. (NOAA Office of Damage Assessment).
- 1989 Participant in the ASTM D-19 committee on development of an ASTM approved MS method for source fingerprinting of petroleum.
- 1989-1993 NOAA/HAZMAT contract chemist tasked with chemical response training of field personal and providing 24-hour, on call, support related to chemical incidents/oil spills.
- 1988-1989 Evaluation of the Pyran Thermal Chromatograph/MS system, Partially funded by the instrument manufacturer Ruska Instrument Corporation, Houston, TX. Developed methodologies for environmental applications of the Pyran instrument including SW-846, METHOD 8275. (PI: E.B. Overton)
- 1988 Development and field testing of a mobile GC/MS for the detection of olefin mixtures associated with ozone formation. (PI: E.B. Overton)
- 1987-1989 Louisiana Offshore Oil Pipeline (LOOP)/Louisiana Department of Wildlife and Fisheries Baseline Monitoring Project. Hydrocarbon and interstitial salinity assessment of sediment samples. (LDWF)
- 1987-1988 Compilation of Background Information and Chemical Characterization of Produced Waters. P/O a larger project funded by the Minerals Management Service and coordinated by Coastal Environments, Inc. titled "Impacts of Outer Continental Shelf Related Activities on Sensitive Coastal Habitats." Provided detailed characterization of production water and assessment of organic pollution impact in the study areas. Contributed to the synthesis of data and writing of final report (MMS 89-0031).
- 1987-1988 Detoxification of dioxin-contaminated sludges using combined microbiological and photolytic degradative approaches. EPA funded project. Provided detailed sample analyses of dioxin contaminated creoste sludges for polynuclear aromatics and chlorinated dioxins and difurans by GC/MS. (PI: R. Portier)
- 1985-1986 T/V *Arco Anchorage* Oil Spill, Port Angeles Harbor, WA. Provided detailed characterization and "fingerprinting" of the spilled oil and assessment of impact in sediments, mussels, clams, geoducks, sea urchins, and dungeness crabs in support of the Marine Resource Damage Assessment (MRDA) Program. Authored the hydrocarbon sections of the MRDA final report.
- 1985-1986 Old Inger: Biological Treatment. Provided GC-FID and GC/MS analysis and interpretation of biologically treated sludge samples from an abandoned hazardous waste, Superfund, site. (Principal investigator, PI: P. Templett and R. Portier)

Experience before LSU

♦Aug., 1984--July, 1985

Environmental Chemist, Enviro-Med Laboratory, Baton Rouge, LA 70806.
Duties included maintenance and operation of two GC/MS instruments (HP-

Resume/Vita: Henry

5985 and HP-5993), QA/QC, and routine analysis of water and sediment samples for priority pollutants using established EPA methodologies.

◊ May, 1983--Aug., 1983

Field inspector, East Baton Rouge Parish Mosquito Abatement, Baton Rouge, LA, 70807. Duties included field sampling, disease surveillance, and larvaciding.

◊ Feb., 1981--May, 1982

X-ray assistant, Rapides General Hospital, Alexandria, LA 70301. Duties included aiding in routine and special procedure x-ray techniques and processing of x-rays.

MILITARY

Service: May, 1976--May, 1980 Active duty United States Air Force. Weapons Control Systems/Radar Technician. Duties included the repair and calibration of airborne electronic systems including radar, air-data computers, and optical sighting systems. Attained the position of shift supervisor before honorable discharge with the rank of sergeant.

Aug. 1982--July, 1985 Honorable discharge from the Louisiana Air National Guard with rank of Staff Sergeant.

Awards: AF Good Conduct medal, AF Longevity Service Award, Noncommissioned Officer of the Quarter, The Louisiana Commendation Medal for Meritorious Service.

EDUCATION

Candidate for Master's Degree in Marine Science Program at Louisiana State University, Baton Rouge, LA 70803 (completed a 30 semester hour program).

May, 1985 B.S. degree in Environmental Science from Louisiana State University, Baton Rouge, LA 70803.

PUBLICATIONS

Bianchini, M., R. Portier, K. Fugisaki, and C.B. Henry. 1986. Determination of Optimal Toxicant Loading for Biological Closure of a Hazardous Waste Site. In *ASTM Proceedings*, 1986.

Winston, G.W., B.S. Shane, C.A. Trynor, L.S. Ortego, C.B. Henry, and D.J. McMillin. 1987. Introduction of xenobiotic activation by channel catfish (*Ictalurus punctatus*) from a contaminated river basin swamp ecosystem. Presentation, American Chemical Society, Division of Environ. Chemistry, 194th National Meeting, New Orleans, LA. Aug. 30-Sept. 4, 1987. (extended abstract published, invited paper) Vol. 27, #2.

- Pierce, R.H., R.C. Brown, E.B. Overton, and C.B. Henry. 1987. Organic pollutant distribution in Mississippi River delta sediments. Presentation, American Chemical Society, Division of Environ. Chemistry, 194th National Meeting, New Orleans, LA. Aug. 30-Sept. 4, 1987. (extended abstract published, invited paper) Vol. 27, #2.
- Portier, R.J., M.A. Bianchini, K. Fujisaki, C.B. Henry, and D.J. McMillin. 1987. A demonstration study on biotreatment of contaminated soils at an abandoned petrochemical refinery. Presentation, American Chemical Society, Division of Environ. Chemistry, 194th National Meeting, New Orleans, LA. Aug. 30-Sept. 4, 1987. (extended abstract published) Vol. 27, #2. (Certificate of Merit Award).
- Shane, B.S., A.M. Troxclair, D.J. McMillin, and C.B. Henry. 1988. Comparative mutagenicity of nine brands of coffee to *Salmonella typhimurium* TA100, TA102, and TA104. *Environmental Mutagenicity*. Vol. 11, #2.
- Boesch, D.F., N.N. Rabalais, C.S. Milan, C.B. Henry, J.C. Means, R.P. Gambrell, and E.B. Overton. 1988. Impacts of outer continental shelf (OCS) related activities on sensitive coastal habitats. Vol. II. Produced Waters. Report to Minerals management service, MMS 89-0031.
- Winston, G.W., B.S. Shane, and C.B. Henry. 1988. Hepatic monooxygenase induction and promutagen activation in channel catfish from a contaminated river basin. *Ecotoxicity and Environmental Safety* 16, 258-271 (1988)
- Henry, C.B., E.B. Overton, and C. Sutton. 1988. Applications of the Pyran Thermal Extractor-GC/MS for the rapid characterization and monitoring of hazardous waste. In *Proceedings of the First International Symposium of Field Screening Methods for Hazardous Waste Site Investigations* (EPA sponsored, peer reviewed). Oct. 11-13, 1988.
- Overton, E.B. and C.B. Henry. 1988. Field deployable instrument for the analysis of semivolatile organic compounds. In *Proceedings of the First International Symposium of Field Screening Methods for Hazardous Waste Site Investigations* (EPA sponsored, peer reviewed). Oct. 11-13, 1988.
- Portier, R., M. Bianchini, K. Fujisaki, C. Henry, and D. McMillin. 1988. Comparison of effective toxicant biotransformation by autochthonous microorganisms and commercially available cultures in the *in situ* reclamation of abandoned industrial sites. *Schr.-reihe verein WaBoL* 80, Gustav Fisher Verlag, Stuttgart, 1988.
- Winston, G.W., S. Narayan, and C.B. Henry. 1989. Induction pattern of liver microsomal m-alkoxyresorufin o-dealkylases of channel catfish (*Ictalurus punctatus*): correlation with PCB exposure *in situ*. *Journal of Environmental Science and Health*.
- Grande, I.H., E.B. Overton, and C.B. Henry. 1989. Analysis of semi-volatiles in soils by thermal extraction-gas chromatography/mass spectrometry. Submitted to EPA for incorporation into a book on field methodologies. (in review--status unknown)
- Henry, C.B., E.B. Overton, T.R. Irvin, T. Junk, J. Nocerino, and L. Butler. 1989. Rapid field screening technology for semivolatile organic compounds in

Resume/Vita: Henry

solids and sludges. Extended abstract (invited paper). 198th ACS National Meeting, Miami, Florida, 10-15 Sept., 1989.

- Daniels, C.B., C.B. Henry, and J.C. Means. 1989. Assessment of the genotoxicity of coastal oil drilling produced waters using chromosomal aberrations. *ASTM Standard Technical Publication*. 1096-1990, pages 356-371.
- Shane, B.S., C.B. Henry, J.H. Hotchkiss, K.A. Klausner, W.H. Gutemann, and D.J. Lisk. 1990. Organic toxicants and mutagens in ashes from eighteen municipal refuse incinerators. *Environmental Science and Technology*. (submitted)
- Michel, J., C.B. Henry, W.J. Sexton, and M.O. Hayes. 1990. The Exxon Valdez winter monitoring program results. In *Proceedings for Conference on Oil Spills: Management and Legislative Implications*, Newport, RI.
- Michel, J., M.O. Hayes, W.J. Sexton, J.C. Gibeau, C.B. Henry. 1991. Trends in natural removal of the Exxon Valdez oil spill in Prince William Sound from September 1989 to May 1990. In *Proceedings of the 1991 Oil Spill Conference*, API, Washington, D.C., pages 181-187.
- Henry, C.B. 1991. "LSU Response Report: Kuwait Oil Fires." Submitted to NOAA's Arabian Gulf Program Manager, John Robinson. IES Technical Publication, 31 pages.
- Henry, C.B. and P.O. Roberts. 1992. "LSU Response Report: St. Eustatius Refinery Oil Spill, Incident #89." Submitted to the National Oceanic and Atmospheric Administration's Hazardous Materials and Assessment Division. Technical Report, 28 April, 1992.
- Henry, C.B. and P.O. Roberts. 1992. "LSU Response Report: St. John Island, U.S. Virgin Islands, Site Visit." Submitted to the National Oceanic and Atmospheric Administration's Hazardous Materials and Assessment Division. Technical Report, 12 August, 1992.
- Henry, C.B. and P.O. Roberts. 1992. "LSU Response Report: Greenhill Well Blowout." Submitted to the National Oceanic and Atmospheric Administration's Hazardous Materials and Assessment Division. Technical Report, 12 August, 1992.
- Jones, R.K., J. Farr, et al. 1992. The Evaporation of Benzene and a Series of Alkanes from Crude Oil. NOAA/Hazardous Materials Response and Assessment Division.
- Henry, C.B. and E.B. Overton. 1993. Chemical composition and source fingerprinting of depositional oil from the Kuwait oil fires. In *Proceedings of the 1993 Oil Spill Conference*, API, Washington, D.C., pages 407-414. Report IES92-05, 13 November, 1992.
- Henry, C.B. and P.O. Roberts. 1993. "LSU Response Report: Sunshine Bridge #6 Spill, Incident #124." Submitted to the National Oceanic and Atmospheric Administration's Hazardous Materials and Assessment Division. Technical Report IES93-03, 18 May, 1993.
- Henry, C.B., P.O. Roberts, and E.B. Overton. 1993. Characterization of chronic sources

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and impacts of tar along the Louisiana coast. U.S. Dept. of the Interior, Minerals Management Service, Gulf of Mexico OCS, Regional Office, New Orleans, La. OCS Study MMS 93-0046. 36 pp.

- Hoff, R.Z., G. Shigenaka, and C.B. Henry. 1993. Salt marsh recovery from a crude oil spill: vegetation, oil weathering, and response. In *Proceedings of the 1993 Oil Spill Conference*, API, Washington, D.C., pages 307-312.
- Mearns, A.J., P. Roques, and C.B. Henry. 1993. Measuring efficacy of bioremediation of oil spills: monitoring, observations, and lessons learned from the Apex oil spill experience. In *Proceedings of the 1993 Oil Spill Conference*, API, Washington, D.C., pages 335-344.
- Shigenaka, G. and C.B. Henry. 1993. Use of mussels and semipermeable membranes devices to assess bioavailability of residual polynuclear aromatic hydrocarbons three years after the *Exxon Valdez* oil spill. 1993. Third Symposium on Environmental Toxicology and Risk Assessment, ASTM STP 1219. J. S. Hughes, G.R. Biddinger, and E. Mones, Eds., ASTM, Philadelphia, PA.
- Roques, D.E., E.B. Overton, et al. 1994. "Using Gas Chromatography/Mass Spectroscopy Fingerprint Analyses to Document Process and Progress of Oil Degradation." *Journal of Environmental Quality* 23 (4). 851-855.
- Scholz, D.K., J. Michel, et al. 1994. Assessment of Risks Associated with the Shipment and Transfer of Group V Fuel Oils. HAZMAT Report 94-8. August 1994. Hazardous Materials Response & Assessment Division National Oceanic and Atmospheric Administration, Seattle, WA 30ppg.
- Mendelssohn, I.A., Q. Lin, et al. 1995. The development of bioremediation for oil spill cleanup in coastal wetlands: product impacts and bioremediation potential. 1995 International Oil Spill Conference, Long Beach, CA, American Petroleum Institute.
- Michel, J. D. Scholz, et al. 1995. Group V fuel oils: source, behavior, and response issues. 1995 International Oil Spill Conference, Long Beach, CA, American Petroleum Institute.
- Roberts, P.O., C.B. Henry, et al. 1995. Fast source-fingerprinting analysis for oil spill response. 1995 International Oil Spill Conference, Long Beach, CA, American Petroleum Institute.
- Shigenaka, G. V.P. Vincente, et al. 1995. Biological effects monitoring during an operational application of Corexit 9580. 1995 International Oil Spill Conference, Long Beach, CA, American Petroleum Institute.

PROFESSIONAL MEMBERSHIPS

- ◇ Member of the Technical Committee mandated under the Firefighters Safety Study Act to provide guidance to first responders and to find methods by which training and equipment can be improved.

KENNETH LEE

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QUALIFICATION HIGHLIGHTS

- o 15+ years of research experience in the field and laboratory.
- o Author of numerous scientific publications.
- o International recognition in microbial ecology, oil spill countermeasures, nutrient, trace metal, and contaminant dynamics in the aquatic systems.
- o Expertise in client support, project management, business administration and scientific presentations.
- o Extensive oceanographic experience at sea.
- o Self motivated, confident, high energy manager.
- o Dedicated professional attitude, committed to getting the job done.
- o Confident and decisive under stressful conditions.
- o Talent for getting diverse groups to work well together.
- o Excellent communications and interpersonal skills.

PROFESSIONAL AND TEACHING EXPERIENCES

1989 - Department of Fisheries and Oceans, Quality of the Marine Environment Division,
Maurice Lamontagne Institute, Mont-Joli, Quebec.

Section Head, Bioremediation and Environmental Assessment

Senior Research Scientist responsible for the development and implementation of original and independent research projects in chemical oceanography that deal with biogeochemical processes in relation to microbial transformations of contaminants and other chemical and biochemical substances in estuarine and coastal environments. Current scientific responsibilities include: 1) research on the development and evaluation of bioremediation technologies; 2) microbiological studies in St. Lawrence Estuary and benthic mesocosms; 3) studies on the natural biodegradation rates of PAHs; 3) the development of microbial exoenzyme assays to assess the impact ocean disposal operations on benthic biota; and 4) the management of regional assessment/contaminant monitoring programs.

Advisor to the department on questions related to marine pollution and environmental impact.
Representative for the department on national and international scientific committees.

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1984 - Kenneth Lee Research Limited, Dartmouth, N.S.
1989

President and Chief Scientist

Kenneth Lee Research Limited, was a Canadian owned consulting and research organization in the environmental sciences. Formed in 1984, the company was unique in that it centered its expertise in chemical oceanography/microbial ecology and employed five professionals. The company had demonstrated experience in multidisciplinary research programs on environmental processes, aquatic toxicology and contaminant monitoring, with government agencies, universities and industries. All contracts (approx. \$1.2M) were completed on time and within budget.

Representative projects:

- o The Enhancement of The Natural Biodegradation of Sable Island Condensate and Hibernia Crude Oil: Department of Fisheries & Oceans, Chemical Oceanography Division, Bedford Institute of Oceanography.

Research Scientist of a project to evaluate the feasibility of enhancing the biodegradation of Hibernia crude oil and Sable Island condensate stranded on and/or buried in sandy beaches, by means of nutrient additions. This study included both field and laboratory components. The fate of hydrocarbons in the environment were monitored and the response of components were evaluated.

- o Significance of Bacteria to the Nutrition of Offshore Scallops: Department of Fisheries & Oceans, Chemical Oceanography Division, Bedford Institute of Oceanography.

Senior Scientist/Project Manager of an unsolicited proposal (UP) awarded to investigate the possibility that offshore scallops (Placopecten magellanicus) may supplement their nutritional requirements by using bacteria sustained natural hydrocarbon seepages or the direct incorporation of dissolved organic carbon.

Oceanographic expeditions were conducted on Georges Bank and Browns Bank to measure rates of bacterial production at the sediment/water interface and uptake of radiolabelled methane and dissolved organic carbon by scallops. Electron microscopy was used to determine if endosymbiotic bacteria were present in the gill tissue of scallops.

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- o Development of Rapid Cost-Effective Sediment Bioassay Procedures Based on Activity of Indigenous Microorganisms: Environment Canada, Conservation and Protection.

Principal Investigator of a project to develop a rapid cost-effective sediment bioassay procedures based on microbial activity (e.g. extracellular enzymatic cleavage of carbohydrates and proteins, incorporation of dissolved organic substrates), by indigenous microorganisms.

- o Assessment of the Toxicity of the Dredged Spoils from Dalhousie Harbour, New Brunswick and the Recovery of the Heron Island Dump Site: Environment Canada, Conservation and Protection.

Senior Scientist for a project supported by the Ocean Dumping Control Act (ODCA) Research Fund to assess the toxicity of dredged spoils on the activity of indigenous sediment microbiota and to collect data on the recovery of the microbial activity at a previously used dump site which may support an application for its reactivation.

- o Oceanic CO₂ Measurements: The Transport of CO₂ from the Atmosphere to the Deep Ocean: Department of Fisheries and Oceans, Physical and Chemical Sciences Branch, Bedford Institute of Oceanography.

Program Coordinator of a project to assess the rate of transport of CO₂ from the atmosphere to the deep ocean in the context of climate changes that will occur as a result of the increase of CO₂ and other radiatively active gases in the atmosphere. The proposal was funded to design and develop instrumentation and procedures to make high precision measurements of total carbonate, chlorofluoromethanes and carbon tetrachloride in seawater.

- o Biologist in a multidisciplinary research program with the Atlantic Geoscience Centre/Chemical Oceanography Division, Bedford Institute of Oceanography, to study natural submarine oil/gas seeps off Baffin Island in the Canadian Arctic. Responsible for biological studies using the PISCES IV submersible and the design/deployment of a sediment trap mooring.

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1982 - NSERC Visiting Scientist with the Ocean Chemistry Division, Institute of Ocean Sciences, Sidney,
1984 B.C.

Research projects included.

- o A joint study between The People's Republic of China and Canada, funded by IDRC; evaluating the role of microorganisms in the biodegradation of crude oil and oil dispersants in pelagic ecosystems, using SEAFLUX enclosures.
- o Field studies under the Ocean Chemistry Division's Carbon Fluxes Program to measure diurnal and seasonal rates of algal and bacterial productivity and sedimentation processes in Saanich Inlet.
- o A cooperative study with the Ocean Ecology Division utilizing the research submersible PISCES IV, to study microbial processes at the sediment-water interface of Saanich Inlet.

PROFESSIONAL AFFILIATIONS

Sep 1992 - Associate Professor, Department of Oceanography, University of Quebec at Rimouski
Present (UQAR).

May 1990 - Associate Professor, Division of Life Sciences, Scarborough Campus, University of Toronto.
Present

Oct 1990 - Associate Professor, Department of Botany, University of Toronto, cross-appointed to the School
Present of Graduate Studies.

Feb 1995- Thesis supervisor for D. Mueller (M.Sc.), Department of Civil Engineering, Texas A&M
University.

Sep 1994 - Thesis supervisor for L. Roberge (M.Sc.), Department of Oceanography, University of Quebec
Present at Rimouski (UQAR).

Sep 1993 - Thesis supervisor for A. Weise (M.Sc.), Department of Botany, University of Toronto.
Present

Sep 1991 - Thesis supervisor for B. Dorval (M.Sc.), Department of Environmental Studies, University of
May 1993 Quebec at Montreal.

Sep 1990 - Thesis supervisor for L. De Marcos (M.Sc.), Institute of Environmental Studies, University of May
1992 Toronto.

Kenneth Lee
Page Five

EDUCATION

Thesis

Effects of vanadium on phytoplankton: Field and laboratory studies, Ph.D., University of Toronto, 1982.

Bacterial and algal heterotrophy in lakes, M.Sc., University of Toronto, 1977.

Diurnal fluctuation in photosynthetic pigments: Bedford Basin, B.Sc. (1st Class Honours), Dalhousie University, 1975.

Scholarships and Awards

Natural Sciences and Engineering Research Council of Canada, Industrial Research Fellowship: 1984.

Natural Sciences and Engineering Research Council of Canada, Visiting Fellowship: Institute of Ocean Sciences, Sidney, B.C., 1982 to 1984.

Elizabeth Ann Wintercorbyn Award in Botany: University of Toronto, 1982.

Natural Sciences and Engineering Research Council of Canada, University Postdoctoral Fellowship: 1981

Ontario Ministry of Colleges and Universities, Ontario Graduate Scholarship: University of Toronto, 1980 to 1981.

Natural Sciences and Engineering Research Council of Canada, Postgraduate Scholarship: University of Toronto, 1977 to 1980.

Ontario Ministry of Colleges and Universities, Ontario Graduate Scholarship: 1977 to 1979.

University of Toronto Masters Fellowship: University of Toronto, 1976 to 1977.

Dalhousie University, Sarah M. Lawson Scholarship: W.K. Kellogg Biological Station, Michigan State University, Summers of 1974 and 1975.

Dalhousie University, Sarah M. Lawson Scholarship: Huntsman Marine Laboratory, St. Andrews, N.B., Summer of 1973.

Kenneth Lee**PUBLICATIONS**

Lee, K., F.-X. Merlin, R.P.J. Swannell, T. Reilly, J. Oudot, P. Sveum, A. Laudousse, C. Chaumery and C. Bocard (1995) Development of a protocol for the assessment of bioremediation agents on petroleum polluted shorelines. In: Proceedings of the 1995 International Oil Spill Conference (Prevention, Behavior, Control and Cleanup). Long Beach, California, February 27 - March 2, 1995. (Accepted)

Lee, K., G.H. Tremblay and S.E. Cobanli (1995) Bioremediation of oiled-beach sediments: Assessment of inorganic and organic fertilizers. In: Proceedings of the 1995 International Oil Spill Conference (Prevention, Behavior, Control and Cleanup). Long Beach, California, February 27 - March 2, 1995. (Accepted)

Lee, K., R. Siron and G.H. Tremblay (1995) The effectiveness of bioremediation in the reduction of toxicity in oiled intertidal sediments. In situ and on-site bioreclamation. In: Proceedings of the 3rd International Symposium - In situ and On-Site Bioreclamation, April 24-27, 1995, San Diego, California, USA. (Submitted)

Swannell, P.J. R., J.E. Lepo, K. Lee, P.H. Pritchard, D.M. Jones (1995) Bioremediation of oil contaminated fine-grained sediments in laboratory microcosms. In: Proceedings of the Second International Research and Development Forum, International Maritime Organization (IMO), May 24-26, 1995, London, UK. (Submitted)

Croft, B., R.P.J. Swannell and K. Lee (1995) The effect of bioremediation agents on oil biodegradation in medium-fine sand. In: Proceedings of the 3rd International Symposium - In situ and On-Site Bioreclamation, April 24-27, 1995, San Diego, California, USA. (Submitted)

Lee, K. (1995) Bioremediation studies in low-energy shoreline environments. In: Proceedings of the 2nd International Oil Spill Research and Development Forum. International Maritime Organization, May 23-26, 1995, London, UK. (Submitted)

Swannell, R.P.J., D. Mitchell, K. Lee, J.E. Lepo, and P.H. Pritchard (1995) Bioremediation of oil contaminated mud flats. In: Proceedings of the 2nd International Oil Spill Research and Development Forum. International Maritime Organization, May 23-26, 1995, London, UK. (Submitted)

Swannell, R.P.J., K. Lee and M. McDonagh (1995) Field Evaluations of Oil Spill Bioremediation Microbiological Reviews (Invited Article: In preparation)

Steel, D.A. and K. Lee (1995) A preliminary environmental assessment of chemical contaminants in the St. Lawrence Estuary and Saguenay Fjord (Quebec, Canada). Canadian Technical Report of Fisheries and Aquatic Sciences (In preparation).

Merlin, F.-X., K. Lee, T. Reilly, R.P.J. Swannell, J. Oudot, P. Sveum, A. Laudousse, C. Chaumery and C. Bocard (1994) Protocol for experimental assessment of bioremediation agents on a petroleum polluted shoreline. In: Proceedings of the 17th Arctic and Marine Oil Spill Program, Vancouver, B.C., June 1994. pp. 465-478.

Swannell, R.P.J., A. Basseres, K. Lee and F.-X. Merlin (1994) A direct respirometric method for the *in situ* determination of bioremediation efficacy. In: Proceedings of the 17th Arctic and Marine Oil Spill Program, Vancouver, B.C., June 1994. pp 1273-1286.

DeMarco, E. and K. Lee (1994) La biodegradation des hydrocarbures aromatiques polycycliques (HAP) dans les sediments du fjord du Saguenay. Dans: *Le Fjord du Saguenay: un milieu exceptionnel de recherche*. J.M. Sevigny et C.M. Couillard (eds.), Rapp. manus. can. sci. halieut. aquat. pp. 86-87.

Tay, K.L., K.G. Doe, A.J. MacDonald, S.J. Wade, J.D.A. Vaughan, A.L. Huybers, K. Lee and R. Larocque (1994) Application of sediment bioassays for the environmental assessment of ocean dump sites. R. van Coillie, Y. Roy, Y. Bois, P.G.C. Campbell, P. Lundahl, L. Marrel, M. Michaud, P. Riebel and C. Thellen (eds.), *Proceedings of the 20th Annual Aquatic Toxicology Workshop*, October 17-21, 1993, Quebec City, Quebec. Can. Tech. Rep. Fish. Aquat. Sci. 1989: 48-49.

Lee, K., R. Siron, G.H. Tremblay and J. Lavoie (1994) Application of the Microtox solid phase test to monitor the effectiveness of bioremediation strategies. In: *Proceedings of the 21st Annual Aquatic Toxicity Workshop*, Sarnia, Ontario, October 2-5, 1994. (In Press)

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University of Cincinnati, Cincinnati, OH 45221-0071

EDUCATION

Ph.D. Environmental Engineering, University of Illinois, Urbana, 1975
M.S. Environmental Engineering, University of Illinois, Urbana, 1973
B.S. Civil Engineering, American University of Beirut, 1971

PROFESSIONAL EXPERIENCE

1990-Present Professor and Director of Environmental Program
1985-1989 Professor of Environmental Engineering, University of Illinois, Urbana, Illinois
1980-1985 Associate Professor of Environmental Engineering, University of Illinois,
Urbana, Illinois
1976-1980 Assistant Professor of Environmental Engineering, The Georgia Institute of
Technology, Atlanta, Georgia
1976 Special Consultant on Computer Simulations and Process Design (Summer),
Snell Environmental Group, Lansing, Michigan
1975 Visiting Assistant Professor of Environmental Engineering, University of
Illinois, Urbana, Illinois

RELEVANT EXPERIENCE

- Principal Investigator on 49 Research Projects and Grants since 1980 with actual expenditures of \$8,689,400.
- Principal Investigator, "Research on Bioremediation of Hazardous Wastes and Chemical Spills," UC/U.S. EPA Cooperative Agreement, April 1993-March 1996 (\$6,000,000).
- Principal Investigator, "Indefinite Delivery Contract for the Army," U.S. Army Construction Engineering Research Laboratory, October 1993-September 1996 (\$2,000,000).
- Secretary, Anaerobic Digestion Group of the International Association of Water Quality, 1987-1992.
- Chairman of the Distinguished Lecturer Committee, Association of Environmental Engineering Professors, 1985-1989.
- Chairman, Unit Operations Manual Committee, 1985-Present.
- Chairman, Research Symposium Subcommittee of the Program Committee of the Water Environment Federation, 1992-Present.
- Member, Standard Methods Committee, 1984-Present.
- Associate Editor, *Journal of Environmental Engineering*, American Society of Civil Engineers, 1989-Present.
- Chairman, Science Advisory Committee Chairman for Regions 7 & 8 U.S. EPA HSRC, 1988-92.

PROFESSIONAL SOCIETIES AND RECOGNITION

- American Society of Civil Engineers, 1972-Present
- American Water Works Association, 1972-Present
- American Society for Microbiology, 1991-Present
- International Association of Water Quality, 1977-Present
- Water Environment Federation, 1972-Present
- Outstanding Graduate Teaching Award, 1991
- WPCF Award for Best Student Paper in the Ph.D. Category (Awardee: my advisee R.D. Vidic), 1991
- College of Engineering Research Award, University of Cincinnati, 1993
- Association of Environmental Engineering Professors, CH2MHILL Ph.D. Thesis Supervisor Award, 1993
- WEF Award for Best Student Paper in the M.S. Category (Awardee: my advisee M. Gupta), 1994
- Unsolicited and Unrestricted Research Grant from the Ford Motor Co., 3-yr, \$120,000, 1994
- WEF Award for Best Student Paper in the M.S. Category (Awardee: my advisee D. Sharma), 1995
- WEF Award for Best Student Paper in the Ph.D. Category (Awardee: my advisee J. Chenga), 1995

PUBLICATIONS: Partial Listing

Author of over 100 refereed publications, 10 books and book chapters, 80 conference proceedings, and other technical publications. Over 20 invited presentations, and 125 conference presentations.

1. Narayanan B., Suidan M.T., Gelderloos A.B., Brenner R.C.: "Treatment of VOCs in High Strength Wastes Using an Anaerobic Expanded-Bed GAC Reactor," *Water Research*, 27(1)(1993):181-194.
2. Vidic R.D., Sorial G.A., Papadimas S.P., Suidan M.T., Speth T.F.: "Molecular Oxygen Effect on the Scale-Up of GAC Adsorbers," *J. American Water Works Association*, 84(8) (1992):98-105.
3. Sorial G.A., Suidan M.T., Vidic R.D., Brenner R.C.: "Effect of GAC Characteristics on Adsorption of Organic Pollutants," *Research Journal of WEF*, 56(1)(1993):53-57.
4. Suidan M.T., Schroeder A.T., Nath R., Krishnan E.R., Brenner R.C.: "Treatment of CERCLA Leachates by Carbon-Assisted Anaerobic Fluidized Beds," *Water Science and Technology*, 27(2)(1993):273-282.

5. Flora J.R.V., Suidan M.T., Biswas P., Sayles G.D.: "Modeling Substrate Transport into Biofilms: Role of Multiple Ions and pH Effects," *Journal of Environmental Engineering, ASCE*, 119(5)(1993):908-930.
6. Sorial G.A., Suidan M.T., Vidic R.D., Maloney S.W.: "Competitive Adsorption of Phenols on GAC - I. Adsorption Equilibrium, II. Adsorption Dynamics Under Anoxic Conditions," *Journal of Environmental Engineering, ASCE*, 119(6)(1993):1044-1058.
7. Sakakibara Y., Flora J.R.V., Suidan M.T., Biswas P., Kuroda M.: "Measurement of Mass Transfer Coefficients with an Electrochemical Method Using Dilute Electrolyte Solutions," *Water Research*, 28(1)(1994):9-16.
8. Vidic R.D., Suidan M.T., Brenner R.C.: "Impact of Oxygen Mediated Oxidative Coupling on Adsorption Kinetics," *Water Research*, 28(2)(1994):263-268.
9. Fox P., Suidan M.T.: "A Comparison of Expanded-Bed GAC Reactor Designs for the Treatment Of Refractory/Inhibitory Wastewaters," *Water Research*, 27(5)(1993):769-776.
10. Khodadoust A.P., Wagner J.A., Suidan M.T., Safferman S.I.: "Solvent Washing of PCP Contaminated Soils with Anaerobic Treatment of Wash Fluids," *Research Journal of WEF*, 66(5)(1994):692-697.
11. Flora J.R.V., Suidan M.T., Wuellner A.M., Boyer T.K.: "Anaerobic Treatment of a High Strength Industrial Wastewater Containing Chlorophenols," *Research Journal of WEF*, 26(1)(1994):21-31.
12. Smith P.J., Biswas P., Suidan M.T., Brenner R.C.: "A Fundamental Approach to Modeling Biofilters Used for the Treatment of VOC Air Streams," Submitted for Publication, *Journal of the Air and Waste Management Association*.
13. Fox P., Suidan M.T.: "A Comparison of Expanded-Bed GAC Reactor Designs for the Treatment Of Refractory/Inhibitory Wastewaters," *Water Research*, 27(5) (1993):769-776.
14. Sorial G.A., Papadimas S.P., Suidan M.T., Speth T.F.: "Competitive Adsorption of VOCs and BOM - Oxic and Anoxic Environments," *Water Research*, 28(9)(1994):1907-1920.
15. Gupta A., Flora J.R.V., Gupta M., Sayles G.D., Suidan M.T.: "Methanogenesis and Sulfate-Reduction in Chemostats: I. Kinetic Studies and Experiments," *Water Research*, 28(4)(1994):795-805.

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EDUCATION

B.S. Chemistry, University of Pittsburgh at Johnstown, 1992

PROFESSIONAL EXPERIENCE

1992-Present Junior Research Associate, University of Cincinnati
1992 Student Laboratory Assistant, Chemistry Tutor, Undergraduate Research
in Organic Chemistry, University of Pittsburgh at Johnstown

RELEVANT EXPERIENCE

- Specialize in the analysis of biodegraded petroleum by gas chromatography/mass spectroscopy.
- Participation in several pre-field projects studying beach hydrodynamics via tracer.
- Participation in field project involving the spillage of oil in order to establish efficacy of bioremediation.
- Familiar with atomic absorption spectrometry, nitrate autoanalyzer method via cadmium reduction, gas chromatography/mass spectroscopy, and fractionation via liquid chromatography.

PROFESSIONAL SOCIETIES AND RECOGNITION

- 3.68 QPA; Dean's List seven times
- Member, Golden Key National Honor Society
- College of Arts & Sciences Alumni Merit Award, 1991
- Lord Scholar Award for Chemistry, 1991
- Scholars in Education Award, 1990

PUBLICATIONS

1. Wrenn B.A., Suidan M.T., Strohmeier K.L., Eberhart B.L., Wilson G.J.: "Nutrient Retention in the Bioremediation Zone of a Sandy Beach," *95th International Oil Spill Conference*, Long Beach, CA (February 27-March 2, 1995).

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EDUCATION

Ph.D. Environmental Engineering, in progress
M.S. Environmental Engineering, University of Cincinnati, 1992
B.S. Civil Engineering, Jesuit University, Beirut, Lebanon, 1988

PROFESSIONAL EXPERIENCE

1994-Present Graduate Research Assistant, University of Cincinnati
1993-1994 Project Engineer, EES, Columbus, OH
1992-1993 Graduate Research Assistant, Department of Civil and Environmental Engineering, University of Cincinnati
1989-1990 Project Engineer, AFRICOF, Conakry, Guinea

RELEVANT EXPERIENCE

- Modelled 3-D transport of radioactive and chemical contaminants in groundwater using various software. Processed the data and sorted it for submittal for RI/FS.
- Investigated subsurface flow contribution to total runoff using unsaturated flow models.
- Explored boundary conditions effects on the numerical stability and the accuracy of the unsaturated flow solution.
- Conducted studies on groundwater contamination and remedial technologies.
- Examined long-term persistency of water level of the Great Lakes. The method consists of coupling synthetically generated streamflows (using Spigot model) with a first order autoregressive moving average model ARMA(1,1).
- Analyzed rainfall and runoff from recorded storm events in Ohio to obtain better estimates of infiltration rates. Estimated infiltration rates using nonlinear optimization schemes.

PROFESSIONAL SOCIETIES AND RECOGNITION

- Quality Team Award, Parsons Environmental, Cincinnati, OH, 1994
- University Research Council Fellowship, University of Cincinnati, 1993-1994
- University Graduate Scholarship, University of Cincinnati, 1991-1993
- Graduate Research Assistantship, University of Cincinnati, 1991-1993 and 1994 -1995

PUBLICATIONS

1. Boufadel M.C., Suidan M.T., "Pilot-scale Evaluation of Nutrient Delivery for Oil-Contaminated Beaches," *EPA Symposium on Bioremediation of Hazardous Wastes: Research, Development, and Field Evaluations*. San Francisco, CA (June 1994).

2. Boufadel M.C., "Unit Hydrographs from the Nash Model," Submitted for publication, *Water Resources Bulletin* (August 1994).
3. Boufadel M.C., "Optimum Loss Rate from Separate Storms Analysis," to be submitted for publication, *Journal of Hydrology*.
4. Boufadel M.C., Buchberger S.G., "Robust Method to Generate Multiple Unit Hydrographs," Presentation at the *ASCE International Symposium on Engineering Hydrology*, San Francisco, CA (July 1993).
5. Buchberger S.G., Boufadel M.C., "Rainfall Losses in the Great Miami River Watershed," final report prepared for the Miami Conservancy District, Dayton, Ohio (August 1992).

Biographical Sketch January, 1995
Alan J. Mearns, PhD, Ecologist
National Oceanic and Atmospheric Administration
National Ocean Service, Seattle, Washington

Dr. Mearns is Leader of the NOAA Biological Assessment Team at the Hazardous Materials Response and Assessment Division (HMRAD) in Seattle. His team provides guidance on resources at risk from pollution and spills of hazardous materials and oil, evaluates environmental benefits and impacts of pollution response technologies including combustion, bioremediation, use of chemicals and no response at all.

Dr. Mearns received his PhD in Fisheries from the University of Washington and his MA and BS degrees in biology and zoology from California State University at Long Beach. As Leader of the Biology Division at the Southern California Coastal Water Research Project, his team developed new understanding about the effects of wastewater discharges and other human actions on the health, diversity and abundance of coastal ecosystems. Working with chemists, engineers and oceanographers, he has developed a variety of empirical models for predicting the ecological effects and effectiveness of pollution control technologies. He also pioneered many of the survey methods now being routinely used to address similar issues around the world.

As lead ecologist for the NOAA MESA Puget Sound Program in the 1980's he help put together the first inter-disciplinary assessments of the health of Puget Sound. next, he headed up a national review of historical trends in marine pollution, providing a foundation for NOAA's current National Status and Trends Program. His team is now conducting a long-term assessment of the recovery of Alaska shorelines following the 1989 Exxon Valdez oil spill.

Dr. Mearns recently chaired the 1994 Annual Meeting of the Pacific Northwest Society of Environmental Toxicology and Chemistry (SETAC) in Victoria. He is a member of the editorial board of the Water Environment Federation, and is past president and vice president of the Washington chapter of the American Institute of Fishery Research Biologists. He also serves as Co-Chair of the Technical Advisory Committee for the Santa Monica Bay Restoration Program and the San Francisco Estuary Institute. He has participated in many international conferences. Recently, he served on the National Research Council's Committee on Wastewater Management in Coastal Urban Areas. He received the 1992 US Department of Commerce Silver Medal and Biology Alumnus of the Year at California State University, Long Beach. He and his colleagues have published nearly 200 papers and reports dealing with coastal marine pollution, environmental quality and clean-up.

SELECTED PUBLICATIONS

Mearns, A.J. and M.J. Sherwood. 1977. *Distribution of neoplasms and other diseases of marine fishes relative to the discharge of wastewater*. Ann. New York Acad. Sci. 128:210-224.

Mearns, A.J. and D.R. Young. 1977. *Chromium in the Southern California marine environment*. 125-141 In C.S. Giam (ed), Pollutant effects on marine organisms. Lexington Books, D.C. Heath and Co., Lexington, MA.

Mearns, A.J., D.R. Young, R.J. Olson and H.A. Schafer. 1981. *Trophic structure and the cesium-potassium ratio in pelagic ecosystems*. CalCOFI Rep. 22:99-110.

Mearns, A.J. 1981. *Effects of municipal discharges on open coastal ecosystems*. Chapter 1. pp. 25-66 In R.A. Geyer (ed). Marine Environmental Pollution, 2. Dumping and Mining. Elsevier Scientific Publishing Co., New York. 574 pp.

Mearns, A.J., E. Haines, G.S. Kleppel, R.A. McGrath, J.J.A. McLaughlin, D.A. Segar, J.H. Sharp, J.J. Walsh, J.Q. Word, D.K. Young and M.W. Young. 1982. *Effects of nutrient and carbon loadings on communities and ecosystems*. 53-65 In G.F. Mayer (ed), Ecological stress and the New York Bight: Science and Management. Est. Res. Fed., Columbia, S.C.

Mearns, A.J. and T. P. O'Connor. 1984. *Biological effects versus pollutant inputs: the scale of things*. pp 693-722 In H.H. White (ed). Concepts in Marine Pollution Measurements. Maryland Sea Grant Publication UM-SG-TS-84-03, University of Maryland, College Park, MD 20742. 743 pp.

Mearns, A.J. 1986. *Biological effects studies at various levels along the U.S. Pacific coast*. pp. 667-690 In G. Kullenberg (ed), The Role of the Oceans as a Waste Disposal Option. Proceedings of the NATO Advanced Research Workshop, Vilamoura, Portugal, April 24-30, 1985. NATO ASI Series C: Mathematical and Physical Sciences Vol. 172. 725 pp.

Mearns, A.J., R.C. Swartz, J.M. Cummins, P.A. Dinnel, P. Plesha and P.M. Chapman. 1986. *Inter-laboratory comparison of a sediment toxicity test using the marine amphipod, Rhepoxynius abronius*. Mar. Env. Res. 19:13-37.

Mearns, A.J. 1988. *The "odd fish": unusual occurrences of marine life as indicators of changing ocean conditions*. Chapter 7. 137-176 In D.F. Soule and G.S. Kleppel (eds), Marine organisms as indicators. Springer-Verlag, New York.

Mearns, A.J., M.B. Matta, D. Simecek-Beatty, M.F. Buchman, G. Shigenaka and W.W. Wert. 1988. *PCB and chlorinated pesticide contamination in U.S. fish and shellfish: A historical assessment report*. NOAA Technical Memorandum NOS OMA 39. National Oceanic and Atmospheric Administration, Seattle, WA, 98115. 140 pp.

Mearns, A.J. 1991. *Observations of an oil spill bioremediation activity in Galveston Bay, Texas*. NOAA Technical Memorandum NOS OMA 57. National Oceanic and Atmospheric Administration, Seattle, WA. 38 pp.

Mearns, A.J., M. Matta, G. Shigenaka, D. MacDonald, M. Buchman, H. Harris, J. Golas and G. Lauenstein. 1991. *Contaminant trends in the Southern California Bight: Inventory and*

Assessment. NOAA Technical Memorandum NOS ORCA 62. National Oceanic and Atmospheric Administration, Seattle, WA.

Houghton, J.P., D.C. Lees, W.B. Driskell and A.J. Mearns. 1991. *Impacts of the Exxon Valdez Spill and subsequent cleanup on intertidal biota - 1 year later.* 467-475 In Proceedings, 1991 International Oil Spill Conference, San Diego. American petroleum Institute Publ.4529.

Young, D. R., A.J. Mearns and R.W. Gossett. 1991. *Bioaccumulation of p,p'-DDE and PCB 1254 by a flatfish bioindicator from highly contaminated marine sediments of Southern California.* 159-169 In R.A. Baker (ed), *Organic Substances and Sediments in Water.* Volume 3, Biological. Lewis Publishers, Inc. Chelsea, Michigan. 332 pp.

Mearns, A.J. 1992. *Contaminant trends in the Southern California Bight: Four decades of stress and recovery.* 5-25 In P.M. Grifman and S.E. Yoder (eds), *Perspectives on the Marine Environment.* Proceedings from a Symposium on the Marine Environment of Southern California, May 10, 1991. 100th Anniversary Meeting of the Southern California Academy of Sciences. Sea Grant program, University of Southern California, USCSG-TR-01-92.

Mearns, A.J. 1993 "Appropriate" Technologies for marine Pollution Control: Controversy in Fitting the Solution to the Problem-Shoreline Cleaning, Bioremediation, Wastewater treatment, Monitoring and Assessment. *Sea Technology*, October 1993: 31-37.

Mearns, A.J. 1995. Confirming response effectiveness: an overview and guide to operational monitoring. 485-488 In Proceedings, 1991 International Oil Spill Conference, Long Beach. American Petroleum Institute Publ.4620.

Mearns, A.J., K. Doe, W. Fisher, R. Hoff, K. Lee, R. Siron, G. Thursby and A. Venosa. 1995. Toxicity trends during an oil spill bioremediation experiment. To appear in Proceedings, 18th Arctic and Marine Oilspill Program Technical Seminar. June 14-16, 1995, Edmonton, Alberta, Canada. In press.

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Education: U.C.L.A., Ph.D., Biochemistry, 1976
Stanford University, B.S. Chemistry, 1971.

Experience: Professor of Chemistry and Biochemistry, University of Alaska Southeast and University of Alaska Fairbanks, School of Fisheries and Ocean Sciences, August 1991 to present.
Associate Professor of Chemistry and Biochemistry, University of Alaska, Southeast, August 1983 to August 1991.
OFCF Trainee in seaweed mariculture, Hokkaido Regional Fisheries Research Laboratory, Kushiro, Hokkaido, Japan, July 1987 to October 1987.
Visiting Associate Research Biologist, Marine Science Institute, University of California at Santa Barbara, July 1986 to June 1987.
Assistant Professor, School of Fisheries and Science, University of Alaska, Juneau, August 1978 to August 1983.
Acting Dean, School of Fisheries and Science, University of Alaska, Juneau, September 1981 to May 1982.
Research Biochemist, National Marine Fisheries Service, Auke Bay Laboratory, Juneau, Alaska, May 1979 to September 1979.
Post-doctoral, Institute of Marine Science, University of Alaska, Fairbanks, April 1976 to June 1978.

Honors Received:

1993 Dictionary of International Biography
1990 International Directory of Distinguished Leadership (1991 ed)
1987 Japan Overseas Fishery Cooperation Foundation Trainee
Scholarship
1986 Who's Who in the West

Memberships in Professional Organizations:

American Association for the Advancement of Science
Phycological Society of America
Western Society of Naturalists
International Seaweed Association

Selected Presentations:

Stekoll, M. S. Coastal Habitat Injury Assessment: Summary of Algal Results. EVOS Trustee Work Plan Workshop, Anchorage, AK. January, 1995.
Stekoll, M. S. and L. Deysher. Restoration of High Intertidal *Fucus gardneri* (Silva) Populations Using Biodegradable Erosion Control Fabric. 15th International Seaweed Symposium, Valdivia, Chile. January, 1995.
Stekoll, M. S., L. Deysher, and T. A. Dean. Seaweeds and the EXXON VALDEZ Oil Spill. 1993 International Oil Spill Conference (Prevention, Preparedness, Response), Tampa, FL. April, 1993.
Stekoll, M. S. and L. Deysher. Coastal Habitat Injury Assessment: intertidal algal

communities. *Exxon Valdez* Oil Spill Symposium, Anchorage, AK. February, 1993.

Selected Publications:

- Stekoll, Michael S.** and Lawrence Deysher. (in prep) Restoration of high intertidal *Fucus gardneri* (Silva) populations using biodegradable erosion control fabric. Submitted to XV ISS proceedings.
- Dean, Thomas A.** and Michael S. **Stekoll**. 1995 (in press). Effects of the *Exxon Valdez* oil spill on eelgrass and subtidal algae. In Rice, S. D., R. B. Spies, D. A. Wolfe, and B. A. Wright (eds). *Exxon Valdez* Oil Spill Symposium Proceedings. American Fisheries Society Symposium Number 00: 00-00
- Highsmith, Raymond C., Tama L. Rucker, Michael S. Stekoll, Susan M. Saupe, Mandy R. Lindeberg, Robin Jenne and Wallace P. Erickson.** 1995 (in press). Impact of the *Exxon Valdez* oil spill on intertidal biota. In Rice, S. D., R. B. Spies, D. A. Wolfe, and B. A. Wright (eds). *Exxon Valdez* Oil Spill Symposium Proceedings. American Fisheries Society Symposium Number 00: 00-00.
- Dean, Thomas A., Michael S. Stekoll, Stephen C. Jewett, Richard O. Smith and Jo Ellen Hose.** (in prep). Eelgrass (*Zostera marina*) in Prince William Sound, Alaska: effects of the *Exxon Valdez* oil spill. Mar. Ecol. Prog. Ser. 00:00-00.
- Van Tamelen, Peter G.** and Michael S. **Stekoll**. 1995 (in press). Population response of the brown alga, *Fucus gardneri*, and other algae in Herring Bay, Prince William Sound, to the *Exxon Valdez* oil spill. In Rice, S. D., R. B. Spies, D. A. Wolfe, and B. A. Wright (eds). *Exxon Valdez* Oil Spill Symposium Proceedings. American Fisheries Society Symposium Number 00: 00-00
- Stekoll, Michael S., Lawrence Deysher, Raymond C. Highsmith, Susan M. Saupe, Zhanyang Guo, Wallace P. Erickson, Lyman McDonald and Dale Strickland.** 1995 (in press). Coastal habitat injury assessment: intertidal communities and the *Exxon Valdez* oil spill. In Rice, S. D., R. B. Spies, D. A. Wolfe, and B. A. Wright (eds). *Exxon Valdez* Oil Spill Symposium Proceedings. American Fisheries Society Symposium Number 00: 00-00.
- Stekoll, Michael S., Lawrence Deysher, and Thomas A. Dean.** 1993. Seaweeds and the *Exxon Valdez* oil spill. Proceedings of the 1993 International Oil Spill Conference. American Petroleum Institute, Washington, DC. pp 135-140.
- Dean, Thomas A., Lyman McDonald, Michael S. Stekoll, and Richard R. Rosenthal.** 1993. Damage assessment in coastal habitats: lessons learned from the *Exxon Valdez*. Proceedings of the 1993 International Oil Spill Conference. American Petroleum Institute, Washington, D.C. pp. 695-687.
- Stekoll, Michael S. and Page V. Else.** 1990. Cultivation of *Macrocystis integrifolia* (Laminariales, Phaeophyta) in southeastern Alaskan waters. *Hydrobiologia* 204/205:445-451.
- Stekoll, M. S. and T. S. Shirley.** 1993. *In situ* spawning behavior of an Alaskan population of pinto abalone, *Haliotis kamtschatkana* Jonas, 1845. The Veliger 36(1):95-97.
- Stekoll, M. S.** 1986. Developing herring embryos exposed to oil in seawater. In C.W. Haegerle (ed) Proceedings of the Fifth Pacific Coast Herring Workshop, October 29-30, 1985. Can. MS Rep. Fish. Aquat. Sci. 1871:135-138.

- Collodi, P., M. S. Stekoll and S. D. Rice. 1984. Hepatic aryl hydrocarbon hydroxylase activities in coho salmon (*Oncorhynchus kisutch*) exposed to petroleum hydrocarbons. *Comp. Biochem. Physiol.* 79C:337-341.
- Stekoll, M. S., L. E. Clement, and D. G. Shaw. 1980. Sub-lethal effects of chronic oil exposure on the intertidal clam *Macoma balthica*. *Marine Biology* 57:51-60.

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET
October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel		\$88.9						
Travel		\$12.5						
Contractual		\$818.2						
Commodities		\$3.8						
Equipment		\$0.0						
Subtotal	\$0.0	\$923.4	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
General Administration		\$42.2						
Project Total	\$0.0	\$965.6	\$600.0	\$600.0	\$45.0	\$0.0	\$0.0	\$0.0
Full-time Equivalents (FTE)		1.3						
Other Resources								

Dollar amounts are shown in thousands of dollars.

Comments: This project is a multi-agency international proposal involving Environment Canada, three Universities, two federal agencies and several private firms all interested in improving bioremediation of sub-surface oil in the Prince William Sound. The project will start where other studies have left off and will ultimately reduce the exposure to affected species in the Prince William Sound. This coordinated study will involve the community of Chenega Village. Local staff hired will be trained to be part of the project through to its end.

This project is not a continuation of any prior project, it will use prior work as a base to build upon.

NEPA cost are estimated at \$15.0.

Because they recognize the importance of this project several of the participating agencies and others are making in kind contributions as follows

EPA approximately \$175,000 over the life of the project

Environment Canada approximately \$65,000 (USA) over the life of the project

University of Cincinnati approximately \$2,750

1996

Prepared:

1 of 4

Project Number:

Project Title: Enhanced Bioremediation Study

Agency: AK Dept. of Environmental Conservation

**FORM 3A
AGENCY
PROJECT
DETAIL**

5/1/95

1996 EXXON VALDEZ TRUS .LL COUNCIL PROJECT BUDGET
October 1, 1995 - September 30, 1996

Personnel Costs:			GS/Range/ Step	Months Budgeted	Monthly Costs	Overtime	Proposed FFY 1996
PM	Name	Position Description					
*	Ernest W. Piper (18X002)	Restoration Chief	24A	1.0	7,113		7.1
	Alex Viteri (187072)	Project Leader	20M	2.0	8,088		16.2
	Dianne Munson (18X019)	Restoration Specialist	18B	9.0	4,732	3,000	45.6
	David Bruce (18X012)	Restoration Specialist	23B	3.0	6,670		20.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
Subtotal				15.0	26,603	3,000	
Those costs associated with program management should be indicated by placement of an *.							Personnel Total
							\$88.9
Travel Costs:			Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996
PM	Description						
	Anchorage--Juneau		444	5	20	150	5.2
	Anchorage Cordova		224	4	16	150	3.3
	Anchorage--Chenega Village		200	8	16	150	4.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
Those costs associated with program management should be indicated by placement of an *.							Travel Total
							\$12.5

1996

Project Number:
Project Title: Enhanced bioremediation Study
Agency: AK Dept. of Environmental Conservation

FORM 3B
Personnel
& Travel
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET
October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
Mail and courier		3.0
Telephone and fax		2.0
Printing and reproduction		3.0
Photography		0.5
Training (Hazmat, etc.)		3.0
Freight and cartage		3.0
Contract with Chenega Village for labor		20.0
Environment Canada contract		165.0
University contracts (Louisiana State, UAF, Univviversity of Cincinnati)		450.0
Federal Agencies (EPA, NOAA)		167.8
Survey firm contract		65.9
When a non-trustee organization is used, the form 4A is required.		
Contractual Total		\$883.2
Commodities Costs:		Proposed
Description		FFY 1996
Consumable office supplies		2.0
Computer Supplies		1.0
Field gear		0.8
Commodities Total		\$3.8

1996

Project Number:
Project Title: Enhanced Bioremediation Study
Agency: AK Dept. of Environmental Conservation

FORM 3B
Contractual &
Commodities
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET
October 1, 1995 - September 30, 1996

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1996
Description				
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
Those purchases associated with replacement equipment should be indicated by placement of an R.			New Equipment Total	\$0.0
Existing Equipment Usage:		Number of Units	Inventory Agency	
Description				

1996

Project Number:
Project Title: Enhanced Bioremediation Study
Agency: AK Dept. of Environmental Conservation

FORM 3B
Equipment
DETAIL

Project Title: Removal of Introduced Foxes From Islands

Project Number:

96101

Restoration Category:

Proposer:

Alaska Maritime National Wildlife Refuge

Lead Agency:

DOI-FWS

Cooperating Agencies:

DOA-APHIS

Duration:

FY-96, FY-97

Cost FY-96:

\$88.9K

Cost FY-97:

\$53.7K

Geographic Area:

Seguam island, Aleutian Islands

Injured Resource/Service:

Black Oystercatcher, Pigeon Guillemot, Common Murre

RECEIVED
May 1996
EXXON VALDEZ OIL SPILL
TRUSTEE COUNCIL

ABSTRACT

Populations of three species of birds injured by the *T/V Exxon Valdez* oil spill will be allowed to recover by removing introduced arctic foxes (*Alopex lagopus*) from Seguam Island. The injured species are: black oystercatcher (*Haematopus bachmani*), pigeon guillemot (*Cepphus columba*) and common murre (*Uria aalge*). Schmidt et al. (1995) demonstrated that oystercatcher and guillemot populations are much lower on islands with foxes than on fox-free islands, and these and other species of seabird populations are known to increase rapidly following removal of introduced foxes (Bailey 1993, Byrd et al. 1994). Although it is outside the area directly affected by the oil spill, Seguam Island has a particularly high potential for restoring populations of the three injured species because it contains substantial amounts of habitat and remnant populations of all three species are present.

INTRODUCTION

Black oystercatchers (*Haematopus bachmani*), pigeon guillemots (*Cepphus columba*) and murre (*Uria spp.*) were injured by the *T/V Exxon Valdez* oil spill (Piatt et al. 1990, Andres 1993, Oakley and Kuletz in press). Few options are available for direct restoration of injured populations in Prince William Sound, but it is possible to take action to cause populations to expand elsewhere in southern Alaska by removing introduced foxes from islands where they have kept numbers of oystercatchers, guillemots, and murre depressed. Restoration project No. 95041 conducted in the Shumagin Islands in 1993 and 1994 demonstrated the capacity for restoration by removing introduced foxes (Schmidt et al. 1995). A

similar project at a larger island like Seguam should restore even larger numbers of oystercatchers and guillemots. Furthermore, Seguam has a murre population that will benefit from release from predation by introduced foxes.

NEED FOR THE PROJECT

A. Statement of Problem

This project is designed to restore populations of species injured by oil spilled from the *T/V Exxon Valdez*. In particular, black oystercatchers, pigeon guillemots, and common murres should increase following removal of introduced foxes.

B. Rationale

Arctic foxes (*Alopex lagopus*) were introduced to a number of Alaskan Islands for fur farming prior to WWII (Bailey 1993). These introduced predators extirpated or substantially reduced populations of native birds. Colonial nesting seabirds and conspicuous terrestrial birds were particularly severely affected. Removal of foxes is a proven restoration technique for native biodiversity in Alaska (Bailey 1993, Byrd et al. 1994). Few restoration options are available for restoring species injured by oil spilled from the *T/V Exxon Valdez*, but removal of introduced foxes is a project that will increase populations of injured species (Schmidt et al. 1995).

C. Summary of Major Hypotheses and Objectives

We propose to remove introduced arctic foxes from Seguam Island, east-central Aleutian Islands to restore populations of black oystercatchers, pigeon guillemots, and murres. The work will benefit not only these injured species but other insular birds such as threatened Aleutian Canada geese (*Branta canadensis leucopareia*), storm-petrels (*Oceanodroma* spp.), cormorants (*Phalacrocorax* spp.), glaucous-winged gulls (*Larus glaucescens*), tufted puffins (*Fratercula cirhatta*), and other species of seabirds and waterfowl. Populations of injured species will be monitored to document increases following fox removal.

D. Completion Date

Work will be completed in FY-97.

COMMUNITY INVOLVEMENT

An attempt will be made to hire one or more local residents of Atka Village, the closest settlement to the project site.

FY 96 BUDGET

This project involves two summer field seasons (1996 and 1997). The first field season will be the primary one for fox removal, thus extra people are required. The second field season involves rechecks for foxes and additional bird surveys; fewer people are needed the second season.

Personnel	32.2
Travel	18.4
Contractual	12.0
Commodities	12.6
Equipment	8.0
Subtotal	83.2
Gen. Admin.	5.7
Total	88.9

PROJECT DESIGN

A. Objectives

1. Enhance populations of black oystercatchers, pigeon guillemots, and murre at Seguam Island by eliminating introduced arctic foxes.
2. Survey populations of injured species as a baseline for ultimately determining the magnitude of increases.

B. Methods

Fox Removal

In May 1996 a Fish and Wildlife Service crew consisting of six individuals will establish several camps on Seguam to eradicate introduced foxes with firearms, leg-hold traps, and M-44 cyanide devices. Methods like those employed at other sites in southwestern Alaska will be used (Bailey 1993). Crews will spend about 2 months on Seguam in 1996 and follow up with a thorough recheck during a 6 week-long period in 1997. Remaining foxes will be removed using methods similar to those employed in 1996.

Oystercatcher, Guillemot, and Murre Surveys

Schmidt et al. (1995) demonstrated that oystercatchers and guillemots attain much higher densities on islands without foxes than on those with foxes. We do not propose to repeat that study, but baseline data will be collected on populations of oystercatchers, guillemots, and murre to document future expansion of these species after all foxes are removed.

Oystercatcher Habitat Mapping

Since black oystercatchers nest on both rocky and sand beaches, all beaches on Seguam will be divided into segments separated by natural features (e.g., cliffs, streams, substrate changes). Segments will be delineated on maps.

Oystercatcher Counts

In 1996, black oystercatchers will be counted by a two-person team cruising the perimeter of the island in an inflatable boat within 50 m of shore during June, the incubation period. This is the period when pairs are territorial and most conspicuous. The best time to count oystercatchers is from approximately 2 hours before low tide until an hour after low tide, the period when they are most actively foraging (Andres 1993). For each oystercatcher observation, the location (beach segment code--see above) and status (single, pair, or larger group) will be recorded. Beaches where singles or pairs are noted will be checked on foot to determine whether nests or territorial pairs are present. All areas will be surveyed at least three times to reduce chances of missing territorial birds.

Guillemot Habitat Mapping

Pigeon guillemots nest in rock cavities along boulder beaches, in cliff crevices, in drift log piles, or occasionally in burrows, especially in close proximity to shallow waters for foraging (Ewins 1993, Sanger and Cody 1993, Drent 1965). Due to this diversity of habitat types it is difficult to map all possible nesting areas for guillemots. Furthermore, comparative data for nearby islands in the central Aleutians are based on bird per km of shoreline. As a result, no attempt will be made to delineate the surface areas of habitats that provide typical guillemot nest sites. Forms like those used by Sanger and Cody (1993) will be employed to document colony data.

Guillemot Counts

Reports in the literature provide differing views on the best time to count guillemots. The relative influence of tide stage, time of day, and time of the breeding season on attendance of guillemots at breeding colonies apparently varies among areas (e.g. California, Ainley and Boekelheide 1990; British Columbia, Drent 1965, Vermeer et al. 1993; and Prince William Sound, Sanger and Cody 1993). For comparison among sites and years, it appears the optimum time to census breeding pigeon guillemots is morning hours during the incubation period (about mid-June to mid-July, Day 1977) and within several hours of high tide. Although peak numbers of guillemots occurred at Prince William Sound colonies during the pre-laying period (Sanger and Cody 1993), Vermeer et al. (1993) recommend counts between early incubation to early chick stages when numbers are least variable. Hence, we will inventory guillemots during incubation. This period also offers a much longer period of time in which to make counts when they are most stable. Due to hourly and daily variation in attendance of guillemots at colonies, at least four replicate surveys should be made to estimate guillemot populations.

Counts will be conducted by two individuals slowly circumnavigating islands in an inflatable about 50 m offshore during periods of good visibility and relatively calm seas. All guillemots within approximately 100 m of shore will be recorded. Island coastlines will be subdivided into segments based on natural features, and counts will be recorded within each segment. Concentrations of four or

more birds on the water or land near guillemot nesting habitat will be delineated as accurately as possible on maps.

Murre Counts

The murre nesting colony on the east side of Seguam (see Bailey and Trapp 1986) will be counted on 5 different days in July to estimate the average number of birds present during the 1996 breeding season. All counts will be made between 10:00 and 16:00 h, the time of day when attendance tends to be least variable (Byrd 1987). The colony will be subdivided into easily identifiable segments for data recording. Counts will be made from an inflatable boat near shore on days when wind is less than 20 kt and swells are less than 3 feet.

Data Analysis

Densities of oystercatchers will be expressed in terms of birds and nesting pairs per km of beach habitat. Guillemots will be recorded as birds per km of coastline. Murres will be expressed as average number of birds present in each segment at the breeding colony. Future comparisons will be made with non-parametric analysis of variance techniques (e.g., Friedmann tests) using survey segments as blocks and years as treatments.

C. Contracts and Other Agency Assistance

Plans are to contract with the U.S. Department of Agriculture, Animal and Plant Health Inspection Service (DOA-APHIS).

D. Location

The restoration program will be conducted at Seguam Island, in the east central Aleutian Islands (Fig. 1). This island is approximately 100 km east of the village of Atka. The island is within the Alaska Maritime National Wildlife Refuge.

SCHEDULE

A. Measurable Project Tasks for FY 96

Start-up to May 1:	Refine study plan, arrange logistics, recruit seasonal personnel
May 1 - May 15 :	Train seasonal employees
May 15-June 7:	Deploy traps and M-44s
June 7-30:	Continue to remove foxes, survey oystercatchers and guillemots
July 1-31:	Continue to remove foxes, survey murres
August 15: -	Leave Seguam
August 20-Sept. 30:	Analyze data, maintain and store field gear
April 1997:	Annual report on FY 96 work

B. Project Milestones and Endpoints

May 96	Fox removal begins
June 96	Oystercatcher and guillemot populations surveyed
July 96	Murre populations surveyed
August 96	Most foxes will have been removed
May 97	Fox recheck begins
June 97	Oystercatcher and guillemot populations surveyed
July 97	Murre populations surveyed
August 97	Confirm that all foxes removed

C. Project Reports

April 96	Annual report on FY 96 work
April 98	Final report on FY 96-97 work

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

This project contributes to the overall restoration effort for oystercatchers (projects 93035 and 94020), pigeon guillemots (projects 93034, 94173, and 95163F), and murre (projects 93049 and 94039). Methods used to monitor injured species in these projects will be employed at Seguam.

The DOI-FWS Alaska Maritime NWR has an existing program for removal of introduced foxes from islands for restoration of threatened Aleutian Canada geese, seabirds, and other native species. However, this program for restoration of seabirds on islands south of the Alaska Peninsula has had little funding, and attempting to remove foxes from an island as large as Seguam would be outside the traditional scope of the DOI-FWS program. Nevertheless, DOI-FWS will match trustee funds by providing the salary for the project leader and project manager and by supplying the majority of the field equipment needed to accomplish the work.

ENVIRONMENT COMPLIANCE

The removal of alien foxes by trapping and shooting from refuge islands was sanctioned by an environmental assessment in 1985 (Environmental Assessment--Proposed Eradication of Introduced Fox on Alaskan Islands. Alaska Maritime National Wildlife Refuge, Alaska. U.S. Fish and Wildlife Service, Homer, Alaska). No additional approvals or permits are required.

PERSONNEL

Steven Ebbert: Project leader oversees all aspects of the field work. In addition, he is the project officer for the contract with DOA-APHIS.

G. Vernon Byrd: Project manager supervises planning, training, data analysis, and completion of reports.

The project leader, S. Ebbert, and the project manager, G.V. Byrd, are well qualified to undertake the proposed action. Brief resumes follow:

1. Project Leader - Steven Ebbert

Steve Ebbert received a Bachelor of Science Degree in Forestry from Purdue University in 1984. The emphasis of his undergraduate degree was wildlife management and he was a cooperative education student with Indiana's Division of Wildlife. As a senior, Steve arranged for a one-of-a-kind special problems class sponsored by an adjunct professor with Animal Damage Control, at that time within the U.S. Fish and Wildlife Service.

Mr. Ebbert earned a Master of Science in Wildlife and Fisheries from Texas A&M University. For his thesis project, Mr. Ebbert tested and improved the effectiveness and selectivity of a new coyote bait delivery system for use with predacides, reproductive inhibitors, markers, or biological agents. He assisted with other research projects in Texas involving predator trapping, radio-telemetry and mammal census techniques.

Starting in 1987, Mr. Ebbert worked with U.S.D.A.'s Predator Ecology and Behavior Project, a captive coyote facility and field station of the Denver Wildlife Research Center. He assisted with research directed at the ecology, behavior, and environmental impact of predators, primarily the coyote, and the development of depredation control techniques. While in Utah, Mr. Ebbert completed over 50 quarter hours of graduate courses in specialized courses such as exotic wildlife management, population ecology, landscape ecology, wildlife competition and remote sensing.

Before working for Alaska Maritime National Wildlife Refuge in 1995, Mr. Ebbert was employed with the Service's Division of Realty in Anchorage. With Realty, he used a geographic information system (GIS) to model wildlife habitat value of private land inside refuge boundaries. He compiled wildlife survey data to update resource maps used in the GIS.

Relevant Publications:

Knowlton, F. F. and S. M. Ebbert. 1990. Developing physiologic markers to identify coyotes that kill sheep or goats. Final Report: QA-090, Denver Wildlife Research Center. USDA/APHIS Science & Technology, Denver, CO. 25pp.

Fagre, D. B. and S. M. Ebbert. 1989. Development and testing of the coyote lure operative device. Pages 235-240 in A. C. Crabb and R. E. Marsh, ed., Proc. 13th Vertebrate Pest Confer. Univ. Calif., Davis.

Ebbert, S. M. 1988. Field evaluation and improvement of a new system for delivering substances to coyotes. M.S. Thesis. Texas A&M University, College Station, 145pp.

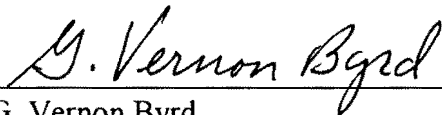
Ebbert, S. M. and D. B. Fagre. 1988. Importance of attractant qualities for improving a new coyote delivery system. Pages 189-194 in D. W. Uresk, G. L. Schenbeck, R. Cefkin, technic. coordins., Proc. 8th Great Plains Wildl. Damage Control Workshop. USDA For. Serv., Rocky Mtn. For. Range Exp. Sta., Rapid City, S.D.

2. Project Manager - G. Vernon Byrd

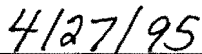
G. Vernon Byrd received a B.S. degree in wildlife management from the University of Georgia in 1968, did post-graduate studies in wildlife biology at the University of Alaska Fairbanks in 1975, and completed a M.S. degree in wildlife resources management (with an emphasis in applied statistics) from the University of Idaho in 1989. Thesis research was on kittiwakes (*Rissa* spp.) and murre (*Uria* spp.) in the Pribilof Islands. Mr. Byrd has worked for the U.S. Fish and Wildlife Service for over 20 years, focusing on studies of marine birds in Alaska and Hawaii. His major interests have centered around monitoring long-term trends in seabird populations, including numbers of birds and reproductive performance at colonies. He has written over 40 scientific papers and 50 U.S. Fish and Wildlife Service reports on field studies, and he has presented over 15 papers on seabirds at scientific meetings. Mr. Byrd currently serves as supervisory wildlife biologist at the Alaska Maritime National Wildlife Refuge, the premier area for seabirds in the national public land system.



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Date prepared

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- Andres, B.A. 1993. Potential impacts of oiled mussel beds on higher organisms: black oystercatchers. Unpublished report, U.S. Fish and Wildlife Service, Anchorage, AK.
- Bailey, E.P. 1978. Breeding seabird distribution and abundance in the Shumagin Islands, Alaska. *Murrelet* 59:82-91.
- Bailey, E.P. and J.L. Trapp. 1986. A reconnaissance of breeding marine birds and mammals in the east-central Aleutian Islands--Kasatochi to the Islands of Four Mountains--Summer 1982, with notes on other species. U.S. Fish and Wildl. Serv. Rep. Homer, AK.
- Bailey, E.P. 1993. Introduction of foxes to Alaskan Islands--history, effects on avifauna, and eradication. U.S. Department of the Interior, Fish and Wildlife Service Resource Publication 193.
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Budget Category:	Authorized FFY 1995	Proposed FFY 1996	PROPOSED FFY 1996 TRUSTEE AGENCIES TOTALS					
			ADEC	ADF&G	ADNR	USFS	DOI	NOAA
Personnel	\$32.0	\$6.8						
Travel	\$12.8	\$0.9						
Contractual	\$0.0	\$0.0						
Commodities	\$7.5	\$0.0						
Equipment	\$2.5	\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$54.8	\$7.7	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
General Administration	\$4.8	\$0.7						
Project Total	\$59.6	\$8.4						
Full-time Equivalents (FTE)	1.0	0.2						
Dollar amounts are shown in thousands of dollars.								
Other Resources								

1996

Project Number: 96101
Project Title: Removal of Introduced Foxes from Islands
Lead Agency: DOI-FWS

FORM 2A
PROJECT
DETAIL

Prepared: 1 of 5

7/11/95
Revised

07-11-1995 04:00PM

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel	\$32.0	\$6.8						
Travel	\$12.8	\$0.9						
Contractual	\$0.0	\$0.0						
Commodities	\$7.5	\$0.0						
Equipment	\$2.5	\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$54.8	\$7.7	Estimated FFY 1997	Estimated FFY 1998	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
General Administration	\$4.8	\$0.7						
Project Total	\$59.6	\$8.4						
Full-time Equivalents (FTE)	1.0	0.2						
Other Resources			Dollar amounts are shown in thousands of dollars.					
<p>Project Description: This project is to prepare the final report for, "Introduced predator removal from islands to restore populations of American black oystercatchers (<i>Haematopus bachmani</i>) and pigeon guillemots (<i>Cepphus columba</i>)."</p>								

7/11/95

Ø7-11-1995 04:01PM FROM

1996

Project Number:
Project Title: Removal of Introduced Foxes from Islands
Agency: DOI-FWS

FORM 3B
Personnel
& Travel
DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET
October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
[No contracts are required for this project.]		
When a non-trustee organization is used, the form 4A is required.		Contractual Total
		\$0.0
Commodities Costs:		Proposed
Description		FFY 1996
[No commodities cost for this project.]		
		Commodities Total
		\$0.0

1996

Project Number:
Project Title: Removal of Introduced Foxes from Islands
Agency: DOI-FWS

FORM 3B
Contractual &
Commodities
DETAIL

07-11-1995 04:01PM FROM TU 02501216110 P.01

1996

Project Number:
Project Title: Removal of Introduced Foxes from Islands
Agency: DOI-FWS

FORM 3B
Equipment
DETAIL